I N D E X

KEYNOTE: "---" denotes inaudible in the transcript.
"*" denotes word was phonetically spelled.

M O R N I N G S E S S I O N
(8:03 a.m.)
Welcoming Remarks
Jesse Goodman, MD, MPH

DR. KUMAR: My name is Sanjai Kumar, and I am a researcher and a regulatory scientist with the Division of Emerging and Transfusion Transmitted Diseases, Center for Biologics at FDA, and it my great pleasure to invite Dr. Jesse Goodman to come up here and give the welcoming remarks and open the workshop. Dr. Goodman.

DR. GOODMAN: Good morning to so many familiar faces. I just want to thank everybody for attending and dealing with this, helping us deal with this important issue. I really want to stand between you and the scientific discussions; and I as usual have to run off, but I am looking forward to hearing more about this. The one comment I would make is I think this discussion here is an example of something we are dealing with in a number of areas, which is kind of the increasing globalization of everything that we do.

With respect to the things that we help oversee in CBER, you know, I sort of see this in two directions. One is that it has often been said we live in a global village with respect to infectious disease threats, and I think we all know that. Even if you look back to HIV, that was really a global emerging infectious disease, although we tend to think of it as US-onset disease, so that increasingly we are looking at things like whether it is avian influenza. Malaria has kind of been a stable or improving problem in a sense. It's been to some degree a success story in blood safety, but obviously what you are all concerned about and we are all concerned about is at cost of deferred donors, et cetera.

But I also don't think we should be too sanguine about it not being a potential transfusion transmitted disease problem or an emerging infectious disease, and I always -- I think people are aware of some of these clusters of transmission of malaria cases in the United States. I saw on one of Hira's slides one of the maps showing the extent of malaria zones at various global malaria zones at various times at history; but I always like to point out that of course CDC was originally, a US malaria control center and much of the southern United States was areas of active malaria transmission. So I think we shouldn't be too -- I mean, with issues like climate change and environmental change, we shouldn't be too secure about malaria.

So there may be advantages to have having improved tools at least in the blood area. I am not talking here about necessarily about implementation strategies, but there may be advantages to having improved tools in terms of our readiness as we try to be prepared.

Then of course there is the issue of travel, and again this has got, again, you got -- the blood bank operators certainly see this in terms of the growth in deferrals, et cetera. My personal story, I think I have affected the NIH blood supply because, you know, I was donating fairly frequently, and then I suddenly couldn't donate because of a trip I had taken. You know, I called up FDA and complained about.

(Laughter.)

But I didn't get anywhere, you know, so don't feel you are alone. But I think there are -- there clearly is on the level that we deal with health and donations, you know, tremendous room for improvement in what we do. Then, as I said, I think there is a -- it is not just a blood bank operation, but there are some public health issues here. Also I am sure many of you in your -- those in blood banking or clinical medicine like I have, you know, all you need to do is see one or two cases of transfusion transmitted malaria and you realize this isn't something trial either in non-immune individuals.

So with that I just say, you know, what I see happening here is obviously we are trying to work with you to assess and bring together a science,

see what some of the possible approaches might be, what some of the policy and practical implications are, and sort of help things move forward in this area. You know, I would be very interested to hear the rumblings, you know, that this might be something that if we can improve or move forward on, you know, could actually be helpful even in the near future to the blood community.

You know, so with all that said, I again wish I could hang out for the day, and I appreciate everybody doing it, and I will look forward to hearing about this. Again, this is an area where I think collaboration and partnership with NIH, with the blood community, et cetera, is really going to be helpful to us and where we hope we can provide rational scientific and policy approaches. So thanks very much. Everybody have a great day.

DR. KUMAR: Now --- Dr. Hira Nakhasi, who is Division Director for Emerging and Transfusion Transmitted Diseases, and he is going to give you an introduction and overview for the workshop.

Introduction to the Workshop

Hira Nakhasi, PhD

DR. NAKHASI: Good morning. Thank you, Sanjai, for the introduction, and also I would like to put my welcome for all of you guys who came from different places, from the country as well as outside the country. I appreciate your coming here because this is a very important issue for us, and we would like to get the input from all of you.

I just wanted to say Jesse, if he is still around, he is not the only guy who got differed. Last two years back human health --- Tommy Thompson went to Afghanistan, you know, as ---, and so he called us and asked whether he can donate the blood, and we had to tell him, sorry, you went to visit Afghanistan so you can't. So anyway, so if it, you know, is some consolation to Jesse.

(Slide.)

Now the purpose of this workshop is --.

(Adjusting equipment.)

I just wanted to give you an overview of what, you know, the -- why we are talking about this --- today's workshop as well as what the discussion is going -- we would like you to focus on. The challenges for the blood and blood products for all of us, including you, is that we need to have the enhanced product safety, purity and potency in order to avoid product shortage, and that is exclusively mutual.

(Slide.)

And how do we achieve that? And FDA's approach to achieving the blood safety is based on five principles. As Jay would call it, five layers of blood safety. The one is the donor screening deferral based on geographical, behavioral, and risk factors, usually by donor education, self-donor, and donor interviews.

Then on top of that and very, very applicable we also have laboratory tests, and based on the outcome of those laboratory tests for particular pathogens, there will be a deferral. Such as currently a deferral for laboratory tests for HIV-1 and -2, HBV, HCV, HTLV-I and -II, West Nile, and syphilis.

In addition to that, we have deferral registries to prevent use of blood from different donors. That is, you know, people if they are differed at one place they can't go other places and donate the blood. Then we have the quarantine controls for the products, which before testing and everything they are not released, and so there is another layer of safety. Then lastly it is the investigations and corrections of deviations.

(Slide.)

Now having said that, you know, the impact on the blood safety because of both the safety and availability. Millions of units, as you know,

are transfused annually, and luckily the risks of transmission of infectious disease has significantly decreased with the introduction of these tests.

(Slide.)

This is just to give you an example here how we have gone from no testing to now with the testing. So for some of these agents for example, for HIV, you know, we are now at 1- to 2-million risk of infection per unit transfused. Similarly for HBV, it is around approximately 1.5/1.6 million, and you know HBV also has one in 200,000. However, because we still have not instituted a highly sensitive HBV --- assay.

(Slide.)

Now to make people understand how we start when a pathogen comes in, you know, in the blood supply and, you know, how do we respond to that. So basically, as many of you know the West Nile story, how it evolved, and we first of all introduced the --- criteria based on epidemiology and determined the risk factors. That is the initial safety measures. Then wherever feasible tests are developed, and then based on the development and testing the people are deferred. So that was the case in the case of West Nile Virus we know. When the epidemic started in 2002, very -- in a broad, big-time epidemic, we first started the donor deferral; and after nine months of that in place the tests were developed, and under IND we have started testing the people and then deferring donors based on the positive test. Then what happens is as the sensitivity and specificity improves on these tests, because it usually takes -- you know, first-generation tests are not as highly sensitive and specific, even though we try to achieve that. However, then based on the risks, we keep both the testing as well as the deferral bases together so that to insure that we have an optimal level of safety.

(Slide.)

The primary goal of this workshop is to seek public input on scientific developments that might support two things. One, if we want to have universal donor screening for malaria; or just reenter donors, which are those people who have got infected or exposed, and then test them for infection, and if they are not then reenter those donors. SO those are the really two major issues here we would like to focus, and I would like to just give you a brief introduction about the malaria background here. I know Sanjai will talk about in detail the biology of this parasite. However, but what I am trying to, you know, give you is set the stage here.

(Slide.)

Transfusion transmission malaria is a public health concern as everybody knows it in the US as well as the world around, and four species of human plasmodium which have been shown to cause transfusion transmission in the US. So you need to keep those factors in mind.

(Slide.)

There is no laboratory test approved, FDA-approved laboratory test to screen donors for malaria infection existing at this time, and currently the blood safety of transfusion transmission malaria is mainly through deferral policies, which you will hear more very in detailed from Alan Williams, a based on the history of malaria infection and travel or residence in malaria endemic countries or regions.

(Slide.)

Now what are guidelines? Currently what we have in place, which is this 1994 memoranda, where we talk about -- we have said that there is a three-year deferral for anybody who has a history of clinical malaria, prior residents of endemic countries, and one-year deferral for a visit to malaria endemic countries by residents who are coming from -- going from non-endemic to endemic. For example, going from here to, you know, Africa or Latin America or Indian for that matter, and identification of malaria endemic areas as provided by the CDC. That is the website.

(Slide.)

So why are we so concerned? The risk of transfusion transmission in the US, each year there are
28 million Americans visiting this endemic areas, and several millions of immigrants from endemic countries to this country, and so they may be harboring some infections sometimes, and there are a significant number of local clinical cases.

(Slide.)

Now what is the impact, what are we doing now, the deferral polices and the impact on the blood supply? Incidents of transfusion transmission malaria in the US is at an all-time low. That is good news. That is down from three cases per year in 1995 to 0.6 cases per year, which is every-other year one case or so at this moment.

However, because of these good deferral policies we also have a significant donor loss because of that because if a person goes and visits these endemic countries and also, you know, also we differ just for the exposure as I told you the guidelines just now. So approximately, you know, from 100- to 150,000 donors per year are deferred on that basis. You know, the estimates vary. Then availability of blood screening tests --- and minimize the unnecessary donor loss, because if we had a test which would allow these people to be tested and a significant number of these people can be re-entered.

(Slide.)

Now one thing, also another point which people have to remember, that what is the biology of this parasite. Briefly, that infectious unit of blood from parasites is very low. That is according to -- you know, this slide was provided to me by Sanjai, and what he tells me is that as one parasite in case of murine, which is P. berghei, can cause the infection, and in the case of the vivax malaria 10 parasites in infected red blood cells can cause the infection. So one has to be --- we have to have an assay to detect that level of infection in the blood. Then on top of that, we have the -- you know, blood products are stored, and parasites can survey under the storage conditions, blood banking storage conditions, up to 20 days. So we have to be careful about that, too.

(Slide.)

So, so what are the topics of discussion I would like to focus you guys on today? And at the end of the day I am sure, you know, at every session we have discussion, and we will welcome from the public, welcome questions from the podium as well as from everybody else. But at the end we will have a round robin, so keep these points in mind. What we want to focus our discussion are what are the main sources of malaria to the US blood supply and how effective are the current safety interventions? You will hear quite a bit. That is the first session today, and you will hear quite a bit about that. Then we also need to keep in mind what are the risks and benefits of donor screening for malaria infection in lieu of risk-based deferrals. Okay? So keep that in mind. Then also we need to focus our attention on what are the pros and cons of universal donor screening compared with testing only for donor re-entry, because I think it is a very important question for us, for everybody here. Knowing what the incidence is, what the --- in this country? You know, we need to discuss the pros and cons of universal donor screening versus testing only for donor re-entry.

(Slide.)

Then with regard to tests. So what are the desirable characteristics of laboratory tests to detect malaria infection in blood donors? We need to discuss that. What kind of -- as I told you, that the infections, you know, what is the dose of infection, and therefore we need to have such tests which can detect that ---.

Then the next step is what are the prospects for the use of malaria antibody tests in the US to screen blood donors or to re-enter deferred blood donors. Because why am I saying that? In Europe and some of the European countries already testing is being done to have our screening people who are exposed in malaria infection using antibody tests. You will hear both from the European perspective as well as the Australian perspective, and also you will hear from some of the makers of this test and what their experiences are.

Then finally we also need mot to keep our mind closed about only the antibody. We would like to see what are the prospects for the use of DNA-based methods as blood screening tests in the US, is there a possibility. Because the reason why I am saying that is because the antibody tests, you know, you have large window period. By the time a person gets infected and develops antibody there is a large window period. The question is is there another --- method where you can look at all of the --- infection and can narrow the window period.

(Slide.)

So with that, I would like to thank you for you attention and I guess I will open the workshop for the discussion. Thank you very much.

DR. KUMAR: Thank you, Hira, for the introduction and overview. If anyone is standing, there are more seats in the front. Before we begin the scientific sessions I have a couple of announcements to make here. First of all, as you can see we are really full, an impressive lineup of speakers here. So to get the most of the workshop we request please, please, all the speakers stay within their time. So, otherwise we will be speaking over somebody else's time, so that is going to create a problem, and I request the session chairs to please keep a close eye on the timing. Also in the interest of time please hold your questions for the end of the session. We have open sessions for discussion in the end. A full transcript of the meeting will be made available, so if you miss something you will be able to catch up with that, and the majority of the presentations will be available online given that we have consent from the speakers. And with that, I would like to hand over the session to Dr. Monica Parise, who is Chief of the Parasitic Disease Program at CDC, and she will open her session.

UNIDENTIFIED: ---.

DR. KUMAR: Yes, and also just one more thing. In case the room gets full here we have an overflow room, Room D, and that is just the adjoining room there. So there will be a live telecast there, but hopefully we will be able to accommodate everyone here.

SESSION I: GLOBAL PROBLEM OF MALARIA AND ITS IMPACT ON THE US BLOOD SUPPLY

Monica Parise, MD, Session Chair

DR. PARISE: Thank you, Sanjai. Okay. We are going to move right into the first session which is the global problem of malaria and its impact on the US blood supply. Actually this is not the first talk in the session. The first talk is Sanjai Kumar's talk on the global problem of malaria. Can we switch the slides? This isn't the first talk.

(Adjusting equipment.)

Global Problem of Malaria, Biology of Malaria Parasites and Implications for Transfusion Transmission Malaria and Detection Methods

by Sanjai Kumar, PhD

DR. KUMAR: Good morning again, and it's me again, so you might see me through the day so you might get used to it. Actually this is not the first talk I want to give.

(Laughter.)

--- the AV person. Well, I just want to give the acknowledgments. Okay. I will do that a little later than. So what I am going to do here in this first talk of this session, I am going to review the global transmission of malaria parasite, the biology of malaria parasite, and acquisition of naturally-

acquired immunity against malaria. We believe this sort of information is key to developing sensible donor deferral policies to identify donors at risk of malaria and also to develop tests that could be used to detect malaria infection in blood donors.

(Slide.)

This is the global transmission of malaria as of today. So with all the numbers here there a couple of things to keep in mind here. Currently there are about 3.2 billion people around the world who are risk of contacting malaria, 300- to 500-million clinical cases of malaria, and 1- to 2-million deaths. I mean, depending on who you are talking to. But there are a few things to keep in mind here. There are four different species of plasmodium. Those are transmitted all over the world, but there are certain things about them to remember. First of all, the vary in their virulence. So about 90 percent of the deaths you see here, they occur from falciparum infection. That is the most lethal malaria, and also the majority of the deaths, about 80 percent of deaths, occur in sub-Saharan African, the south and Saharan desert in Africa, and most of those are children under the age of five. They bear most of the burden of the disease. The other thing about this plasmodium species, their transmission, global transmission, is not even, and you will see that in the coming slides there, and the disease is transmitted almost around the world. There are 100 countries where the disease is transmitted, but still the transmission is not even. You will see that also.

(Slide.)

Okay. These are the representative distributions of the four species, and I guess this is important for us to know where this infection is coming from, where the risk is coming from. If you look in sub-Saharan Africa plasmodium falciparum is the predominant disease. In Asia it is the plasmodium vivax is the predominant disease. In Central America again plasmodium vivax, and South America you get quite a good number of falciparum as well. So if you think of the reasons why this is happening some things can be explained here. For example, in Africa mostly in central and west Africa, and to a large extent in eastern and southern Africa also, Africans there, they lack the --- antigen on the
--- red cell, and that is the receptor used by plasmodium vivax --- red cells. So it makes the vivax is almost absent there, and probably an evolutionary adaptation. Probably vivax --- disease that is --- cause too much problems there, so evolutionary that happened. But still I was rather surprised to see --- vivax is there. But why the falciparum is --- predominant species in most of the Africa and parts of Central and South America, I just do not understand that clearly. Look at plasmodium malariae, and that is one of the causes of major trouble in terms of transfusion transmission malaria, because ---. It is pretty much absent in Asia and big parts of Central and South America. It --- low numbers, but it is still there in Africa. Plasmodium ovale is present almost close to 10 percent in Africa, but also disease there is in the form of co-infections. Most of the time it is not the sole infection. It is present along with falciparum infection, just to keep that in mind.

(Slide.)

So the global reach of malaria, there is no credible transmission of malaria in this country, and Monica will talk about that in detail. But in today's interconnected world this infection comes to us in may ways. The problem of malaria is rising, and there are more cases today than there were 30 years ago, so malaria is going to stay here. So what are the major factors which contribute to the rise of malaria transmission globally? Well, environmental factors, human activities, and I guess the two are interconnected, drug resistance emerging and drug resistance in malaria parasites, and the vector populations. In terms of vector populations, the global population of

the --- species, those high --- capacity for transmitting malaria parasites is globally increasing, and there is good evidence for that.

(Slide.)

So, but all is not gloomy about malaria. So this is the malaria mortality data which I got from this extract from a paper by Carter, Richard Carter and --- Mendis. If you look at the beginning of the 20th century, approximately 10 percent of all deaths globally used to happen because of malaria, but now those figures are significantly lower. So in Europe and North America close to one percent deaths used to happen because of malaria. It's almost --- now. But there is one disappointing place here in sub-Saharan Africa. So with all the advances in modern medicine, that has not helped people in Africa in terms of when it comes to malaria. It's still 10 percent of all deaths today. This is data from 1997, and I don't think figures have changed. They are still because of malaria. So that is mind boggling. The world minus sub-Saharan Africa used to be eight percent before, and the numbers have come down to close 0.1 percent now. So this is one place where the majority of the problem is in terms of mortality. Unfortunately I could not find similar sort of data for malaria transmission 100 years ago and now, but this is in terms of mortality of malaria.

(Slide.)

Okay, so making the case in point. So the majority of the malaria ---. It started in the late mid '50s and it was --- intervention through ---. So making the case in point in Indian here, around 1955 before DDT was introduced there were close to 10 million cases. With the introduction of DDT malaria almost disappeared in India, so it went from 10 million cases to very few cases. I am sure the transmission was still going on, but very ---. With time and the lack of --- and malaria education programs were lifted, and the resurgence of malaria began in the early '70s, and then the transmission peaked around here to six million more cases around 1975. Then interventions were introduced again and they came down again, a now there is a rise, and I did not put the --- here. This --- but the numbers are rising again here. But it makes a case of point here how malaria has made its comeback also and malaria intervention methods work out successfully also. This is simple matters by controlling mosquito populations. What was not happening in sub-Saharan Africa was they were completely left intact. DDT was not introduced during that time. Most of the efforts were placed in India, in --- and around that area and really paid off.

(Slide.)

So this is a part of the global environmental changes, what some might want to call inconvenient --- here.

So if you look, this the prediction in 2020 based on a projected two percent at two degrees Fahrenheit rise in global temperature. So how malaria may be introduced in the parts of the world, in the majority of Europe also where it is not there, and how the intensity of malaria is expected to change, and that is given that there will be no human interventions made, successful interventions made to control malaria. So if this prediction is true so you might brace ourselves as well to prepare for this.

There is another point I would like to make here. There are some --- studies that increase in global temperature will lead to increase in malaria parasite. Because clearly the data shows that a two- to three-degree increase in in temperature in laboratory conditions facilitates the faster development of malaria parasite in mosquitos, and eventually that will lead to more malaria transmission.

(Slide.)

This is the picture, global picture today of the

drug --- drugs. The drug resistance is everywhere, and probably that is the major factor that is contributing to a rise in malaria, and that is why the intervention of --- in Africa have not been very successful.

(Slide.)

So looking at the life cycle of malaria parasites here, this is for those who don't think about malaria issues everyday. So this is upon inoculation. The parasite enter into your cell and the infection commences there. From single human this parasite develops into 10 to 40,000 ---. Those come out in --- cells where the parasite develops, and each parasite develops into somewhere between 16 to 32 blood
--- and the infection continues here, and then it gets transmitted again into ---.

So there are several things here about malaria biology --- that are part of building on transfusion transmission malaria. First of all, the --- cycle is somewhere between six to 14 days, depending on the species, but there are several exceptions, and I will talk about that in a second. The blood stage continues for 48 to 72 hours, depending on the species. So here is this incubation period that is around 21 days. Then the primary infections, a person will experience different sort of clinical diseases, varying from the most serious form of malaria to this fever; and then there is the story of these adults from endemic areas who develop partial immunity, but they carry low-grade parasite ---, and those are the most important group of people for us to watch out for.

So looking at the malaria biology and immunity again, so the definition of incubation period, the time that it takes between the infection to the first appearance of blood form parasite. That varies between the species. In the cases, I just said, it can be somewhere between 14 days to 21 days. But in the case of plasmodium vivax and ovale they have the --- forms in the liver, and this all could take sometimes a month to a year, and some --- strains of plasmodium vivax are known to just do that, so this period could be up to nine months very easily.

So when you are talking about this primarily, malaria ---, we have to keep this in mind. The chronicity of infection in the case of plasmodium malariae, the parasites have been known to have been demonstratively present up to 40 years. In the asymptomatic carries in multiple exposures in individuals who are either born in endemic countries or expatriates who live for a long period of time develop partial immunity. They have no clinical disease, but the carry. The --- develop ---- immunity as well and they carry low-grade parasite burden. So something I will keep talking again and again in talk, the parasite burden in asymptomatic carries is not known how many parasites they carry. That presents a big problem in terms of their identification and exclusion. Here I mentioned the infectious does of blood from malaria parasites is very low.

(Slide.)

So talking about travelers, so they have no prior immunity. Infection can be acquired shortly before departure from an endemic area, and the come and become infectious while in this country or in a non-endemic country. Infection with a strain of plasmodium with prolonged latency, I talked about that. Residents born in an endemic area or had a prolonged residence, they become asymptomatic carriers, a good number of those, and parasite burden is not known. Then history of clinical malaria, those could be still infectious because of inadequate treatment, and then there is still the possibility of relapse from liver form parasites in the case of vivax and ovale malarias.

(Slide.)

What are the laboratory methods that have been used over time? Direct demonstration, microscopy or DNA detection is we have to worry about the window period in this and also the parasite burden in asymptomatic carriers. The other method is look for a surrogate exposure such as looking for the

antibodies, but then you have to worry about the time lapse between the parasitic exposure and first appearance of parasites, and there are few early reports on --- presence and data that show that it doesn't take very long after the first appearance of blood form parasites to the seroconversion. Then the a screening test to whatever we might want to implement should be able to detect at least the predominant plasmodium species that are known to cause transfusion transmission malaria.

(Slide.)

Detection methods I will just pass through quickly. Microscopy, thick film, quantitative buffy coat method, then antigen detection, methods of --- look for circulating parasite antigens, nucleic acid --- detections, and then antibody methods.

(Slide.)

This is my favorite slide I like to show. This is Sir Ronald Ross. He published this paper in 1903, an improved method for the microscopical diagnosis of intermittent fever; and 103 years later this is the method that is used around the world to detect malaria parasite, and there has not been much improvement since then. The staining may be a little fancy. The filter may be a little nicer.

(Slide.)

So this is a thick smear. You can see the parasites here. This is the gametocytes here. There are the rings here, and the sensibility depending on the hands holding the --- could be five parasites per microliter to 500 parasites per microliter if it is done by --- experienced technician.

(Slide.)

This is the quantitative buffy coat method here. So it is simply it is a hematocritic tube where you apply the infected blood. Then it is centrifuged in a hematocritic centrifuge at a high speed, and based on the bugs here, the different cells and the densities, the infected red cells, the form a ring just next to the white cells and on top of the uninfected red cells under a microscope. In a florescent filter you can see the parasites there, --- gametocytes and different --- here. So this is modification of the --- microscopy, but from what I see here it is clear to be as sensitive ad microscopy, but it is not very much preferred by people out in the field

(Slide.)

There are antigen detection methods. So there antigens there --- malaria and there antigens from the --- parts where there are --- between different plasmodium species. So, but there is a word of caution. So there are all kinds of methods there, and there are commercial companies who make and cell these tests. But as a word of caution here, these antigens could be circulating around four weeks after completion of chemotherapy. So presence of these antigens does not always equate to the current infection. The sensitivities vary depending on who is testing them, and they are sold by many commercial companies. Just to avoid the appearance of any endorsement, I am not showing any commercial. But none of those are FDA approved, but they are available in the market in different countries nonetheless.

(Slide.)

These are DNA methods here. This is our own data here using a small subunit of ribosomal RNA. We can detect up to 0.2 parasites per microliter. That equates to 250 parasites per ml of blood, and this method, I will talk about this in much more detail in my second talk. But this is at least 20-fold superior to thick film method. You will see all kinds of these kinds of publications in the literature. There is another word of caution I would like to have, but which I will talk about in my next talk.

(Slide.)

Then there is antibody-based detection methods. So is the florescent IFA method here. It s a method of choice. It is sort of a gold standard that is used by different laboratories, and some countries, including France, until very recently have used to screen for at-risk malaria donors by IFA. But it has sort of becoming outdated now because the sensitivity in travelers with a single one-time infection is questionable. The method is cumbersome, and it is probably difficult to be adopted in an automated format. But nonetheless, the method has been used very effectively and the CDC still uses this for malaria diagnosis.

(Slide.)

This is the data I would like to show here from my own laboratory here. This is an ELISA, and you will hear all kind of ELISA later in the workshop, but I would like to show this. So one of the concerns we have is the pan species recognition. The test should be able to detect at least the major species of plasmodium, and this is the data we ---. This is falciparum --- protein two --- proteins here. These are recombinant proteins. These were provided to us by ---, and this protein was produced for clinical use. There is a highly --- protein here, so that is why we get very little background ---. This is a test done at --- dilution.

But I would like to show here the recombinant CS protein recognized down to only --- falciparum, but also recognizes all samples in vivax with one exception for OLER samples from plasmodium malariae. The same was true for AMA1's. The majority --- recognition across these species and then --- to us from Marianna Wilson from CDC.

So we were rather kind of surprised because if you look --- sequence homology at protein level we really --- similarity at least for CSP and the --- protein here. When you mix the three proteins in the same --- in ELISA everything gets recognized in a very nice way.

(Slide.)

Probably some people here in the audience would be very pleased to see this data, and again in Western here this same sort of thing appears here. It recognizes -- I am not sure how I am doing on time. This thing is jumping. Okay. Okay, I will done in five seconds.

So recombinant CS recognizes all PF samples, and also these recombinant proteins recognize vivax sera, malariae sera, and ovale sera. So that is sort of good news that if you have highly --- recombinant proteins from one or two species and in combination are --- even if we have done --- characterization. But again the word of caution here is probably it would be much better for these assays to work with the endemic sera where people have multiple exposures. But we have to be a lot more careful when we are dealing with primary infection. The person has seen only one time a parasite, so sensitivity may not be seen.

(Slide.)

My collaborators here. They are the people from my lab. Hira, who has been always supportive, Dr. David Haynes always provides good parasites, Monica Parise and Marianna Wilson, we can't do without them, and then NIAID Malaria Vaccine Development Branch for giving us these wonderful recombinant proteins; and I will stop here.

Malaria in the United States
by Monica Parise, MD

DR. PARISE: Thank you, Sanjai. We are going to move right into the next talk on malaria in the United States.

(Slide.)

Okay. So today I am going to talk about traveler's malaria, locally-transmitted malaria and the special case of locally-transmitted malaria, that caused by transfusion-transmitted malaria. Thank you to Sanjai and the organizers for asking me to give this talk.

(Slide.)

The points that were already mentioned I am going to go over sort of rapidly. 1,000 to 1,500 cases reported in the US every year, 99 percent of them are imported infections in travelers and immigrants, and we have up to about 10 cases a year of locally-acquired infection.

(Slide.)

So locally-acquired infection can be induced by transfer of blood as occurs in blood transfusion or by the other methods listed on the slide. It can be congenital transmission, and, as was mentioned, we occasionally have outbreaks, usually very small, of local mosquito-borne transmission in the United States.

(Slide.)

This map shows where local mosquito-borne transmissions occurred over the last 50 years or so. It is not surprising. The vector is really almost everywhere in the United States except in the Rocky Mountains and up in the very northern part of New England. So these outbreaks are scattered around and they do tend to be particularly concentrated in areas where there are a lot of travelers and immigrants as one factor.

(Slide.)

This is data from the National Malaria Surveillance Summary that we put out at CDC every year. This is from '73 up to 2004, numbers of reported malaria cases. The total number is in red, the numbers in foreigners in green, and the number of travelers in magenta. Really the main points here is there was a large increase in the '70s due to immigration from southeast Asia, and then the cases have really been trending up over all, but there has been a recent decrease in the last couple of years. That decrease has been proportionally more in foreign residents than in US residence.

(Slide.)

SO we looked at that and asked, well, why might that be, and here is a graph that shows immigration to the US over the last two centuries or so. One thing you can see is in very recent years there was a very step drop-off in immigration. That is really since 2001. That is probably related to restrictions after 911. So that may be one factor that is causing this recent decrease in cases in foreign residents that we see in our overall surveillance.

(Slide.)

These are rates. So now I am going to move onto travelers. These are rates that we calculated using data from -- on denominators on numbers of travelers and malaria cases through our US surveillance system. There is one point, just an overall point I want to make about any of the rate data I am going to show today, is that you have to remember that the data in the travelers and then in the transfusion cases, those are really rates in the face of a program, probably a very effective program. For travelers, a lot of people take chemo prophylaxis. We have a program to limit risk in the blood supply. So these rates could very well be much higher, you know, in the absence of those programs.

So here you can see really there is an order of magnitude with the lower risk among travelers to Mexico. Highest risk is in the green in Africa, and then sort of intermediate is what we see in the rest of the Americas and in Asia. Sorry. Well, there is the intermediate.

(Slide.)

This is really the same. This is a snapshot of just '99 data. The main point I wanted to make here is that this is the risk we see in immigrants, refugees, asylees, versus travelers, and the rates are significantly higher in the immigrants versus the travelers, as one would expect, because they have less exposure probably.

(Slide.)

This is really more of the same. Highest risk in Africa. These are all travelers, immigrants, and refugees, and, you know, very low in Mexico, which you see in the light blue.

(Slide.)

This brings just one country case up. This is Mexico. Much higher risk in your immigrants as opposed to your travelers.

(Slide.)

Now, this is a meeting on basically donor screening by lab tests, but these are questions we always get, so I am just going to sort of address them up front. We are always asked. "What about the resorts? They have such low risks. Can't you just leave us alone on these resorts in the Caribbean in Mexico?"

Well, I guess a couple of points. You know, the risk is in resorts, but really largely this is resorts in rural areas. It is not all resorts. An except there is La Altagracia Province in the Dominican Republic. We have heard a lot of arguments. "Well, is a rural versus an urban resort? You guys are calling these rural resorts things that are big and urbanized."

We used to give an anecdote. I went on a consultation in Mexico about a year-and-a-half ago to a large resort. I won't name it, but we went. You know, this would have been an argument that there is no malaria here, but we saw cases in the town. So I think all of us really argues for the need for the best evidence-based recommendations we can do. You know, certainly as long as we are using the questions and also on the lab tests.

(Slide.)

So as far as, you know, just to give I think or opinion at this point on these resorts, you know, these are areas that we consider risks high enough to recommend prophylaxis for travelers. Now at some point we may change that recommendation and think about standby treatment for very low-risk situations, but we are not there yet. So we are putting travelers on preventive measures. It would seem risky not to defer blood donors to those areas.

This also, when we talked about these very low-risk situations, I think it raises a couple of interesting questions that we have really not dealt with adequately completely yet. So, you know, these questions are would you base recommendations on the endemic situation, which is what we do now, which is the conservative approach, or use traveler or transfusion data, for example. Well, if you chose to use sort of the latter, traveler data, given the micro-epidemiology of malaria, you need to consider that the assumptions based on an average traveler might not hold for all travelers across the board; and then we would also have to grapple with, well, what would be an acceptable level of risk, you know, both for travelers or for transfusion, whatever group you are talking about.

(Slide.)

Okay. So back to US data. This is malaria cases reported in the US by species. Highest number from falciparum, followed by vivax and then small percentages of the other species.

(Slide.)

This is trend data, and we really see a rise in the trend of more falciparum and less vivax over the last, you know, decade or so. Why might that be? Well -- oh, sorry. Anyway, just to hypothesize why might that be, one thing is probably, you know, the progressive loss of sensitivity of chloroquine. It is just not working anymore, and people are still using it. Two, travel to areas where, you know, countries are seeing more vivax, more falciparum. An example is there is India. Travel to India has increased. The ratio of falciparum to vivax in India has increased, so that would contribute as some possibilities.

(Slide.)

Where are people acquiring malaria? About 60 to 70 percent of our cases in the United States are acquired in Africa followed by Asia, and then Central America and the Caribbean.

(Slide.)

Travel to Africa accounts for only about 0.6 percent of US travel, 2003 data. Yet about two-thirds of all malaria infections and 86 percent of deaths of all falciparum were acquired in Africa, and in the last 15 years or so not surprisingly 93 percent of US deaths were due to falciparum; 73 percent of those were acquired in Africa.

(Slide.)

There is a lot of travelers to the Americas, including the Caribbean. So this data, this just shows our surveillance data species-wise, and pretty much what we see is, you know, we are seeing all these other species, falciparum, vivax and malariae, really from most of this area. This is probably an error. I think that is probably a mistaken diagnosis basically based on if you look at PAHOO data they also certainly report malariae in the Americas, no mention of ovale, and a big review article put out by Bill Collins last year was that in extensive literature review there was no mention of ovale in the Americas.

(Slide.)

The special case of VFR is these immigrants who return to their homeland to visit friends and relatives. This accounts for a lot of travel. This was the purpose of travel in 44 percent of the 26 million trips made in 2002, and the geosentinel network of travel medicine reported that these travelers have about eight times higher risk of acquiring malaria than tourists.

(Slide.)

Okay. I am going to move on to transfusion-transmitted malaria now. From 1963 to 2005 we had 96 cases reported, so these are the cases. About two-thirds were over 40 years old, about half male. A fatal outcome in 10 percent. That is much higher in comparison to what we see as sort of the overall rate of fatality rate due to malaria infections in the US, which is about one percent. Where we were able to implicate a blood product, whole blood, packed cells, platelets, this is skewed because remember this is data way back to the '60s. Certainly this would be shift more toward packed red cells now. Range of one to 192 donors per case with a mean of seven, and from -- these cases are not picked up right away. I mean, here you go. Time from symptom onset to diagnosis up to 180 days with a median of about nine. People just don't think of this because there is no travel history.

(Slide.)

Where we were able to implicate an infective donor this is sort of the profile: about 75 percent almost between 20 and 40 years old, 90 percent male, 60 percent foreign born. Area of acquisition, this is again the overall series, about half from Africa. I will show you recent data sort of at the end of the talk. This has shifted to more towards Africa now.

(Slide.)

We were able to do a complete investigation, this is some records, in about 62 cases. In about 60 percent there was a failure in the donor screening process, so that is why the case happened. In about 40 percent even, you know, the process as it is supposed to work worked fine, but cases slipped through. This is largely due to the biology of malaria, P. malariae. Two-thirds of those were P. malariae that could come up decades later as the issue there.

(Slide.)

This just shows sort of what I just told you in a flow diagram, and the point I wanted to make here is, you know, where the screening process worked. Again, two-thirds malariae, about one-third other species. If you calculate what the risk is here for where the screening process worked as it was

supposed to, the risk is about 0.09 cases per million units transfused, and I will show you data overall on incidence in a minute.

(Slide.)

Sort of a special case, as I mentioned, are these VFR travelers, these prior residents of endemic areas. We were asked a couple of years ago to look at, well, should they be deferred longer. I mean, these people that now live in the United States, but used to live -- so they are US residents now, but they used to live overseas. Could these people, you know, be semi-immune and be carrying the parasites for longer? Should they be deferred for longer than on one-year US resident deferral?

