any risk.

So this is the same reentry algorithm that has been described already today. The idea is to obtain a followup sample and test it with either the same or an alternate anti-core assay. If the sample is again repeat reactive, then the donor remains indefinitely deferred.

But if the followup sample is not reactive with the alternate or anti-core assay, then that followup sample would also be subjected to HBV DNA testing with the sensitive NAT assay. And if the NAT test is reactive, the donor would be -- remain deferred. If the NAT test is non-reactive, the donor could be reentered.

And the required sensitivity that has been proposed is less than 10 copies per mL. So now I want to talk about the COBAS AmpliScreen HBV test, which is currently under review. The test has two sample preparation methods -- a standard method which is used for individual samples and has a 200-microliter sample input, and that has a sensitivity that is a 95 percent limit of detection at 16 international units per mL, which is about 80 copies.

And then, the MultiPrep method, which is used for pooled samples, which has a larger sample

input of one mL, and uses a high-speed centrifugation step, to concentrate the virus. And then, after that, the steps are essentially the same as the standard specimen preparation method. And that has a more sensitive limit of detection at 4.4 international units per mL, which is about 22 copies, which is still not as sensitive as the proposed requirement for using that in a reentry algorithm.

So what we tried to do is identify a method to lower the 95 percent limit of detection to under 10 copies per mL, with minimal procedural changes.

So at such low titers, the sensitivity of a test is significantly affected by the limitations of sampling. So if you take a one mL sample that might have three detectable DNA molecules in it, and apply the MultiPrep procedure, so centrifuge it and then eventually recover the sample in 200 microliters, the PCR test is done on 50 microliters of this material.

So the 50 microliter sample that is tested may not contain any of the targets. So a non-reactive result might be observed, even though there were some targets in the sample. So low titer samples can appear negative, just due to sampling error.

And you can increase the odds of detecting

that just by testing more samples. So if we start with the same sample that's recovered in 200 microliters, but then use three aliquots of it into three separate PCR reactions, it's likely that at least one of the three aliquots will contain one of the targets.

So in this cartoon, two of the three aliquots contained the target and were reactive, and the interpretation of the result is that if at least -- if one or more of the tests are reactive, the sample is positive. And if all tests are negative, then the sample is considered negative.

So we can do some calculations. As I stated, the limit of detection of the test is 4.4 international units per mL. That's the concentration that's detected 95 percent of the time. So using Poisson distribution, one can calculate what the hit rate would be for lower concentrations.

So a 65 percent hit rate, for example, would be observed on a concentration of 2.4 international units per mL, which is about 12 copies per mL. So just looking at the statistics, if one had samples of 12 copies per mL, and did -- and analyzed one sample and conducted three PCR reactions on it, and considered the result to be positive if at least

one of the three were reactive, then the sensitivity would improve from 65 percent to almost 96 percent.

And this just illustrates that the probability of getting a reactive result at 12 copies per mL with the test is 65 percent. So if you just do one test, you can pick it up 65 percent of the time. But if you do two more replicates, then you have a much greater chance.

You have eight possible outcomes. Seven of them are reactive. And if you add up all of the probabilities, you have a 96 percent chance of picking it up, and only a 4 percent chance of missing it.

So we tested these calculations out with the study, and the experimental design was to take the HBV international standard and make dilutions to 30, 10, 3, and 1 copies per mL, and then to do 40 -- to analyze 40 replicates of each of those levels. And on each replicate one one mL aliquot was extracted, and three PCR reactions were conducted.

So I don't expect you to read this, just look at the colors. These -- this table shows all -- the results on all 40 samples at 30 copies per mL, and there's two columns for each replicate. One shows the target result, and one shows the internal control result. But what you should look for is where the red

is, so the red indicates a non-reactive result.

So out of the three replicate tests done on the 40 samples there were a few non-reactive results, but all 40 samples had at least one positive result. So the overall cumulative detection rate was 100 percent.

And then, at 10 copies per mL, again, all 40 samples had at least one of the three replicates detected. So the cumulative detection rate at 10 copies per mL was also 100 percent.

At three copies per mL, we're starting to see, again, that the sampling issue prevents many samples from being detected. But 29 out of the 40 still had at least one of the three replicates positive for a cumulative detection rate of 72-1/2 percent.