So we find eight cases in this database, and five of them came up within a year anyway. Two of them came up over three years. They were P. malariae. Even if you changed it to three years you wouldn't get them. Then there was one, it had been one case that if the deferral guidelines had been changed you would have picked it up.

(Slide.)

Species involved in transfusion-transmitted malaria, overall most -- you know, more falciparum followed than by vivax and malariae. Not surprisingly where the screening process didn't work you see more falciparum. Where it did work, because malariae doesn't follow the three-year rule, that is the most predominant species.

(Slide.)

Here is species involved by region, and basically here what you see is from these three species we are really seeing from all -- you know, all of these three major regions, whereas in the transfusion cases we have seen the P. ovale come from Africa.

(Slide.)

There has been quite a change in sort of implicated donor profile, infective donor profile over the years, with over the last 20 years by far most of this is from immigrants.

(Slide.)

This slide shows the incidence of transfusion-transmitted malaria again over the last 40 years or so. The denominators come from data either published in the literature that I got from Lou Katz from -- actually from a donor. You guys know the data from the donors better than I do. I'm a malaria person, but -- but anyway, but what it shows is basically that the rate has come down and it has remained at a very stable low rate in recent years.

(Slide.)

So the last 15 years, he is what is happening most recently. We have had 16 cases from 1990. We have been able to implicate a donor in 14. I have to say the 2005 case where there were about 150 donors we have still not been able to complete the investigation. It is like pulling teeth to get all those donors back in, so we don't have an answer on that one yet. Twelve cases in immigrants, two of those were really people who were born here but have lived long periods oversees, so we group them there. One US traveler to Kenya, one VFR to Africa, 86 percent of these in the last 15 years acquired in Africa, two in China due to malariae, 71 percent P. falciparum, and a failure of the screening process in about 70 percent.

(Slide.)

So in summary, we are seeing a low incidence of transfusion-transmitted malaria using the current guidelines. We calculated a rate of about 0.23 cases per million units transfused; and, as I mentioned, where the screening process works it is about 0.09. Highest risk, as I mentioned, in immigrants and exposure in Africa.

(Slide.)

Acknowledgments to people who helped me technically or gave me data, and thank you. We will take questions after the session.

So we are going to move on to the next talk, which is going to be current deferral polices to reduce the risk of transfusion-transmitted malaria and their impact on donor availability by Dr. Alan Williams.

UNIDENTIFIED: Actually --

DR. PARISE: Oh, you switched? Oh, okay. Oh, I am sorry. I am very sorry. I --- up here. So we are going to have malaria in the United State military and its implications for safety of the blood supply by Dr. Chris Ockenhouse.

(Adjusting equipment.)

Malaria in the United States Military and Its Implications for Safety of the Blood Supply

by Christian F. Ockenhouse, MD, PhD

DR. OCKENHOUSE: Good morning. It is a pleasure to be here. I think I might be one of the lone voices out here. I don't have an agenda this morning. I don't represent a company that is trying to market an antibody or a nucleic acid based therapy, but I would like to maybe talk to you about what the situation is of malaria in the United States military, and specifically for blood transfusion-transmitted malaria.

(Slide.)

Now as all of you know, malaria infection in the US military has a long history. We spend a lot of money, millions of dollars each year, to develop new drugs and vaccines against malaria. Malaria, because of the deployment of our troops worldwide, is a constant concern. We take a lot of effort to try to prevent infection in our troops with proper prophylaxis, and it doesn't always work. We have about 35 to 100 cases of reported malaria every year. It is probably a little more. A lot of cases, we have malaria that don't get reported to the proper channels. Approximately about 60 percent of the cases are P. vivax and about 40 percent of the cases are P. falciparum.

Now historically, as many of you know, malaria has been in the US military, many militaries, because of the deployments around the world in Viet Nam, World War II, Somalia. In plasmodium vivax 300 cases occurred after troops returned to upstate New York, 300 cases after a couple of months in Somalia, and they all had P. vivax, and that is peculiar to east Africa. Just a couple of years ago a contingent of Marines went for only 100 hours into Liberia, Monrovia, to quell a disturbance. 126 went there for 100 hours, and 80 of them came down with malaria. Now that is a lack of command control on proper prophylaxis, and that commander should have been relieved of duty. He wasn't.

(Laughter.)

But that takes a lot of effort and a lot of -- because a couple of those Marines were deathly sick. We also have malaria in Iraq and Afghanistan. There isn't any actually in Iraq. It actually occurs from soldiers who pick it up when they are elsewhere, and most of the cases come from if they have been deployed to, for instance, Republic or Korea. Several months later they show up in Iraq and they come up with a --- illness. It turns out to be P. vivax and it can be genotyped as coming from Korea.

If Afghanistan there is transmission of malaria, and in the last five years we have had cases of severe and complicated malaria, a couple of deaths. But, you know, all these cases it has been found they have been non-compliant.

(Slide.)

But what does this tells us about the safety of the blood supply? How safe is the blood supply against malaria infection for US military personnel? I think it is extremely safe. I believe that the deferral policies, the policies that we have now, work. US military follows the recommendations on the donor deferral policy. The only exceptions are for fresh whole blood in emergencies in mass casualty. In Iraq when we have mass casualties and we don't

have enough fresh blood, we will line up soldiers and ask them to donate blood. It happens, but we will test them for obviously HIV, hepatitis B and C.

And when I was asked to talk about this I didn't know very much about transfusion-transmitted malaria in the US military, ad we have a lot of -- we have a large blood program, Army blood program, ad we track all cases of infectious disease in transfusion over the last 10 years. Over the last 10 years we have found three cases of malaria that was of infection that was coincident with transfusion, and they are listed there, case one, two, and three. All the cases occurred -- two of the cases occurred after transfusion, 149 days and 720 days. The other one was coincident with blood transfusion from falciparum. But all the cases had a history consistent with malaria exposure. I don't believe that any of these cases were from transfusion-transmitted malaria. So our donor policy, our deferral policy works. We use the same deferral policy as recommended by the FDA and the American Association for Blood Banks.

(Slide.)

So this is deferrals by type. The Army Blood Banking Program gave it to me. In orange about six percent are deferrals. This is one year, from March '05 to '06, just in this past year. About six percent of the deferrals are due to malarial exposure of having traveled to malarias. As you can imagine, we have a lot of soldiers that have been deferred. However, if you look in the purple, actually a lot of these deferrals are low hemoglobin and hematocrit. The fact is if we are really worried about retaining the deferral pool, maybe we ought to look at individuals who have anemia; and there are other reasons for deferral.

(Slide.)

So let me go on. Is there evidence that the blood supply poises an unacceptable risk for malaria using the current policies of donor deferral? I would say that it all depends on what your level of risk is. This is where if you are risk adverse and you believe that there is no acceptable risk for a single case of malaria, then you think that all blood should be screened. However, if you feel that our current deferral policies work, then what is the problem? Is the problem not the safety of the blood, but is there something else that we should be concerned about? I think that is where some of the discussion today may lead.

Are we at risk of permanently losing potential blood donors; and, if so, how can you minimize that risk? How can you get some of these people who are deferred for one year or three years back into the pool? You could actually screen for malaria if you had a proper test, either a passive or active, but perhaps what you could do is you could change the policy, the current policy for deferral to retain potential donors that pose a low risk.

(Slide.)

Now the active -- I am not going to get into this. There are several talks later about the active detection. You know, there is obviously many groups are working on nucleic acid testing and testing for enzymatic reaction of parasite products, antigen detecting, or passive detection of malaria specific antibodies. Much of this in the US military's effort is for diagnosis, not for the screening of the blood per se.

(Slide.)

But, you know, we have looked at passive detection. What we do at Walter Reed in the context of a lot of drug and vaccine trials, we actually tend to use --- challenge of malaria in volunteers who hare non-immune volunteers, and we infect them with malaria as a part of vaccine and drug studies and control individuals as well. And we follow them daily, and as soon as we see a single malaria parasite, even if they are asymptomatic, we treat them. So these are individuals who are very, very low parasite densities, about approximately one parasite per microliter, much lower than what you would observe in an

endemic area. We then continue to follow these individuals up, and we collect plasma and do thick blood smears to make sure they are negative.

But, you know, a couple of years ago we said, you know, if we wanted to measure the exposure of malaria in an individual who had already come down with malaria, who was potentially clinically ill, an AA non-immune clinically ill, could you detect antibodies and how quickly could you detect those antibodies. The answer is using a variety of assays, and there are many assays. We use a couple of recombinant proteins. We can detect the exposure of malaria in 96 percent of the individuals, 75 subjects, within certainly 30 days.

But does this help you in a non-immune population? You know, who is at most risk for transmitting malaria in the United States? My emphasis is in the United States.

(Slide.)

Now, non-immune travelers I believe are not high risk for transmitting malaria. It is highly unlikely that a non-immune asymptomatic blood donor carries malaria parasites in their blood. The only caveat is if that parasite threshold for density is less than the clinical threshold, and they would have to be in that window period. I think that is very highly unlikely.

So if you look at the current donor policies, who would be at risk in this country of transmitting malaria in their pool? Would it be a traveler, a non-immune traveler? Which think would be greater than 99 percent of the individuals who travel anywhere in the world. Are they at high risk for transmitting malaria in their blood? I say that they are really not.

Now residents from endemic countries that either live here or go back home may be asymptomatically infected with malaria, could be. It is usually mostly in children, and asymptomatic adults and adults who carry malaria parasites in their blood for prolonged periods of time is actually quite rare. Now if you really wanted to do something, you could actually continue the deferral policy for those individuals. Or you could alter the deferral policy. What is so special about the one-year deferral for travel or the three-year deferral for a clinical illness? Or you could do the easy thing, other people think would be the easy thing, screen all the blood if you had a good test for malaria. But there is a cost and there is a benefit. The costs are not only costs of losing permanent individuals from the pool of potential donors, but there are financial costs as well. In an era of increase in medical costs we need to be cognizant of trying to control medical costs, and perhaps we should be looking at deferral policies first before we undergo a prolonged screening of blood for all donors.

I think that is the last of my comments. I look forward to a discussion later. Thank you.

DR. PARISE: Thank you very much. Okay. Now we are going to move on to the last talk of this session, and then we will have a panel discussion. Now Alan Williams is going talk about current deferral policies.

Current Deferral Policies to Reduce the Risk of Transfusion-Transmitted Malaria and Their Impact on Donor Availability

by Alan Williams, PhD

DR. WILLIAMS: Sorry. I appear not to have gotten loaded here. This should just take a second. But in the meantime I am going to get started. I think in the previous talks you heard some interesting presentations about some of the scientific and epidemiologic parameters surrounding donor deferral policies, and all of these are obviously quite relevant and ---.

(Technical problems with AV equipment. Speaker still presenting, but not being recorded.)

There have been I think a total of four blood product advisory committee discussions related to malaria. I am going to present a couple of the

key discussions that were voted on. In June 19th of 1999 the committee considered an exemption for daylight travel, particularly travel to Mexico on cruise ships and visiting resorts. Some of the discussion surrounded the fact that trying to distinguish between rural versus urban exposure and dusk to dawn versus daytime exposure would greatly increase the subjectivity of the donor screening questionnaire in context of which trying to screen for a travel history is complex as it is. Also certain of the vectors have certain times of the day that they feel one simply cannot totally rule out mosquito exposure during the fringes of daylight hours, again increasing some of the subjectivity for a donor to try to distinguish whether they were there only in the twilight or evening hours versus during the day. So the committee at that time in response to the question did the committee members support a change in the current blood donor policy to allow for travel when travel exposure was limited to hours of bright daylight did not vote in favor of that recommendation by a vote of nine to five.

(Slide.)

In June of 2000, the agency issued draft guidance entitled "Recommendations for Donor Questioning Regarding Possible Exposure to Malaria." That guidance, draft guidance, it was proposed that the 1994 recommendations be modified. One, to provide a definition of residence defined as five years in an endemic country, again not specified as cumulative or continuous. The draft guidance did provide specific donor travel questions. This included a capture question format to be followed and followup of a travel history. It did not distinguish time of day and rural/urban exposures. In other words, even on a cruise ship a passenger who made a daytime trip to an endemic area and then back to the ship by the evening was subject to deferral. And it provided a provision related to this concept of partial immunity from individuals who were residents in malaria endemic countries who then returned to that country might in fact be asymptomatically infected by the virtue of partial immunity. It provided that immigrants, refugees, citizens or residents of malaria endemic countries would be deferred for three years after the last visit to the endemic country.

(Slide.)

There is an additional discussion in March of 2001 around the plasma risk issue. The committee considered the question of are there available data sufficient to conclude that it is safe to prepare frozen plasma products for transfusion despite a history of malaria risk in the donor.

The committee did not support that on a routine basis plasma prepared by separation from whole blood should be exempt from malaria donor criteria. With some differences when that plasma is collected by automatic apheresis our specifically by an Autopheresis C procedure, which was a target of discussion based on a variance that was projected to be requested of the agency by an individual blood collection establishment.

The committee also voted whether the agency should continue to allow use of FFP when personal information from a donor indicates a history of malaria risk, and the committee voted in favor of that policy continuing.

(Slide.)

In terms of current considerations, there have been through the years a lot of discussions, a lot of interaction certainly with our colleagues at Centers for Disease Control and Prevention and with our blood collection colleagues, and recommendations that are currently under consideration and hopefully will publish soon as again draft guidance I am following because this will emerge as draft guidance. There will be plenty of opportunity for discussion and comment, but these are the current considerations.

That there is a distinction to be made between countries considered endemic for malaria versus areas within a country endemic for malaria, and this distinction comes --- considering residence versus travel. Travel to an area that is not endemic for malaria within a country that itself has endemic areas

would not be a cause for deferral, whereas travel to an endemic area in a malaria endemic country would be cause for deferral. On the residency basis, residency defined on a country basis due to microchanges in the epidemiology of endemic malaria so that residence is defined on a country basis rather than an area basis.

The recommendations for specific donor travel questions is proposed to be removed from FDA recommendations, and instead because of the difficulty of collecting travel histories and the great deal of work that has gone into streamlining and improving the validity of the donor questionnaire process as is being done in several other donor eligibility areas, the agency is prepared to accept the organizational taskforce questionnaire which in fact uses a tiered approach to collecting travel history starting with capture questions and then working down to individual travel exposures. So instead of dictating questions, the FDA is prepared to accept that questionnaire and its subsequent modifications. --- time of day and rural/urban exposures are not distinguished.

One change is that immigrants, refugees, citizens or residents from malaria endemic countries are deferred for one year after last visit to the endemic country. You notice this is a change from the previous draft. This is acknowledging the point made by Monica that in fact most of the cases that returned to countries in fact would not have been -- the post-transfusions cases would not have been eliminated by a three-year deferral. So due to the difficulties of trying to distinguish this information as reported by a donor, this is simplified to match the residence of non-endemic countries as proposed.

Any future guidance will extensively reference the CDC travel website. The URL is provided here, and my understanding is that the CDC is working hard on a long-awaited geospacial map that could be used to actually pinpoint on a map hopefully electronically where a donor has traveled and then have a drop-down in terms of not only current, but hopefully historically exposure to malaria, which would create a pictorial screening environment that hopefully would help the confusion aspect of this screening procedure quite a bit.

(Slide.)

As mentioned, the revised recommendations would again be published as draft guidance. The recommendations would apply to cellular blood components for transfusion, red cells prepared for immunization of source plasma donors or manufactured injectable products, as well as plasma process from whole blood collected by apheresis and intended for transfusion or for the preparation of cryoprecipitate, but would not apply to source plasma. This is a specific exemption in the regulations.

(Slide.)

I am not going to address in great depth the concepts of donor loss related to deferrals. I know there are several speakers today that have data from their own experiences. Just borrowing from one figure that was put forward by Marcus Blood Centers, at least in their system there is a --- of about 1.2 percent overall loss of donors for potential malaria exposure. I think one thing to keep in mind in looking at donor loss issues really for any donor screening event is deferral onsite for which centers certainly have very good information, but doesn't necessarily address the larger group of donors who know they aren't eligible and simply don't come to the blood center, potentially a much larger population. But the range of the estimates provided from ABC, 0.2 to 3.1 percent, and as you saw even higher and double that in areas of the military.

This would translate to in the range of 120- to 150,000 potential donors per year known to be deferred. I believe that is onsite deferrals, representing up to 180,000 donations. There have been certainly a lot of observations as well as some publications that donors who once deferred are difficult to get back to the blood center. They don't fully understand that

they are not eligible or they consider this as being, you know, an unfortunate and unpleasant experience in interacting with the blood center and they are simply tough to get back. So that combined with the fact that many of the deferred donors for some of these travel deferrals are repeat donor populations, some of them very valuable apheresis donors, is very costly from a donor loss standpoint.

Deferrals currently certainly reflect increased travel to malaria endemic countries that historically have not --- previously frequented, specifically areas of southeast Asia that are now becoming in vogue for travel. So that you are seeing some changes in the epidemiology of the deferrals as well.

There is a recognition now that deferral of repeat donors not only has the loss of the immediate loss of that donor, but a cumulative effect down the line that actually makes it more difficult years hence to maintain an intake donor population due to loss of well-established repeat blood donors. There was a publication in 2004 from Transfusion that travel deferrals disproportionately impact male donors in the 25- to 39-year-old age range, probably in large part due to business travel, and then that flattens out for older donors. So really impacting in large part a major portion of the current donor base.

(Slide.)

Some of the screening challenges that you have heard alluded to before. Travel histories are just difficult to obtain precisely, and I think in many cases to try to make the criteria as simple as possible helps improve the validity of a screening as well as prevent some of the false-negative and false-positive deferrals that plague some of these questions.

The definition of residency has been somewhat vague in current considerations in the agency. We are trying to simplify that to a one-year continuous exposure which should be an easily understood definition. Clearly donor screening doesn't capture all the exposure possibilities. Even considering that a proportion of post-transfusion exposures are due to false negative screens where he donor did not self-defer appropriately, there are some situations where one simply cannot identify exposure. An interesting case is published in Transfusion in 2001 by Charles de Gaulle Airport. A cabin mechanic --

(Laughter.)

Okay. A cabin mechanic was diagnosed with falciparum malaria. Sorry. I guess I hear that. As sometimes happens at international airports, this mechanic served flights coming in from Africa and specifically flights from Angola, and this individual had managed to contract falciparum malaria from working inside an airplane that had returned. Anecdotally he had been a blood donor eight days before he was diagnosed with his illness. That blood was retrieved and destroyed, but it shows some of the situations that it is simply impossible to control.

Obviously despite use in other areas of the world, currently in the US there is no test approved at this time for reentry of donors deferred. Formulary exposure, this would certainly be a great improvement to the situation and hopefully this workshop will take us closer to that reality. Absence in up-to-date mapping utility makes it difficult for blood establishments to actually sit down with a donor and pinpoint exactly where they traveled and whether or not they had exposure, and as most of your post-donation information related to trying to get a fairly complex travel history triggers costly operational measures. This can mean product retrieval and quarantine, consigning notifications, biologic deviation reports that need to be submitted to the agency when the --- is released, and all in the context that endemic areas for malaria are subject to change. This is not a static environment. It is a micro epidemiology where new areas of endemicity do pop up now and then.

(Slide.)

And this actually happened just this year where CDC in contact with the Bahamian government was made aware that there was an outbreak of P. falciparum malaria in Great Exuma Island in the Bahamas. It was actually documented for a couple of weeks during the month of June. Eighteen cases were identified, four of these were in travelers, and it was a self-limited epidemic. No cases since June 19th. This is in contrast for the past six years in Great Exuma where only one case had been identified. So CDC recommended prophylaxis to travelers to this area and contacted FDA and the blood community to make sure this information was known, and then AABB on one of its latest news reports just reminded establishments that a 12-month deferral for donor travel to an area identified by CDC as having endemic malaria was appropriate and defined April 1st as a conservative starting point for this outbreak.

That is all I am intending to present. I look forward to questions.

DR. PARISE: Thank you, Alan. Okay. I just now would like to ask all of the panelists who talked in this session to come up to the front and we will have the panel discussion.

Session I Panel Discussion

DR. PARISE: Okay. We will open it up for questions. Hello? Can you hear me? Okay. We will open it up for questions. A few of the questions that the FDA had asked us to specifically address and people may have questions or things you want to comment on that about these subjects. What are the main sources of malaria risk to the US blood supply, and how effective are the current safety interventions?

DR. KUMAR: Well, I guess we would welcome questions from the audience first. Yes, Tom, please. Maybe if would be nice if we just move up here.

DR. PARISE: If you can identify yourself just before you speak.

DR. McCUTCHAN: Yes. My name is Tom McCutchan from the NIH. How statistically sound -- actually it was in Hira's talk -- is the evidence that we are actually having an effect over the last 10 or so years because of a deferral policy? I mean, one case in 100,000 or two cases in 100,000 shows up nicely on the graph, but I am sort of naive with regard to how statistically sound that is.

DR. PARISE: I guess that is my question. I haven't done any kind of statistical analysis saying comparing the right now back to 1960. I mean, if you look through, there clearly were more cases per year in those earlier years than we see now.

DR. McCUTCHAN: Well, in '60 there are, but when did the deferral -- I mean, what I am talking about is the last several years. We claim that it is a success, but I am not -- and I am sure it is, but statistically how sound? I mean, those curves flatten out pretty nicely. They are on a downward slope, but --.

DR. PARISE: I mean, I -- we haven't really analyzed defacto. You know, there are probably a number of factors. The donor policy hasn't really changed since '94, and if you look back, you know, it didn't even change that much since then for a number of years before. I can't remember. We had a table in the paper we wrote in the New England Journal several years back that defined what the ---. Well, at the very beginning it was just, you know, donor will be free of any communicable disease, and then, you know, later it became more malaria-specific, and there is probably a lot -- you know, there probably are different factors. I mean, if the donor policies as well as how well the blood banks are doing at implementing those policies. I mean, it is not just the specific change in policy per se. But all I can say is we are clearly seeing less cases than we used to see induced by transfusion.

DR. McCUTCHAN: Right. But what I am asking is, is that real?

DR. PARISE: I guess I would say yes. I think it is real.

DR. McCUTCHAN: Okay.

DR. SUBIANCO*: Sal Subianco from America's Blood Centers. It is a little bit of followup to the previous question. If about 60 percent you indicated of the individuals that transmitted malaria had failed or the questionnaire failed to catch them, and is there a reason for us to assume that at least 60 percent is focused in one group? Couldn't we consider that 60 percent of the questions are ineffective in general and that this would be an indication in a certain way that now despite this failure of 60 percent were are still having an extremely low incidence of malaria by transfusion? And as part of that, that is a question for you obviously, but as part of that question for Dr. Williams, if we have the evidence that the problem is smaller, if it is clearly related to immigrants from areas of very high prevalence like Africa, west Africa, why are you considering more rigorous donor deferral criteria like reducing the residence for five years to one year? What was the driver maybe for that?

DR. WILLIAMS: I am not sure I necessarily understand the first part, but I guess if, you know, a proportion of your screening processes are successful and you apply that to the relative proportion of donors capable of transmitting malaria that is -- you know, if a higher proportion are transmitted by prior immigrants and you have a certain failure rate, you have a higher likelihood of seeing, you know, a failure result in transmission than you would in a travel deferral where the potential to transmit is much lower to begin with. So that might be at play there.

In terms of the change in the residency definition, Sanjai might wish to comment on that further, but I think it's basically two considerations. One is scientific and the other simply trying to simplify the questioning process. From the scientific basis, I think it simply relates to the fact that one year in an endemic country is sufficient time to have multiple exposure to malaria that would result in partial immunity. That is the basis of the concern. From a screening standpoint, a five-year residency definition that could be variously interpreted as continuous versus cumulative is somewhat more complex to administer to a donor than a one-year continuous period, and at the same time the one year is somewhat more rigorous, but also would capture the individual who had been in fact a resident versus a five-year residence considered continuous where the donor may have gone somewhere for a short period and wouldn't have been caught by that residence.

All of these are, you know, basically best estimates of a deferral criteria that will give us the best ability to distinguish, but that is some of the thinking that went into that. Sanjai, do you have any additional --?

DR. KUMAR: Yes. Just comment very quickly on five years versus one years. Five-year deferral apparently comes from mostly children in sub-Saharan Africa who die of malaria. They die within the first five years of their age. So that was taken as an indication of evolution of partial immunity of clinical immunity, and along with that comes with an asymptomatic low-grade parasitemia. But there is very little that looks at how immunity evolves in adults and if you are worried about long-term residents, the adults who donate blood. So whatever linkage that is there it says that within one to two years continuous residence in a malaria endemic area is sufficient to induce partial immunity, and all it requires is sometimes somewhere between two to four exposures to malaria parasites. So immunity in adults is known to evolve a lot faster than in children, and that is why I think the five-year might be scientifically erroneous for adults.

DR. PARISE: Did that answer your question? Okay. Jerry, go ahead.

DR. HOLMBERG: Yes. Jerry Holmberg, HHS. Actually my question, I have multiple questions on one topic; and that is, you know, what we are really dealing with is addressing a geographical deferral, and we all know that geographical deferrals not only include malaria risk, but also other parasitic

or viral risks such as --- or chick virus and some of the other emerging type of concerns that we might have. What I am concerned about is, first of all, the tiered question that you referred to. How would that -- Alan, you referred to the tiered question in the uniform standard questionnaire. Would that -- how would that affect some of the other organisms that we are concerned about? Then also I would like to follow up with you commented also on the yellow book and the webpage. How often is that updated? Is the yellow book still available and is the webpage updated? And then also what is the time table for the GIS improvements?

DR. WILLIAMS: So I will go ahead first with the question about the capture question approach. It really should reach the same end, and it should reach it in a more efficient manner. So that instead of asking a long list of potential travel areas, one starts out with a question, for instance, "Have you been out of the United States in the last three years?" This would then eliminate a large proportion of donors who would not have to go through the subsequent questions as far as, you know, where they have traveled, where they have resided and so forth. Given that some of the agents that you mentioned are also included, some of the --- viruses and other emerging agents from sub-Saharan Africa, we have seen, you know, potential protections from lhasa, ebolas, other agents which may or may not have asymptomatic carrier states. But the question process should certain get at these individuals who have had that exposure at least as well as the previous way of administering the question. In addition, these questions I think importantly for the first time in -- since the genesis of the donor screening process have been looked at by the National Center for Health Statistic for donor understanding of the question and have been modified to try to optimize that.

DR. HOLMBERG: So has that been the validation process?

DR. WILLIAMS: What process? I'm sorry.

DR. HOLMBERG: Is that the validation process?

DR. WILLIAMS: It's a cognitive validation. It is not a validation from the standpoint are you really capturing the information you need to capture. That is much more difficult.

DR. PARISE: I will take the questions then on the updates of the yellow book and the website. We systematically -- and Paul --- is here who really knows more about this than me from the CDC. So, Paul, wherever you are, if you want to --

UNIDENTIFIED: Here.

DR. PARISE: okay. If you want to add anything if I get anything wrong, please feel free. We systematically update the yellow book every two years, and the website, too. I mean, we look at every -- across all countries and look at the data we get from WHO and other sources. If something comes up in the interim, for example as has happened in the Dominican Republic in the past or in the Bahamas now, you know, that would be cause for an immediate update. And although we can't alter the hard-copy yellow book, I mean, that would trigger changes to anything we put up on our website on the various pages, which include, you know, the website yellow book.

I guess as far as the mapping project, there are going to be discussions I think at lunch on some -- you know, in detail on that with a group of people. But one of the barriers at this point has been just identifying the resources to sort of take it to completion, and hopefully we have solved that problem. I no longer work in malaria actually, and Paul can correct me. I think they are going to try to get this up by the end of the year. I guess I would say, I mean, I think it is important that we are going to have a pilot phase to that first.

We have made some assumptions. You know, these data are not black and white, and what really isn't black and white is the urban versus rural, and it is hard to really completely ignore the urban versus rural because the risk

areas in some cases are really defined by urban verus rural. I mean, the information we get from WHO and what we put out is that, you know, there is malaria risk in urban areas and none -- I mean in rural areas and none in urban areas. So we will have to define where cutoff is. That has been very difficult to do. We have made some assumptions, and I think the pilot is going to be important to make sure that that is working and that, you know, we get input from people before it totally goes live. Paul, anything to add?

UNIDENTIFIED: Sure. I will just add the time line partly for that, we are aiming at December, but we will see as time goes on. Another thing to add with regard to the book, certainly as there are temporary recommendations for
--- prophylaxis such as there were for the Bahamas, those are expected to be very short, so those do not get the -- the outbreak notice goes up onto the website, but it does not change the yellow pages section of the online yellow book when there are -- there has been one reprinting of this current addition of the yellow book, and we were able to add additional changes to the actual print version as well as to the online version such as we did with the Dominican Republic because that will be a longer term change to the --- nations.

Monica is correct. Every two years it does undergo a comprehensive change, and --- question is the book available, absolutely, including it is now available not just through government sources, but also through private sources. You can find it from online retailers as well as in book stores now.

DR. SAYERS: Merlyn Sayers from Carter Blood Care, Dallas, Fort Worth, and Waco. This may be a little big to the panel discussion, but I am prompted to ask the question because of the known risk in question one. I don't think it really matters whether one is talking about --- or --- disease or malaria. We are talking about transfusion-transmissible risks which are so rare that each new event inevitably justifies publication. So against that background, Monica, just to refer back to a point that you made which had to do with how you would base recommendations on the resort issue and would those recommendations be based on the endemic situation, which would be the conservative approach, or would you look at the traveler data, which would be the less conservative approach. Then you went on to say but then you would have to grapple with the question of acceptable risk. So I just wonder if any of the panel members think it would be worthwhile having a workshop in the not too distant future where we might address what exactly acceptable and non-acceptable risk is and then perhaps use those discussions to guide some of our deliberations about what new interventions might be to reduce the risk of remote likelihoods of infectious disease.

DR. PARISE: Let me just say one thing quickly, and then I will defer this to the FDA, who really has purview over this. I just again want to remind people the risk is low, but that is the risk in the face of effective programs. Then I guess as far as the question of acceptable risks I think I will defer that to Sanjai or Alan, if you have anything to --

DR. WILLIAM: Unless Jay wants to address it. I would say typically from a regulatory standpoint we make decisions on a scientific basis. Define the risks, the cost-benefit of interventions, and acceptability of risk often is determined at different levels.

DR. EPSTEIN: Well, I think that there is no magic number. That what we try to do is achieve the safest feasible blood supply, which means driving the risk as low as is practically attainable. So we are always looking at the sources of threat and the available interventions, and we are always constrained by issues of practicality. You know, there are limits to what can be done. But, you know, what is possible to do and practical to do we think should be done, even if the absolute level of the risk is low.

I think where the argument gets engaged is that there is an undefined domain of public health tradeoffs if you look at the, you know, dollar

costs per infection or adverse event prevented in the blood system. We know that it is out of proportion to that kind of cost effectiveness metric applied in other areas of medicine, but that gets you into the whole question of what does society expect.

You know, we continue to respond to at least a perceived demand for the highest achievable level of blood safety, so I don't have an absolute answer except to say that it is not driven by an absolute number. In other words, there is no threshold for action that has ever been set or agreed upon. Dr.ÿOckenhouse, you seem to want to answer also.

DR. OCKENHOUSE: Well, yes. I happen to think that is the reason we have this problem. I mean, based on that philosophy you will exclude one case of -- if a solution was possible, let's say a technical solution of finding a parasite of blood in a transfused unit, we would exclude -- if there was one case that we would exclude we would institute that policy. I am not sure that is in good public health for the -- you know, a good public health practice, and I get back to I think there are two issues here, and I am not sure. One is safety, and I think the blood supply is quite safe for malaria risk, and I base that is if you didn't have that, you know, is this at a -- are individuals at risk for death? Perhaps, and why would they die? Probably in my opinion as a clinician probably poor medical care. They should have been diagnosed with malaria. This is a totally treatable illness. This is unlike some of the viruses.

On the other hand there is another thing, is this deferral and if this keeps coming up permanently losing individuals permanently from the deferral pool, and this comes up because there must be some type of -- what is driving this? What is driving this, one, we are not having enough blood -- if we defer individuals permanently that suggests that we are -- let me back up here. I'm sorry.

It seems to me that the impetus to retain blood donors is because we want to use those individuals for additional blood donations. So that is what drives a lot of this policy. I think that if we -- you can retain individuals by changing the policy. You know, who are at risk for transmitting malaria? It is for every donated blood in this country, it is -- for instance, if I was -- I am non-immune malaria. I go in to give a unit of blood. They will ask me have I traveled somewhere, and I say, yes, I have traveled to Africa last month; and they will say, "You are now deferred for one year." What is the evidence that that is good policy? I think there is very little scientific evidence that that is good policy. Perhaps it is very good that I was deferred for one month, because the chances of me if I was exposed to malaria coming down with malaria are very high in the first month, but not in the sixth or seventh month. So instead of losing me permanently for deferring me for a year, you may only need to defer individuals for a shortened period of time.

You could retain a lot of those individuals and keep the cost down without having to screen a lot of units of blood for malaria. On the other hand, if you think that the safety of the blood is not to the best that we can achieve, then you would argue that we need to sample all blood for antibodies of nucleic acid of the presence of parasites. It is this type of dichotomy, what is driving this issue. Is it the safety or is it deferral policy?

DR. KUMAR: Maybe I would like to one thing here. I think we have already --- deferral policy has been. If you go back --- numbers are not like not to be completely cited, but some of the data that is available there for --- days. The cases in transfusion-transmitted malaria used to be as high as 60 or so in each year, and those were days when there was much less travel to malaria endemic areas and much less immigration from endemic countries. We have to bear that in mind.

DR. PARISE: Go ahead, Jay.

DR. EPSTEIN: Well, I actually was going to raise the same issue, hoping that Monica would respond. Dr.ÿOckenhouse, you have basically put a challenge in front of us. You have said is the deferral policy for travelers overkill, and I think that is an important question. There is a scientific basis for the deferral period, which is the information about time to illness in travelers who become ill with malaria, and what we have done is we have tried to maximize safety by looking at about the 98 percent capture. Now it is true when you look at that curve, you know, the majority of cases are a lot earlier than one year. It is just that there is a tailing end, and so then the question is, well, do you address that or do you accept the risk.

So it begs two questions really. The first question -- and these are the things I had originally stood to ask. The first question is what do we know about the malaria risk in persons we defer, and I know there are some data. I don't know, Sanjai, if you were going to bring it up later, but we didn't hear it in the first panel. You know, do we have an estimate of the malaria incidence in travelers and in particular travelers who might qualify as donors, because that is important. It is a measure of the, you know, efficiency of the deferral.

Then the second question, which I think is even more provocative, you are really saying if a non-immune traveler contracts malaria, do we really think that there parasitemic and still asymptomatic. In other words, what is the likelihood that they actually will transmit, and I think that that is an answerable question. It comes back to how carefully have we scrutinized the donors who actually transmit it to find out how long was it from their exposure and was there any early evidence of symptoms. You know, were the actually donating say between febrile intervals.

So I think that those two pieces of information would help us put, you know, a scientific framework on the very provocative and reasonable question that you have asked. So I am hoping Monica can answer those questions.

(Laughter.)

DR. PARISE: I guess one thing now, you know, as Sanjai mentioned before and Jay has brought up before, I mean, we are basing this really on cases of malaria. So these are asymptomatics, and that is really being extrapolated to the asymptomatics that we don't have a handle on at all. So, you know, that is just one limitation of all of this. But we do know, and I didn't bring the slide, that when you look at people that live in non-endemic areas, I mean US residents, and look at how many of those, what percent of infections come up over a year, it's two percent. So I wasn't here when, you know, these donor deferral policies came up, but I am assuming that, you know, when the cost -- when the risk-benefit ratios come up numbers like what was discussed and this one-year and three-year were brought into being that those kinds of things were taken into consideration. The number for what comes up in foreign residents over three years is considerable lower, and I believe it is about 0.2 percent. I do have that calculation, but I have not looked at it for a little while.

I guess one just point that came up to me, you know, as we look at the deferral policies and who is ---, I mean, one thing that is possible that I don't know how much we know about either is that, you know, it just may well be true that, you know, these -- we are seeing most of these, you know, cases come across in immigrants as opposed to African. Part of that, you know, many of them that come from Africa, and one factor there could also be that the donor deferral guidelines are functioning better in our travelers and they are functioning in immigrants where there may be language barriers and other things.

I guess your last question there, what do we know about these donors and their symptoms. You know, unfortunately we don't know very much. It is a good point, and it is probably something that we should incorporate into the questions that we suggest be asked in these investigations. I think that is good idea. You know, much of this data we just took from surveillance reports.

You know, we only have a little more detail on the donors I would say from the last decade when we really started, you know, to try to do things more systematically, and we do have a list of somewhere between eight and 10 questions that we at CDC suggest that whoever is doing the investigation, the blood establishment or the State Health Department or in conjunction, ask to those donors. We have not asked them about their symptoms, but we certainly could do that.

DR. GORLIN: Gorlin, Minnesota. Alan was far too kind. I was responsible for the May 15th, 2001, submission to BPAC showing up from Minnesota in my snow boots since I forgot my shoes, and was very pleased to have the positive answer from BPAC suggesting that growing donors by plasma pheresis would be okay if they had gone to deferral areas since there are simply zero cases of transfusion transmission from a frozen plasma pheresis product. Only when I returned did I find out that the computer system had absolutely no way of accepting a donor for one product but deferring them for everything else, and so my QRA department chased me into the corner where I was of course -- had no courage to actually submit the variance.

We have just updated our computer system, which apparently we think now could do this. I am little well bereft that the contemplated guidance update is not going to accept that plasma pheresis exception, so I am just a little curious as to your current thinking.

Then a quick question for the military. I would -- I am naive as to logistics as to getting product routinely and rapidly into endemic areas, and if there were a large-scale engagement in an endemic area wouldn't it be a logistic advantage for the military to be able to draw soldiers in those areas and have a testing methodology? I'm just curious.

DR. OCKENHOUSE: Well, for your second question, that is exactly what we do. We draw -- the Army blood banking program has two components. They have a frozen packed cell component which has been going on for 20 years, and I don't think we have actually drawn on that. All the blood that we use for transfusion worldwide is either flown in on a daily logistical basis or is collected in country and screened using the currently accepted screening procedures.

DR. GORLIN: But which don't include a malarial screen.

DR. OCKENHOUSE: Right, no.

DR. GORLIN: So wouldn't it be an advantage if you were collecting in country to have a malarial screen?

DR. OCKENHOUSE: Well, not really. No, I don't think it would be an advantage at all. I don't think we would do that policy. I mean, if we are deployed in a malaria endemic area our soldiers will be on malaria prophylaxis. Malaria prophylaxis, there is a very small chance of them transmitting malaria, so I don't think we would do that.

DR. GORLIN: And Alan?

DR. WILLIAMS: I will just comment on your first question. I am glad to have the extra time here. In fact as you mentioned, the variance was never submitted, so the issue -- the question wasn't asked. The blood products advisory committee discussed that question carefully and took a vote. The nature of the variance request is a variance to a regulation, so --- regulation takes some time; and, you know, basically we didn't have to face the question at that time, but it certainly could reemerge in the future, and that might be a discussion for another day.

DR. EPSTEIN; If I could just add to that. It is true that we thought the issue mute because the variance request was withdrawn. It is also true that in reflecting on the discussion at the advisory committee we felt that there were two pieces of data that were missing. One is the residual red cell count in a variety of apheresis products prepared in different ways, and of

course linked to that is the uncertainty about the minimum infectious dose of plasmodial, which might be an extremely small number.

Then the other issue is that a lot of the data that committee reviewed had to do with non-transmission by plasmas that were being infused in I guess the Minnesota area, but what we didn't have is an estimate for how many of those donors might have actually been incubating malaria. So we didn't know what to do with the non-transmission data because we couldn't answer the question of whether a malaria-infected donor had made -- been a donor of FFP what would have happened in a recipient. So, you know, we were a little bit disquieted by the gaps in the information, and so, you know, these just gave us cause for pause.

DR. NAKHASI: Thank you. I have two questions. One is for Dr. Ockenhouse. I just -- you know, which is basically following up on to what Jay and others asked. So when you were mentioning that there is no immune -- non-immune travelers are not at high risk, and it is highly unlikely that non-immune asymptomatic blood donors carry malaria parasite in blood. What kind of an evidence do you have, because that dovetails with the question you were posing to ask as an FDA, why should we different with people, you know, if they are only exposed ---.

DR. OCKENHOUSE: Well, I base that on the biology of the malaria parasite. It is I would think that if someone is coming in to donate a unit of blood they are going to come in probably fairly healthy. All right. They are not going to be ill. They are not going to have a fever. If they are, you will defer them on sight because they have a fever.

So the question then becomes if I am donating a unit of blood what is the likelihood that I am carrying a malaria parasite? I am a resident of the United States. The likelihood of me carrying a malaria parasite is extremely small because I am healthy, unless the level of parasites in blood are below the clinical threshold. The clinical threshold is about one parasite per microliter. All right?

Now, when would that occur? That would occur within a period of exposure. I could have been exposed two weeks ago. I could have traveled two weeks ago to Africa. I could have been bitten, and I could have malaria parasites in my blood. Yeah, and I come in now two weeks later and I am here for my regular donor time. I am going to give a unit of blood. Now I could have a malaria parasite. All right? And I could donate that blood, and that blood could be transfused and the recipient could come down with malaria.

However, now the alternative is if now I am six weeks out or two months out and I said I traveled two months ago, could I have a malaria parasite? It is highly unlikely, because 90 percent of the cases of malaria come out within the first 21 days because I am non-immune. Now for immune individuals who have been exposed previously all bets are off.

So the question comes, and I don't know the demographics of people who give blood in this country, how many of them are non-immunes? I suspect that the vast majority of individuals are non-immunes. They haven't been born outside of the country. They haven't lived in an endemic area for quite a while. So how many of those individuals that we defer really impact the blood supply and -- you know, these are demographic type of questions which I don't know. It would be very interesting. Because, you know, you could say we are going to defer individuals specifically on a few questions. You don't need a lot of questions.

The second thing is that I don't think geography makes any difference. GIS, you know, that is all well and good, but I could go to the most heavily endemic area in the world, western Kenya where I go all the time, and I could come back and I am off prophylaxis, and I am ready to give my unit of blood. The changes of me transmitting malaria is extremely small, and so if somebody goes to Mexico on a resort and we are deferring those individuals I

think that is a ridiculous policy. You know, that is a personal opinion, and I would like to see the evidence to say that we are hurting the blood supply by deferring those types of individuals because I think that is the issue here. People say why are we doing this. It is a deferral issue. We lose people in blood bankers' case. We are deferring all these people. We are removing them permanently from donating, and I think part of that is by overkilling the policy, and I think we ought to use evidence-based policy based on the biology of the parasite. Now if other malariaologists in this room disagree about that, that would be very important. But I would like to find out if anything thinks if you are a non-immune that you are asymptomatically carrying malaria parasites. I would like to find you and study you, because that just doesn't occur.

DR. KUMAR: Chris, let me comment on something just very quickly here for what you said about a trip to western Kenya and --- prophylaxis. You presented that yourself a few minutes ago giving a military campaign in Liberia, 126 Marines came down with it 100 hours after landing there. Okay? What are the chances? I mean, how much can you trust a 21-year-old young man going to western Kenya that he will follow the chemo prophylaxis and how much compliance will be there? Certainly the issue is a lot more complicated than you ---.

DR. OCKENHOUSE: No, that has nothing to do with non-compliance. They could not ---.

DR. KUMAR: If the US military cannot enforce compliance on malaria chemo prophylaxis how can one be sure that the normal population, especially most of these young men and women going to --- will follow up and will consider --- simply they have been considered --- malaria prophylaxis?

DR. OCKENHOUSE: No, I wouldn't even ask that question. I don't think it matters one bit whether an individual has been on prophylaxis or not. I would just ask how long have you been out of the country. If you have been less than a month your chances of having a malaria parasite incubating is probably higher than if you were out three or four months.

DR. NAKHASI: But the question is, which is again a question to the blood bankers here, how feasible is it to ask that question and how effective is that when people remember that he was one month back, there are three months ---. That is the question, and as Jay mentioned there is a risk versus benefit in that ---.

DR. OCKENHOUSE: Sure. I mean, if you ever donated blood you go in and, you know, some people just ask questions. They don't even listen to the answers. So, you know, that is obvious. All right? So --

(Laughter.)

DR. OCKENHOUSE: But the question is how serious do you take this. All right? And how serious is the training of the individuals who work in blood banks? All right? I can't control everything, but what I can control is understanding the biology in the context of how likely malaria is to be transmitted.

DR. NAKHASI: I would like to hear from biologists, too, how effective is that when it is one months or two months or three months.

DR. PARISE: Let me just say one thing. We have got about one minute, and then we are going to cut this off at 10:15. If people want to chat, then after --

DR. NAKHASI: Maybe we should hear John Barwell.

DR. PARISE: Yes, and then we will stop, and then people can come here and talk among themselves if they want to continue.

DR. BARNWELL: The exception to that is vivax and ovale, and that is because people going to endemic areas are on prophylaxis, and you are killing off your first malaria parasites that come out and you are worried about the relapse phenomenon. If in depending upon the strain of the parasite, the relapses could occur within one month, it could be six months, and it could be

nine months to a year. That is one of the reasons I think that probably some of those policies were instituted for deferral of non-immune -- you know, from a non-endemic area for one year. Now if you look at the recent one in the last 15 years, there hasn't been a case due to vivax and there is one case of ovale.

DR. OCKENHOUSE: So that is my point. In the last 15 years we have one case of malaria transfusion per year; 10 years, you know, 10 cases, not one case of vivax. I would even say, you know, vivax is a non-fatal illness. It is an illness. Matter of fact, you don't even have to treat it. You can probably -- they are going to be sick. Individuals are going to become sick, and it is self-curing eventually. All right? Now the point is are you going to institute sampling and testing of every unit of blood to satisfy one case of malaria? And I am not sure that you need to go to that length when you might be able to just change in order to retain donors. Relook at this one-year versus three-year policy, and look at and base it on real evidence. Certainly you can go, you know, with the first couple of months and then, you know, double it, but --.

DR. NAKHASI: Yes. I think we will have more discussion on that at the end of the day. Just a quick question for Monica. I think, you know, I was surprised, maybe that is my naivety, that 90 -- you said out of the 69 implicated donors 90 percent were male. Why not females?

DR. PARISE: I don't know.

(Laughter.)

DR. PARISE: You know, I don't know.

UNIDENTIFIED: ---.

DR. PARISE: Some of that. I guess if I separated that out by decade that might be what comes out, because a lot of the earlier ones were in military from Viet Nam. That is probably what it is. I guess I just want to close with one thing. I mean, there are two percent -- we may be missing the first primary infection as John just said, but as you can consider this one year, I mean, two percent of these non-immune US residents come up over one year. So, I mean, that has to go in the equation I think.

All right. We are going to close. I am sorry. You know, we need about a 15-minute break, right? Okay, and if you have questions, I mean, there is going to be a larger panel discussion.

UNIDENTIFIED: The purpose is questions for the whole audience, not to come up to the front.

DR. PARISE: Well, I guess I am just trying to say that it is time for a break. We will have a time at the end of the day for another panel discussion, so you can get your questions in.

UNIDENTIFIED: I do think that you have a major misperception that perhaps the audience should know if they don't, and that is that perhaps the decline in cases of malaria in the last decade, that is probably totally unrelated to the screening policies. Because as most of the audience knows, we have been screening geographically for malaria for more than 30 days. This didn't start with the 1994 FDA policy. We have tweaked it a little bit. We actually have screened people for whether they took prophylaxis, different geographic areas. But screening has been going on for more than 30 years, and I think it would be foolhardy to believe that changes over the last 10 years have resulted in a decline of transfusion-transmitted malaria. Clearly there are a lot of other issues.

(Whereupon, a break was taken at 10:21 a.m.)

SESSION II: TESTING FOR MALARIA INFECTIONS

Roger Dodd, PhD, Session Chair

(Technical problems with equipment. Introduction of first speaker not recorded.)

Developing a Test to Detect Malaria Infections in Blood Donors

by P. Nigel Appleton

MR. APPLETON: I have to say thanks for being invited. Sorry. I will get used to this technology eventually. I have to say thank you for being invited. It is an honor to be here and a pleasure. I am always rather surprised not to be booed and hissed because I'm commercial.

(Laughter.)

Thank you for that. Okay. To put me in context, New Market actually is a diagnostics company operated from the rural end of eastern England, and I am reliably informed that I keep my hands to myself.

(Laughter.)

I am reliably informed that the part of England we operate from is going to be thick with enough of these mosquitos by 2025 or dripping parasites, but there we go. Here I am.

(Slide.)

About 2001, the local blood transfusion service or the local outpost of it suggested to us they were losing a lot of donors under this long deferral system they had for these people they felt might be at risk of transmitting malaria through transfusion. About 60,000 a year was mentioned, and that is an awful lot to replace in a voluntary donor system, and of course we, being England, we have a high immigrant population and we travel. We are inveterate travelers. So that is a lot to make up. In any case, the deferral system it was suggested to us was not 100 percent effective, and that was illustrated later when a gentleman I believe of --- extraction who had been in the country for a large number of years and had not been out, not visited his homeland, actually managed to transmit malaria by a blood donation. He had just, as far as anybody knows, just been asymptomatic for all those years. Had he been tested with the test I am about to describe he would have been picked up, but nobody knew that at the time.

(Slide.)

So the task was to develop a test to allow or to help to allow quicker return of deferred donors into the donor system to try and stop this seepage of good blood from the system. At the same time of course we want to at least maintain or preferably improve the safety of the blood supply. That goes without saying I suppose, and for those people that wanted to use a laborious --- antibody test we would like to replace that. I think many people here will recognize that it is a laborious test. It is very subjective in its interpretation to say the least, and the operator variations are usually quite unacceptable. The reproducibility is unacceptable, and so on.

So, but I need to stress that this, what we were asked to do, was not a malaria diagnostic. It is an aim to strategy attempting to maintain donor numbers. That is where we started from, and since it was a good customer that was asking us we thought we had better go along with it. So we had a number of criteria to meet.

(Slide.)

We have a number of criteria to meet. Whatever we develop has to be reasonably compatible with what is already going on in laboratories. It is not the time for a small company like ours to try and impose some kind of weird or fancy new testing, however good it might be, something new and fancy on the using laboratories. Some people tried. We don't.

Simple and cost effective goes almost without saying, although it is something kind of surprising how people's definitions of what is simple vary. We use simple as simple.

(Laughter.)

High sensitivity and specificity, well, they do go without saying, and we can usually find a consensus on what those two terms mean. Amongst that of course we should have it reactive with all four plasmodium species affecting humans. So, I mean, none of this is any surprise to you. This may become more important in the UK because there is some anecdotal evidence that the rate of

importation of infections with the two rarer species, malariae and ovale, may actually increasing. Not sure yet, but there is some indication there, so it may become a bit more important.

(Slide.)

So we were involved in this as well. We are the developer of this test, and we have our own targets and criteria and agenda to meet, so we have to make as much use as possible of existing knowledge and reagents. When we looked at it, the actual existing published knowledge in malaria serology, it wasn't an awful lot. The actual reagents available to us were actually quite limited. We also have to be commercially viable with respect to costs. It is nice if you can get IP protection, but that is not a major aim. We always choose a microplate format to start. It is widely spread, widely used, widely trusted, the machinery to handle it is out there, so those are most of our criteria.

(Slide.)

And we have problems. The first problem we came across, the numbers of sera we have to test to validate what we are doing is small if you compare it to anything else. Syphilis, any of the other diagnostics we are involved in. We have tens of samples rather than the hundreds we would normally want to look at in this, and many of the samples we had access were years old.

They had questionable storage histories. In some cases storage histories were just always being in a freezer for 10 years or we think it has been a freezer for 10 years, but it might have defrosted itself a couple of times. Many samples we had access to have unknown or inadequate case histories, and particularly that time scale of when was the patient exposed or infected, when was the sample drawn. We often don't know, still don't know. It is getting better.

We have no seroconversion panels available to us. I think I would like to talk to the US Army. I think they might have much better material available then we have. There you go, and the only --

(Laughter.)

There is no true gold standard test for malaria serology. All we have got is the IFAT, the immuno-fluorescence test. Some of the results there are not always reproducible. On a stored serum you can have a stored serum associated with an IFAT result of any particular titer, say 80. That sample is resurrected a few years later from storage. It is retested, and the titer is 20 or 320 or significantly different, and you are never sure whether it is because it is a deterioration sample, which is possible, or whether it is just observer variance, which is equally possible with the IFAT. So all these are difficulties.

The other problem of course is we want a test that is pan reactive. The only one we can cultivate, the only species we can cultivate, is falciparum. Now the information available suggests that some antigens of plasmodium falciparum cross-react with antibodies induced by the other species as well, but we don't know which, what, where, or why. So that as a source of antigen, that is not particularly good. We did try it. Crude --- of the cultivated plasmodium gave us huge background readings and very little discrimination. Low single to noise ratio, so we very quickly abandoned that.

(Slide.)

We turned to our friends in the academic and research institutions. The National Institute for Medical Research, various universities and so on, to help us identify some highly conserved and immunogenic antigens. We hope to make use of prior research into candida vaccine molecules, because obviously people who have been researching into vaccines would be biased towards immunogenic molecules, which is obviously a good thing. At the end of all that then lots of discussions. The MSP's, some of which have already been mentioned, the merozoite surface protein showed the most initial promise and had an

advantage to us in that the recombinant versions of these were readily available.

(Slide.)

We did of course look at the common antigens, the pan-malaria antigens, the ---, dehydrogenase, and in our hands they are not particularly immunogenic. Many patients do have antibody titers to them, but they are very low, very variable. We are not sure whether the recombinant versions of these enzymes that we used were correctly folded, exposed to correct epitopes. We have no real way of finding out. As it turned out, the MSP's are immunogenic because they are exposed at the surface. They are exposed to the immune system during replication of the parasite and even portions get shed into the blood. So there is plenty of chance in the immune to hold of these and recognize them, so we concentrated on those.

(Slide.)

We have some technical considerations, too. If we are making a test we want to make a good test, and the format we prefer is this recombinant antigen sandwich where we have recombinant antigens on the solid phase and in the conjugates because we get very low backgrounds with that. Very high signal-to-noise ratio, so the discriminatory power is excellent. It does make very economic use of materials. We generally get very good stability, very robust in guard-band studies. That is important to us.

We found that whether you are doing these tests or whether some laboratories do these tests manually or use liquid handling systems of one sort or another, somebody somewhere will find a way of doing it wrong. They will use the wrong temperature or the wrong time, whatever. Sometimes the machines don't reach the temperatures they are programed to reach, in spite of the fact they tell you they are reaching those temperatures, and so on and so forth. So the test has to be really robust and take account of those variations.

With this format we can also use undiluted samples, which helps with the sensitivity, eliminates one --- and one sample dilution step, and we can use an automated way of verifying the sample presence in the well. So you can't do that if you are using antiglobulins as conjugates. You can only really do that with the recombinant conjugates, and of course we can in theory at least detect all classes of the immunoglobulins.

(Slide.)

To cut a long story short, we concentrated on the MSP recombinants, various fragments of MSP's. Not to our disgust, to our not very great amazement one recombinant is not enough. This was the first one we tried, is a plasmodium --- from recombinant. That is not enough. Add another one in, things look better Another one, things look better still. Add in a vivax MSP recombinant and we are getting somewhere close to where we want to be, and we now are using three from falciparum and one from vivax.

Now an interesting thing -- well, it is interesting to me because I have to develop these damn things --

(Laughter.)

About this format of test is that we haven't yet decided what is the limit of the number of different recombinants we can coat the same micro --- plate with. Because the amount of protein needed to give a result is very, very tiny and nowhere near the saturation limit of the plastic. We have had up to 10 so far in experimental studies which we can coat simultaneously on the plate and still get results for all of them, so that is quite promising. It makes for a complex and difficult control manufacturing procedure, but that is what my production manager is for.

(Slide.)

Okay. We have a test. We put it out to a blood bank pre-launch to see what results they would get. Those are the results they got, which we were quite pleased with. I have to say the malariae and ovale, the number of samples

here I think were something like three in seven respectively, so it is not really statistically significant. That is the best we could do with that.

(Slide.)

Post-launch, and I have to confess the big error of the morning here was as P. ovale. It should read P. vivax. It is the English spelling of vivax.

(Laughter.)

Dr. Ceed and his colleagues down in Australia got those results, which again were very pleasing, and they calculated some extra risk factors. They thought if they reduced their deferral periods, use this test in their own environments, then the risk were as you can see there. The additional risks of infecting recipients with these plasmodia were there, and they obviously think these risks are acceptable because they are using the tests down in Australia, as they are in the UK -- as they will be soon in Holland.

(Slide.)

So what are we doing now? Well, we still want to improve the performance. We are assuming that the performance with ovale and malariae in particular need to come up to near 100 percent. So we found some very cleaver people at the London School of Hygiene and Tropical Medicine who know about things about bioinfomatics, and so they are helping us out there, identified some candidate genes there. We also think there may be a possibility of identifying some more useful antigens for the diagnosis of malaria by antigen detection. That is a bit of a spinoff as far as we are concerned. We are concentrating on antibody detection.

(Slide.)

And the ongoing problems, still limited numbers of samples, particularly for the two rarer species. We were thinking of calling for volunteers to go and be infected, but we haven't had any yet.

(Laughter.)

And it --- more intensive work on the genomes of these two particular species. It is really only just beginning to the same extent as has already been done on falciparum and vivax, so information is beginning to come in thick and fast so we are hoping to do better.

(Slide.)

Lastly I have to acknowledge all the people that helped, including Peter Chiodini here and his colleagues, and many, many people contributed to this development process with the results that you see. Thank you.

(Applause.)

DR. CHIODINI: Okay. Thank you very much for getting us off to a good start. In the interest of time I am going to if you agree keep all questions until the summary discussion at the end. So I suggest we now move on with Freddie Poole's presentation, antigen/antibody diagnostic assays for malaria.

Antigen/Antibody Diagnostic Assays for Malaria
by Freddie Poole

MS. POOLE: Thank you, Peter. Good morning. In the Center for Devices and Radiological Health we look at diagnostic tests a little bit different than in the Center for Biologics. We look at the diagnostic tests as a method for aiding and diagnosing or in diagnosing infection.

(Slide.)

This morning what I am going to do is go over a little bit of the regulatory background so you can get an idea of how we formed our position on diagnostic tests and provide an outline of the types of information that we believe that you would need to demonstrate to us that the device is safe and effective.

(Slide.)

In 1938 the Food, Drug and Cosmetic Act was enacted and then 1976 the Medical Device Amendment where the Center for Devices and Radiological

Health gets its authority was enacted, and most of our authority can be found in section 513(A). Then it was amended again in 1990 with the Safe Medical Devices Act and in 1997 with the FDA Modernization Act, and that is where we got different paradigms for doing our regulatory work.

(Slide.)

In the Food, Drug and Cosmetic Act devices were classified as Class I devices, and Class I devices were those that could by just using general controls, that is things like good manufacturing practices, now they call quality system regulations, registration, and listing and record keeping, manufacturers could demonstrate that their device is substantially equivalent or just as good as another one on the market before 1976. And those that went into Class I were most of the devices for which the test result didn't support life or prevent impairment of health, or it didn't present an unreasonable risk of illness or injury if the result was a false positive or a false negative.

(Slide.)

The second type are Class II devices. With Class II devices the general controls are insufficient. We require special controls, and those tools such as guidance documents or what we call now special controls documents, or label regulations or other standards that were developed.

(Slide.)

The other devices are classified as Class III devices. Those are the devices for which the test is critical in the diagnosis of disease. The test result could present a risk of misdiagnosis which would lead to illness or injury, and for those type of devices valid scientific evidence is required. That valid scientific evidence comes from well-controlled clinical trials.

(Slide.)

Because we have not approved or cleared a device as yet and they have not been classified, devices for detection of plasmodium antigen or antibody are still considered Class III devices because the test results are of substantial importance in diagnoses and treatment of life-threatening illness. They will remain Class III until we receive the first device, and because of the critical pathways that we have on strategic plans now that we have, FDA is willing to relook at the first device, and we believe that we could get sufficient information. There is a new way from the medical FDA Modernization Act of doing automatic classifications and putting devices into a lower classification if it is the first on the market and it has not been previously classified. That process is called a de novo process.

(Slide.)

There is a system that was cleared by FDA. In fact it was approved, the QBC system. It was approved as a ClassÿIII device for automated differential cell counter, but we didn't consider this a predicate because it was a malaria tune that a stain, the acridine orange stain. In fact that was a stain for parasites, and so this was not considered to be adequate or appropriate predicate for antibody or antigen detection devices.

(Slide.)

We have seen different types of assays published in literature, and we have looked at them and other speakers have mentioned them, so I wouldn't belabor those.

(Slide.)

I will now provide some of the information that we would look for for a device for the first of a kind. We ask for non-clinical study. We like to see characterization of components, a description of the antibody used or the antigen and other controls that were used to produce the device. We look at limits of detection of the assay, how the cutoff values were set and how they were validated. We also look at reproducibility and position studies, cross-reactivity with other plasmodium species and other parasitemias, interference

from either endogenous or exogenous substances, and also the stability of the device, which should stress storage and shipping conditions.

(Slide.)

The clinical studies depend totally on what type of claims that you are going to make in your indication for use. That is what would the test be indicated for. Would it be indicated as a presumptive diagnosis or a confirmatory test, would it be indicated for use and differentiate in the different species of plasmodium, and those are the types of information that we require and we ask. In the clinical studies when you develop them, all of those parameters or what we call claims must be demonstrated to show that they are safe and effective and support whatever indications and conditions for use you claim in your package insert.

(Slide.)

With the clinical studies we should see that the probable benefit of the test results outweigh any foreseeable misdiagnosis. You should present a unified multi-site study protocol, because it is very difficult to analyze data when each study used different protocols. Foreign studies are acceptable if they were done following the Declaration of Helsinki.

(Slide.)

In clinical studies we look for appropriate clinical sensitivity. You should have clearly-defined populations, endemic and non-endemic, a clear description of disease status and how it was defined. Most of us still consider the thin and thick blood smears as the gold standard. We would accept IFA also if you clearly defined which method you used and which antibodies were used. We also want a clinical protocol that describes all the methodologies used in the clinical studies, and it would also include exclusion and inclusion criteria and the types of quality control used.

(Slide.)

Clinical specificity should include patients with microscopical other evidence of plasmodium species other than those being detected by the assay or other parasitemias that are present in blood, and other conditions with similar symptoms.

(Slide.)

In clinical studies the cutoff values should be validated with the appropriate target population. The study should represent a spectrum of patients presenting for testing for infection.

(Slide.)

You should include a statistical analysis, and the studies should provide an adequate representation of all type of cases, sub-types, any co-infections, and 95 percent confidence interval, and our statisticians at FDA are always available to comment on any appropriate model that would demonstrate statistical sufficiency.

(Slide.)

For more information we are also free to accept what we call a pre-IDE. It does not mean that you need to do -- have an IDE approved. It means that we would look at your protocol before you start your studies, and we will also give you information on what we consider appropriate type of a study that would support your claims, meaning your intending use. Thank you.

(Applause.)

DR. CHIODINI: Thank you very much indeed. Very useful background, especially for someone like me from a different healthcare system, so thank you for that. Now we are moving on to life at the sharp end. Marianna Wilson works in a reference lab that at the end of the day has to decide whether someone does or doesn't have malaria, and then is investigating hopefully very rarely cases of TTM. So it will be very interesting to hear your perspective.

CDC Experience with the Laboratory Tests Used to Investigate Incidents of Transfusion-Transmitted Malaria

by Marianna Wilson, MS

MS. WILSON: Good morning. I would like to talk to you about the diagnostic tools that we have used at CDC for an awful long time. Certainly blood film examination, antibody detection by IFA has been -- was established at CDC in the mid '60s, and we have used it continuously since then. DNA detection was established in the mid '90s.

(Slide.)

Just a little word about the malaria IFA test, because I am sure a lot of you don't know much about it. The antigen is a washed cell thick smear of red blood cells infected with a species of antigen. There are four species, as you have heard earlier today, and we have been very fortunate to have access to all four species at CDC. I am not sure that is available anyplace else in the world as far as that goes. So we have always since we have had access to all four species used all four whenever we did --- test.

(Slide.)

Sensitivity of the IFA test, as determined a very long time ago, was 98 percent based on patients with P. vivax and P. falciparum infections. Certainly through the years we have continued to add cases of P. malariae and ovale, and the sensitivity is the same. Sensitivity of course is dependent upon the type of patient, the stage of infection, and the time infection, so that is a very loose figure. Specificity at the time is 99.5 percent. One patient with syphilis cross-reacted in that first initial evaluation, and then in the '70s babesia microbia became a problem in the United States, and babesia will cross-react in the IFA test for malaria. Patients with babesia will cross-react, as will patients with malaria will cross-react with babesia antigens.

(Slide.)

We have some data with human experimental infections on the malaria IFA test, and --- parasitemia precedes antibody by anywhere from two to six days. This is a little bit different than we heard a little earlier, but I am sure there are reasons for that. If the parasitemia is suppressed through drug therapy, the patient may still develop detectable antibody.

(Slide.)

This slide is not really complex, but it was a little bit -- let me explain a little bit. The red line is from US military personnel who were prisoners of war in Viet Nam. The black line is from Nigerian -- people of Nigerian origin in the United States. The green line is from experimental infections, and the blue line is from US military personnel in Viet Nam for usually one year who came back to the United States and developed -- came off their prophylaxis and developed malaria rather quickly in the US.

This data shows the time interval of titer levels of patients months after therapy. Everyone was treated. The initial levels were relatively high for all different groups. But as you can see, there is a difference between people from endemic areas with life-long exposure, people with long exposures without prophylaxis, and people who have only one or maybe two infections at the most. Titers will decline in this group fairly rapidly down to six months. About 50 percent were still positive at six months, but at very low titers, and again still this was about 50 percent also. Still positive at 12 months, but still at very low titers. Unfortunately we didn't have the opportunity to follow everybody at a longer time period except for the Nigerians, who maintained their titer after a small initial drop, but maintained their titer, and who knows how long they continued to maintain their titers.

(Slide.)

This is a slide just to show you typical malaria IFA results. In the first across the type are the different antigens coming down this way, slide diagnosis here.

In this case a patient with P. falciparum has high titers to falciparum and malariae, fairly high titer to ovale, and a little bit lower titer to vivax. This is what we consider to be the typical African pattern. Almost inevitably you test an African who has been infected and this is the type of result you will see, regardless of how long it has been since they say they had their last malaria attack.

In this case this is a patient with P. falciparum, but it was a US adventure traveler who has a primary infection with P. falciparum, reacts only with P. falciparum and not with the other antigens.

This case was again the type of thing, but a case of P. ovale. Reacted only with P. ovale, but not with the other three antigens, and then we do see the in between situations.

This is a case of vivax, a little bit of reactivity with ovale, a higher titer to vivax. But this would be a patient who has only had a few infections, one or two.

A case of malariae diagnosed by site as malariae, but I would assume he most likely has had falciparum also at some point in time. High titers to both falciparum and malariae, lower titers to ovale and vivax. The point to this slide is certainly with the IFA test you have got to have the specific species antigen in order to detect all of the individual cases.

(Slide.)

We also used PCR for malaria at CDC. It is a two-step nested PCR using gene-specific primers followed by species-specific primers, and Sanjai will talk more about PCR so I will leave the details of that up to him.

(Slide.)

The PCR as evaluated at CDC has 100 percent sensitivity, 100 percent-plus because, yes, this is based on smear results, and so there are certain individuals who react in PCR who are detected by PCR as being positive, but not on smear. Very definitely PCR is more sensitive than smear results. Specificity is also 100 percent, including looking at babesia patients.

(Slide.)

The algorithm for laboratory testing of suspect donors is that, number one, we screen with serology because that is the most sensitive assay to detect antibodies at any point in time. For those who are IFA-positive, we will use PCR to see if we can detect current nuclear material. In the end just because we are a good old diagnostic lab, we still do blood film examinations on the IFA-positives, too. These are almost usually negative in the case of donors.

(Slide.)

Now for donors from '63 to '98 this is prior to the use of PCR, but the majority of donors were detected by the use of serology, 72 percent. Then there is a section of serology in blood smear, 14 percent, blood smear alone by 10 percent. Those are the ones we didn't get serum specimens on, and then four percent were sole donors, and so they weren't tested at all.

(Slide.)

If you look at the overall numbers, the implicated donors where the information was available, 98 percent had positive serology, 33 percent had positive blood smears. Actually that number is going down these days.

(Slide.)

These are the last five cases of transfusion-transmitted malaria that we have investigated. Of these five, the fifth case in 2005 the investigation is not completed. As you can see, there were 150 donors, and at this point in time we have only tested about 100. The two blood banks involved have not been able to get a lot of the donors back in for testing. So we did have in that case one positive early on in the investigation by IFA, but that particular person was negative by PCR and his history was he denied any sort of travel history or any sort of malaria history. So that is an ongoing process.

For the other four cases, previous four cases, you can see that the IFA test detected one positive donor in each of the different cases. In the two older cases, PCR was positive. In the two newer cases, PCR was negative.

(Slide.)

Just looking at it in a slightly different way, because I was interested in looking at the date of the transfusion versus the collection data of the donor specimen that we tested to see if there was some indication of problems there. In the first case, the slide result of positive, the PCR was positive, and the specimen was drawn about four months after the original donation date. The second case we actually were able to -- we think. We think at that point, that was the actual donation sample tested. Slide results were negative, PCR was positive. For the other two cases, the collection date of the donor sample was approximately a month or two after the initial donation date, but the PCR was negative as is the slide. So no rhyme or reason from this very small sample as to why the PCR was positive or negative.

(Slide.)

This is a slide which actually was shown at a previous FDA workshop, and I borrowed it from a coworker, so take these numbers just as an example. At the time they seemed to be relatively decent numbers for the detection rates of the different tests, but those certainly are subject to a great deal of debate I am sure. But just to give you a little perspective, antigen detection tests are the least sensitive of the tests for parasites or parasite products. Microscopy is next. PCR certainly picks up more than microscopy, but it is still relatively insensitive. If you look at the numbers in terms of consider the fact that you might have only 10 parasites per unit, and we are way below the detection rate of the current PCR.

(Slide.)

With that, because a lot of this is old data, I threw in a couple of old references in case you are interested in looking at the original data. Thank you.

(Applause.)

DR. CHIODINI: Okay. Thank you very much indeed. You have partly I think answered at least one of the questions that is coming up. When I talk to virologists they say to me, "For goodness sake, Peter. Why don't you just do a PCR like we do for hepatitis C, and what's all the fuss about with this malaria?" So, Sanjai, would you like to tell us?

Prospects for DNA-Based Tests to Detect Malaria Infections

by Sanjai Kumar, PhD

DR. KUMAR: So getting right into this, you find from Dr. Nakhasi's talk that blood safety has so much improved for HIV, SCV, and more recently West Nile virus by introduction of DNA-based tests, and they have become the mainstay of donor blood safety for the viral infections. So why is such a test not available? I mean, if we can do the same thing for malaria parasites our problem will the solved. All those Mexican and Carribean travelers, they can all come back and donate blood.