And at one copy per mL, 10 of the 40 samples had at least one test result positive for an overall detection rate of 25 percent.

So if you take this data and use Probit statistics to determine the 95 percent limit of detection, it would be six copies per mL.

So the conclusion is that using the MultiPrep specimen preparation method on one aliquot per sample, and doing three PCR reactions on each

aliquot, and calling a sample positive if one or more of the reactions are reactive, then the cumulative 95 percent limit of detection for the COBA sample screen test is under 10 copies per mL calculated to be six copies per mL.

So if a followup sample from a deferred donor was non-reactive with an alternate anti-HB core test, coupled with a negative test result with a highly sensitive NAT test, this should provide sufficient data to safely reenter donors who were previously deferred.

And I'll be happy to take questions.

ACTING CHAIRMAN ALLEN: Thank you.

Dr. Hollinger.

DR. HOLLINGER: Yes. I'm just a little confused. Is there -- why is it that it doesn't work with the 200 microliters? Why don't you just test the 200 microliters which has all the particles in it, instead of doing three replicates? What am I missing here?

DR. HERMAN: The reaction can't accommodate that large a volume. So I can't take the 200 microliters of the extracted sample and put it all into the PCR reaction. The PCR reaction is designed -- that would be developing a whole new assay.

DR. HOLLINGER: I thought initially you showed that you had looked at 200 microliter samples, though, in the very first slides. And you looked at --

DR. HERMAN: Oh, no. Let me go back to the -- we have two different sample preparation methods -- one that uses one mL plasma, and it's a more sensitive method but it's less convenient because it requires this high-speed centrifugation step. And when you do the high-speed centrifugation step, and then extract the pallet, the pallet -- the recovered -- DNA is eventually recovered in 200 microliters of an assay reagent with specimen diluent.

And then, 50 microliters of this material can be brought into PCR reaction. Regardless -- and with the standard sample preparation method, one starts with 200 microliters of sample, skips the centrifugation step, and just extracts that whole volume. But that still gets recovered in 200 microliters of specimen diluent.

With both methods you end up with a recovered DNA in 200 microliters, and only one-fourth of it -- 50 microliters -- can get into the PCR reaction.

Does that clear it up? Maybe --

DR. HOLLINGER: Thank you.

DR. HERMAN: We have things that we've thought about to do large volume PCR reactions. With infectious disease testing, the -- one of the main limitations is, how much sample can you get into a test? And one way of doing that is to make better sample processing methods that can concentrate big plasma samples into very small volumes, and there are many factors that limit that.

And the other is to make a really giant PCR reaction, and there are factors that limit that also.

DR. HOLLINGER: Just for information, how fast are you spinning this down?

DR. HERMAN: It's 23-1/2 thousand G's, I believe.

DR. HOLLINGER: For just an hour?

DR. HERMAN: For an hour.

DR. HOLLINGER: And that's in serum?

DR. HERMAN: That's in plasma.

DR. HOLLINGER: I mean, plasma.

DR. HERMAN: It doesn't pallet 100 percent of all the virus particles.

ACTING CHAIRMAN ALLEN: Other questions or comments?

Okay. Thank you very much, Dr. Herman.

The schedule now has us moving to our open session. We're going to actually modify our agenda very slightly. Earlier this morning we did not have the presentation of plaques and recognition of the BPAC members who are -- for whom this is the last formal meeting, let me put it that way, and we will do that now.

We will then have a break of 15 minutes and come back and move into our open hearing.

So, Dr. Epstein.

DR. EPSTEIN: Well, this is always a bittersweet moment at our Advisory Committee meetings. On the one hand, it's a very special privilege to be able to thank our BPAC members for their service to the committee and to the FDA, but, obviously, it's a sad moment when we have to ask those people to step down because they've completed a term of service.

We value greatly the advice that we receive from the Blood Products Advisory Committee, and we're fully aware that it requires a very special effort to digest the materials that we send to you and to pay close attention during the course of our very detail-oriented meetings.

And so it's my pleasure, my privilege, and

with sadness, to thank these specific persons. Dr. Kenrad Nelson, who completed not just a term of service at the last meeting but also his tour as Chairperson; Dr. Jonathan Goldsmith, committee member; Dr. Michael Strong, who has been our member as a -- the industry representative; and Dr. Charlotte Cunningham-Rundles, also a voting member.