So why is it not happening? I can tell you one thing for sure. This not for the lack of effort. The number of publications in malaria that describe DNA-based tests, so for routine diagnosis, for monitoring the efficacy of drug trials, for genotyping, for more of a population sort of a study and looking at the antigenic variations, and more recently for emerging vaccine efficacy. The --- parasite inoculations are coming ---, and how many blood form parasites are coming out. So the --- measuring efficacy of --- vaccines. So all of this is happening while we still don't have a DNA-based test to screen blood donors.

(Slide.)

Okay. So what are the challenges here? In the beginning Hira talked about that. I talked about that. Malaria parasites are highly

infectious. --- in 1943. So by induced infection to plasmodium vivax in human volunteers 10 blood form parasites were sufficient to cause violent infection, and they went on to write in that paper that conceivable a single malaria parasite is enough to cause --- infection. They --- believe that would not happen. In mouse malaria that had been shown. So that is the problem with one there. So imagine being singly infected --- present in a transfusion unit and how would one detect that.

Number two, window period. You cannot -- one cannot screen a blood donor a day after coming back from a foreign trip. So there is a window period to consider, at least 21 days in most cases, so that is another issue, and how the DNA tests are affecting testing that in non-immune travelers. I will talk about that.

So for me the biggest problems lies right here. The minimum parasite burden in asymptomatic donors is not known, and therefore we cannot set a sensitivity limit, sensitivity criteria of what one is looking for actually. The number of parasites that survive between the time of donation to transfusion is also not known. Just keep in mind that malaria parasites are highly --- inducible. --- parasite cultures as you know, it is very difficult to keep them alive, and how to detect a few parasites potentially present in a unit of blood. So it is more of a sampling issue.

(Slide.)

Okay. So there are a number of methods now. Some of them are reaching --- level. But if you look at them, they are all basically PCR-based DNA amplification method. The primary and nested PCR's as Marianna talked about. TaqMan is a real-time PCR, and microarray test, and I will show you a sample of all of those.

(Slide.)

So coming --- looking at the first ---. I would say that most groups, those who are looking at diagnosis, but not for genotyping, they use the --- gene as the primary target. The first --- genes --- by McCutchan in 1983, he is here among us in the audience. Then he went on to publish the first paper in 1989 with DNA detection using --- in the gene, and in that paper in 1989 he demonstrated they can detect 10 falciparum parasites in that test. So the sensitivities --- there defined 16 years ago, 17 years ago. This gene is probably one of the most suitable genes to target. There are seven to eight copies per genome present there, so that gives high amplification. The gene is very stable, always present --- genome and contains both conserved and semi-conserved regions. So that allows both --- species identification using this target.

(Slide.)

Okay. So I always like to show that of my own lab so they ---. This is a TaqMan assay. We can detect a single malaria parasite in one microliter of spiked normal blood.

(Slide.)

This is the microarray test again using --- gene. We can detect falciparum, vivax, malariae, and ovale, and again we can do a species differentiation. So TaqMan assay works, microarray works, others --- real-time PCR we have antibody sensitivity, which I will show you in a second.

(Slide.)

Okay. So now we are coming to reaching the sensitivity where it could be used in a meaningful way for donor blood staining. This is again our own data. In the whole blood ---. Others can do slightly better than this if you use the whole blood. If you just take spiked blood and boil it directly, so no fancy DNA --- involved here, you can detect 0.25 parasites per microliter. But if you --- it is 250 parasites --- of blood are potentially a little more than 100,000 parasites that could remain undetected in a unit of blood. If you

do a little more work here, if you do DNA isolation, you can detect two parasites in an ml of blood, but it is still that potentially --- 900 parasites undetected in a unit of blood.

(Slide.)

So what is this telling us here? Okay. So let's look at the slide and then I will talk about that. What others have done best in terms of sensitivity, in this paper in 1991 in drug efficacy trial in a PCR-based hybridization method that makes it a little bit more complicated and probably difficult for a blood screening, but they could detect 20 parasites per ml of blood. In a quantitative real-time PCR --- vaccine efficacy they can also detect 20 parasites in real-time PCR. I wonder if that probably is slightly more sensitive, but it was again a genome DNA extraction we can detect two parasites per ml here. So what is the problem now here?

(Slide.)

The problem is sampling now. No assay or no good assay should detect a parasite if the parasite is not present there. Importantly one parasite can cause --- in a unit of blood. How is one going to find that parasite? So the challenge is not in the sensitivity of these DNA tests. The challenge is how to fish out that parasite and detect it now. Okay. So we are looking at some issues regarding the -- how the DNA tests could be useful in screening blood donors who are naive blood donors, the ones Chris Ockenhouse was talking about again and again.

How can one detect those? It has been claimed that DNA tests, real-time PCR, can detect a parasite five days before microscopy can. So, and usually if you talk to people in the field they will say it takes about 11 to 14 days after --- before we can detect by microscopy. So potentially it is six or seven days after one can detect a parasite by these methods. So then it is reasonable to predict based on that that real-time PCR could be used to detect malaria parasites in the major of non-immune travelers two weeks after departing from an endemic area. I would say the majority of donors because then again the issue of latent --- parasites in --- vivax and ovale.

(Slide.)

But then again the question becomes more murky when one is talking the asymptomatic carriers here because simply we do not what is the lowest parasite burden. I think we will be setting -- if we set the criteria that we have to detect a single parasite in a unit of blood we are setting ourselves up --- criteria, but the problem remains what we are looking for also. No one has a good sense, and maybe somebody in the audience knows the answer. What is the minimum parasite burden? I am not talking about the maximum parasite burden. What is the minimum parasite burden in clinically-immune asymptomatic carriers? So those who were residents in endemic area and --- long-term residents. So where the solution might lie, maybe technology for parasite concentration. I mean, some of this has been seen recently in the literature because malaria parasite or because of characteristic of utilizing --- as a food source ---. So people have been trying to concentrate malaria parasites using magnetic ---. But I think there is a problem there also because in falciparum at least most of the transfusion
--- happen recently. One --- immature --- forms or living forms, and they don't carry much iron content there and not much of ---. So I don't know if that method would work there. Again, in a unit of blood if you can fish out a single parasite that remains to be seen. The technology needs to be developed, and then again I would --- again and
--- minimum parasite by the infected donor. To me that seems to be key now that could allow us to determine the --- sensitivity.

(Slide.)

I would just thank my collaborators here and stop.

(Applause.)

DR. CHIODINI: Well, thank you very much, Sanjai. Lots of points there that I think will cause discussion. So may I ask the presenters from this last session to join me at the front so we can now have our panel discussion please.

Session II Panel Discussion

DR. CHIODINI: It is probably easier if I do the chairing from here, and I think probably if I take alternate questions from each microphone that is probably the most equitable way to do it. So since there are two there I will take yours first.

DR. McCUTCHAN: Thank you. Tom McCutchan, NIH.

P. knowlesi I think is fairly clearly now established as a fifth human species, and it worries me that we are talking about four species when basically knowlesi could really get loose. I see John Barnwell on the panel, and -- or Sanjai or anybody, but knowlesi is important I think, and I think we should address it. Also there is another ovale. There are two ovales.

DR. BARNWELL: Yeah. Tom, knowlesi is potentially another human species. We know that a number of the simian malaria species will infect humans, and they probably do periodically. But if we are worried about blood donors from Borneo -- because what is going to have to happen for knowlesi to get out of its present ecology is for it to be able to go into another mosquito vector. So we should keep an eye on it, but I don't know if we really need worry about it at this time.

UNIDENTIFIED: With eco terrorism and everything it seems like a potential, and basically if you are worried about the blood bank you are not worried about mosquitos.

DR. BARNWELL: True. True, in that sense if you have a lot of eco terrorism in that part of the world, but I think presently if you are looking IFA tests for instance it is going to cross-react.

DR. KATZ: This is a really short question.

DR. CHIODINI: Could I ask you just to identify yourself because I'm an alien ---.

DR. KATZ: Yes. I'm from Iowa, Louie Katz.

(Laughter.)

UNIDENTIFIED: Where?

DR. KATZ: Marianna or Dr. Chiodini, I am very interested in the relative -- the range of sensitivities for the smear that I see published five to 500. Now I always thought that the five was in UK labs and the 500 was in my lab, but I saw you quote five as well.

(Laughter.)

DR. KATZ: I am just trying to put all the tests in perspective. Talk a little bit about the hands -- the eyes that look at the smears.

DR. CHIODINI: Marianna, do you want to go first?

MS. WILSON: Go right ahead.

DR. CHIODINI: Okay. Well, I think there is a range, and it is lab dependent. I mean, we have had a lot of discussion recently in another context, John Barnwell and myself, with microscopy versus malaria diagnostic tests, rapid diagnostic tests, and we came as a consensus with a reliable microscopy test with expertise should be able to get down to 50 parasites per microliter. Now when you put thick smears you know you put five microliters on, and so Marianna's figure of five parasites might be the very best microscopist will get there on a good day. That's the sort of sensitivity. Below that I think it is very iffy. If you reread the smears you might or might not pick them up again, but five parasites per microliter, I don't think many people would exceed that. That probably is around the limit actually.

DR. DAWSON: George Dawson, Abbott Laboratories. Enjoyed the presentations. Very nice. For Nigel Appleton I had a couple of questions. One is your assay format has the potential to detect IGM class antibodies and

thereby reducing the window. Do you have evidence that you detect IGM class antibodies in that window period?

MR. APPLETON: Yes, we have very limited evidence because with having no timed samples. We did purify the IGM from a few of what we thought might be fairly early samples, and run those, run the purified fractions in the test, and it is certainly somewhat positive. We have confirmed there is no IGG or IGA in those particular fractions, but that is all the evidence we got. It's not really scientific.

DR. DAWSON: Okay. Thank you. The second question is when you showed the detection sensitivity for plasmodium ovale and malariae, are they individuals who are solely infected with those plasmodium species, or could it be cross-reactivity due to previous exposure or co-infection with falciparum and vivax?

MR. APPLETON: It could indeed be cross-reactivity, or it could represent earlier infections with the other species. We have not sufficient data to be able to rule that out I'm afraid. No, that's all we can say. We don't have sufficient data to rule it out. There are just know to us as samples from patients with those particular infections.

DR. DAWSON: So you will be making efforts to try to find singly infected individuals or ---?

MR. APPLETON: Oh, yes. We always make an effort.

DR. DAWSON: Okay. Okay, and a question for Marianna Wilson is you mentioned in your presentation that the time between detection of antibody and parasitemia is two to six days or four to six days. What is the time between exposure to the parasite and parasitemia? So in other words, what is the total window between the time you are infected to the time you make antibodies? Because I would think that parasitemia phase is not going to happen immediately. There is going to be some lag time between the --

MS. WILSON: I'm going to pass that to John.

DR. BARNWELL: Oh, yes. That is going to be a varied by species, and it is going to vary by a lot of other factors. It is going to vary by the intrinsic incubation in the liver stage. It is going to vary by how many --- are released from the liver and therefore how low your infection is when it starts out and how long it is going to take until you get to the point where they were detecting a parasitemia. I could guess anywhere from in that type of situation you are talking in most cases anywhere from nine to 21 days or more.

DR. DAWSON: Okay. So then the total time between average time or range of time between exposure and antibodies would be nine to 21 days plus six days.

DR. BARNWELL: Plus, plus six.

DR. DAWSON: So essential 30 days you should be able to in most cases that you know about, there should be an antibody response for a person infected.

DR. BARNWELL: Yes, hypothetically.

DR. DAWSON: Hypothetically, and are these all based on -- have you done animal studies and human observation cases? Or is it just the mix of things, or is -- does your data come from one set or another? Does it come from experimental infections where you know the time of injection, or do you do from some kind of, you know, ---?

DR. BARNWELL: It comes from a wide variety of data, including experimental infections and patients. A lot of Marianna's came from experimental infections, and also knowing, you know, patients that you have a fair amount of data about.

DR. DAWSON: Good. Thank you.

DR. SUBIANCO: I'm Sal Subianco from America's Blood Centers. I want to jump the gun a little bit in terms of this meeting and ask you, the test

experts, what would you expect in a test for blood donors, not for diagnosis, that we should be using? That is, what would you want from that test?

DR. CHIODINI: I will have a go at that first, and then we will see what the rest of the panel say. Okay. What we are trying to do is we are trying pick out those who might be asymptomatic but parasitemic. That predominantly is going to be the semi-immune donor. I mean, Chris Ockenhouse's was well made earlier about the traveler who gets their first attack of malaria. Apart from a window period when there is about seven days where they can have parasites in their blood roughly before they get symptoms, asexual parasites, they will be symptomatic. So that situation you are looking for the semi-immune donor. Either a person who has gone from a non-endemic area to live for a long time in the tropics or was brought up and lived there.

Therefore, what you want is something that is going to pick up antibody purely as a surrogate marker that lots of antibody would indicate that there is a risk that they might have parasites. We know that many of those with antibody won't have parasites. That in a sense doesn't matter because it gives you a margin of safety. So you want to pick up anybody who is able to be walking around, get through a donor session, answer the questions about health, say they are perfectly well, and be allowed to donate who could still have parasites in their blood. A good marker for that is antibodies, and I have got a presentation later that perhaps will give some more evidence to that.

As sensitive as possible, but also specific. You don't want -- I mean, cross-reactive for babesiosis. We see a few. We don't see many in Britain. Obviously you see more here. So you don't want it to be compounded by other possibilities and exclude donors unnecessarily, but sensitive and specific for antibody would be my criteria. The NAT debate will come in a moment I am sure, so let's open it up to the rest of the panel.

MR. APPLETON: Well, I'm very lucky in this position because all I have to do is to produce a test that satisfies my customer. If the customer says, "That's good for me to use," I'm happy. If they say, "I want to increase the sensitivity of specificity," then I have to work. But as long as they are happy, I'm happy.

(Laughter.)

DR. SUBIANCO: Dr. Nakhasi just asked me if I'm happy.

(Laughter.)

DR. CHIODINI: You are allowed a supplementary. When the question the British Prime Minister they are allowed one supplementary, so you are allowed a supplementary.

(Laughter.)

DR. SUBIANCO: No, I'm not happy yet, because I think that I just wanted to call the attention that we are trying with questions to identify donors that even you with all the expertise and all the technology that you have can't identify in terms of risks. There are misses in all your systems, the same way that we have misses in medical history. Just by attempting to make them more restrictive, more restrictive, we may not come to that level of sensitivity that we would love to have on a theoretical basis.

DR. CHIODINI: Okay. I'm going to take leave to disagree with you on that if I may and to say that really I think the first thing we need to do is keep it simple. I have heard a lot of discussion about risk and within country risk and so on. I think the more complicated a strategy, the more likely the risk for human error surrounding it.

So what we are saying is that it should be multi-layered. So you have history taking. We know there are imperfections both in individual face-to-face clinical practice and in donor sessions. Secondly, time exclusion, and again that depends on an accurate history. Thirdly, geographic history, which again not everybody knows which tropical countries have malaria or even necessarily have an understanding that the country they have been to is in the

continent of Africa for example. So you do have to be careful on that. Then finally, laboratory assays like antibody detection, which really rely upon another is yet to be debated to give you that multi-layered approach. That way you are minimizing the chance of error.

I think to expect no error is unfortunately without -- unless you don't transfuse at all it is not possible. But I do think we can minimize error, and what we are hearing here is very critical appraisal of the tests saying, well, it doesn't quite detect or it doesn't quite detect that, and how long before the window period has ended. Those can all be addressed, and I think it is right to be critical, but these criticisms should give the impression that things aren't safe. It is already very safe without antibody testing, and it could be much safer when you -- well, when and if you add antibody testing. Are there other comments?

DR. BARNWELL: I think what you are asking is it depends on whether you want a direct test or you want an indirect test. The antibody is an indirect test. It is going to rule out more people than have actual infections, and I think that with a little work that you can get down to the sensitivities that the customers are looking for. I don't think they are there quite yet in some of the species.

The direct tests I think Sanjai raised the question is what we don't know is what is the level -- in all these donors that have given transfusion malaria, what is the level of parasitemia that we need to detect to be sure that we are not going to -- and I don't think we know that, because by definition they are not patent and we don't know what their level is.

DR. CHIODINI: Okay. Good. I think probably we will just move it on a bit and take a question now from the microphone over there, and then we will come back to you in a moment.

DR. GORLIN: Sensitivity -- Gorlin, Minnesota. Sensitivity and specificity are great, but what we know from the HIV test is that positive predictive value is also important in the blood donor population. To whit, the HIV antibody test is great with greater than 99 percent specificity, but at the Red Door Clinic, our STD clinic, its positive predictive value is greater than 99 percent. Fifteen blocks down the street at our blood donor center it is less than one percent. So a question for our friends in the UK is now that you have experience in blood donors, what is its positive predictive value in people that have never left the UK?

DR. CHIODINI: Okay. I will show you data later on for that. The first generation assays, the positive duration tropical area donors were 0.45 percent. In the latest assay the work we did there were no positives in non-tropical area donors, but I will show the data after lunch.

DR. NAKHASI: Hira Nakhasi, from FDA. I just have a couple of questions for Marianna. You mentioned that the window -- you know, the serological tests, IFA tests, you see parasitemia preceding four to six days. My question is you also tested them later on with PCR. How much can you narrow down that window period there?

MS. WILSON: We have not had the opportunity to do that with our PCR. Those experimental infections were done in the '60s.

DR. NAKHASI: Oh, okay. So did you -- and also one of the charts you showed that there were IFA in the '63 to '98 there were 71 percent serological positive. Did you go back on some of the samples and test with PCR and could see? No?

MS. WILSON: No, because we didn't get the --- blood at the time. We only got serum specimens.

DR. NAKHASI: And lastly, the question I have is regarding this. I am little bit perturbed by the fact that sometimes you see this IFA positive, PCR positive, but not sometimes. Is that because there is some change in the

parasite mutations are occurring? That the PCR technology, the primers you are using are not picking them up or something?

MS. WILSON: No. It is most likely due to the number of parasites that are there. The test is not sensitive enough to pick up the parasites.

DR. NAKHASI: Oh. So it is the number of parasites, load, that in -- let's say for example the 1996 sample you show IFA positive, a donor I guess it is, and PCR positive. The number of parasites are much higher there?

MS. WILSON: Yes, because you could see them on the blood film.

DR. NAKHASI: ---?

MS. WILSON: Yes, much higher.

DR. NAKHASI: But then 2002, malaria, --- 10, and you know you still didn't pick up the PCR. Okay. All right.

DR. CHIODINI: Do you want to just interject there, Monica?

DR. PARISE: There is just one limitation I wanted to put on the data that we have from the US data, is that these test often aren't done in real time. You know, this donor passed on this infection and then we hear about it, you know, maybe a month or two later. I mean, the patient has to get sick, it has to go through the system. So then sometimes we do those. You know, we don't always have a sample from the time of donation, so you are dealing with stuff two months later, which is also another factor why they could be negative. We don't know what happens in real time.

DR. CHIODINI: And parasitemia can fluctuate.

DR. PARISE: Yes.

DR. CHIODINI: If you did an NAT test on somebody with -- a semi-immune with falciparum at weekly intervals you might get positive, negative, positive, and the parasitemia does fluctuate. It sequesters remember, falciparum. So when you are near the limit of sensitivity of the assay it is quite possible, the cutoff point, to get in sequential samples from the individual of positive, negative, positive. That reflects that you are getting close to the limit of the sensitivity of that particular assay, and I think a lot of the NAT discussion here really centers are sensitivity, how low can we go in relation to how many parasites in the bag of blood.

DR. NAKHASI: No, I was -- that was a good point, but I was trying to go in the direction that if there was an NAT test and from the time of infection, can you get the antibody response? Would it be appropriate to use that, because you should be seeing parasites first and then antibody response.

DR. CHIODINI: Well, that was the interesting thing about the French airport malaria reported in 2001 when they got hold of the unit. It was PCR-positive, but the person was asymptomatic. They were in that window period, and I think from vaccine studies NAT's have been positive five to seven days before appearance of blood films. So there is a window there of about five to seven days where it can be detectable by NAT but not by blood film, and then of course they will reach the disease threshold.

DR. HOLMBERG: Jerry Holmberg, Health and Human Services. I was the one playing the Mozart there, so --

(Laughter.)

DR. HOLMBERG: I get fined. I have a followup question and then another question. Actually a followup comment really and a question in regards to Dr. Bianco's comment about what would the panel be happy with as far as donor screening. Dr. Chiodini, you mentioned that you would like to -- that it is basically a surrogate. So from what I am implying or what I hear you saying then is that it really should be a test that has both capability of IGM and IGG. Correct? Is that -- would that be a correct assumption?

DR. CHIODINI: It would depend how you wish to deploy it. The way we deploy it is with a window of at least six months after the last possible exposure to malaria or after last illness that night have been malaria, by which time we essentially would be okay with an IGG assay, but it has already been

indicated. The assay which we actually use is capable of detecting all classes. So we would be happy with an IGG assay, but in fact the assay we are using as Nigel has described detects the other classes anyway.

DR. HOLMBERG: Okay, and then Mr. Appleton. You commented that what you had a problem with, not having a panel available, a sampling panel available. Is this widespread throughout the world? Is there a panel available from CDC? So this is a deficiency across the ---

MR. APPLETON: Not as far as I know. No, apparently. Yes, it seems to be common throughout the world. I mean, I don't know what studies have been done over the years on the course, the time course of malaria, but I haven't been able to find anybody who has got nice freezers full of longitudinal serolized samples at all unfortunately. Perhaps it is too common a disease in the places where it is common for them to bother.

DR. KUMAR: --- the samples they collect is too little volume for laboratories to ---, but people do publish sometimes hundreds of samples. Looking at the evolution of natural immunity and stuff like that. I just don't know how hard it would be just to --- it back with those studies and generate samples. Yes, go ahead.

DR. EPSTEIN: Yes. I think it has been made clear that blood can be infectious before any of the available assays is positive. I am just wondering since it has been pointed out that animal species can be infected and that P. falciparum cultivable, is a human experiment feasible in which you could infect a human volunteer and follow the course of blood infectivity with a bioassay? In other words, infecting an animal or doing some other in vitro cultivation with -- you know, and point dilution titration to figure out titer and answer this question of how long can there be infectivity in the asymptomatic individual and at what titer. In other words, are bioassays available that could answer that question, and is that a feasible experiment?

DR. CHIODINI: John, do you want to take that?

DR. BARNWELL: I don't human experimental infections are possible anymore under those conditions. Ethically it is not possible, and the animal models really aren't going -- they are models, and therefore I don't think they are going to reliably mimic exactly what goes on in humans.

DR. EPSTEIN: I haven't said an animal model. What I have said is an animal bioassay for the infectivity in human blood. In other words, can you titrate the infectivity in human blood by inoculating an animal?

DR. BARNWELL: Yes, you can. But I think if you go back in the literature you are going to find a lot of these studies have already been done to a certain extent, and you can subinoculate from humans into animals or other humans. They have been done since the '20s, '30s, '40s and '50s. You can find it in the '60s. You can find it in the literature, and -- you know, you can subinoculate from these individuals and produce a bioassay, essentially produce a malaria infection, and you can produce it when the levels are so low that you can't find them in the blood by the current assays. But I don't understand why you want to --?

DR. EPSTEIN: Well, because the question is what is the parasite burden in those subclinical infections, and if you have a bioassay then in principle you can also titrate.

DR. BARNWELL: Sure you can.

DR. KUMAR: Maybe, John, I can. Maybe I can comment a little bit further there. I think a person who can address that most efficiently though --- Ockenhouse. They have the access to these --- challenge experiments in humans in looking at drugs and vaccines, so --.

DR. OCKENHOUSE: So we do. We have done over 500 human challenges over the last 15 years. These are in the context of drug and vaccine in non-immune individuals who come in, and the experiment is fairly simple. We challenge them with P. falciparum sporozoites. Only P. falciparum now, and

sporozoites from mosquito, and we follow them every single day. Now if you know the biology of the malaria parasite, the malaria parasite has to reside in the liver for the first five to six days before it emerges into the blood. It is in the liver it is totally asymptomatic. So you can transfuse someone in the first couple of days and they are not going to get malaria because the parasite is not in the blood. So we follow these individuals, and we don't follow them because we are interested in the natural history, although as a scientist of course you try to learn, but we want to see if they become positive by the standard gold standard, which is a thick and thin smear.

Now we would have as the military has learned over the last five years that that is a very, very poor gold standard. It is absolutely the training of reading smears. This is diagnosis. Now let's put that aside, but we have experts that we take blood, a very small aliquot of blood, maybe a drop or a couple of drops ml. And we make a thick smear every single day, and we can monitor when they become slide positive. All right? From the time of exposure of a mosquito to the time they are positive in the smear. When they are positive in the smear we treat them, totally, and we abolish.

Now that period of time between exposure of the mosquito and the time they are smear-positive in the model that we use is about nine to 11 to 12 days. It varies. Okay? Now if you say that the parasite is in the liver for the first five days then it is in the blood between day five to the time we find it on a thick smear. Okay.

So are these individuals asymptomatic? In fact, about half the individuals by day nine are totally asymptomatic. They means they have malaria parasites in their blood, but they feel well. But we are able to say on a thick smear we found the malaria parasite. So what is the level of malaria parasite? About one parasite per microliter. So that would be 1,000 per ml, and figure out how many in a unit of blood. That is a lot of parasites. So asymptomatic. All right?

Now, let's say though -- and this is where it becomes -- that is for diagnosis. You can develop. These are -- nucleic acid tests ought to be great for diagnosis. I believe we are going to have FDA tests in the near future. The issue becomes, and this is it, what level of parasitemia in a unit of blood is sufficient to cause an infection in the recipient.

Now, you know, if you have one viable parasite that can multiply every 48 hours, that is -- and so what are you going to test? If you are a blood banker, what is the volume of whole cells? It is not virus. We are not testing plasma. What is the volume that you need to test? So this is where it becomes a technical obstacle to overcome, and what, you know -- so that is the challenge. The other half of the individuals are symptomatic, very low-grade fever, backache, myalgias.

DR. DAWSON: Could I ask a question about that study you just described? Do you do antibody testing on those experimentally infective people, and how uniform is the antibody ---?

DR. OCKENHOUSE: Oh, yes. Right, right. Yes, absolutely. Now this is where it is different than the CDC. In the CDC, what happens in those cases is those are symptomatic individuals. People who come in with clinical malaria are going to have very high levels of parasites. They are usually in a clinician probably on the low end a hundred, on the high end a couple thousand parasites per microliter. Of course you are going to see antibodies a couple of days later because the antigenic load is very high.

In our individuals we also collect blood during the course and after treatment. I have never seen a positive -- I have looked at 75, so there are a number that N equals 75. The earliest that I see antibodies is approximately seven days after the onset of treatment. Okay?

So we have the individuals come in, one parasite per microliter. We treat them. We follow them. Seven days later, bingo, antibodies, and occur in

most all the individuals. And it is true they will go up, but because they were treated at such a very low density the half-lives of antibodies come down.

So you could actually have, you know -- and it is unlike Nigerian example, because those individuals from Nigeria, Viet Nam, probably had malaria growing up and they were boosted, and then they have a persistent antibody response.

DR. DAWSON: Once you get an antibody response during the ramp up of the parasitemia, you are not going to see it. But once you see antibodies, do the antibodies persist? And this is for the general panel. Do the antibodies persist during the total parasitemia that is observed in an infected person?

In other words, is antibody a good marker for once a ramp up has occurred will you detect after that ramp up all the people who would be parasitemic, assuming you have got the right antigens? Is a antibody test a good thing?

I am asking as Abbott Laboratory's representative. Is an antibody test a good thing to detect parasitemia with the exception in that early ramp up phase? So I would like to hear opinions about that. Okay? And I know you may need to -- you don't have everything you need perhaps for ovale and malariae, but for falciparum certainly it looks like you are detecting, you know, 99 percent. Is that in general true of, you know, the infection?

MS. WILSON: As far as I know, the only data really on antibody persistence is the data that I showed today, and that data unfortunately was never published. So that is about all the data we have got. Certainly somebody who is parasitemic, particularly like a Nigerian or somebody who is going to remain antibody-positive for a very long time and will maintain antibodies as long as he has the parasitemia.

DR. DAWSON: And even when the PCR result would fall below detection the antibodies should maintain their reactivity.

MS. WILSON: Yes, and that is how in our donor, in our infected donors, that is how we pick out the infected donors. The antibody is still positive. The PCR is not though.

DR. CHIODINI: Yes, that is the critical point. It is a very good marker for those who might still, not necessary do, but might still be carrying them.

DR. KUMAR: I would like to add to that. I don't think anyone is saying now that the antibody is not a good tool to detect those infected donors from who are being asymptomatic carries. Especially those who have low-grade parasitemia, because those parasitemias --- low antigenic stimuli. They will keep the antibody low boostered and at the same time keep them asymptomatic also. The day parasites are clear the antibodies will drop. So for a very --- test I think it is a bonus from two sides. I mean, when the antibody drops those get reentry, and if the antibodies are present that means the person, even conceivably after some
--- non-endemic area, antibodies, there is a good chance that parasites are still present there. What we are not addressing --- enough now, and I don't know if people have
--- to ask the question. What will the sensitivity of these antibody tests to detect primary infections in travelers, and I think that is where it might get tricky. Because I think even if most laboratory tests will not --- sensitivity and specificity will pick up most of the -- I mean, any --- probably can develop --- and within a few --- that will detect close to 100 percent of the --- come from endemic areas --- stimulation. But it will get tricky to establish sensitivity in non-immune travelers who have seen a one-time infection while the infection is still rising.

UNIDENTIFIED: Along those same lines, if you have these ELISA tests which are picking up on these people that are coming in from these foreign countries, and even though they have been treated, they are going to retain a

fairly high level on the EIA. Would you then defer these people? I mean, because if you can't rely on the NAT test, because the NAT test is going to be positive/negative based on the paroxysms. You know, so where would you -- I mean, are you going to rely strictly on the EIA?

DR. KUMAR: Well, I guess the data is really limited on ---. I mean, they have the experience for this --- the problem, the number of sero positivity in at-risk donors is less than four percent. So, I mean, if somebody gives you 96 percent on donors who have been deferred anyway and you are losing only four percent I will take that assay anytime.

DR. CHIODINI: That is your insurance policy. A lot of those four percent, in fact it is down to nearer two now as it got running further on, we know don't have parasites in their blood, but we know that if we exclude them we have got that margin of safety. We are very worried about NAT and felt that because literally one parasite in a bag of blood is theoretically able to transmit an infection that we are not prepared to go with it on that basis. We can't put our hands on our heart and say to a recipient there is no malaria in this bag because your NAT is negative, and that is the critical point with the NAT's. So the positivity rate as Sanjai said is not high enough for us to be wasting a lot of donors. We know we waste some, but we get an awful lot back.

UNIDENTIFIED: Right. Well, in the case like a vivax donor where they may have dormancy in the liver, that is the concern there. Because you may have their EIA may be elevated. I don't know what your studies show on that. But the dormancy, and then all of a sudden the time they come in to donate there may be some that are just getting out in the blood, but they still haven't gone into the temperature range yet.

DR. CHIODINI: Yes. I mean, that is what John was talking about. Do you want to elaborate on that a bit further, John, the point you made before? --- I think.

DR. BARNWELL: Yes, and I think that is probably the main concern. I think the point of it is that in most of these cases what you are trying to do is to save a large proportion of what would normally be deferred, get them back into the system. So you are going to have to take some losses on these, and I think that those are going to be the smaller numbers.

DR. McCUTCHAN: I am not sure how important this is, but one of the questions that seems to be of importance is how sensitive we need to be to detect asymptomatic carriers. There was sort of a comparison with the trials Chris is doing, and I wonder if there is any relationship between what being asymptomatic, a naive person being asymptomatic during a trial and the question really being asked, how sensitive do we need to detect carriers. Whether there is any, if that isn't apples and oranges.

DR. KUMAR: Tom, maybe I would like to address that, at least my ---. I do not think we need to worry about how early. If you are talking about DNA tests, or even antibody tests, any test can wait for a few further days and a naive person if he is going to come up like --- which has been hit by --- strains of plasmodium, it doesn't become positive by day six. It is not a diagnostic test. It is not a test that will be used to treat people. It --- pass on day six when you can pass to day nine or 12 or day 15, and I think that is sort of where it can be always allowed. So the sensitivity in terms of window --- I do not see as an issue.

DR. CHIODINI: It is also being done in with the time exclusions. I think it is back to Chris Ockenhouse's point that most of the falciparum in the non-immunes are in the first, what, month. So if you have got a period of time, say six months before you offer them an antibody test, they have either had or pretty well not going to get malaria if they are non-immune. The vivax is a caveat to that, but that would also address that point.

UNIDENTIFIED: Terrific.

DR. NAKHASI: Hira Nakhasi, FDA. You know, because in order to get -- I think there is a lot of discussion and confusion going on here, when the antibody comes, when the PCR comes up. I think I would like to request Dr. Chris Ockenhouse here if he can make a chart here for example when you have the person getting infected; and he pointed out to us that there is a five, six days they remain inside the liver and then they come out, and after four or five days after they come the antibody comes out. Is there some kind of a graph you can show us when the PCR positivity comes up, when the antibody positivity comes up. Then it can give a perspective to the people how, which test and how sensitive a test is needed.

DR. CHIODINI: I think that is a very useful point, but you must also I think add to that equation a time exclusion. I don't think anybody is even beginning to consider that you are going to do them in the first month when that is the time that this would all be happening, or even in the first three months. You are talking longer, when that has all happened. So we are not talking about trying to get donors in the first month when they will be parasitemic but asymptomatic. That would be an act of great folly. We are talking about waiting I think a period of time to enable those who unfortunately are going to get malaria to have got it and declared it before you then test them. But certainly Chris' data is very interesting and I think should be, if he agrees, sort of summarized for the meeting in some form. I think that would very valuable absolutely. Yes, I agree.

(Multiple conversations.)

DR. LEIBY: Peter, David Leiby, the Red Cross. Maybe you will cover this this afternoon, but I have heard you mention in the last few minutes that you had some antibody-positives that didn't have parasitemia. We just heard that there were issues with nucleic acid testing for obvious reasons because of the waning of the infection or difficulties in detecting low levels of infection. How confident are you that individuals who have antibodies do not have an underlying parasitemia?

DR. CHIODINI: Okay. I mean, the direct answer to your question, Alan Kitchen*, whom I think you know quite well, is about to do this with us. We are going to get all the antibody-positive donors that are currently deferred and put those, their samples, through nucleic acid testing to answer that question. Certainly if someone like me who goes to the tropics but does not have malaria, if I had had it before, one attack, my titers would be quite low and I would be pretty confident in that situation. I am very unlikely because I am non-immune to be harboring parasites, though I have got a little bit of antibody. Those who have got very high antibody levels I think are much more likely to be parasitemic, the semi-immune donors from the tropics. But the direct answer of how many of the travelers are parasitemic and tropic area donors are -- sorry, the semi-immunes are parasitemic and antibody-positive we will answer directly by doing that experiment. We don't yet know.

DR. LEIBY: The real answer is we don't know.

DR. CHIODINI: No, that has got to be done either in your own healthcare system as well as in ours actually.

DR. LEIBY: Oh, absolutely.

DR. CHIODINI: What we are doing is we are saying if you have got antibody you are out, but we know that at least -- well, as far as you can do it with NAT you will say it. At the threshold of NAT they all are not positive. You then have a debate about what happens below the threshold.

DR. LEIBY: As you know, in many parasitic systems the presence of antibody is actually indicative of the parasite still being there. Many years ago back when I was doing Leishmania work at Walter Reed one of the underlying thoughts was that Leishmania infections actually cleared, and Hira can probably state this as well, that Leishmania provided sterile immunity. But then there was some rather novel experiments in which they took mice that they thought they

could not detect the parasite in, could not show the parasite. When the immuno-suppressed the mouse, just like you would see in HIV infections today, you saw a blossoming of the infection and a return.

DR. CHIODINI: Yes. We certainly see that in our Leishmania cases, yes.

MS. WILSON: I would like to just -- David, the slide I showed on antibody persistence was post-treatment and these patients were followed for some of them up to 18 months afterwards and certainly had no signs of clinical malaria at all. So we assumed -- it is an assumption. We assumed that they were cured at that point, but the antibody levels remained high.

DR. LEIBY: Well, that was precisely the point with those previous studies of Leishmania. Everyone always assumed that they hadn't cleared the infection. There was no clinical disease. There was no signs or lymph nodes of infection, but the parasite had been sequestered in locations where you could not identify it. So then we you treated and immuno-suppressed them, you got the infection back. The same way with toxiplasmosis* and a whole series of other infections which we continue to see once an individual becomes immuno-suppressed, and I propose that perhaps they are sequestered somewhere where we can't detect them.

DR. KUMAR: Well, one thing we already know is for radical cure to malaria how soon antibodies will drop. There is definitely not a precipitous drop. So one thing we know for sure is that if radical cure of malaria parasites lead to loss of immunity very soon in --- studies once you treat the -- once you give radical cure to adults, and I am talking about adults, clinically immune adults, within two months they all become positive in endemic areas. So it is the presence of that parasite that maintained their immunity, and also it --- immunity --- stages. The immunity --- the blood is --- immunity that is there. So parasites are gone and immunity is gone as well.

DR. CHIODINI: I think the analogy perhaps between the Leishmania and --- to that as well. Toxiplasma* is slightly different from malaria. Once it is out of the liver you are essentially dealing with erythrocytic* stages only, and I think that is perhaps not the best analogy to go for the Leishmania immuno-suppression model in this context. I think particularly with modern radical cure for malaria it would be very unlikely that if they had been adequately treated the parasitemia would remain dormant for some considerable time. Okay. Yes?

DR. GRESENS: Hi. Chris Gresens, Blood Source, Sacramento, California. I am finding this exchange of information fascinating and I appreciated the recent discussion about implications to transfusion recipient safety.

I would like to go back for a moment to donor resources and ask you please, Peter, in your experience in the UK allowing at-risk donors after six months who test antibody-negative to reenter the donor pool. Have you done any comparisons as to how many donors come back under this new system versus what used to happen with the one-year/three-year deferrals? I would be very interested to see what impact this might have.

DR. CHIODINI: Yes. So we did ask at one point, you know, if there was information on that, and unfortunately I don't have figures for you. That actually would be a very important thing to have. All that we do get from the donor center is that they say, first of all, donors are somewhat aggrieved to be deferred and, secondly, very often don't return. But that is very anecdotal, and I am sorry to say I am unable to produce figures to that, but it is not for want of trying. Okay. Any more questions?

(No response.)

DR. CHIODINI: We are pretty close to the lunch break. So I think unless there are any, we have had a very long and very vigorous discussion, I will thank all the speakers very much and we will adjourn for lunch. Thank you.

(Applause.)

DR. KUMAR: Before we break, I think there could be a little bit of a snafu. We were supposed to eat in the cafeteria upstairs, but there are bigger forces working here. Directors ---, so they have closed down this cafeteria here. I just heard that, but I haven't seen that with my own eyes. So if that is the case we will need to cross the street and go to the ---. That is just up the hill like three minutes walk from here. So may need to do that, and we will definitely --- everybody is back here for the third session by 1:30.

(Whereupon, the luncheon recess was taken at 12:16 p.m.)

A F T E R N O O N S E S S I O N

(1:30 p.m.)

SESSION III: PERSPECTIVES ON TESTING FOR MALARIA INFECTIONS IN BLOOD DONORS

Roger Dodd, PhD, Session Chair

DR. DODD: I would like to get started again. We have done a pretty good job of keeping on time. Let's hope I can keep it up. I an Roger Dodd. I am from the Red Cross, but don't hold that against me.

(Laughter.)

Sanjai asked for just a moment to make a couple of housekeeping announcements.

DR. KUMAR: Well, I just have two. I hope you all survived the debacle regarding the cafeteria. Those who need any taxi or any --- please let --- know at the registration desk to make sure she can call up now, and please don't forget the evaluation forms. It is part of your packet. If you could --- leave it with one of us when you leave that would be appreciated.

DR. DODD: Thanks, Sanjai. We are going to get down to the real questions now. We have a session on perspectives on testing for malaria infections in blood donors, and we will hear experience from a number of parts of the world and some quite brand new data that I think is going to interest you and some --- approaches to the issue of dealing with malaria potential infectivity in donor. I would like to go right ahead and ask Peter Chiodini to tell us about the United Kingdom experience. Peter.

United Kingdom Experience Regarding Malaria Antibody Tests and Their Contribution to Blood Safety

by Peter Chiodini, MBBS, PhD

DR. CHIODINI: Thank you very much, Roger. I am essentially going to share with you the experience we have had in determining our policies for screening donors for malaria infection. It may or may not be suitable for your own healthcare system. This is simply our experience that we hope you may draw something from. It is certainly not indicated as a way that you should perhaps proceed.

(Slide.)

Now there are very few things that Britain has more of than the United States, but the depressing fact is we have a lot more malaria than you do. Despite having a quarter of your population, in absolute terms we have about twice as many malaria cases returning to the UK as you have reported in the United States. As you can see, it is predominantly a problem with plasmodium falciparum. Until 1986 we had more vivax than falciparum, and the non-falciparum species predominated. But there has been this inexorable rise in falciparum over that period, so that in 2005 and similarly in previous years about three-fourths of the cases have been plasmodium falciparum.

(Slide.)

Now these are the 2005 figures in a bit more detail, the lowest ever total of vivax since we began keeping this data set in 1977. That has been quite a dramatic change for us. The other is ovale predominantly, African one, and malariae, but really you are talking falciparum with a minority presence from the other species.

(Slide.)

Where do they come from? Well, west Africa predominantly for the falciparum. This is a 2005 data set by country, geographic area. There is --- falciparum of those cases of the 1338 to take off 300 where it was given. You are dealing with a vast number from west Africa, and if you look at the malariae and ovale it is a similar sort of picture, west Africa. Obviously some from east or southern, and then with vivax in contrast it is a problem of people returning from Asia.

(Slide.)

There are also the group we have already heard about with VFR, visiting family in country or friends in country of origin. It predominates again with all the species really, but particularly with the falciparum. Again, visiting family usually in Africa.

(Slide.)

And in terms of ethnicity the depressing fact is it is the black African population that suffers the bulk of the malaria cases. In terms of ethnicity, African or Asian descent also accounts for a number of cases. These are a neglected group. It is another talk, but a neglected group in terms of access to preventive measures.

(Slide.)

So who then are the risk groups for transfusion-transmitted malaria? First of all the travelers. We have heard a lot about those this morning in debating what their risk was. These are people who have almost no immunity to malaria or none at all, and that is always going to be symptomatic if parasites are present and that is excluded from donation, provided of course you wait long enough and you are not worried about that one-week window period when they are out of the liver and in the blood before you get a symptomatic threshold. Almost all of the falciparum in this group is over and done with in the first two months, and there are virtually none presenting after six.

(Slide.)

Residents in contrast are a very different group. Almost all brought up in sub-Saharan Africa in terms of blood transfusion risk. They are still partially immune to malaria parasites; and thus may be asymptomatic, but parasitemic, and may harbor P. falciparum for some years.

(Slide.)

I am going to take you through five cases of transfusion transmitted malaria that we had in the UK from '86 to 2006. They were all due P. falciparum. The first donor was a semi-immune European, a white missionary in fact who had spent 10 years in Africa. Long enough clearly to become semi immune, but it was a fairly of history taking at the donor session. The history of travel was not given, and thus the person was allowed to donate. Retrospectively looking back, the blood film was negative, but crucially the indirect florescent antibody test was strongly positive. So here is one that would have been excluded had we had an antibody test in operation and a malaria risk had been identified.

The second was a west African. The blood was collected for plasma usage in those days. This was the mid '80s. We were allowed to collect the blood and use it for plasma fractionation even if there was a malaria risk. That is no longer the case. But it was before there was a proper computerized system, and as a result an error was made and the unit was issued as whole blood. It had been stored for 19 days. It reinforces the point made this morning about days of storage and thus produce transfusion-transmitted malaria, which was -- and I looked after the recipient with one of my colleagues, not noticed until the oncologist looking for leukemic blasts found 50 percent parasitemia in the red cells. But there had been parasites present for some days. It goes to show that you find what you are looking for. But again, had we had an antibody test in operation that would have been presented because the patient was strongly sero-positive.

(Slide.)

Another west African donated two months after a visit to that particular geographic area. Travel history not elicited, but again IFAT by that time and ELISA was available. It was also positive.

Here is an interesting one. African donated two-years-and-ll-months after travel. At this stage our exclusion period for residence was very similar to yours of three years, but I would take some convincing that the parasites would have decided to disappear when it is actually three years were up.

(Laughter.)

As a result there was a transfusion-transmitted case, and both the ELISA and IFAT were positive, strongly positive on donor serum. Following that three-year episode, the deferral period was increased to five years.

So in the last case, this west African donor was perfectly allowed to donate. They fulfilled all the criteria. Back for five years as a residence, thus you can donate, and the eight-year travel history was checked with the donor at interview later and was confirmed at eight years. IFAT and ELISA positive and PCR positive on this particular one. The previous four were not running PCR at that particular time. So my case really is all five would have been prevented had we had an antibody test and it had been deployed.

(Slide.)

So our risk factors to transfusion-transmitted malaria in the UK are failure of history taking, and that is always going to be a risk whatever you have, and tightening up on history taking has to be done whether or not any healthcare system implements antibody testing. Administrative error. Again, human beings involved. Less likely now we are fully computerized, but theoretically still possible.

But the big group of concern, the semi-immune individuals. They are not looked after well in terms of prevention, but also are not necessarily picked up subsequently. So they can be asymptomatic but parasitemic, both long-term residences, such as Peace Corp for example, or of course long-term expatriates or indeed residents.

(Slide.)

So what about our strategy? These data that have just shown inform how we developed our strategy. The first thing we say is that complete

prevention may not be possible, I think is not possible with current methodology. We have to acknowledge that risk and work with it. I think it is unhelpful to think that we are going to eliminate risk. What we are talking about is minimizing it. So with the aim of minimizing it we then want to also not exclude potential donors unnecessarily, and donors are very precious to us. We are short of them.

(Slide.)

So the triad then, history taking, time exclusion, and screening donor serum for antimalarial antibodies. You will note that PCR does not appear here. As you will have gathered this morning, I don't think it is sufficiently sensitive and I think its use could actually confuse things.

(Slide.)

History, we are not going to spend time on this, geographic location, length of time, et cetera, and history of past malaria. All the kind of obvious questions you would need to assess malarial risk.

(Slide.)

There is a problem with history, and I was trying to summarize this. --- Slinger and colleagues from Canada had summarized it much better than I could by bullet pointing it. So I just invite you to read this for a moment, and that I think makes the point that no matter how good the history taker you have got a non-medical donor who is under stress probably. They are waiting to be bled. You may not necessarily get an accurate history of malaria exposure given all the other questions that they are asked to answer.

(Slide.)

Now time exclusion. This is our data from 2005. It would be typical for the last few years. Going back to Chris Ockenhouse's point, most of the falciparum is out of the way in the first month. These would be the non-immune travelers, and by one to 5.9 months inclusive we got nearly 99 percent of them have been clinically unwell and have been diagnosed. There is a --- even at 11.9 months, so there are still a few that haven't presented. These will be the semi-immunes. There is a longer --- for vivax and ovale which have ---. So even by 5.9 months there are still only three-quarters of them having had their malaria and had it diagnosed. That will always be a problem. It is inbuilt into the life cycle. After --- discussion I wish I had put my life cycle slide in, but I left it out for the sake of time. The --- can you give you a delayed primary attack nine months later quite commonly and in some cases over a year after leaving the endemic area. That will always be a risk which we have to acknowledge.

The P. Malariae are all out of the way by 5.9 months, but I think there is a caveat to that. These were symptomatic ones. The oligo-symptomatic or asymptomatic ones that can go on 40 years would not be in this group. These are the group that actually had clinical malaria.

(Slide.)

Now what about antibody testing? Well, a seminal study for this was by Chris Draper, who Marianna knows fro the London School, and his colleagues back in 1980. They looked at UK residents and immigrants with malaria, and they had serum from them taken a week after the onset of symptoms. All right three-quarters of the UK residents, those who essentially would be malaria naive, had antibodies detectible in the IFAT. All the immigrants were sero-positive.

(Slide.)

The other point, which I think is of great relevance to our need to exclude the semi-immune donor, was that the immigrant patients had higher mean IFAT titers. The antibodies persisted longer, and there was greater cross-reaction with other non-falciparum malarial antigens.

(Slide.)

Now we published in 1997 work with a commercial ELISA, which as you will see in the next slide is no longer available, but the initial stages -- at

any rate, it performed actually rather well. It was P. falciparum antigen-based from --- antigen, and in non-tropical area donors 0.45 percent sero-positivity. Putatively false-positives, because these were people that shouldn't have had a malaria risk as far as could be ascertained, and 1.5 percent sero-positive in tropical area donors. So we got 98.5 percent of our donors who had tropical exposure and thus potential malaria risks back as blood donors at that stage. That meant we could safety retrieve about 40,000 red cell units discarded each year unnecessarily.

(Slide.)

We were extremely pleased with this, and unfortunately considerably dismayed by '99 when we had to stop using the assay because we were concerned that it had sort of lost some sensitivity. But so important with these donors, too, and so important to get these tropical area people back into the donor pool, we introduced the IFAT in the National Transfusion Microbiology Laboratory at Collindale in north London, and ran it in parallel with a new ELISA which you will see in a moment. So we ran the IFAT and we are using alternative assays in parallel.

(Slide.)

Now the malaria assay was the New Market Lab, which we have already heard about this morning, so I will skip this slide and to onto this one.

(Slide.)

These were the serum samples we tested. Now we were debating this morning about serology panels, and I just want to tell you about our panel. This is from our own patients at the Hospital for Tropical Diseases who are not bled in addition to this, but we get ADTA samples and we also get serum samples sometimes from them. So these are from what we get for diagnostic use. They are then anonymized, and we have the species and the number of days from treatment and so on, but we don't then have them identified to individuals. But the prevalence of these I think is really quite tight because these are our own patients. We know the species has been confirmed in our own laboratory, and thus there isn't a question about have they been stored and how long and all the rest of it.

So acute samples were those within the seven days of the first positive blood film. Along the lines of the Draper time period, and --- followup is greater than or equal to eight days after the first positive film. We also looked at routine blood donations who shouldn't have a malaria risk and malaria risk donations, or so-called TA's, tropical area donors who had returned a minimum of six months, between six and 12 months ago. The group that you would pull in if you have a six-month exclusion with antibody test, but you wouldn't get if you waited 12 months.

(Slide.)

This is published. This is from our --- paper from a few years back now. The numbers are not enormous, but bear in mind what we said about the difficulties getting a panel. I don't think there are many labs that would probably get a panel much bigger than this one. The P. falciparum if you look here. These are acute remember, within seven days, so that is really quite early. Compared to IFAT, the commercial ELISA was more sensitive, and there were no IFAT-positive commercial ELISA negatives in that group.

Vivax, we only ran a P. falciparum IFAT. We don't have sufficient antigen from the others to prepare an IFAT. So this is again to falciparum IFAT, but certainly as you would expect because this has got vivax epitopes in the ELISA. The ELISA outperformed the IFAT. The numbers are small. Only 10 here, but for all it is worth ovale wasn't much different, but perhaps with a tendency which you will see later to be less sensitive in the ELISA than it was by the IFAT. There were not enough malariae to make a comment.

(Slide.)

On the followup, this is 46 here, so that is a reasonable number. We would like 46,000, but that is simply not achievable as you can imagine. ELISA-positive now to 5.7. They are all positive by IFAT, what you would expect. It is a falciparum antigen, multiple epitopes ---, so not too surprising. The recombinant actually did quite well. There were two IFAT-positive New Market negatives, and one was at day nine and day 31. It is too early that you would be reinstating them as donors anyway, but it is worth pointing out that the ELISA did miss two in that particular set. In the vivax again small numbers, but a little pickup as you would expect. Ovale again for what it is worth, slightly less good in the commercial ELISA. Now I have got some followup with sera we have added since, which I will show you in a moment.

(Slide.)

The low risk, these are non-tropical area donors who shouldn't have any antibody, and they were put through by ELISA only. They were not done by IFAT simply because of the numbers, but one of them were positive in the commercial ELISA. The tropical area donors that we collected we had a higher pickup rate, 5.4 percent to the commercial, than we did with the IFAT. Now if you go through groups, IFAT negative, commercial positive, we were okay with those. IFAT positive and commercial negative, we wanted to ask ourselves some questions about both positive we were comfortable with.

(Slide.)

With regards to the IFAT positive/ELISA negatives you have to take account I think of a learning effect with the IFAT. These are data from the North London Center after it started using it, and they couldn't be blinded to what they were testing. They knew they were going to be testing people from their antibodies by IFAT, and that obviously would influence whether they went in the donor pool or not. In talking to the slide readers they were quite clear that if they were in doubt as to whether it was positive or not at the screening dilution they would opt for the positive. So there is an inevitable bias towards the positive, which is understandable given the seriousness of letting malaria through.

Look at the decline. This is January and March, 2002, April and May, 2003. Both positive 2.5 percent. It comes down to about 1.5 percent. In the early days they were more semi-immune type people that were going through than subsequently because they had all been put back in by the time we got to here. ELISA only again the positivity rate goes down by about the same ratio, 4.5 to about 3.0, but a considerable reduction of about 14 fold here from 9.0 percent down to 0.6 percent as the readers got more and more and more confident reading the slide. So I think this is genuine training effect as they got more comfortable with it, but it also highlights one the inherent defects of the IFAT, and I certainly would not commend it to you as a way to proceed.

(Slide.)

Now we also looked at the ELISA -negative/IFAT- positive donor sera over a three-month period, January to April, 2003, and looked at the titers. As you have seen from Marianna's presentation, those who are the semi-immunes or even the acute malarias will have much higher titers. Our screen dilution is one in 20. For this study they were also looked at in one in 10, but in my routine lab we screen at one in 20. They go to one in 40. So all of them are one in 60 or less, which to my mind would not put them in the semi-immune category. In my lab, the semi-immunes are about one in 320, one in 640. So you dealing with very low levels, and based on that and the learning effect I was quite comfortable to recommend to them not do to the IFAT, but just to proceed with ELISA.

(Slide.)

Here is some data now on the non-falciparum species with sera that we have added from malariae, ovale, and vivax since the Voxx paper. So we managed to get 30 malariae out, which is no mean feat given the small numbers in

only 29 a year in Britain, and the IFAT outperformed the New Market. It is interesting in Marianna's data the IFAT for P. malariae and falciparum seem to go together. So we have got less pickup with the commercial one, but it has got no malariae epitopes in it. It outperformed for vivax, but that we have already confirmed, and for ovale on a rather bigger number, 36, it doesn't seem to be as sensitive as the IFAT. So the areas for improvement would be to malariae and ovale if suitable epitopes can be found.

(Slide.)

On the followup, these are very small numbers, and I don't think you can do more than look at a trend. Again, the vivax better for followup as well as for the acute. Ovale, a tendency to be not as good as the IFAT, and there was only one P. malariae, so you can't really make any reasonable comment on that.

(Slide.)

How many are we testing now and what is our peak reactive rate? In 2004 we tested 42,000 odd. In 2005 because we were getting tropical areas donor back, particularly semi-immune people that would be excluded permanently that we were keen to have back and they were keen to be in the donor pool, it went up to 67,000 nearly. Then per quarter now we are doing around 12,000, so this year if that is the January to March, 2006 total, if that is replicated, it will be about 48,000 screened. The repeat reactive rate, that decline I think is genuine. Again, in the early data we are getting donors back in, so we are now around the two percent mark. Not too different from the 1.5 percent that we had with the commercial ELISA that eventually had to be withdrawn, so it gives us confidence it is a similar sort of range of positivity.

(Slide.)

Now should we screen all of them or just target it? I just talked of the March, 2006. Those are the last figures I have from NBS on numbers of donors tested, but new donors in that month, 19,000 were tested and there were all, because they all are tested for hepatitis B surface antigen. But because we target the malaria testing we tested only 4,000. If we undertook universal testing for antimalarial antibodies we would be talking about nearly 15,000 more tests per month, which we don't consider in the cost-benefit ratio to be a worthwhile activity. You could do it, and I have sort of heard a few arguments about the weakness of history-taking that might make people consider it. Our on policy, we felt sufficiently confident that we could just target the donors to test.

(Slide.)

What are our selection guidelines? Well, implemented in November, 2005, a donor must not donate if they have ever had malaria or they have had an undiagnosed fever that could have been malaria whilst abroad or within six months of leaving a malaria endemic area.

(Slide.)

They mustn't donate if they have lived in any malarial endemic area for a continuous period of six months or more at any time of life, and you have got a year. I don't think that is unreasonable at all, and yet at any time of life I think is very important. Adults can get semi-immune to malaria as Sanjai said really perhaps quicker than children, so certainly that is an issue to not have the period just confined to infancy. They mustn't donate if they are less than 12 months after last leaving a malarial endemic area. That is the without testing. That is the travelers particularly.

(Slide.)

Now discretionary, donors who have had malaria diagnosed. If more than three years have passed since antimalarial therapy has been completed and the symptoms are resolved and so on, they can be antibody tested and accepted if it is negative. That may change because our initial position on this one who have had malaria diagnosed was to go for six months with an antibody test, but

we were overruled by an EC directive that the authorities have since been back to the Council of Europe that has accepted a shorter period. But it takes a while to go through to the next directive, so at the moment we are obliged to follow this directive; but we would prefer and I think we will in due course get a six-month period with antibody testing, because they are no different from the residents. You are saying the residents are semi-immune because they have had malaria lots of times, whether or not they recognize it as such. If somebody says they have had malaria even if once is out for three years it is not logical, so we felt that six months for both would be reasonable.

(Slide.)

For other donors, it is six months. If at least six months has passed since the date of the last potential exposure or the date of recovery, a validated antimalarial antibody test can be performed, and if negative they can be accepted as donors.

(Slide.)

Two slides now of conclusions. The first thing is what I am definitely not saying is that it is all antibody testing and nothing else. This is additional. This is belt braces in another belt. It provides a safeguard that is additional I think and complementary to history-taking and to time exclusion, and is not a replacement for either of them. Both of those need to be rigorously applied, which requires considerable skill in history-taking.

(Slide.)

I do believe that the UK positive selective antimalarial antibody screening of donors who might have a potential malaria exposure actually facilitates earlier donor reinstatement, which is important to us because we do need the donors back in the donor pool. But I also think it does it without detracting from the current safety of the blood supply. Thank you.

(Applause.)

DR. DODD: Thank you very much, Peter. I am sorry. I forgot to put the time on. Next we are going to hear the experience from France where there is also testing of donors for malaria antibodies, and Olivier Garraud will speak.

French Experience with Malaria Antibody Testing
by Olivier Garraud, MD, PhD

DR. GARRAUD: First of all, thank you for inviting EFS and myself of course to share the French experience with you. I am ---, so you may try ---. It is an evening exercise you may try to pronounce correctly. It is very difficult. I am member of the board of directors at the EFS, which is a national blood bank system.

(Slide.)

Actually what is the situation in France regarding malaria risk? There are more and more immigrants, especially from sub-Saharan Africa. There are more and more travelers for business or leisure and coming or going from and to tropical areas, and malaria is currently the first cause of fever in returning travelers.

(Slide.)

Keep in mind that France is not restricted to the old Europe. There are locations which belong to France but which are located in all continents where in some places there is malaria suspicion.

(Slide.)

In metropolitan France malaria is mostly imported, while in French Guiana or in the Comores Islands it is endemic, and these endemic cases contribute to approximately 10 percent of imported cases in France.

(Slide.)

Here is a representation of imported cases in France, and as you can see the huge majority is imported from west Africa, formerly the French-speaking part of Africa. It is essentially P. falciparum.

(Slide.)

Indeed, France reports most imported cases than any other European country. More than 7,000 cases are recorded per year, but we do know that it is largely underscored. A lot of Africans, for example, don't go to hospitals and have traditional medicines. So we think that we can double that and it is probably more close to 15,000. It's an estimation. As you can see, most cases indeed come from west and central Africa. Very interestingly, more than 75 percent of cases occur in Africans residing in France, and these are essentially as I said P. falciparum cases and five percent are severe cases.

(Slide.)

Now let's move on to transfusion-transmitted malaria, which is the topic of the day. Actually in France as in other European countries and probably in the US, this is incidental. So it doesn't occur frequently as said this morning. It is very different to the situation which is encountered in endemic countries where it is extremely frequent and almost impossible to correctly evaluate. Anyway, when transfusion-transmitted cases occur it is always severe and often lethal.

(Slide.)

This has been taken from a paper which appeared in --- last year from Reesink in the Netherlands, and have added a couple of observations which I picked up some other publications. Over the past 10 years we recorded one case in France that have been confirmed, two cases have been recorded in England, and I understood from this morning's session that we can say that there were 10 cases in the US. It is interesting to see that there were seven case in Italy by this time.

(Slide.)

The history of transfusion-transmitted malaria in France, from the '60s to the late '90s more than 100 cases were recorded. Since the early --- we encountered only one case in France, but we have also to note that the one case in '93, one was doubtful in '98, there was one near miss case in '99, so there was information --- donation so we could retrieve the bag and lend it to destruction, and there was one little case in 2002.

(Slide.)

Well, excuse me. So what about --- in France and blood collection first? Briefly, blood collection in France is --- in one government organization termed EFS.

(Slide.)

Blood collection concerns more than 2.5 million labile products. Some of them may be at risk for P. falciparum transmission essentially.

(Slide.)

So transfusion-transmitted malaria prevention in France is based essentially on three issues. Information and self-exclusion for the donors, a questionnaire and medical interview, and third on lab testing which was formerly EFA and as I will show you we turned. We recently turned to ELISA.

(Slide.)

Was that safe enough? Probably not, because as I said one case passed through with all series of malaria ---, and there was one little case, a 2003 case, which occurred in the UK. It is very similar to the 2002 French case as I read actually, or I talked about that with Dr. Kitchen.

(Slide.)

Well, so EFA, which was the former test which was implemented, has been performed since '95, for more than 10 years. Initially it was based on a non-standardized assay, and there were several difficulties that --- can rate.

(Slide.)

If you don't mind, here are my personal thoughts on serology when applied to plasmodiae, and I think I am very much in line with Chris Ockenhouse

as understood from this morning. So I won't comment further on that, and those folks who might be interested may refer to the quotations.

(Slide.)

But actually what are the actual needs? We need tests that show some robustness, reliability, sensitivity, specificity. Automation would be an advantage of course, and because this is not -- and this has been said this morning. It is important to re-insist on that. This is not clinical biology. This doesn't aim at, I mean, deciding if somebody gets sick or not. This is to improve the safety of blood donation. So false negatives absolutely are not permitted. False positive can be.

(Slide.)

After the 2002 little case we went to revise our current measures in two main lines. The first line was revision of a deferral policy which complied with the implementation of the European directive as been said by Dr.ÿChiodini a couple of minutes before, and the second line was to implement an ELISA-based serology. I will comment on that.

(Slide.)

So first the revision of a deferral policy. It is based on three different epidemiological situations which leads to different attitudes leading to deferral or non-deferral of a blood donor, and rehabilitation is based on serology as shown. I don't know if it is --- on that maybe a time -- I mean I have not time enough, but everything, you have that on the slides and I can comment during the questioning after.

(Slide.)

So we went also to revise the lab testing, and we first had an observation period so we tried to test for antigen, NAT, different kits for different EFA's, different ELISA's; and we went now to a test period in 2005, so we evaluated two tests in France. It is probably the same in England's. Those tests must be CE marks. CE is for European Community, so it is pretty much the same that FDA approved in the US. This has absolutely to be CE marked to be implemented, and very few producers go to the CE mark because that costs a lot of money and the market is not very strong, so they don't have money back. So that is a real problem for malaria, but not only for malaria. That is a real problem for --- disease whatsoever.

So we went to select one technique which was an ELISA and one kit, which was the one made ---, which is a Swiss-based company, and the selection was made on a series of -- oh, sorry -- a series of performance which are --- on the slide.

(Slide.)

I have one slide which doesn't show up. Anyway, never mind. So the decision tree, so the specificity was approximately 98 percent and the sensitivity reached 100 percent in this selected test.

(Slide.)

So we went to draw a decision tree, as shown. So if negative or a non-repeated positive they are -- it is considered as negative, so on the serology it is negative, and the labile product can be transfused to patients. If there is any repeated serology it goes to destruction.

(Slide.)

We do a second test based on EFA, but this test is to give appropriate information to the blood donor. As I said, this is not clinical biology, so it is what do we do with a blood donor then after we tell him or her you can't donate blood because there was some marker which was positive. We have to explain a little more, because it is not possible to leave him or her at the blood center without some appropriate information. So we do this test, but this is not a rehabilitation test. So the decision is made on the ELISA only with the EFA is only for better information to the blood donor. So it is to

tell him or her that is probably a false positive or a false negative. So this doesn't requalify the testing.

(Slide.)

Again this has been said this morning. Interpreting serology is tedious in P. falciparum infections, especially because there is a blind period in the incubation time which is labile. Because there is a problem of immune or semi-immune, especially for former residents having been living in endemic countries, and because for P. vivax the antibody level is never that big, so it is very tedious to interpret to serology.

(Slide.)

In our experience -- oh, I understand. This is the first set of slides, Sanjai, but if --- it doesn't matter. So in our experience 3.5 percent of samples undergo testing in France probably, and all in all such exclusion, medical interview and lab testing discard more than five percent of all blood donations in total, which is pretty high and which questions the shortage in blood supply. One day five-percent exclusion is almost unaffordable actually, especially during summertime because we experience some shortage in France right now. So it is not 10 percent, I have revised my calculation. It is more five percent than 10 percent.

(Slide.)

So the measures taken in France are almost sufficient because we did not experience another case, but statistically speaking this doesn't mean anything because we knew before revision of these policies that one case of transmission of P. falciparum infection occurs so rarely that it is a very difficult to say that our measures are more effective than the previous ones. So when time has elapsed we can see if we were effectively good or if we have to revise again our politics.

Something which I would like to comment or so on is that we have great expectancies on procedures for pathogen reduction. We have implemented these measures in France. We have an intercept procedure which is authorized by the ---, the equivalent for FDA. So in some places, incidently my place for example, we have intercept treated platelets, so this is efficient on bacterial viruses, parasites of course, and leukocytes. There is another one which is in trial which is riboflavin, vitamin B2, which is tested. This will probably come into trials for --- and we are still awaiting some efficient procedure for red blood cells which is supposed to be in the pipeline but probably won't come before the next five or 10 years.

So it is my feeling, my personal feeling that these procedures might be something very important to take into consideration, because this will probably eradicate any living pathogen in labile products, so including the parasites. That is something which is good to think about, especially by the time of ---.

(Slide.)

So I would like to thank people who helped me to some of this information. Professor Candolfi did the sum up of ELISA tests. He tried more than 15,000 sera samples from exposed individuals. This was an independent study, independent for EFS, so we committed him to do this study based on the European study. And I got some information from Dr. Kitchen and some of my information from Dr. Klueter in Germany, and I thank also all the biologists who are in charge of qualifying blood donations at EFS. We have 14 labs in the French territory to do that, and thanks also to Hemovigilance Network in France based EFS and the hospitals. Thank you.

(Applause.)

DR. DODD: Thank you very much, Dr. Garraud. Next we have Susan Stramer from the American Red Cross. She has changed her title a little bit to issues with malaria screening in the US, but she is also going to cover data

from Australia, and in recognition of that she is going to have extra time to talk. That is --

DR. STRAMER: --- the end. It is good to start at the end.

DR. DODD: Yes, that is the end. Yes, I was going to say you got the -- okay. Okay.

(Adjusting equipment.)

Australian Experience with Malaria Antibody Testing and
Feasibility of Implementing a Malaria Test for
US Blood Donors
by Susan Stramer, PhD

DR. STRAMER: Thank you. I would like to thank the organizers for inviting me. I do have a complex title, and to add to that a complex talk, so let me get started.

(Slide.)

What I hope to cover today is in the US focusing on data from the American Red Cross is the way we do our current donor questioning, including the process for determining donor eligibility. I will also talk about post-donation information which also influences donor eligibility and product qualifications, donor deferrals over the period of time from 2000 to 2005, that six-year period. As Roger mentioned, I will cover the Australian experience, which is similar to that that has already been reviewed for the UK and French, the testing in algorithm, but the Australians as one would expect have an interesting twist in the way that they do it. Then what are some US testing options after all of that.

(Slide.)

So we start with donor questioning. As has already been discussed, have you ever had malaria. No, you are accepted; yes, how long has been; and if it has been greater than three years, yes, and no, we defer the donor.

(Slide.)

Then we go into more serious and more complicated questions relative to travel. Have you traveled anywhere, yes/no, where, how long has it been? So we discriminate then between the travel deferrals and the residence deferral. Now this is quite complicated. I am not reading the slide. You all know the processes, but two rules to help us get through this process include what we give to our health historians and at the Red Cross we have over 7,000 of these individuals on our different mobile and fixed sites. They have reference listings. They give them countries by region to assist in identifying general area. They have electronic reference tables that don't include all countries, but they have countries with no risk listed and alphabetized listings of countries of risk with destinations within a country. I said it wasn't all inclusive, so they do have to refer to the CDC's yellow book. Many countries have cities of the same name in several different provinces or states, which adds to some confusion.

If more information is needed they consult another atlas, the Hammond Atlas, and if they can't determine what to do most likely what happens is the donor is deferred because in the meantime while they are trying to decipher all this information the donor is waiting and waiting and waiting. So the donor then becomes deferred, experiences a relatively poor experience with their donation, doesn't come back, and then we are left with a consult with headquarters to determine what should have been done.

(Slide.)

This slide shows you over a month of occurrence for the last two years how many times the donor questioning process has failed, how many times we have accepted an ineligible donor due to malaria questioning, and without looking at the numbers it is about a frequency of one in 10,000.

(Slide.)

Now look at those same numbers with the other problems that we have relative to behavioral deferrals, it is a total of 2,520, and half of those are for malaria. I also showed you where we implemented UDHQ in March of 2005, and that has not done anything to change the patterns of malaria. All of these slides are by month of occurrence, and you see trends that appear to be decreasing. Now I don't really believe these are real and they are probably related to reporting lags as I will show you in a subsequent slide.

(Slide.)

So not how many of these acceptance of ineligible donors have resulted in products being released that would be termed unacceptable or violative? So we have had about 12 percent or exactly 12 percent, 144 of 1,176 of our acceptance of ineligible donors over the last two years resulting in a BPD. So that is about one in 100,000 presenting donors.

Now where have these individuals come from? These were now just focusing on the BPD's, the 144; 42 of them came from Mexico, followed by China, Dominican Republic, et cetera, et cetera. So I am going to give you two examples of complications related to the questions we ask, one from Mexico and one for China.

(Slide.)