So if each of these people would come up in turn, I'll be happy to award a certificate and a plaque as a token of our appreciation for all your effort on our behalf.

Okay. First, Dr. Nelson.

(Applause.)

And did you have the photographer ready? Okay. Notice this spontaneous setting.

Okay. Next, we'd like to thank Dr. Charlotte Cunningham-Rundles.

(Applause.)

Okay. Next, we'd like to thank Dr. Jonathan Goldsmith.

(Applause.)

Now, Dr. Michael Strong, thank you.

(Applause.)

So perhaps one round of applause for everyone together.

(Applause.)

So, Jim, do we get our break now?

ACTING CHAIRMAN ALLEN: Yes. We'll take a 20-minute break. Please be back here at, well, 10 minutes after -- 10 minutes after 11:00.

(Whereupon, the proceedings in the foregoing matter went off the record at 10:50 a.m. and went back on the record at 11:14 a.m.)

DR. SMALLWOOD: We're in countdown mode. Dr. Allen?

ACTING CHAIRMAN ALLEN: Thank you. We will now move into the open public hearing. Just got new stuff put on my papers here, so I -- I've got two speakers who want to speak at the -- on the reentry of anti-HBC donors, Dr. Andrew Heaton and Dr. Steven Kleinman. I need to, first of all, read the open public hearing statement.

Both the Food and Drug Administration and the public believe in a transparent process for information-gathering and decision-making. To ensure such transparency at the open public hearing session of the Advisory Committee meeting, FDA believes that it is important to understand the context of an individual's presentation.

For this reason, FDA encourages you, the open public hearing speaker, at the beginning of your written or oral statement, to advise the committee of any financial relationship that you may have with any company or any group that is likely to be impacted by the topic of this meeting.

For example, the financial information may include the company's or group's payment of your travel, lodging, or other expenses, in connection with your attendance at the meeting. Likewise, FDA encourages you at the beginning of your statement to advise the committee if you do not have any such financial relationships.

If you choose not to address this issue of financial relationships at the beginning of your statement, it will not preclude you from speaking.

Okay. Dr. Kleinman, may I call on you first, please, to present a combined statement from AABB, ABC, and ARC.

DR. KLEINMAN: Hi, and good morning again. The AABB and other blood banking organizations have been working with FDA over the last several years to develop an algorithm for reentry of donors who have been deferred due to reactive anti-HBC results.

Blood banking organizations believe that

the proposed anti-HBC reentry algorithm will be a major benefit to deferred donors, will increase the number of donated units, thereby improving blood availability, and will not compromise blood safety.

AABB, ABC, and ARC support the FDA proposed algorithm and urge BPAC to endorse its use. Approval of this algorithm is only the first step in moving anti-core donor reentry forward. The next step is for manufacturers of HBV NAT assays to work with the transfusion medicine community to design and carry out the necessary studies to establish that their testing system can be used for anti-HBC reentry.

The blood banking organizations are committed to this project and will provide the needed donor specimens. We urge the manufacturers to promptly meet with FDA, so as to devise the appropriate studies to obtain an anti-core reentry claim. Furthermore, we urge the FDA, under its critical path initiative, to encourage HBV NAT assay manufacturers to participate in pursuing this reentry claim.

Another necessary element for anti-core reentry is the availability of an FDA-licensed more specific anti-core assay for routine donor screening. Compared to the non-specific assays used at the end of

the 1980s and in the early 1990s, one such assay currently exists, and the data suggest that another core assay currently under FDA review may have even greater specificity, thus potentially increasing the yield of donors who could be reentered.

It is the goal of the majority of blood collection agencies to move forward with anti-core donor reentry soon after licensure the implementation of the PRISM anti-core assay, provided that one or more HBV NAT tests are approve for this purpose.

AABB, ABC, and ARC believe that there is widespread consensus that anti-core reentry will be beneficial to deferred donors and to the blood system and urge that all involved parties find a way to expedite its approval and use.

Thank you.

ACTING CHAIRMAN ALLEN: Thank you, Dr. Kleinman.

Any questions or comments pertinent to Dr. Kleinman's presentation?

Okay. Dr. Heaton.