If you had been to China, and maybe Alan remembers what provinces he has been in, many of us, I don't remember what provinces I have been in. Donor historians are asked for certain provinces to search for the rural areas and determine by those rural areas if a donor should be excluded. So mind you these are 7,000 individuals in the field all doing it the same way, giving their ham and apples, their popdown screens and everything, and they have to look at ---, altitude and latitude. I can even say this as I look, you know, memorize this, let alone these individuals with all the tools they have, so stuff is going to happen. The complexity leads to deferral in these kinds of situations. Those donors would be deferred, and we wind up with lots of questions to headquarters.

(Slide.)

The question that I have for you is do you know how many states in Mexico have a city named Monterey? Well, the answer is seven. There are many, many other complications like this, but what makes it even more confusing is some of those states have malarious risk and some of those do not. Well, if you have been Monterey you better memorize your map when you donate blood.

(Slide.)

The other issue we have is post-donation information. This isn't necessarily callback as we think of with routine PDI's, but these are donors who come to donate subsequently and there is a forward. And they say, "Well, wait a minute. I came in before and you accepted me. What changed?" Well, nothing changed. We either made a mistake or the information you provided to us wasn't clear, or somewhere the system broke down. Well, this frequency here for PDI is about one in 2,000.

(Slide.)

Here is PDI against the other bureau history PDI's. So again, after UDHQ we really haven't seen a change in the number of malaria PDI's by month. Again, this is month of occurrence.

(Slide.)

This is that same information, just taking a subset of it, squeezing the X axis a little bit so that only do you see month of occurrence, but also months that these were discovered. That is what this line is with the N's. So there is about six-month lag time between occurrence and discovery. So if we are making changes to improve the process, whether it is corrective actions to problem --- or just process improvements, it takes about a half a year to realize whether those are effective.

(Slide.)

Now I am going to go to donor deferrals. The next two slides will show in a much more simple form, but I have all the bells and whistles here of all the numbers. The green line shows you -- now let me just say this is six years of Red Cross data, about 24 million presenting donors. The green line shows you the number of donors deferred due to travel. So it ranges from in 2000 0.8 to 1.3 percent 2005. So that is on the increase significantly so. What is decreasing is the number of residents we have to malaria areas and the number of people who have had malaria, and most of your populations run together in a number of data sets that I will show you. Travel accounts for 90 percent of our deferrals for malaria.

(Slide.)

Here is the data on a table. David will also show this. If you take the number, the percentages of deferrals are shown in the second column, times the mean donation rate, the annual mean donation rate, you get about a half-a-million lost donations over the six-year period of time.

(Slide.)

Focusing just on the most recent year where we have had a total of about 1.4 percent lost donors, that accounts projecting it by the annual donation rate of 1.725 in 90,000 totally lost donations, 86,000 of those due to travel. Similarly when we collected the ABC data, they also had about 83,000 malaria travel deferrals. So in total that is about 175,000.

(Slide.)

So what does this represent as opportunity losses lifetime? I just showed the data for 2005. I will show you in subsequent slides that most of these are first-time donors in all categories, over 80 percent, and most that is greater than 90 percent. Well, less than 10 percent will never return over the observed period of time. So I asked the question and we did an analysis over a 10-year period to determine how many first-time donors we convert to repeat donors and then over lifetime how many donations do they make in their career and over what period of time. So if we look at the travel deferrals I showed you for 2005, the 50,119, multiply that by the mean number of donations per career. We are looking at about 300,000 career donations that are lost for travel alone in 2005. One year, and that is just for the Red Cross.

(Slide.)

Now looking at the demographics of some of these lost donors. Again, green is travel, red is residents, and yellow with the blue line is malaria. The age groups where you see -- the age groups are on the X axis with the first symbol being less than -- oops. I am pointing at myself. That's useful -- less than 24 years to greater than 60 years. Most of those who resided in or had malaria fit into the less than 24-year age group, the youngest, and ages are equally distributed among those who have traveled.

(Slide.)

I am going to run through some history of the first-time repeat status and the percent returned. For those who have had malaria in six years we have had a whopping 492 deferred donors. Three of those donors have been deferred twice. Although they have been deferred they have come back and tried to donate. 98 percent of those are first time, and of the repeats only two donors had two prior donations; 60 percent are males, and of those who have returned there have only been four after the deferral period with one to two donations per donor. So this isn't a very productive group.

(Slide.)

If we look at those with residence, it is fairly similar, although the numbers are higher; 167 deferred previously two to three times. First-time donors, again 96 percent, repeat donors, very few prior donations, 60 percent lost, and how many have returned? 1.1 percent with two to seven donations per donor. So again in six years, 338, not a very productive group.

(Slide.)

Now if we go to our travel group, 4,300 of the 237- donors have been deferred two, three, or four times previously, and they are still bound and determined to donate, which is good. Here we still have a lot of first-time donors, 83 percent. Of those repeats we did have 12,000 donors with many prior donations. The number of males, percent males, is comparable to the normal donor population at 50 percent. Now where we have an opportunity lost here is that only six percent of these donors have returned to make two to 23 donations per donor.

(Slide.)

Where are these donors deferring from? If you look at the numbers by collection, percent collections, the highs for us at least occur in California in all cases. Our northern California region hits every target, infection, residence and travel, and LA hits two of the three. Oakland has took the high ranking with 2.4 percent for travel. If you look at low by collections, Puerto Rico takes the price. Puerto Rico in the last six-year period of time has never had a donor deferral for malarial infection. Now whether that is true or not, those are the data.

(Laughter.)

Perhaps they have had --- or other things, but not for malaria. Okay. The data we got for ABC shows similar patterns. In the southeast they have the low, in the Gulf coast area 0.7 percent. Unlike our west coast, they have east coast the highest numbers of travel deferrals.

(Slide.)

Where does malaria rank with the other deferrals that we have within our system? It is about five percent of the total with the highest already as mentioned. Things like low hemoglobin, unacceptable blood pressure. But hemoglobin obviously is the winner here -- or the loser.

(Slide.)

These were very interesting abstracts submitted for this year's annual meeting, and I asked permission of Brian Spencer to show this data. The abstract was accepted. It will be presented in Miami this year. It is data collected from six REDS-II centers, and what they did is compared travelers to malarial areas with imported malaria cases and then looked at where donors are being deferred. So if you look at the pink, just let's go across this draft, 84 times more people in the United States travel to Mexico than to Africa, but 94 times more malaria is imported into the United States from Africa than is Mexico. So that translates to a rate per million travelers of 8,000 times more risk for Africa than from Mexico and 91 times more for the Americas relative to Africa. Then the yellow boxes show actual deferrals for the six regions that 80 percent of the travel deferrals were related to the Americas with 40 percent being Mexico.

(Slide.)

This is published data that has been alluded to, but from 1963 to 1999 where is the risk in transfusion-transmitted malaria? It is not in US travelers, even though we are all answering the same questions. Somehow either the risk is lower in travelers or the questions work better in travelers.

(Slide.)

This is also previously published data, and it is relevant as I dive into the Australian data because the issue comes up do we need all four species of plasmodia. So in this slide if you focus on the yellow cross, what is in the center is the number of malariae and ovale cases in the Americas, and the numbers are very small. In North America for example there are none reported for 2002.

(Slide.)

Now I am going to take you through the bullets that I have on these slides from the Australian Red Cross Blood Society. These were very graciously shared and discussed with me by Clive Seed and is fellow coworkers at Perth in

western Australia, Tony Keller and Sally Thomas. So their last case of transfusion-transmitted malaria which was a fatality occurred in 1991. At a frequency of one million donations per year, their risk is believed to be less than one per 10 million. Clive have updated risk estimate differences for falciparum and vivax. Let's just leave it as less than one per 10 million.

Their risk comes from travel to southeast Asia, and it is more vivax, and so their calculated risk is higher for vivax than it is for falciparum. Their donor deferral rates due to the combined travel questions is five percent, but I am going to urge you to use donor deferral with a grain of salt as I will explain. So they lose about 20,000 donors a year out of their 1.1 million donations. It represents 50 percent of their current red cell discounts, and any donor that answers yes to a question in all cases they will still collect blood. They do not throw away plasma. Australia has tried to maintain self-sufficiency forever, so they send their plasma to the fractionator. They just discard red cells. So when you are talking about deferral in Australia we are talking about the --- of distribution of cellular components. They are maintaining the relationship with their donors and their donors are continually coming in to donate.

So what you see yellow under the deferral codes is what their yes responses are, 95.5 percent in travelers, 4.4 percent in residents, and less than 0.1 percent are those that had previous infection. They --- like in the UK a resident is greater than or equal to six continuous months, and I have highlighted within the past three years because that is going to change soon to be consistent with the Council of Europe to ever. So if you have ever had six continuous months you would be considered deferred, which means your red cells would be pitched.

So Clive's impression of all these processes is, and it answers some questions we were asked this morning, any process that is reliant on --- soliciting and recording accurate information has a significant error rate. Now the Australians ask questions much more simply than we do. They don't granulize China. Have you been to China? You are out. It is just that simply, unless you are in Hong Kong or ---, so they do have a couple of exceptions.

(Slide.)

So July 17th they implemented their testing-in strategy for any deferred donor, and it was modeled after the UK system that was described. Rather than using six months, however, they did a search and convinced themselves and their regulators that four months was adequate. So greater that or equal to four months has passed since leaving a malarial area, a recovery or cessation of symptoms, a prior infection, blah, blah, blah, they will tested with an antibody test. Testing is not needed if greater than three years has passed. They will collect the donation. They have systems in place to quarantine the red cells until the antibody testing is done.

They use the New Market test, the recombinant Pf/Pv test that has been described this morning. We have talked about cross-reactivity. They base the data on what was published by Kitchen, a 50 to 75 percent cross-reactivity to ovale. At the time of implementation New Market told Clive "We tested one malariae sample and it was positive," so that is good.

(Laughter.)

But their lost malaria P. malariae case was in 1956, so it is not too big a concern, and malariae and ovale represent a very small percentage of their import in malaria. They took all the data to TGA, which is their regulator, and it was approved based on the low frequencies they see of PM and PO. Cross-reactivity and their four-month, as they call it, embargo. If the test is negative the red cells are released for transfusion. The donor deferral is removed there is another reason for deferral.

Clive wanted me to emphasize that staff training is critical and an IT system is critical. They still don't have Queensland up on their system,

which is 20 percent of their collections, between Queensland is not up on their most current computer system yet. But before they had implementation in the other four states excluding Queensland they did have one large process recall, which again is why it is so important to be automated.

(Slide.)

What are their results? 26,000 donor screened, including all deferred groups. Testing is centralized in Sydney. Since testing began 77 percent of the donors have come back to make subsequent donations. The repeat reactive rate is consistent to what has been shown for the UK at about 2.3 percent and is consistent with their pilot study in western Australia. I will talk more about the repeat reactive donors, but each repeat reactive is tested to determine whether they have evidence of parasitemia. Now granted these tests may not be the most sensitive, but this is what is done.

They use an antigen dip stick from Binax that has sensitivity and specificity as indicated, again just for falciparum and vivax. It is a whole blood test, and with the centralized model with everything going to Sydney it is a challenge to get the repeat reactives tested on the strip within 72 hours. They also do a PCR, a commercial PCR assay from Artis.

If there is positivity on antigen with our without PCR positivity since that takes longer they refer to their physician for clinical assessment, and they are termed as probably parasitemic. Each repeat reactive donor is notified by letter so that the donors have a letter to share with their physician if in fact they develop any febrile illness. Even if they are repeat reactive and notified, they are still okay to donate plasma. Don't want to get rid of that plasma. Even if they are positive by PCR or antigen, we are still collecting their plasma.

(Slide.)

So just this week I got an email from Clive saying we have had our first probably parasitemic donor, an Indian resident who migrated to Australia in 2005, completely asymptomatic, had a sign signal on the New Market test, was repeat reactive, had a faint band on the Binax strip antigen assay that was repeatable. It was PCR negative. Followup is pending. The donor was referred to their specialist, and according to Clive's analysis the number one within about 40,000 tested is within their predicted rate.

So what I have now here in yellow, this five to four category, how many donors they have had that have been repeat reactive over the total number of donations tested. So for travel they have about one percent repeat reactive for antibody. Again, none of them were parasitemic or antigenemic. Nine percent for residents and 30 percent for infection, which I asked the question of Clive of why bother. You have 50 donors, 15 are antigen. Just get rid of them. Well, you know, they want their plasma.

(Laughter.)

Anyway, as far as recovery of indexed red cells to the end of May it is 32,000, and that is considerably higher than their predictions, and they believe their predictions may not have been accurate because they weren't capturing all those donors that they really had as frac donors, and they refer to those as their hidden deferrals.

(Slide.)

So now we get into the question of what should we do. Universal screening is obviously screening all donations, and we have already discussed should it be by antibody test. Is an antibody test adequately sensitive, or do we also need NAT? Crucial is the testing-in model, and we have seen various flavors of that today. So in yellow I have just my own schematic what you have for NAT. Here you would have the parasitemic period, whether it is seven to 14 days, nine to 16 days, that whole discussion. Or if you go all the way to four to six months, is the testing algorithms used? That is all well and good. This is when you probably can most reliably detect a donor for DNA, but as time goes

on we have already discussed these tests will not be positive because of the low levels of parasitemia. So then within this window period when does antibody appear? What is the length of this window period? Is it really days, and if it is days is that adequate? And then how long is antibody? We certainly believe it is not lifelong unless you are chronically stimulated with antigen as we have seen from sub-Saharan African residents.

(Slide.)

So what do we think about it, a model of the testing-in model? So I listed pro's and con's. An advantage is it shortens the deferral period. That is a no-brainer, from 12 months or three years to four months. In the Australian model a donation is adequate for the followup, and if it tests antibody negative the red cells are used and the donor is automatically reinstated. If we used such a model in the United States, would we have to do a sample only at four or six months, or would it be a donation? And if the donation tested antibody-negative, would we use those components?

What is against the testing-in model? In our system, which is already fraught with complexity, it just --in my opinion and many others, it just adds another layer of complexity. It does not alter our current questioning process. The donor loss would likely be equivalent, and then we have the whole issues of cost which embed process in that. So we have computer costs, process development costs, including how we manage donors, development of a testing algorithm. And another big question is the cost of reagents, and if we are doing so few tests would any of the US manufacturers who would have to go through FDA, would any of them be interested for the small of the market, … la HTLV confirmatory testing. We just still don't have a licensed confirmatory test. So when the market is small these companies generally aren't interested.

(Slide.)

Going to universal screening, and I am focusing on antibody only, universal screening will only be practical in the United States if we can eliminate the travel deferrals, which would bring back our 1.2 to 1.3 percent of presenting donors. This would definitely increase donations. Could we convert those 83 percent first-time donors or those repeat donors now who don't come back into lifetime career donors? It would certainly I believe improve donor retention. I mean, we are obviously not losing them for travel. It reduce self-deferrals and I think it certainly would increase goodwill at the blood centers.

What are the con's? We don't know what the required algorithm would be. As I said, would it be antibody only? We certainly don't know the costs. We don't know the test performance, although David is going to show us some test performance. Would the tests be required to be sensitive to all four plasmodial species versus cross-reactivity for ovale and malariae? What is the specificity? Would we lose donors unnecessarily? But would the donor loss be smaller and perhaps more specific than those we do today because of travel? How would we manage our donors? Would there be some deferral period or as long as you are antibody-positive you are history? Then what else would we do for confirmatory testing?

(Slide.)

So here is a possible scenario. You ask the donor a question. Have you had malaria, blah, blah? Yes. I would suggest no test. Why bother? That is a permanent deferral. We could do something similar for those who have been residents. In the past three years have you lived -- well, greater than five years outside the United States or Canada. You would have to have drop-down menus and programs to determine malarial areas as we do today. But if the answer is yes we could either test with a deferral period, you know, yes with deferring those that are antibody-positive. But I would almost argue not to test and defer. Either three years as we do currently or perhaps permanently since these donors very much resemble those who have had malaria. Then we will

test our other donations with the open question. Is it a four-species test or a falciparum-vivax specific test, depending on cross-reactivity for the other two species. One thing that I think is key, but will definitely stir discussion, is what will we do about the window period?
A) ignore it, B) do NAT, which I think would be unreasonably expensive; or do we ask the donors with recent travel to self-defer during what we consider a reasonable window period of time. I have already mentioned additional testing as Clive does in Australia. Would we want to do a PCR test or an antigen test just for better donor counseling?

(Slide.)

With that, I would like to thank my collaborators. A special thanks to Ed Notari, Kathy Waldman, very, very special thanks to Clive Seed, and thank you for your attention.

(Applause.)

DR. DODD: Thank you very much, Susan. I think there is lots to think about there, and I hope we will have some productive discussion a little bit later.

Next we have David Leiby, also from the American Red Cross ---, who is going to talk about enhancing US blood availability by testing for plasmodium species infection. David has some real data to show us, too.

Enhancing US Blood Availability by
Testing for Plasmodium Infection
by David Leiby, PhD

DR. LEIBY: Unfortunately I don't have a whole lot of data, Roger.

(Laughter.)

I was asked to give a talk on the study design that we are working on in the Red Cross to get at the deferred donor issue. We have only started this study a month ago, so you are only going to see a month's worth of data, but I think it is rather worthwhile. The other challenge speaking so late in the session is that everyone has already used my slides.

(Laughter.)

Or if I am using their slides they have already shown them. All the topics have been discussed. There is not sense talking about malaria biology. There is no sense talking about the New Market test, and Sue covered donor deferral ad nauseam.

(Laughter.)

So I will go my own way.

(Slide.)

But I will start with some data that we actually presented at the AABB meeting and actually the Trop Med meeting which generated some publicity back in the fall. This is similar to data that Sue just showed as she suggested, but it really gets at the donor deferrals, about 1.1 percent that the Red Cross has lost from 2000 to 2005. Once again, you can actually do calculations of the number of lost donations, 456,000 over a six-year period. This is really a conservative estimate. As Alan mentioned, this doesn't take into account the people who self-deferred, and there are a large number of donors who never come to the donor center because they know they are going to be deferred, or their friends say, "Well, you were on the cruise ship with us. We're not going to be able to give blood, so don't go." They never come. Once again, it is really the type II deferral. It is the travel deferral that is the large bulk, or 96 percent of the deferrals or the amount of donations we lose.

(Slide.)

When you graphically show it, this is the same graph that Sue showed. I go with the less is more as opposed to more is less.

(Laughter.)

I am much more simplistic than Sue I guess. But anyway, it is the same trend Sue talked about with those with malaria and those living in endemic areas. Rather low contribution, actually a declining value here for those endemic areas, and it is really this value up here with this regression of 0.9. The travelers, that is increasing. That is really of interest to us, and that is where are our lost donations and that is why I think we are all here today, is because of the travel issue alone.

(Slide.)

Now this was already presented by Alan, so this is nothing new as well, but I think it brings up an important topic that I want to talk about for a little bit. This came out of the CDC, and this was their travel --- about cases recently in the Bahamas and the fact of the need to look at for this, defer donors and everything else that is associated with that kind of issue. We see these all the time. Every year they are somewhere else. It is the Dominican Republic, now it is the Bahamas, where will it be next? I don't know.

(Slide.)

So where I really think this leads to, and I would like to propose from now on, this is really the princess and the pea phenomenon. This is how we have dealt with malaria for years and years and years. Each time there is some other problem, some other issue with travel, someplace else malaria occurs, we throw another mattress on top of the pea.

Now if you remember the story of the princess and the pea in the fairy tale, you have to really stretch back to when you were a kid perhaps, it was all about determining whether or not the princess was really true royal blood. It was her ability, because she was truly a princess, to be able to feel that pea.

Well, what we have done, we have added so many layers on top of this malaria issue that our princess can no longer feel the pea. The pea is stuck down here. We have buried the pea completely. I would ask you, and I have posed this question to other people before, if malaria was a newly emergent disease would we go through all this effort to try to prevent its transmission? I don't think we would at all.

Can you imagine if you took this same approach for West Nile virus four or five years ago? The first question would be in the last three years have you traveled to Oklahoma, Nebraska, Texas, or Kansas? Were you there between the months of April and October? Did you get off the tour bus?

(Laughter.)

Was it between dusk and dawn, or was it in full sunlight?

(Laughter.)

Were you within 100 meters of a pasture full of horses? And most importantly, was there a dead crow on the side of the road?

(Laughter.)

All kidding aside, I don't think that is the way to approach the issue, and I think what has happened through this princess and the pea phenomenon is we have taken what should have been a blood safety issue, what really was always a blood safety issue, and we have piled so many mattresses on top of it that it has become a donor availability issue. Somewhere we have lost sight of what the true issues really are.

(Slide.)

In our study at the Red Cross, and as I said we are only a month into this, we are trying to address some of these issues, some of the things we have talked about today. As you have seen, there is very little data actually available about donors themselves; and I think that is an inherent problem, and I will raise that as well.

First of all, are deferred donors actually infected? Well, certainly some of them are. We have heard data that transfusion cases have dropped over the last 10 years, but we really don't know if that is due to donor

deferrals, if they are due to another phenomenon, or why that is occurring. This is why we need some of these types of data.

The type of deferral, is that associated with certain infections? Certainly people who had malaria might be the ones who are more likely to have it. Or might it be other people who have traveled to certain regions? What about the effectiveness of the EIA assays in general? Can we find an assay that works? Is IFA okay, or are there other options which we can use? What about the specificity and sensitivity of these very tests? Certainly we talk about this being of primary importance for any test we work with.

What about the usefulness of PCR/RT-PCR followup? We have spent a lot of time talking about this, and I think that is something that will come in probably discussions at the end, and I am not sure there is really a very clear answer as of yet.

What about interventions? And likely everyone else I am going to throw out my own interventions, and I am sure we will come back to these. We have the possibility of just testing alone and testing everyone. We could test the subset, at donation or as done in Europe and parts, we could test months later after a three- or four-month period. We could have a permanent deferral for a subset of donors as Sue has just suggested, or we could just say, "Well, we are doing fine as it is. We are not going to change a thing."

(Slide.)

In our study at the Red Cross, as I said, in its infancy is designed as follows. First of all, we are going to use serologic testing with the New Market EIA. New Market seems to have the market cornered right now, they are really benefitting by all of us using their test and generating data.

The design of our study was first to take approximately 3,000 non-deferred donors from the greater Chesapeake and Potomac region or Red Cross, which is located in the Baltimore-Washington area. That is really to determine the background, the EIA background. We assume that there are not many infected individuals in the donor pool in order to differentiate those infected who are deferred from the background. You have to test a significant number, so we are testing 3,000 non-deferred donors. Then we are going to test malaria deferred donors from greater Chesapeake as well as the Penn-Jersey regions. In fact, I think we got our first samples in this week, so we are on our way.

When we have positives by the New Market EIA, we have arranged with CDC and Marianna. She is going to test them by IFA so we get some kind of understanding of what the IFA means -- the EIA rather, not the IFA. We are going to test the samples by PCR and RT-PCR, and hopefully we will be able to confirm some of the infections, but also be able to identify the species, because I think that is important for the test itself because we know it only picks up two of the species with great reliability.

We are also bringing these positive donors to ask them various risk factor questions. A short questionnaire about -- for malaria deferred donors as well as anyone we find as positive if they are in fact non-deferred donors.

(Slide.)

Well, there are a number of challenges in this type of study. Anyone in here knows quite a number of done studies with deferred donors. It is not the easiest thing to get large numbers with. So first of all, we are challenged by obtaining sufficient numbers of deferred donors and --- if we want to look for particular types of deferred donors. So at least for now we are working with a sample size of convenience. We are taking all comers we can get through the two regions.

As I just mentioned, one of the challenges with the EIA is that -- I don't know if bias is the right word, but it picks up primarily two species. There is some cross-reactivity with the other two. I have my own questions of whether it is really that important in the United States about the other two species, and I think we will discuss that a little later.

With PCR and RT-PCR parasitemia may in fact have cleared. We have heard data like that. We can't see the parasitemia. There is also sampling and concentration issues, and I want to bring this up because I often hear that, and this falls for a lot of parasitic infections. People say, well, the PCR aren't sensitive enough. Well, it is really not the PCR's. They are fine. This goes for --- disease. This goes for Babesiosis. You are comparing them too many times to a viral NAT test in which there are plenty of copies of the virus. The issue, and this has been brought up here and pointed out, it is really that there aren't that many parasites circulating in the blood. So it is really a sampling issue, a concentration issue, and I think that is where we have to determine if we would incorporate NAT testing how we are going to concentrate the sample in order to increase the testing capabilities. Then lastly the accuracy of the questionnaire.

(Slide.)

So our first part of our test, and this is where -- actually the data that I can show you today before I get into some of my own thoughts of how one might handle the testing deferral issues -- was the EIA testing of non-deferred donors. We had a group of 3,229 donors. These are non-deferring donors. They were sequential, randomly selected in the greater Chesapeake-Potomac region that were tested as per the product insert of the New Market kit. Initially reactive was 21 out of the 3,200, 0.65 percent, and then repeat reactive there is 11 or 0.34 percent.

Now when we did that we thought, well, the repeat reactive rates are not too bad. That is about 99.6 specificity rate, if one assumes that all those are in fact false positives. Keep in mind these are donors who were not deferred. They were acceptable as blood donors.

What was interesting is when we went back to the regions and said who are these 11 individuals. Who are these 11 repeat reactive donors?

(Slide.)

Much to our surprise, or not to our surprise, the first two had no travel history. Now these are just based on information on the BDR. We are in the process of having these donors fill out the questionnaires, and we are going to get more information from them so we know exactly what are their travel history or their life histories, and so forth. This is just off the BDR. Two of them had no travel history. Two had reported European travel history only. One traveled to the endemic --

(Laughter.)

--- we are safe from the viruses. One of them traveled to an endemic area of India for a long period of time. That is a malaria endemic area. Two in fact were born and lived in Africa for extensive periods of time; and then what was really the clincher, four of them previously had been diagnosed or treated for malaria greater than three years ago or had lived in Africa -- and had lived in Africa.

So among our non-deferred donors the New Market kit was very good at finding donors who had malaria before, which raises issues about the question itself, who these donors are, how long antibody titers are maintained, what those antibody titers mean. Now try to understand what this means, what the test is telling us.

(Slide.)

We sent the samples down to Marianna. Are you still here, Marianna? We sent the samples down to Marianna for some additional testing, and she ran her IFA on these. Normal cutoff at the CDC for IFA positivity is one-over-64. She shared this morning she runs all four species. I won't go into that. We also sent her 10 negatives, and these were actually donors who tested EIA negative, so they are somewhat slightly selective, but they were negatives out of the non-deferred population. Out of the initial 11 at the one-in-64 level, Marianna found two that were positive. One was falcip and one was vivax. She

also found one of our negatives as positive. It turns out it was ovale, which may not be picked up by the New Market test kit. We don't know.

So we weren't sure. We were a little disappointed. We thought we would get more positives, but that okay. We don't what a lot of the tests mean and how they compare. But in discussions with Marianna, and actually she is the one who proposed this, if you went down one dilution and you looked at a one-to-sixteen dilution we found that two out of the 10 negatives were positive, but eight out of the 11 were repeat reactive. I mean, eight of the 11 were positive by IFA. Now let me come back to this and what this perhaps might mean, and what it is really showing is that her IFA and the EIA seem to be picking up something similar. We don't quite know what it is, and the negatives were quite good.

PCR's we did in our hands, and nested PCR was all negative. Again, not surprising considering these are probably very low levels of infection. These donors were probably infected many years ago. So what does this mean? What does it mean to have 11 repeat reactives, perhaps a fair number of positives?

(Slide.)

I think what this says and the implications at least from my perspective is that there is a significant number of donors with past malaria exposure. We know that. There are people, and Sue showed the numbers as well, people who admit to having malaria exposure. What we are not seeing is those who are beyond that three-year period who aren't captured by the question. So these aren't captured by the travel history question. What I would suggest, that perhaps these have some type of long-term antibody titers.

(Slide.)

And another slide you have already seen, Marianna's slide once again; and she pointed this out quite clearly before, that individuals who were recently infected with experimental infections within 12 months their antibody titers rapidly decline. Those from perhaps Nigeria or those who were prisoners of war who have repeated exposures, the titers remain elevated. But here she was only out 18 months, and I think what is lacking is where would they be four years from now? They would be way over here somewhere, and I would expect these antibody titers probably to wane with time. So what perhaps what we are picking up with the New Market kit and at a one-to-16 dilution are maybe antibody titers from people who have malaria, but they way down. They are dropped considerably down.

But that raises the question that what does that antibody titer mean in someone who is non-deferred. Are they semi-immune? I don't know. They have antibody. They have had exposure. We know that. But we also don't know what that relationship is transfusion transmission. Certainly those individuals who are semi-immune are the ones who seem to be involved in most of the cases of transfusion transmitted malaria. We also don't know what these individuals' infection status is. They have some type of antibody. Are they still infectious? Has the parasite cleared? I am not sure of the answers.

Of course then the caveat is, as we talked this morning, we have seen very few transfusion cases in the last 10 years. So even if we have these individuals in the pool, what does that mean? What is their implications? And I don't have the answers there either.

(Slide.)

But let me walk through some approaches perhaps to enhancing the availability of blood since that is what we are talking about, and some of these have been talked about, too. First of all, if you have had malaria -- and I think these are really the primary core of the blood safety issue, these individuals who had malaria. Certainly, as Sue suggested, we could just go and permanently defer them. That is very easy to manage. It is already done in several countries. There is also a precedent with babesiosis. We defer anyone who has had babesiosis indefinitely. I am not sure I necessarily agree with

that either, because in many ways babesia donors mimic some of these malaria donors. Some individuals will rapidly clear, antibodies are gone. We find some babesia donors have elevated antibody titers for years. What does that mean? I'm not sure. We could test and defer those with persistent titers for a defined period, or we could accept those with baseline titers, those who have returned to zero.

(Slide.)

The second group is the residence group, and these are probably the secondary corp of blood involved in the secondary corp of blood safety issues, and certainly this group has a very close relationship to the "had malaria" group. I think Sue proposed that as well. Probably the ones who have had malaria are the ones that had long-term residence, so they are really closely related. One could go with a limited deferral, but we have talked about that, a limited deferral due to question strategies. It is really difficult to manage, and we have talked time and time again about the issues of these questions and the difficulty with specificity and sensitivity issues. Again, you could go through the same type of testing procedure.

(Slide.)

Lastly the bulk of where the problem arises today and I think the core of the blood availability issue is actually our travel donors. We could ask a limited deferral, but these questions really I think are not very effective, and I think it gets back to that princess and pea phenomenon. We will keep adding new questions. Each year we will find something new to layer on top, and we will just take more donors out of the pool Again, once again, we could go with a test.

(Slide.)

So then to summarize where I think we are, I think we are really at a unique opportunity to address the malaria issues, and I think by having this meeting today I think it is really a great opportunity to realign the blood availability safety dynamic that has gotten I think somewhat skewed. We spend so much time layering this questions that we have lost track of what we are really here to do.

I think the other important thing that will come out of this meeting and these discussions actually is the fostering of additional research, and certain topics like immigration and donor demographic issues. This has been brought up earlier, too, about how things have changed in the last 25 years, how we have become much more global, and we have to address issues of travelers as well as immigration issues. We have to work on the retention and reentry of deferred donors. Very few of these deferred donors every come back, and I would even question -- I am not sure, but maybe some of the Europeans can provide some data as well -- if you defer a donor for three or four months, are they still going to come back for testing, or are they gone as well?

What is the rate of infectivity in travel deferred donors? Hopefully in our own study we will have some data before too long. What is the significance of long-term antibody titers? Are the parasites there? Are they capable of transmitting the infection? And lastly, what about the declining rate of transfusion transmission? What does that do, too? All these together will hopefully stimulate test development, too, although I am sure New Market would rather have the market all to themselves.

(Laughter.)

I will stop there. Thank you.

(Applause.)

DR. DODD: Thank you very much, David. Again more to think about. Now we will end up with -- I supposed I was going to call it a double whammy, somebody who represents statistics and the FDA. Estimated risks and benefits of blood donor screening for malaria compared with donor deferrals for geographic exposure, Steve Anderson. Thank you.

(Adjusting equipment.)

Estimated Risks and Benefits of Blood Donor Screening for Malaria Compared with Donor Deferrals for Geographical Exposure

by Steven Anderson, PhD, MPP

DR. ANDERSON: All right. I think everybody is getting a little tired out because it is the afternoon after lunch, so I am going to try to keep this as interesting as possible. My name is Steve Anderson as mentioned. I am from the Office of Biostatistics and Epidemiology, and I will try not to bore you too much with equations, numbers, et cetera, although I think some of the numbers we will show you from the results from the model will shed some light on some of the discussions going on now. So we are here to share some of the modeling work that we have done recently and for evaluating the risk and benefits related to the possible implementation of a malaria blood screen test.

(Slide.)

So before I launch too much into the probabilistic modeling I just wanted to emphasize what the current US policy is. It includes deferral on travels who are from malaria endemic countries in the last year, immigrants of course in the last three years, and donors that had malaria and were asymptomatic for less than three years.

The issue here for this modeling exercise that I am going to talk about is this really is sort of the baseline example that we are using, the current policy, and then what we are going to do is use our probabilistic model to evaluate a couple of different scenarios. In addition to the current policy, we are going to weigh the advantages or disadvantages of a universal nucleic acid test and testing scenario. Or using another scenario, which would be universal antibody testing.

I just wanted to mention that these are independent of each other. So don't assume that we have the current policy and on top of that we are doing either of these. These are all independent, so we are talking about current policy, or NAT testing, or antibody testing. So assume independence here.

(Slide.)

Just to sort of refresh people's memories or introduce them so be it to probabilistic modeling, the issue here for us is that rather than using single numbers or point estimates, we are employing statistical distributions for some of the input data and parameters. What we are doing there and the reason we are doing that is we are trying to represent some of the uncertainty of that data that are going into the model.

Just sort of a thumbnail sketch, you could spend a whole day on Monte Carlo methods, but we are using the sort of simulation methods to choose a value from each of the distributions that we set up in the model. We do that for one iteration. It generates an output, and then we keep running the model. For instance, we have run the model for this particular exercise 10,000 times, and what we get out of that is one single aggregate output distribution. I am going to be showing you the results of that distribution in some of the final slides.

(Slide.)

Another issue for us when we are dealing with probabilistic modeling is of course the underlying uncertainty in some of our data. Where does that come from? Well, it arises from the lack of or limited data for a particular input parameter. We also have a tendency to employ assumptions in our models occasionally when there are no data, and that adds to the uncertainty of the model. I am not going to go through all of the different types of uncertainties that are in the malaria model, and there are many more than this. This is by no means sort of a comprehensive list, but it just gives you kind of the idea of some of the things that add to uncertainty.

We are proposing some theoretical or hypothetical test sensitivities NAT, as you will see in the model, and for the antibody testing. But again, that is going to be determined by whatever policy or testing strategy that could potentially be introduced; and the advantage I should say of this type of modeling approach is that it can be very easily changed in the model and the model updated. What we are doing then is representing this uncertainty in our competence intervals about the means estimated outcomes, and you will see some of that in the final results tables near the end of the talk.

(Slide.)

I am not going to go through too much of this. I think you already know a fair amount just from previous talks from Dr. Parise and the CDC people. I think again just to emphasize, most of the cases seem to be imported. The range I believe this morning that was given, we are using 2004 data in this particular model, but the range could be 1,000, too. I think 1,500 was quoted this morning. An important thing, too, for the model is 50 percent of the cases were plasmodium falciparum. Again, that is adjustable in the model. We used I believe 63 percent at one point in the model, so this is a changeable factor. Again, we have other species represented in the modeling as well, and I will show you some of examples of that later in the talk. Just to remind people, the transfusion-transmitted malaria rate is quite low. We are estimating, and these are the traditional estimates, 0.25 cases per million units collected.

(Slide.)

Just to sort of keep the talk moving, I thought I would just outline some of the risks or costs. We sort of bunched these together because costs tend to be viewed as risks as well. But you can sort of glance down these and see, and I think the one important thing is to emphasize I think that -- Dr. Stramer mentioned this as well -- there is cost of testing the entire blood supply, cost of retesting units, the loss in blood donors and blood units is a critical thing, cost for recruiting donors, et cetera. But there are benefits potentially to be gained by alternative strategies, and that might be you may gain additional donors, you may detect additional malaria units, et cetera.

(Slide.)

Again this is by no means a comprehensive list, but just a few things to keep in mind.

(Slide.)

The overview of the model and the model components. The first component really we look at the size of the donor pools, and really what we are doing is fractionating the donor pools down to really what the lowest populations within that donor pool and the high risk populations. So for malaria it seems from the talks today that the key high risk groups would be the subpopulations that were travelers in the last year or so and then immigrants in the last three years or perhaps longer.

So those are the high risk groups, but then we are also keeping track of all the remaining donors in this model as well. We want to learn about the prevalence in those high risk donor groups. That is how many malaria units are generated, and then the type of screening method that is going to be used to eliminate any possible malaria units or eliminate as many possible units before they head into the blood supply. The questionnaire, really there are questions about the effectiveness of that. That is a focus of the model as well, and then for testing we are focusing on sensitivity because that is a driver for risk as well.

Again, the outputs in the model are outline here in yellow at the bottom. The risks are costs. It will feed us outputs for those, and then feed us of course outputs for benefits as well.

(Slide.)

I have the advantage, too, that most people have talked about a lot of the data in this slide as well, but let me point out some of the things about this slide. US travelers to malaria countries estimated at 27- to 29 million, and what we do is multiply those numbers by 60 percent, which is the population qualified to donate times five percent, the donation rate of the general population. Then you get that as an estimate down here of the donors per year that travel to a malaria country estimated at about 880,000. I am showing these as simple point estimates right now, but we do have confidence intervals about these. But just for a simple example I am just showing you the point estimates right now.

We have data that shows donors that are deferred by questionnaire, 150,000. If you subtract these two then you can get those donors that are self-deferred, and that is just a total subtraction based on the simple data that are available.

(Slide.)

Slide sort of two and three, the second and third components of our model are sort of merged, so I am sort of mixing prevalence again with the effectiveness of testing or the effectiveness of this questionnaire screening in this slide. Our interest is the effectiveness of the questionnaire screen for current policy. We estimated that at 95 to 99 percent, and I think the one thing to remember is that these sort of interventions effectively lower the malaria prevalence in donors and prevents fewer donors from donating perhaps or prevents any sort of positive donations from entering the blood supply. So it is a critical portion and element to consider here.

Okay. As model outputs we estimated in the model again. I am going to show these in summary form in a table. Potential mean malaria donors per year were 42, so a fairly small number, and that works out to about 71 donations. Three malaria units that potentially enter the blood supply, possibly more with the uncertainty about those.

(Slide.)

Testing scenarios. Moving onto universal nucleic acid testing. So specifically this type of approach means that we are testing all donors with a nucleic acid type of test. Specifically the highest donor risk to consider, though are the these traveler populations that travel to malaria countries have been back less than year, and then the immigrant populations that have been in the United States less than three years.

We assumed in this particular model that there was a one-month window period for the donations, for these donors and donations, and that means that malaria would not be detected in that one-month period. For all other donors we essentially assumed there was no window period, and our hypothetical test sensitivity was 99 to 100 percent, although I think there are a lot of issues and I think that could be updated given the sort of concentrations of the parasite in blood. Sort of challenges to detection using this methodology.

(Slide.)

All right. Moving on then to the universal antibody testing. Again we are testing all the donors that we collect blood from, but specifically we are interested in the traveler and immigrant populations once again. Again for travelers that have been in the country less than three months that travel to malaria countries we assumed a three-month window period for the antibody test. Again, I think based on the information and discussion this morning this could be reduced probably to about one month, but we used a conservative assumption here of three months. Then greater than three months we assumed just the test sensitivity, whatever that may be, and I will talk more about that in the next slide. We assumed no window period for immigrants less than three years and all other donors no window period.

(Slide.)

All right. So just continuing to talk about if a test were going to be implemented what would be the sensitivities. These are the hypothetical test sensitivities for an antibody test that we were looking at perhaps for falciparum. The range would be 94 to 99.5 percent. For vivax a little bit less, and for other strains, ovale and other strains, it would be ranging from 50 to 75 percent.

Now we have information on where people in the United States did travel, so we could weight these prevalences based on where individuals traveled because certain species are more prevalent in certain regions. So for instance, travelers to Africa 82 percent of infections are P. falciparum, fewer are P. vivax, and seven percent are others. But you see for Asia, America and others there is a heavy emphasis and predominance of P. vivax to be concerned about. So if you weight it based on these, the weighted test sensitivities would come out more like this. Again, we have ranges about these, but these are the types of things that you can do with the model. Again, this is hypothetical, and this could be adjusted for whatever the test sensitivity is of whatever test is developed and used ultimately in screening if that be the case.

(Slide.)

Okay. Now just to look at the potential risks and benefits of the current policy. Just to orient people on the slides, we have risk and we have the 5th and the 95th percentiles expressed for many of the numbers below them, the benefits, and then going down the left side other current policy. We are considering individuals that were self-deferred, questionnaire-deferred, and then we are adding those together in the bottom line.

So starting over here with malaria units, we moved the benefits. Additively we are eliminating about 67 units predicted by the model. Again most of those are self-deferred, but there are a few, 10 percent of those or so, that are being deferred by the questionnaire. Malaria units not removed, again it ends up being about three predicted by the model with a range of one to five. Donors removed, again about 150,000 with the questionnaire and slightly more self-deferred, adding up to about 880,000.

(Slide.)

All right. So taking the current policy and going along the bottom line of the previous slide it is represented here, but then comparing the results for NAT testing and antibody testing. You can see the malaria units removed. It is very similar, 67, 66, 61 predicted by the model. So in some cases the antibody testing may not be perhaps as good as current policy. Again, we can adjust the window period, and that may adjust these numbers, adjust sensitivities. That may have an effect on these numbers. The big issue for benefits, again are potential donors gained. Basically you probably gain from those in the current policy that are deferred. Here it was 150,000 plus 730,000. So we would guess you would probably gain donors anywhere from 150,000 donors to perhaps up to 880,000, which the total with self- deferred and the questionnaire-deferred.

Again, I think costs really are less of an issue for the current policy. There may be costs for additional recruiting, but generally other costs are thought to be low. But then for NAT testing and other types of tests you are going to be testing 14 million or more units, so that could be a significant issue. Retesting of units will be an issue as well.

Probably the biggest difference again here besides, you know, the donors gained are the removed. So from the population again these 880,000 for the current policy, but then for NAT testing it is much fewer. Perhaps 40 that might test positive, so that would be viewed as a benefit. Perhaps a few more with antibody testing. Again, these numbers are going to be dependent ultimately on what type of policy is implemented, so the numbers could vary depending on any changes in this policy.

(Slide.)

All right. So key uncertainties, overall there is uncertainty for a fair number of the components of the model for many of the model inputs. Malaria prevalence in donors who travel to -- with a travel history of less than a year, I think we need more information on that, so that is a fairly significant uncertainty portion of the model, so a lot of uncertainty associated with those prevalence estimates. There is also variability in species by region over time. Today I think, you know, falciparum is really the predominant species. That may change in the future and it may be another species. So over time that certainly may be an issue in these types of models. Sensitivity of tests that would be used again, you know, we put up hypothetical examples and estimates and use those in the model, but again we could just change those depending on what type of testing or testing regimen was used -- but a fair amount of uncertainty.

(Slide.)

Conclusions from the malaria model, basically the current policy many donors are deferred, about 150,000 perhaps at the door Perhaps as many as a total of 880- if you included self-deferrals, 880,000 if you include the self-deferrals. With antibody testing or NAT testing for that matter the numbers that are deferred actually would be even fewer, but again you have to weigh that against the costs, and those are significant costs of testing and retesting more than 14 million units a year. Again, but the testing scenarios, there may be a net gain of at least 50,000 it would seem like based on the current deferrals, and that could go upwards of 880,000, although I doubt it would reach this as Dr. Stramer referred to in her previous talk.

(Slide.)

Some further conclusions, I think definitely you need an explanation of cost for each of these different options and different policies that might be considered. Validate assumptions in the model with data on the test sensitivities based on real life results for each of the tests, and then again complete the model and do a peer review of the model, the assumptions of data used, et cetera.

(Slide.)

And I wanted to acknowledge most of this work for the modeling was done by colleague Hong Yang in our group with help from Sanjai Kumar and OBRR, and then a fair number of the OBRR staff contributed as well. Thank you.

(Applause.)

DR. DODD: Thank you very much. That was really -- stay up here if you would, Steve. I think we have got 15 minutes for some discussion with questions, questions from the floor. Then we are having a break and we are having a bigger roundtable, so if I could have the speakers along with Celso, Louie and Steve.

Session III Panel Discussion

DR. DODD: Okay. Well, we have got a couple of questions to speak to, but we will follow on and we will take questions from the floor, if there are any. Yes, Sanjai.

DR. KUMAR: I've got two questions. The first one is for Dr. Leiby. Where you are here? In your testing of about 3,000 samples you were showing reactivity of 0.34 percent for whatever reason. I mean, looking in --- you found that people had a history of malaria that traveled, but assume that -- let's assume that the current safety for transfusion-transmitted malaria, the safety is not acceptable but satisfactory, one case every-other year. So the question is whether this antibody testing will improve blood safety, and that remains to be seen, and if your numbers are showing 1.2 percent of deferral across the board how much do we gain? There will be less than the one percent gain will be there if your background is coming to 0.34. I mean, given that background may have reasons why it is there. It may not be true background, but

nonetheless, those donors will be deferred. So the gain may not be as much as, I mean, one may have assumed.

DR. LEIBY: How many donors do we defer now?

DR. KUMAR: Well, I mean, the numbers that we hear from you guys, 1.2 percent.

DR. LEIBY: Okay, and then how many with the test?

UNIDENTIFIED: You do the math.

DR. KUMAR: Well, it will come down to less than one percent. That is what I am saying.

DR. LEIBY: It is 1.2 versus 0.34, right?

DR. KUMAR: Yes.

DR. LEIBY: That is a big savings. Now the assumption then is the safety margin the same, and that 0.34 keep in mind were donors who were donating blood already. They were non-deferred donors and which there are some question about perhaps their suitability. What we have, don't forget, is we have a question strategy in place. We have seen very few transfusion cases. Is that due to the strategy that is in the place? I don't think anyone has tested that.

DR. KUMAR: Well, I guess the self-deferral could be another issue there.

DR. LEIBY: And self-deferral as well as Alan has raised, and I think some of the numbers. I hadn't seen numbers that large before. That is not my field, but self-deferral numbers as well aren't included. So I think in the long run if you had a test that you could assume its sensitivity and specificity, you could guarantee safety. I understand that is critical to the FDA, and I understand that. If you guarantee safety of the blood supply to get that many donors back --

DR. KUMAR: Well, that remains to be seen I guess.

DR. LEIBY: Right.

DR. KUMAR: Okay. The other question is for Dr.ÿStramer. The countries we defer permanently for a history of malaria, they are doing it only when they are having antibody testing now, so nobody is deferring without testing for antibody. So if we work on that model in the absence of -- if you work with that model without any antibody, without any testing in place here in this country, the donor loss in terms of anybody who was born south of -- Saharan Desert in the African continent will probably run into millions of permanent deferrals, so just to keep in mind that policy.

DR. STRAMER: You are talking about those individuals who had malaria?

DR. KUMAR: Yes. Yes.

DR. STRAMER: We have only deferred 492 over six years.

DR. KUMAR: No, no. What I am saying is the model you presented. Defer anybody --

DR. STRAMER: If we defer people who had malaria?

DR. KUMAR: Yes.

DR. DODD: No. Anybody who had been born in a malarious area or had lived in a malarious area. I think your suggestion was that perhaps you could defer all of those permanently.

DR. STRAMER: Well, I said that is one possibility and one thing for discussion. If their demographics are similar to those who have had malaria and we are talking about safety, I think it is one of the things that should be discussed.

DR. KUMAR: Well, that is what I am doing, trying to discuss --- safety and availability. So some of the safety is rather satisfactory right now. The availability probably would not help.

DR. STRAMER: Well, if we are doing testing we have two options. We could test those individuals.

DR. KUMAR: Absolutely.

DR. STRAMER: Right, or we don't have to. I mean, those are the two, we need to look into those. I just presented all possibilities. Granted we may have more deferrals.

DR. DODD: Peter wanted to speak to that, too.

DR. CHIODINI: Thanks. I just wanted to say something on the back of this issue about the diagnosis of malaria. Diagnosis of malaria in the tropics is notorious inaccurate. It is either made on clinical grounds, and that may be 50 percent accurate, depending on the prevalence in the area, or even if it is made on slide diagnosis again it can be very inaccurate. So I think it is not helpful to get too hung up on diagnosis of malaria versus qualifying as a resident of an endemic area, because a resident has to get semi-immune by getting malaria. All you have got is one person who has been told they had malaria. So I think this matter of lumping them together in terms of what you are do with them, we are not allowed to at the moment, but we would like to. Certainly they have very similar risk factors. They are very similar to groups of people who said they have had malaria versus the semi-immunes.

DR. KUMAR: But I guess one thing you have to keep and bear in mind there, that children who are born in sub-Saharan African, and if you believe the data from the people who have done field malaria --- all their lives, most of the people believe that there is not a single child in many of the countries of sub-Saharan Africa who has not experienced clinical malaria by the age -- by the time they are one-year old. Just keep that in mind. So we don't confuse it between the people who had history of malaria --- born there. So I guess it may be a fair assumption that anyone who was born in that part of Africa had a clinical malaria, and obviously by the time -- I mean, they grew up there. It certainly would be a fair assumption to make, that is all.

DR. DODD: Thanks, Sanjai. Jed.

DR. GORLIN: A quick question for Steve. We heard from Dr. Parise that two-thirds or so of transfusion-transmitted malaria the questions failed. I wondered how you included the failure of questions in your model.

DR. ANDERSON: Basically that seems to just be -- I mean, Alan, you may be able to answer that better than I can, because that is the effectiveness of the donor screening essentially. Because most of that data we have gotten from Alan in the past, the 95 -- we used a 95 to 99 percent effectiveness of screening.

UNIDENTIFIED: Yes, depending on the complexity of the issue generally we have estimated between 85 and 95 percent or higher.

DR. ANDERSON: But I think, you know, the data presented here, we could adjust that, you know, based on, you know, failure of questions further with that type of information in hand. It would be probably lower our estimate at least a little bit, down to 90 or perhaps lower.

DR. DODD: Alan.

DR. WILLIAMS: Questions for David. I was intrigued by the control data obviously, and I am searching for explanations.

(Laughter.)

DR. WILLIAMS: To what extent has this EIA been qualified in terms of cross-reaction with other protozoan, parasitic infections, bacterial infection, as you might expect someone from -- you know, spent time in developing countries, might have had a lot of exposures?

DR. LEIBY: I think Nigel could speak to that. But, I mean, the major concern I would think would be with babesia, and we are actually -- I mean, as I said, this data we have just generated and one of the things we are going to do is test those 11 donors for babesia for as well since we can easily do that since babesia and malaria are the -- you know, they are the same species -- not the species -- same groups of parasites. You would expect that to be perhaps the greatest chance. As far as other cross-reactivity, I am not aware. Nigel?

MR. APPLETON: (Away from mic.) We haven't done extensive testing. We wouldn't know what to look for. We tested against --- for instance is no cross-reactions --- and whatever infections there are in the general population at any given time. There doesn't seem to be any cross-reaction, so I think we are using recombinant --- generally fairly safeÿ---.

DR. LEIBY: Peter might be able to comment on this, too.

DR. CHIODINI: No, we didn't put the assay through panels of sera for things like --- and so on, but we wouldn't expect cross-reactivity with those. I would agree with the comment about babesia. They are very similar organisms and there is cross-reactivity certainly in the IFAT. That is very well documented, and I think it is reasonable that we could -- it would not be unexpected, let's put it that way, if it cross-reacted with babesia, but fortunately that is a relatively small population group.

DR. BIANCO: But I don't recall if you said that in your presentation, but what is that background? What number would you expect from that assay doing an experiment similar to that done by David?

DR. CHIODINI: Okay. Well, we put 880 sera through from non-tropical area donors and didn't get a positive. We for the previous assay, the one that failed, the positivity rate was 0.45 percent. So had it been a similar value that would have been acceptable for practice. We would then of course try to analyze what they were due to, but certainly on this latest ELISA no positives in 880.

MR. APPLETON: Can I answer that please?

DR. DODD: Yes, use the microphone please.

MR. APPLETON: Every lot of cases we produce --- several hundred randomly selected donors, plasma and sera. We have yet to find a positive in any of those.

DR. DODD: Those of you who went out at lunchtime, you realize that we can call this a non-tropical area here.

(Laughter.)

DR. NAKHASI: Thank you, Roger. I just -- Hira Nakhasi, FDA. I just want to, you know, focus the panel on these questions which are there. So in respect to that, I would like to ask three separate questions. The one, first one, is in regard to Peter. You presented some scheme of IFA or IFAT you called, EIA and both. What would be the ideal way of testing it? You know, which would be --- pro's and con's. Could you emphasize what would the ideal way of testing, screening or confirmation of the test?

DR. CHIODINI: You mean as a strategy of deploying serology?

DR. NAKHASI: Yes. Right.

DR. CHIODINI: Well, I am not saying it is ideal, but I would advocate first of all keeping it as simple as you possibly can. Use as few criteria for serological testing as you can with one particular time frame. What you don't want is are you a resident, did you have malaria, and what country was and what part of the country. The simpler, the better. So our bottom line recommendation for Britain which we took back to the Council of Europe was that if you are going to test serological go at six months. That allows for a window period, it allows for the early cases in the travelers. Test at that point for all comers. We were, as I say, instructed that we had to have a three-year deferral for a past history of malaria, but I think that will be revised.

As I said, very simple, and then do you use one assay or two assays? We debated that and decided that you got very little --- confirmatory assay. So if they are repeat reactive, if they are reactive in the transfusion microbiology lab, they are retested. If they are again reactive they are excluded from donation. If they are antibody-positive they are offered if they wish to come back retesting at three years. I don't know, because it hasn't

been going that long, this particular assay, how many will come back, but they are offered that.

What we also plan for donor care as much as for answering David Leiby's question, is those who are strongly positive in the EIA we will recommend NAT testing, because if we can find parasites it is beneficial to them that we clear them, but that is for donor care.

DR. NAKHASI: Thank you. This question is for Sue. With regard to the Australian study, you said they tested a person, if it is positive they keep the plasma, and then, you know, so -- and they keep on collecting the plasma. Did you find out from there that there is any problem of contamination? Because with these positive samples that are hanging around, you know, you were saying 15 out of 50 they found out, and then your suggestion was just forget about it. But the question is, you know, in our setting here if we have lots of them around for example, what would be the problem with contamination?

DR. STRAMER: Well, they have really until this last month have only had one that has shown any reactivity on an antigen test or PCR. In that case the donor was negative by PCR. So the question is are these antibody only with very low levels of parasitemia. They have problem. They box them up and ship them off to their fractionator, and that has been their practice for a very long time if not forever. The don't consider --

DR. NAKHASI: Okay. So they don't have any problem with that?

DR. STRAMER: No, I specifically asked Clive, "Well, what did you do with the one plasma unit from your Indian immigrant who was repeatedly dipstick positive?" And he said, "There is no process to stop that plasma from being shipped." So all plasma is shipped to the fractionator and the methods used by the fractionator if there is any residual parasites will be inactivated.

DR. NAKHASI: Okay. This question is for the gentleman from France. You told us that there are a significant number of positive malaria cases, 7,000 or so per year in 2005 I guess, and yet we found only one transmission transmitted case. So why is that disparity?

DR. GARRAUD: Actually most of these people who are infected are Africans residing in France, and most of them are not candidates for blood donation, so that is probably the reason why. Nevertheless, we would like to have more Africans giving blood for a few type reasons, because we also have a lot of Africans. This is a topic which has not been addressed today, but this is something which much be addressed, especially in France I think. We have in hospitals a lot of people with varieties of phenotypes, especially African phenotypes which are not found among the French population. So we also to have a variety of blood donors to transfuse those people.

DR. BIANCO: Just what were the reasons why those donors would have been deferred if it was not history of travel to a malarial zone? What were the other reasons?

DR. GARRAUD: There is no other reason. I mean, they just don't come today because they are not used to and it is not traditional for Africans to candidate for blood donation.

DR. DODD: Okay. George.

DR. DAWSON: Very interesting data that David Leiby showed on the blood donors having the 0.3 percent sero-positivity, and what turned out is that your normal confirmatory test, your gold standard, could not confirm those at the 164 dilution. So I wonder if you have tested known pedigreed samples serially diluted to show that the ELISA is actually more sensitive than the IFA at one to 64. Then secondly, it may lead you to think you need a newer confirmatory test which perhaps may dissect the reactivity that you are seeing with three or four recombinants into individual reactivity so that you can say. You know, seeing reactivity to two independent gene products is a strong case for confirmatory when an IFA perhaps cannot confirm it. So have you done any of those types of studies yet? I know it is pretty early.

DR. LEIBY: Marianna just gave me those results last week, so --
(Laughter.)

DR. LEIBY: She got the sera last week, too. A Herculean effort by her just to get those done for me. I don't know the relationship between the EIA and the IFA, is more one more sensitive than the other, and I am not going to go out on the limb and say that. I think part of the issue here is let's assume for example that those four donors in fact had malaria and they are positive, and if they have had malaria for at least three years, it has been at least three or five years since they were infected, what does that type of individual look like in an EIA and IFA. I asked that Marianna that question, and she said the only data they had was the slide that she showed, too, which tracked donors out to 18 months. So what would a test look like or what should it look like picking up a donor who is that far out? Is there any significance? I am not sure either. That is something Marianna and I can talk about, comparing the two.

UNIDENTIFIED: I just think as you do these types of studies you are going to continue to turn over these type of samples and you will have to come up with some kind of algorithm to ---.

DR. LEIBY: Oh, sure. I mean, once you get starting --- we are going to raise more questions than provide answers I am sure.

DR. DODD: Okay. We are running out if time, but if we can get through these questions quickly I will listen to each of you at the microphone. This Chris and then that Chris.

DR. OCKENHOUSE: Okay. A quick question to Dr.ÿStramer. I thought you presented some very compelling evidence that it is not the individuals with malaria or residents, but the individuals who have traveled that are contributing to the large deferrals. Is that correct? Taking that into account, maybe 90 percent, what do you think is the real added value -- or Dr. Chiodini, of using malaria antibodies in that population? What are added value are we going to get to either retain those individuals exclude those individuals? They are non-immune travelers. They are not residents and they hadn't had malaria before.

DR. STRAMER: Well, the assumption is if we did antibody testing on them and they were negative as the testing-in model show after the window periods that are used in the testing-in models, that these individuals have not been exposed to malaria, they are not infected with malaria. I think the larger issue is trying to avoid the point we are getting to that we have to test these individuals. What do we do with the questioning process to streamline it and avoid -- it is not only the donor deferrals. It is all the post-donation information, the one in 2,000 that results in post-donation information, the one in 10,000 that is a donor-related error, that is we have accepted an eligible error, the one in 100,000 violative BPD's we file. So there are a lot of other costs associated with the questioning process itself. It is not just the deferred donors, although that is most directly related to blood availability. So there are a lot of advantages, and what we should or could or do today is looking at other creative ways to maintain safety, but trying to bring about better message to these donors and increase availability. So I think antibody testing in those populations would be very helpful. A testing-in model in our system would be far to complex.

DR. DODD: It is going to be hard to tell if it has worked if you only expect about 50, you know, malaria donor in a year though. I mean, it is going to be hard to assess the validity of your approach. But I think your hidden suggest was let's just take them anyway. I think that is what I am hearing.

DR. OCKENHOUSE: That is exactly right. I like that.

DR. DODD: Yes. You have been saying it consistently all day. I like it.

(Laughter.)

DR. OCKENHOUSE: If you are making an assumption that a negative -- most of those could be negative, right?

DR. DODD: Yes.

DR. OCKENHOUSE: You are adding on another layer, another blanket of testing, and you are not increasing safety.

DR. KATZ: Roger, can I make a point?

DR. DODD: Yes. Please do.

DR. KATZ: One thing that came out of the data, it has been emphasized a lot, but it has been in a couple of presentations, is that the risk come from -- if there is risk from travelers it seems to come from travelers -- not all countries that we defer for have equal risk, and it seems like the risk is highest from Africa per traveler. Now clearly there are a lot more deferrals for risk to Mexico, and I guess you would have to build a model around that, so even at a lower risk there are more people. But it seems to me highly unreasonable to be extending this personal opinion now to be having the Mexican travel deferral when we have had only six or eight imported cases, clinical cases reported to CDC. I really think that deserves -- I mean, there are probably a lot of subtleties about this that have to be looked at, but I think we could take care of a lot of the deferral problem if we didn't include Mexico as a malaria risk area, even though malaria occurs there. I would encourage, you know, further discussion on that. I realize we can't answer it here.

DR. DODD: Well, you just ---.

DR. KATZ: Well, yes. I mean, you guys have heard me say this for years. Give me back Mexico. That is all I want is Mexico.

(Laughter.)

DR. KATZ: We are hemorrhaging blood and it is silly, and I think Sue's numbers are quite striking that if you multiply the numbers out of there, recognized cases versus the number of travelers and numbers of deferrals, about an 8,000-fold difference I am guessing in the risk of the sub-Saharan donor versus the Mexican donor. Well, that is fine, but -- and that is the simplistic approach, and maybe you could give me back the Bahamas soon and --

(Laughter.)

DR. KATZ: Everything but --- and some other places in the Americas, and just leave me alone and let me draw donors. That would be fine. The other thing that has been mentioned a couple of times and hasn't been emphasized is there has to be market for the assay, and in order to have the market for the assay the sales of the assay have to satisfy the manufacturer that it is worth it to jump over the CBER hoops, and so there is the crux of the matter. So at the end of it all, assay or no assay, give me back Mexico.

(Laughter.)

DR. DODD: Well, Lou, you just banned yourself for the rest of your life from visiting the Alamo.

(Laughter.)

DR. DODD: Chris, final one.

DR. GRESENS: Thanks, Roger. This question is to Dr. Stramer please. In your second-to-last slide when you proposed a potential algorithm for testing and historical screening -- and I am not challenging this at all. I just want to understand a little bit better. You mentioned for donors who previously have had malaria that potentially they should just continue to be deferred, no testing done, and for donors who were former residents of endemic areas that we either continue to defer for three months without testing or just defer forever. Is there a particular reason, or is it just that there is not much meat on that donor bone so to speak, or is there another reason why you didn't suggest testing earlier?

DR. STRAMER: Well, you could go either way with that group. I believe demographically as Peter has mentioned, too, these sorts historically

and demographically with those individuals who have had malaria -- perhaps not properly diagnosed when they lived in the malarial area, but at least from or demographics they look no different than a donor who has had malaria. Again, we are depending on a lot to say if had malaria that the donor knows that accurately, although you would probably remember from the symptoms.

DR. DODD: Okay. I am going to close it down now. Thanks very much. Sanjai, a 15-minute break, is that still okay? Okay. A 15-minute break, and then we will get down for real meat for the business and Jay's guidance.

(Whereupon, a break was taken at 4:13 p.m.)

DR. KUMAR: I know everybody is very tired, but the sooner we start the sooner we finish from here and go home, yes? So we --- from what we have on the agenda. I am just dying to acknowledge the people who helped us to put this workshop together, and after that Professor Chiodini is going to have a brief presentation about the --- standards for NIBSC, and that is an issue that is becoming more and more important to all of us. So after that Dr. Epstein will go to moderate this session, the last session. That is the roundtable discussion. Okay. So I will be very quick.

So Dr. Jerry Holmberg, he is from the Office of Pubic Health Sciences at the Department of Human Health Services. He is here, and he was very generous to provide us partial financial support for this workshop, and also his office is the cosponsor of this workshop. Dr. Tom McCutchan from NIAID, he is here also. He generously provided this conference facility. Our CDC colleagues, I don't know what I would do without them. They always respond to my frantic calls for last-minute help. Our DOD colleagues, Dr. Ockenhouse is here. Several members of AABB, who were very helpful, and many of them served on this program committee panel. Jennifer Scharpf, she is Associate Director for Policy, has been extremely helpful, going beyond the call of her duty. Rhonda Dawson, the administrator the workshop and also she provided a lot of logistics, a tremendous amount of help. Olga Maximova from Dr. Asher's lab, she helped me with the design of the poster you saw. Bobita Mahajan from my lab was tremendous help, Dr. David Asher. So although we have a transcript service here, Dr. Asher is generously being the semi-official reporter of the meeting, and now I said it in public so he has to help me. Now he is obligated. And then in the end, but foremost, the program committee members. They have been extremely helpful and they are the ones who set the agenda of topics that were discussed here and helped us finalize the various speakers here in the workshop. So it was not just us who came up with the idea of what should be discussed and who should be talking here. It was the program committee and many of those members are here today. So with that, Dr. Chiodini, it is all yours.

WHO International Standards for P. Falciparum
by Peter Chiodini, MSSB, PhD

DR. CHIODINI: Thanks very much. I shall be very brief. This really should be presented by Sally Baylis from NIBS, but the have a major inspection of NIBS going on as we speak -- or probably it has just finished. And so she unfortunately couldn't attend this meeting, so I am kind of doing a Dan Rather and I am reading this on behalf of somebody else. So this is a study where we looked at producing an international standard for nucleic acid testing for plasmodium falciparum, which I think you will agree is probably long overdue.

(Slide.)

Now, why do we need it? Well, first of all, if you survey parasitology labs they all tend to have different assays. My own lab has the smooney* method that was referred to earlier plus the padley* method which was developed later, but many of the other labs have their own in-house assays, and there is no agreed standard for sensitivity. In transfusion practice I think it could be used to examine those who have been deferred for positive antibody

testing as has been discussed. Investigating transfusion-transmitted infection I think it is very valuable, and also it will validate assay sensitivities. Diagnosis of malaria is an obvious one, and we already heard about the use in vaccine studies as well, so I think the case is quite clear.

(Slide.)

So how did we set about this? Very rarely it is necessary to undertake and exchange blood transfusion to treat severe falciparum malaria with very high parasitemia. It happens in my hospital probably no more than about once a year now. But a few years ago we had a patient who underwent exchange transfusion. We routinely take the equivalent of six units and the whole blood exchange, and the bags of blood were then made available to us and the spouse of the patient gave consent for the material to be used for research. The patient was under a different clinical team from my own, so there was no conflict of interest. Having got permission, the material was then sent to David Padley at NIBS, and the parasitemia was estimated at 10 percent.

(Slide.)

Four preparations were prepared. Sample AA was freeze-dried blood from that preparation. BB was a liquid preparation of P. falciparum cultured material from the London School of Hygiene and Tropical Medicine to about 10 percent. Another of the bags of blood from the patient had seven percent parasitemia, and that formed another sample. Then it is three-fold, a 1,000-fold. That is three logs dilution of sample CC as a reagent for routine use.

(Slide.)

These are the performances in the light cycler, the amplification curves, and AA, BB, and CC are all clustered together there on the left, and the 1,000-fold dilution came up as you might expect after rather more amplification cycles. There was no loss of titer following freeze-drying of the patient material. Again, it was about 10 percent parasitemia.

(Slide.)

So then there was a collaborative study, and we literally contacted friends and colleagues that we knew that were in the malaria business of doing diagnostic work and asking them if they would be interested in receiving a sample of this standard. NIBS prepared about 2,000 ampules of the freeze-dried preparation AA and each laboratory was asked to participate, test the samples in four independent assays, 10-fold dilutions and half-log dilutions around the endpoint. From those labs we eventually got 20 data sets. Interestingly to see that only two were quantitative assays. The majority of the labs are still using qualitative assays. That I think is one of the things the malaria community will rectify before long, and the NIBS collated and analyzed the data.

(Slide.)

This is how the samples clustered. These are results from the lab, and you will see from the table they come out clustered around the 8.5 log area for the four. That is AA, BB, CC, and then DD, which of course is more dilute.

(Slide.)

This is the data perhaps in a more readily-accessible form. The mean as I say, about 8.5 logs for the first three preparations, and then because it is 1,000-fold less you are down by three logs for DD. That is the range and standard deviation, and you all have seen the range of course on the previous slide. My own lab is in there. I am not going to tell you which one, but our two different assays did agree actually really quite well, but there was a fair amount of cluster. Potency is relative to AA. Remember, that is the international standard. B and C were extremely close, and D was three logs below as you would expect as it had been designed as such.

(Slide.)

What about stability? Clearly if it is an international standard it has got to be there for some considerable time, and NIBS reckoned their 2,000 ampules have got to last 10 years, so it is freeze-dried and then stored at -20.

No loss of potency compared to the initial starting level after eight and 12 months at -20, and you will see of course a falloff at +45, but it is not obviously going to be designed to survive at that temperature.

(Slide.)

So what do we think? Well, first of all, we feel from that data that mean log 10 is 8.5 log 10 equivalence for sample AA and very similar for BB. The C -- sample DD, I'm sorry, has been coded as NIBS cod 04112 as the working reagent, with A as the international standard. We think it is going to be very stable and certainly will last 10 years in the storage conditions when it is freeze-dried, which it is, and the proposal is that this should be established as the first international standard for NAT testing with the NIBS code of 04176. It is proposed that the potency be defined as 109 international units per ml, and since this protein is batches of the equivalent of half a ml of the material that would give us five times 108 per unit.

Now we have submitted that to the World Health Organization ECBS, Expert Committee of Biological Standardization. It went in at the end of June, and obviously we await feedback and liaison with them as to whether further work is required and what the likelihood of it being accepted is. I think our position is it is very clear there is a need for standardization. Whether or not NAT testing is accepted for transfusion practice in terms of reducing risk to donors is another discussion. Whether or not it is, it is still needed for diagnosis, and I think it is high time that we of the malarial community had a standard against which we could calibrate our assays.

In terms of the spread, which is one of the questions I am sure you will be asking, the NIBS people tell me that when they first put the international standard out for hepatitis C that spread of results from the labs was very similar to what we have here. I think it indicates the need for convergence of methodology and practice. I don't think it indicates variation in the standard. I think probably it is a good point to stop. Thank you.

(Slide.)

The participants, I'm sorry, just one final slide. Many of you will recognize the usual suspects of malarial labs here. We have the CDC represented of course as you would expect. We have Kevin Kain's lab here from Toronto, Swiss Tropical Institute, and the EFS in France. So many sites, and we would of course be receptive to more laboratories receiving the standard to add to the data set. Thank you.

(Applause.)

DR. KUMAR: Thank you very much. Now I would ask Dr. Jay Epstein, who is our Office Director for Blood and Blood Products to come in and moderate this session. This is the last roundtable discussion. Dr. Epstein.

Session IV: Roundtable Discussions

Jay Epstein, MD, FDA, Moderator

DR. EPSTEIN: Thank you. Thank you very much. I see that our panelists have noticed their names on the program and seated themselves, for which I'm grateful. I don't have to read everybody's name. This is the last session of the day, and our hope is that we will have a discussion that can round out those discussions that have occurred throughout the day since necessarily the issues raised here will recapitulate what you have been listening to. Just by way of reminder, the way the program was structured, we first learned about the global problem of malaria and its impact on the US blood supply. We then took a look at the test technologies for detection of malaria infection, both direct detection and antibody detection. We then heard about experience outside the US, in the United Kingdom and France and Australia with use of these newer technologies to refine the strategy for donor deferral and reentry, and then now we are hoping to have what might be a summary session which we hope will illuminate the various pro's and con's of the core questions.

In selecting panel members for this roundtable we had quite a difficult challenge, because really there is no reason to exclude anyone. It could have been any and all of the speakers and any and all of the participants. But we tried to tilt perhaps a little bit toward the views of the public health experts, the regulators, and the blood banks in the hope that we can have a broad perspective on how things might play out.

So for the convenience of the participants we have projected the questions for our esteemed panel, and the format that we are going to follow here is first to have a discussion amongst the panelists and then open it to the floor for further deliberation. So let me just read off the questions and then we will go back to them one by one.

So first, what are the desirable characteristics of laboratory tests to detect malaria infections in blood donors? But here I think it has become clear that we are not talking one scenario. We are talking about the potential scenario of pre-donation screening, which might have itself the strata of screen everyone versus screen people with risk factors -- caveats of course. Then we have within that stratum, you know, could you use antibody or would you have to use a direct test such as nucleic acid technology. Then we have really a separate question, which is, well, what are the desirable characteristics for a test that would be used only for reentry. That is to say, to potentially reduced the deferral period for individuals otherwise deferred based on their risk histories.

We then would like to discuss what are the risks and benefits of donor screening for malaria infections in lieu of risk-based deferral. This is sort of the risk and benefit analysis that has been touched on by a number of people.

Third issue, what are the prospects for the use of a malaria antibody test in the US, either to screen donors, that is pre-donation screening, or to reenter deferred donors. Then lastly, what are the prospects for the use of DNA-based methods as blood screening tests in the US, and we didn't think we should be asking about use of DNA for reentry because we know that there is the false-negative problem.

Okay. So for the panelists then to start, let's open the discussion on the first question. What are the desirable characteristics of laboratory tests to detect malaria infections in blood donors? And let first frame that question, if we are entertaining pre-donation screening what would be your answers. So we are really asking you, well, how good does a NAT test have to be to be a donor screen and do a deferral and is it conceivable that one can use an antibody test in lieu of deferral given the recognized issue of a window period. So who wants to open the discussion?
Yes, Peter.

DR. CHIODINI: I'll step up. I think the first thing to say is that I would be very concerned about having an antibody test instead of something. I think it is great merit in adding it to the other --- of history taking and time deferral. The second point is as we do it we actually test the samples in real time. So at the initial encounter that unit is usable if it passes all the other safety checks, including the malaria. So we don't send them away, test the serum later, and then call them back, so that now we are doing it in real time so there is not a loss there. I think NAT testing is a non-starter. Unless you extract the whole unit, you cannot say there are no parasites in it, and I think theoretically one would be enough, 100 certainly would be I think, and therefore the sensitivity at the moment of the current assays is simply not adequate to rely upon that. You couldn't look the recipient in the eye and say there is no malaria in that unit.

Then we come to antibody testing. In a sense it is helpful if it is a bit oversensitive to be honest, and that way you have got the safety net that you want. Certainly highly sensitive, highly specific. It is not over-

sensitive that is even better, but would tolerate a bit of over-sensitivity to make sure we capture people in. Even with that with the current practice you lose very few donors by having that technique in place. Antigen detection by dipsticks is not enough. It is not sensitive enough. Blood film is not sensitive enough. There is work going on that I am aware of looking at HRP2 assays, and I don't know how they will match up yet. I think the experiments and studies are still being done, and that is something worth looking at. Whether it is better than NAT or as good as I don't know, but that is something that could be evaluated. So I think in summary my position is that testing is in addition to, and I would clump as we have done for antibody testing.

DR. EPSTEIN: Would anyone else like to comment? Dr. Nakhasi.

DR. NAKHASI: I just want to again sort of push this argument further --- Peter. What would be the -- we discussed about the various antigens --- species. What would be your idea of having all the four, two, three, which one would --- because your experience told us something. So I would like to know how to enhance the sensitivity and specificity of everything or we should focus on first two, three, --- falciparum or ovale -- or vivax, sorry.

DR. CHIODINI: Okay. I think it boils down to what you are prepared to accept in terms of sensitivity against the various species that might be there. The one that is going to kill recipients predominantly will be falciparum. There was a fatality last year in the 2005 figures from the UK with plasmodium vivax, so even in those who are not recipients of blood transfusion vivax occasionally is fatal, so we shouldn't dismiss it. But predominantly it is falciparum in a very defined risk group, so falciparum and vivax, good sensitivity, which I think we have with the current assay, is to us in our practice acceptable. I think you must decide whether the current sensitivities for ovale and malariae are good enough. We would all like them better. I think there is no dispute we would all like even more sensitivity for those species, and I would endorse that. I think it is for your authorities to decide whether at the moment that sensitivity is acceptable enough for them given the risks of the donors coming in.

DR. EPSTEIN: Roger.

DR. DODD: Well, for me the simplistic answer is always sensitive, specific, inexpensive, and in this case available, and I think that one of the issues is that we are probably not going to get the test we want unless we make a commitment to uniform screening. Having said that, I am very uncertain about the relationship between the cost of doing that and the opportunity costs of continuing to do what we are doing now.

But I think the nightmare scenario for most of us in the US in blood banking is a mixture of the same sort of questioning that we are doing now plus testing, because I think we have seen a fair amount of evidence that we are doing a pretty good job. So I would actually back up to what is perhaps a more sensible answer, which is that the properties of the test should be such that they meet the FDA paradigm, which is that we could replace our questioning strategy with something that was at least as safe.

Now that begs a question, but I think the answer is we are going to have to do quite a bit more work, and I am glad it is starting, to understand what those parameters are going to need to be to get the replacement test. In a sense I am talking from an area of practicability and blue sky ideality for us. I mean, I certainly can't challenge Peter's position that the objective is maximum safety, but in fact in order to make the big leap we have to give something away and -- or in order to get Mexico back we have to give something away.

(Laughter.)

DR. EPSTEIN: Could I, Peter, ask you to clarify a point? You stated that you test in real time. So I understood that to mean that for a

donor who based on history otherwise would be indefinitely deferred a test will be done at that donor setting. Is that correct or incorrect?

DR. CHIODINI: If they present for donation and let's leave aside the history of previous malaria, but for the other groups, the travelers and the residents, if they present to the donor session and they have already been back for six months they can be tested at that point, and if they pass with a negative antibody test and all the other tests then they are in. Unfortunately at the moment with a history of previous malaria they would have to be --- about three years. That is a temporary wrinkle I think.

DR. KATZ: But you do collect a unit at that time, is that what we are understanding?

DR. CHIODINI: No. If they have been back less than three years with a history of previous malaria they will not collect then. There is not point.

DR. KATZ: No, no. The travelers and residents.

DR. EPSTEIN: For the travelers and the residents though you would collect the unit?

DR. CHIODINI: That is correct, we would collect the unit.

DR. EPSTEIN: And you would then do an antibody test concurrent with the donor ---?

DR. CHIODINI: That is correct, and currently we lose two percent of those with antibody positivity. So we are getting back 98 percent if they have been back six months. The merit for us of going for six months with antibody tests rather than 12 months without is we get them back six months earlier and they --- chance in their second donation we will obviously then be six months earlier, and so on and so forth.

DR. EPSTEIN: So let me perhaps challenge the blood bankers on the panel. Is any strategy of, if you will, selective testing thinkable in our environment?

DR. KATZ: Well, yes, it is. But I think all of us are very nervous about a scenario where we are doing essentially manual screening in a certain sense, and then we have to segregate a sample to be tested for something that is outside the routine. That is how we kind of keep running up against universal testing. So we have to make our computers work, which is easy to say and hard to do. I have to have a way that when my donor tells me they went to Mexico that I can't label that unit until there is a result in the database that says it was tested and was negative for malaria. We need to be able to do that I think in order to make sense out of this. So, but the perfect shouldn't be the enemy of the good. We can probably get there eventually and do this for the whole world with a four-species sensitive assay, blah, blah, blah. So, I mean, that is where I would like to get, where that donor history enters the computer and won't let me label a unit I've got that result, and I think that is the concern that you are hearing from Sue and from me and from everybody else that doesn't like sending BPDR's to Jay.

(Laughter.)

DR. EPSTEIN: But surely we appreciate the attention you know.

DR. KATZ: Yes. Short of that, I would like to be able to say Mexico ain't a risk, or something equivalent to that.

DR. EPSTEIN: All right. Well, perhaps it is not formally on our set of questions, but we could talk about modifications to deferral strategies. But for the moment we are focusing on the issue of test characteristics, and I think, you know, Roger, what you clearly stated is the ideal test for screening, pre-donation screening, would be that which can supplant the questionnaire. But if we were to shift gears and ask, well, what are the necessary characteristics for a reentry test, now again mindful of the fact that reentry testing is already in use and there is some practical experience from, you know, France,

the UK, and Australia. How do we see it here? Alan, you wanted to comment on another point?

DR. WILLIAMS: Well, you actually shifted gears before I got a chance to answer the question. There still is an inherent attraction for being able to test directly for the agent that causes the infection, so my question to the lab group is how far away technically is it to be able to concentrate a sample so that sampling is not is an issue for PCR, that one could feasibly have 100 percent detection of nucleic acid of the agent? Because that potentially could solve numerous problems.

DR. CHIODINI: I think the problem is the parasite comes in a packet. It is not uniformly distributed across the unit in low concentration. It is in -- if you have got one parasite it is in one red cell, and 10 are in 10 red cells, and so on. We were discussing this in the ---. You have sampling issue. If there are 10 parasites in the bag or 200 in the 200 mls of packed cells, can you sample enough to be sure you have got it? And obviously if there is only one or 10, in terms of the sampling volume it is simply not practical. So whilst I am extremely attracted to NAT and we do further things in my lab, I am somewhat depressed at the thought, well, we probably won't for the foreseeable future be able to pick out one or 10 parasites in 200 mls of packed cells, and that is my concern. What you can do is get to the threshold, and we have seen what the sensitivities were, and what we don't know is how many of the antibody-positive donors would be above the NAT threshold. It is useful finding out those, which is what we want to do for donor care, and if you added that in as well, you would have an extra layer of safety. But what would you say to someone who say you have got antibody-positive but NAT-negative on the unit, are you going to use it? I think at the moment you would probably have to say no.

DR. EPSTEIN: I think that question also goes back to the scientific uncertainty about the level of parasitemia associated with --- in humans. You know, what we heard was that in mice, at least in one model, one parasite is enough, you know, in one red cell. But we don't actually know what is true for human transfusion, and we certainly don't know what is true for stored blood. So I guess one way of approaching that question is how important are experiments that could define the infectivity, minimum infectivity of a contaminated blood, or are simply forever stuck with the problem that because there might just be one parasite per unit we can never use the direct test.

DR. KUMAR: Can someone make an outside comment?

DR. EPSTEIN: Yes. Well, we are going to open it very shortly, but, Sanjai, in your case, yes.

(Laughter.)

DR. EPSTEIN: Use the mic please. Please use the mic.

DR. KUMAR: If we try to -- okay. I will be quick. If we try assume that a single infected red cell will be present in a unit of blood and that is the sensitivity we try to achieve, then the total given blood volume of 10 liters we will assume 20 parasites will be present in the entire donor. That sort of numbers may not be actually viable for the parasite itself. I mean, there is no --- that it will be 10 parasites or 20 parasites can even be --- and survive. So the key is to get that sort of data actually from the field.

DR. EPSTEIN: Okay. I think another point of view on that is kinetics. In other words, how quickly does parasitemia rise. Because, you know, the risk will be related to the duration of the sub-detectable infectivity, and that may be a very brief time. So that is just another perspective.

DR. KATZ: Well, Jay, there are some questions that haven't been talked about, but the number of scenarios that we get in the donor room is so huge. Well, I took all my methylquin* like a trooper. Well, I skipped a couple of days of my methylquin, a couple of weeks of my -- I mean, so I think the answer to that question about the duration and the level of parasitemia is all

over the board, and I tend -- I would love to agree with --- that I think direct detection is a non-started in the donor setting because of the 12,000 variables we are trying to deal with with these people.

DR. EPSTEIN: Okay. So let me in the interest of time move to the question of reentry testing, and I think that the core question that has been posed, at least in the US, is that the available test has antigens for falciparum and vivax, but we have experience with transmission of malariae and ovale. So, you know, could we consider a test as currently available? I think the issue of antibody sensitivity is minimized by the waiting period, although we have seen this debate about, well, how does it compare to an IFA as a gold standard and do we really believe it is better. But I think we ought to focus on the question on the antigen specificity of the test. So who would like to field the question of, well, what is the necessary characteristic of a reentry test, at least in our environment? Would we use a two antigen test?

DR. DODD: I'm going to turn that one back to you, Jay, because I think that your agency has some fairly rigorous requirements for making claims, and obviously the claim that one would want to make with a reentry test is that this donor is now suitable to give blood again. And if there is an assumption that maybe the donor infected with ovale or malariae, it appears that we wouldn't have a test that is firmly labeled to detect that. So where would you go with that question, if I may, or your staff, if I may turn it around?

DR. EPSTEIN: Well, I think the scientific part of the question is what is the risk coming from malariae and ovale, and I think that the available data suggest that that risk has been decreasing, and so the bottom line question is, well, how low should it go before you stop worrying and how confident could you be given the fact that global epidemiology changes. So I don't know the answer, Roger, except that as long as we continue to think malariae and ovale are a threat we wouldn't want to reenter a donor based on a two-antigen test without knowing what species of malaria had caused the infection in the first place. So, you know, I think that is the way I would frame it. But again, you know, we are here to gather information and hear opinion.

DR. KATZ: Well, can I turn the question around? And that is all countries are not equal, and I think we have heard that. So right now what we do is we look at the yellow book in some iteration, and we say, well, this, this, this, and this. But if the FDA said, well, the yellow book is fine, however we are going to say that X, Y, and Z, or vivax and falcip. So they are not included anymore if you have good test, like we do for type O. Okay? We have to defer donors from sub-Saharan Africa if we don't have an HIV sensitive for type O and it works okay I suppose. But can the FDA see a situation where we would say there is minimal or no ovale or malariae in this list of countries that is in the yellow book, so they are okay if you are using a test that is sensitive for what is there.

DR. EPSTEIN: Okay. Well, I think that is a suggestion we can think about. Shall we move on? Other comments about test characteristics, or are we ready -- yes, please.

DR. GARRAUD: Something which is puzzling me a little bit is that we can't qualify for blood donor or a blood donation, because what matters actually is if there is any danger in the bag, not necessarily in the human. That might be different, especially when you think about malariae, which may be quiescent? Is that correct? For years and years and not appear in the bag until let's say five years or something like that. So that is a real question, is that the qualification of the donor or of the donation.

DR. EPSTEIN: In our regulatory system we qualify the donor, but of course it is based on our understanding of the safety of the unit as well as the safety of the donation. But, yes, I think you raise an important point. There are different risks associated with the different species.

I think let's try to move on to the second question. What are the risks and benefits of donor screening for malaria infections in lieu of risk-based deferrals, and we are starting to touch on this, and you heard at least one effort to model the relative risks and benefits. That was Dr. Anderson's presentation. I think the take-home message was that the residual risks might come out about the same under certain reasonable assumptions, and maybe even a little better if window periods are shorter. But certainly the costs, the cost profiles are different. So what do our panelists think about the relative risk and benefits were we to try to move to screening in lieu of risk-based deferral? And I think as we approach that question we ought to bear in mind that we have heard that risk-based deferral is rather inefficient, that, you know, historically as much as 61 percent of actual case of transmission occurred due to failures of screening. And I think that that point was brought home, you know, very strongly by the data that we heard presented by David Leiby where, you know, a group of about 3,200 donors, presumably from a low-risk region, actually had 11 EIA positives and a large number of persons who actually had risk factors. So how do we see this tradeoff? Basically perhaps equivalent residual risk, but higher cost and potential for lowering residual risk. How do we see the tradeoffs? Any takes? Yes, Peter.

DR. CHIODINI: Could I just ask a question? Within this if you looking at savings and the cost of producing an assay, will there be any saving in staff time at all removing the risk-based assessment? I mean, I don't think it should be removed, because you are in a donor session being asked multiple questions. Would you actually reduce costs if you dropped it?

DR. EPSTEIN: I think the blood bank representatives might answer that better than I can. Lou, do I have a taker?

DR. KATZ: Yes. Certainly, and I think, Peter, you understand this. It would lower the aggravation factor incredibly. The amount of time in the donor room, and more importantly with post-donation information and BPDR's there is a lot of time and effort that goes into the documentation and submission and remediation of the times when we find out it fails. So there is a big deal there, plus we don't know for sure what it costs us to replace a donor. We think in our center that our recruitment cost is about between 70 and 80 percent is getting a new donor in door, that 20 or 30 percent is to get a repeat donor to come back. But we have to replace everyone of those donors with a new donor we think, so we can't quantify it, but it is not cheap. I can't give you a number.

DR. EPSTEIN: And again we shouldn't forget that on the benefit side there is potential to expand the donor base and that it wasn't just avoiding the onsite deferrals. It was potentially recruiting in the large cohort of, you know, what? You estimated, what, over 800,000 who are self-deferred per annum. So we should factor that into the discussion. Did I see a hand up? Was it Matt?

DR. KUEHNERT: Yes. I was just going to make a broader comment maybe that I am having trouble when we talk about risk and benefits trying to weigh these, because we don't really have an adequate framework with which to judge. You know, I see these numbers. We might be able to gain 800,000 donors, but I don't know what that means in terms of both, you know, cost savings, but also in terms of lives saved in terms of recipients because those units are not available. So, you know, this applies to, you know, any strategy that is employed where we really need a framework to be able to evaluate, and we have sort of missed that opportunity in the past with leuko reduction and bacterial screening to be able to evaluate a strategy before, during, and after it has happened. So that would be my plea, is to be able to develop a framework for that. I think with the model Dr. Anderson presented we really are on a good road towards looking at sort of theoretical, but trying to plug numbers in isn't possible unless you have that framework.

The other thing I wanted to just throw out there is, you know, this seems to be more an availability issue rather than a safety issue. We certainly don't want to decrease safety, but it sounds like more we are looking at, you know, tradeoffs. You know, in looking at the numbers, you know, 0.25, let's up to one per million, we are looking at about an equivalent risk to bacterial contamination in red cells, which we currently don't do anything about and perhaps could even argue the risk of gram-negatives, which cause some high morbidity. So, you know, not to diminish the importance of all this, it is very important, a very interesting issue, but I think it really is more an availability issue rather an issue about safety and that would maybe better frame the issue when you thinking about costs and benefits.

DR. EPSTEIN: Other comments on this question? Yes, Jerry.

DR. HOLMBERG: Yes. I just want to follow up on what Matt said, and one of the comments that I am very concerned about is that I -- well, first of all let me preface that. What David presented was outstanding as far as the control group and, you know, some of the information there that was gleaned on just the effectiveness of the questions and really the time that you went back to go back and look at some of those donors that fell out. However, what I am concerned about is the geographic, getting rid of the geographic question all together, because we do rely on a certain amount of safety there in picking up or some of the emerging diseases, and Leishmaniasis is a good example. You know, we don't worry about that right now with in Kuwait primarily, you know, because it's a malaria area, too, I mean, so, you know, there is -- I am just concerned about the ripple effect if we would completely alter the scheme and do away and say, well, there are problems in the way the question is asking and that we are not getting the accurate information. I think that possibly we need to revisit the tier approach and how that is addressed in getting the information.

I think one of the things that we have heard very loud and clear is that there is a difference between the traveling to other places in the world versus the Americas, and I think that in our country where we have seen a great influx in the Hispanic population I think we have to be sensitive to that and we have to be able to tease that out. Especially with the benefit of Hispanic population adding to our donor pools and especially what is the increase or the benefit of the savings in terms of the number of -- well, I will just use group O red cells for instance because the Hispanics would tend to have a higher percentage of group O's. So, you know, is there an advantage of targeting that population?

So I think that there are some things that we need to do. I think that, you know, I would encourage a real review of the differences in the travel and the visit and maybe take a look at the questioning.

DR. EPSTEIN: Thank you. Other comments? Yes, Peter.

DR. CHIODINI: May I make a very brief comment about geographic risks? Of course we looked at this in a bit more detail when we were formulating our guidelines, and in fact for the definition of a resident what we wanted, but were not permitted by Council of Europe, was we actually wanted just sub-Saharan Africa and --- New Guinea to be the areas where we would considered that you would get semi-immune relatively easily and focused on those areas. In the end we were instructed that resident had to cover all the other areas, but that would have put them into more like the traveler category. So I can see where you going on the Mexico one and have some sympathy with that.

DR. EPSTEIN: Okay. So I think we have actually been discussing the next two questions and their subparts, but I will raise them formally. What are the prospects for the use of a malaria antibody test in the US either to screen blood donors or to reenter deferred blood donors? I think what I have heard is that the prospect for screening is limited, but that it may be a useful adjunct, that the criterion to use it in lieu of screening should be that it produces no

less safety, --- is overriding principle. But is there more to say about it in terms of prospects? What about just from the technology point of view? Yes, Lou.

DR. KATZ: I don't think we know the answer, and I was at a meeting recently with some of the people in this room where the enthusiasm for universal screening was like logs more than I ever dreamt it would be. So if this was to be let's say on a big automated machine on the sixth channel of a big automated machine and costs us $2 a donor, then I got to get out my pencil and figure out whether I can pay for it. So I am not sure universal screening is a dead issue at all. There are all the complications that we have talked about, about how many species and all that sort of thing.

DR. EPSTEIN: Other thoughts? Roger?

DR. DODD: Well, I think that one thing that we heard --

UNIDENTIFIED: Use the microphone.

DR. DODD: Sorry. I think that one thing we heard loud and clear throughout the day is that the prospect of detecting an infectious individual within say a couple of weeks of then infecting event is relatively dim, but nevertheless that period -- well, that period could include infectivity via blood. As Susan pointed out, probably if we were to go that route we would want some other mechanism to dissuade or self-dissuade individuals who have just traveled from even presenting, and I don't know how rigorous those procedures would be or whether one could, you know, do it in a fashion that didn't lead to all the agony of deferral if we could put it that way. So I think that that is an issue that will need quite a lot of thought before we were to move ahead with this, but I don't think it is insoluble.

DR. EPSTEIN: And what about reentry? How do see the prospects? Again, we have discussed this quite a lot, the issue of different species and test sensitivity. I mean, I guess the ultimate test in the European theater is whether there will be any transmissions from donors who are duly reentered with negative antibody tests, and we just may not have had enough experience to know.

DR. CHIODINI: I think --- being a hostage to fortune and saying this, but I think we are fairly confident that we have got a handle on our risk groups. It is very clear that it is the semi-immune person who we particular want to capture and get the antibody testing done, and they characteristically have had very high antibody levels, and I think I would be I think it fair to say astonished if someone who was semi-immune to P. falciparum was negative in the antibody test to P. falciparum. So we feel confident that we would get those out, and I think that is par excellence if you are looking at safety --- they group to go for.

DR. EPSTEIN: Other comments about prospects for reentry testing? I am sure -- I assume no one wants to introduce NAT for all the reasons discussed.

(No response.)

DR. EPSTEIN: Okay. Well, let me extend this question though, because what we have heard through the day is that there may in fact be opportunities to improve the strategies for geographical deferral and, Lou, you were suggesting that. There is the issue of recognizing the species predominance in geographic areas and perhaps using that as a way to augment the deferral strategy, perhaps basing that on the ultimate disease risk to the recipient I think we heard. We heard the pleas about Mexico and the Americas, which is linked I think to the challenge Dr.ÿOckenhouse put in front of us, which is just accept the traveler. The argument that we have low rates of transmissions, very low rates from travelers, and it may be because of the biology. It may be that because the travelers simply because they are not semi-immune are not parasitemic, or at least certainly not for very long when they are asymptomatic. So there is that issue, and then I think we have also heard that, well, why not focus the deferral strategies where the risks come from and, you know, clearly it is Africa and particular sub-Saharan Africa perhaps with

changing dynamics from Asia as, you know, we saw in Australia. I don't know what the future is in the US.

So are there any thoughts about the deferral issue as an alternate approach? In other words, if we don't introduce testing, either for screening, augmented screening, or reentry, do we think there is a place to go refining deferral?

DR. KATZ: Yes.

DR. EPSTEIN: Lou says yes.

(Laughter.)

DR. EPSTEIN: Well, we will think about it. Okay. Lastly, what are the prospects for the use of DNA-based methods as blood screening tests in the US, and I think Dr.ÿChiodini advised us that is a non-starter and I didn't hear a lot of dissent. Are there other thoughts about that? Essentially the problem is, you know, you can have very low parasitemias, particularly in semi-immune individuals. You have a sampling problem. We may never be able to overcome it. Does anyone think that current technologies offer hope in that direction?

(No response.)

DR. EPSTEIN: No, and I think that this discussion actually mirrors a discussion that happened two years ago at Council of Europe where there had been movement toward use of DNA and where the experts in the Council of Europe basically said, well, that is just not scientifically sound given what we know. So I think what we are hearing is that is still the state of the art.

Open Discussion

Jay Epstein, MD, Moderator

DR. EPSTEIN: Okay. So at this point let me just open this dialog to the floor, and please feel free to comment, raise questions to our panelists, and otherwise speak your minds. Steve.

DR. KLEIMAN: Steve Kleiman, AABB. Jay, just for clarification. I think it is kind of understood, but it came out in the panel discussion, and that is about your question about reentering deferred blood donors. I think that if by reentry we mean what we generally mean by reentry in the US -- that is you are here today, go away, come back at some point time, we will collect a sample, test it, then tell you that you are eligible as a donor -- if that is what we mean by reentry, it is generally worthless in this circumstance because if we have to defer somebody for six months and then bring them back between month six and month 12 and qualify them on a sample in order to just get a few months more of eligibility, I don't see that as practical. But if what you mean is what they are doing in the UK, whereas if the person comes in today and tells you "Seven months ago I was in a country. You know, I traveled to a country," and you say, "Okay, you can donate. We accept the donation and we will run it through this test and we will throw away the red cells if it is positive," then I think Lou's comments are correct that we need some computer systems to be able to do that, but it is not out of the question. But I would like to rule reentry as we classically identify it as not a helpful solution. At least that is my opinion.

DR. EPSTEIN: Thank you, Steve. Fair point. Please introduce yourself.

UNIDENTIFIED: Jerry ---, FDA down in Richmond. Yes, I was talking to colleagues here from the UK and France at the break, and they -- we were talking about the fact that they also use the EIA testing on their tissue donors and organ donors, and we here in the United States also link a lot of that testing as the same testing which we do for tissue donors. I just wanted to ask question, was do you go ahead and defer those donors based on the titer level on their EIA testing? What do you --?

DR. CHIODINI: If they are positive and it is an organ or a tissue they particularly want to use, then an individual risk assessment is done on that particular donor to decide whether those results mandate that the organ or

tissue shall not be used or not. It fortunately has been in very few cases, and I think give the critical shortage of organs it is justifiable to do an individual risk assessment.

DR. EPSTEIN: Thank you. Jed.

DR. GORLIN: Gorlin, ---. It turns out that there are more Lutherans in Tanzania than there are in Minnesota, and I found this out when I got to serve the CDC on a --- grant to evaluate the blood supply in Tanzania which was quite terrifying, but I also had the opportunity to travel to a number of the world Lutheran missions; and the typical scenario is the traveler comes there and does work for two to four weeks, often in some pretty uncontrolled and highly malarial endemic areas. So I have considerable concern about just discarding the risk to travelers because I think not all travelers are the same. So I am very attracted to Lou's concept of the traveler to Mexico could be treated one way, but the traveler to sub-Saharan Africa I would think the status quo would be well advised.

DR. EPSTEIN: Thank you. Merlyn.

DR. SAYERS: The spotlight seems to be on comment that Lou made. I would like to agree with everything that he had to say about the selective screening of donors and the challenge that that is going to be if we are looking at malarial antibody screening, the challenge that is going to present to our information management systems when it comes to teasing out a certain select segment of the donor population for discreet testing. My comment, though, is that I sincerely hope that is the direction we are going to have to go in because of other diseases on the horizon. I would really like to think that it is selective testing that we are going to employ for --- and that is going to be selective testing that we are going to have to invoke once there is a preon* test on the market. It would make no sense given the epidemiology of those two diseases to test individuals who are at risk for those conditions repeatedly at each donation.

DR. EPSTEIN: Thank you.

DR. GRESENS: Chris Gresens, Blood Source, Sacramento. A quick question getting back to the specificity of the antibody tests and our prior experience with other tests, and I will mention ALT, which I realize wasn't FDA's doing, and hepatitis B core antibody even in our own center. We lose a lot of donors, and until we moved to a new system which was more specific we lost very large numbers of donors for what we believe to have been false-positive results. I am looking at Dr. Anderson's analysis and I am wondering how firmly we believe the loss of donors really will be across the United States on the order of a couple 1,000 per year versus 20,000 or 200,000 per year. Could you perhaps, Dr.ÿAnderson, or others comment on false-positivity please?

DR. EPSTEIN: Well, I think that false-positivity of the New Market test of the IAF is better answered by those who are using it. Perhaps Olivier or Peter could comment on the experience.

DR. GARRAUD: Yes. Actually, yes, we do know that a lot of false-positive in the -- you know, in France that
--- sensitive that we do not afford any risk. We have invented the precautionary principle you know, and so as far as we have any doubt we can accept the donation. But one of the reasons why we try to implement this ELISA-based test which has minimized the false-positives or so, that was not the primary aim. The primary aim was to detect the -- what were false-negatives with EFA. But, by the way, we have also minimized the false-positives. Did I answer the question?

DR. EPSTEIN: Is there a number you could put forward for the false-positive rate of EIA's?

DR. GARRAUD: Yes. I think it is probably 1.4 percent I think of the tests.

DR. EPSTEIN: Peter, would you agree with that figure?

DR. CHIODINI: That isn't what our data showed. When we did 880 we got no positives, and thus we are only running at two percent positive for the whole donor, selected donor population at the moment. We feel we can live with that positivity and we don't feel it is cost effective to tease out in detail those two percent, albeit the question of NAT testing, which is a separate issue, to see if they may be semi-immune.

DR. EPSTEIN: All right. Harvey.

DR. ALTER: This is Harvey Alter, NIH. This discussion has given me shaking chills.

(Laughter.)

DR. ALTER: You know, the whole --- was that an antibody test or a test would replace the questionnaire, but I am getting the sense that that is not going to happen, and certainly if that is not going to happen then doing universal testing has little appeal to me. So the appeal comes to reentry if it can be done, as Steve said, simultaneously. Because if you could do the reentry, not only would it reenter, but I think it would allow you simplify the questions. In other words, the thing that Sue presented of not only are you in this country, but when were you in this country and how long were you in this country, where in the country were you. So the question could be basically were you in this country, and then hopefully you could narrow down the number of countries and give Mexico back to who -- but not break it down so precisely, because you are going to have the antibody test as the backup.

But the main point I wanted to make is that this whole discussion would be moot if we had a preemptive strategy in place to get rid of all these agents with some pathogen reduction method, and I don't want that to die despite all its problems. I think we have to dedicate ourselves, because this is just a continuing spectrum of new agents, relooking at old agents, and I think we have to do the NAT approach to pathogen reduction. Say this is something we want to achieve. We are going to make a major effort. We are going to put government support together with industry and see if we can do the pathogen reduction what we did with NAT testing in a five-year time span. So that is my point.

DR. EPSTEIN: Thank you, Harvey. Sue.

DR. STRAMER: I just want to reemphasize, and perhaps it doesn't need reemphasis, but the keep-it-simple approach. Because what we have now in the field are less and less specialized individuals, less and less training of individuals, and as we complicate the questions, complicate the process, as David Leiby showed, just add another mattress to the pile, it doesn't really help what is going on in the field. The complication in accepting ineligible donors, increasing numbers of post-donation information reports, and frankly protecting us for the future. I mean, you know, it may be convenient to say we have excellent blood safety with respect to transfusion-transmitted malaria, but we don't know what the future will bring. Unless we give a message to the manufacturers now to get working on something and not to kill the prospect of universal screening, if the situation does come up and more --- cases of US borne cases of malaria occur we won't be prepared.

The testing-in strategy really from my money does nothing. It just shortens the deferral period. The reason it works in Australia and other parts of the world is because their questioning process is infinitely simpler than ours is. When I went through our information and what we do with Clive I almost had to resuscitate him.

(Laughter.)

DR. STRAMER: Because he said there was no way, there was no way that they could do in Australia what we do here and possibly do anything like the test-in strategy. The reason it works for them is because, as I said, the maintain the relationship with their donor and the continue to collect plasma. Their questions are very simple and they do believe the New Market test has excellent specificity. You know, whether it is in people who have traveled for

one percent false-positive -- we don't know if it is false-positive, but one-percent repeat reactivity in travelers to 30 percent in people who have had malaria. So, you know, again the plea is just keep it simple, and I don't believe we should dismiss universal screening because, one, it may be something we can use in tandem with streamlining the questions. Maybe not eliminating the questions, but at least look at a much simpler and more effective for the field approach to questioning. But again, if we send a message out of no, we will have nothing in development.

DR. EPSTEIN: Thank you, Sue. Any other comments or questions?

(No response.)

DR. EPSTEIN: Okay. Well -- oh, Steve.

DR. CURR*: I was going to do this last. Steve Curr with FDA. I wanted to sort of agree with everybody. I love coming to these workshops. I used to think I was the smartest guy in the world, and I found out --- I am probably dumber than dirt. I love the workshops for a lot of reasons, but I also want to reiterate one thing. I remember coming to one of the first FDA workshops I came to was discussing this problem about NAT, and 1990 they are not going to do universal screening with NAT because it is impossible and won't be profitable, it is not cost effective. By 1994 they said, well, we are going to do it, but we are not sure how we are going to do it, and they developed the pooling system. The next thing you know, now it is licensed.

So I have not really answered anything or even questioned anything, but I love the workshops because all the information does come out and the ideas that we -- we don't want to discard anything, and the ideas and the reason we get so many different people in here is you see all these different views, and I think that is what is more beneficial in these workshops because we do get these things. And the other question, answer just to refer to what Sue was saying is the idea that, yes, you know, where is the money going to come from, and then, you know, you are talking, what, microarray analysis. People come along with the more tests you can get onto this thing the more beneficial it is to have. The old history of this thing is we had a donor that came in, or millions of donors came in, and you did one test, and then it was two tests, then it is five, and HTLV, ---, you add it and we keep on adding it. It is obviously going to be happening that pretty soon we are going to have one donor and it is going to be a million tests against that one donor.

(Laughter.)

DR. CURR*: And maybe that is what we are heading for, so the microarray analysis is great. So I am really not saying anything other than thank you very much for having the workshop and I love these things.

(Laughter.)

UNIDENTIFIED: I think once every seven years ---.

Closing Remarks

DR. EPSTEIN: Thank you, Steve, and you have made my job as the summator much easier because my task here is really to thank everyone. First I would like to thank our many fine speakers, panelists, and moderators, especially those who have come long distances to share their experience with us. We very much appreciate that, and also to commend everyone for keeping to time. It is quite rare for us to have a workshop and actually finish ahead of schedule. Fifteen minutes seems like a very luxurious margin, and especially on behalf of the sponsors, the Food and Drug Administration, our Center for Biological Evaluation and Research, and the Health and Human Services Office of the Secretary, Office of Public Health Sciences. We are very appreciative for the participation of everyone who has come and thought about these issues and shared information and opinion so that we can move forward with the best possible information and with the ability to consider all of the credible options. So we thank you very much for your participation and wish everyone a safe return journey home. Thank you very much.

(Whereupon, the meeting was adjourned at 5:18 p.m.)