DR. HEATON: GEMProbe, Incorporated and Karon Corporation have submitted a biologics license application for the procleics ultria blood screening assay to the U.S. FDA on September 29th. Both Karon

and GEMProbe do support the FDA request for BPAC advice on a reentry algorithm for the application of HBV nucleic acid testing to allow reentry of donors previously deferred for HB core antibody serology tests.

Specifically, and in addition, Karon and GEMProbe also support the AABB proposal for a validation study of 3,000 recalled anti-HB core deferred donors as a means to establish HPB NAT as a required component in an anti-HPC deferred donor reentry algorithm. The companies fully support the transfusion medicine community in their desire to pursue HB core reentry.

Thank you.

ACTING CHAIRMAN ALLEN: Questions for Dr. Heaton?

Okay. Thank you.

Are there any other comments that anybody wants to make during the open public hearing?

Okay. We will close the open public hearing and move to the committee discussion. This was stated to be -- or the next presentation was the FDA perspective and questions for the committee. I understand that, in fact, there will not be formal questions for us to discuss. But Dr. Kaplan will

present the FDA perspective at this point.

DR. KAPLAN: Okay. So what you have heard is that we have a -- we're proposing an algorithm, or the Blood Committee is proposing an algorithm to reentry repeat reactive anti-core donors. This algorithm, at the current time, cannot be validated because two big elements -- mainly one big element is missing, is that the testing with a more specific anti-core test of -- it's not available.

Sue Stramer mentioned that she is conducting a trial, and that she -- she's in the process of collecting that data. So basically we don't have formal -- as the Chairman said, we don't have formal questions for the committee. However, we would like the committee to -- if they have any comments on the proposed algorithm, if they can do so.

ACTING CHAIRMAN ALLEN: Okay. I guess we don't have a slide or a piece of paper that shows the formal algorithm. Well, we've got lots of paper. We don't have a --

(Laughter.)

-- slide that shows the formal algorithm.

DR. KAPLAN: Yes. Let me see if I can pull it forward.

ACTING CHAIRMAN ALLEN: Okay. You know,

we certainly heard a lot of data presented by blood collection organizations. We have statements from the organizations themselves in terms of the way in which they would like to see this proceed. I think we've got lots that we can discuss and provide guidance to the FDA in terms of its moving forward on this process, even in the absence of specific questions.

So with that as background, let me open the floor to discussions, questions, comments, or whatever, on the issue before us of reentry of donors that test repeatedly reactive for anti-core.

Jonathan. Dr. Goldsmith.

DR. GOLDSMITH: I was just trying to figure out about the impact of this whole system. Have any of the blood collectors surveyed these donors who were deferred to learn if they would actually come back as donors again if they were reentered through some kind of algorithm?

Do we have any information about that? Or are these people who have had a test, it came back negative from their point of view, and, therefore, they are going to drop out of the blood donor pool from that point forward? Do we have any information about these people? Have they been surveyed?

ACTING CHAIRMAN ALLEN: And I would add to

that, I would be interested, in particular, from, you know, the Red Cross' perspective what has been -- I mean, certainly, in looking at the data that Dr. Stramer presented we see a much lower return rate by those donors that received that letter. Even though they're invited to come back in eight weeks, we I think saw a much lower return rate than was true for donors who didn't get any letter of notification.

And I will just throw it open. Are there any representatives from blood collection agencies that would like to address the question that Dr. Goldsmith raised?

Dr. Kleinman.

DR. KLEINMAN: Yes. I don't have a good answer to your question. I don't think that that specific type of survey has been done for anti-core positive donors. I was going to relate a similar phenomena, though, and that is for ATL deferrals that occurred prior to the change of criteria.

Centers have tried to access donors who were deferred for ALT and reinstate them, since there is no longer a deferral criteria. And I think the yield has been -- and I don't have any numbers, but I think anecdotally the yield has been reasonably satisfactory, whatever that means.

I mean, the centers who do it say that they find it a worthwhile process, but we don't have the actual numbers to help you there.

DR. STRAMER: Regarding a survey for anti-core, we don't have that. And for ALT, for reinstatement, I don't have the numbers off the top of my head. But we did see higher rates for those donors that were more recently deferred. The numbers that I do have are for P24 antigen in which we did do reinstatement, and about 30 percent of the repeat reactive donors who were eligible for reinstatement did return and were successfully reinstated. So for P24 antigen it was about 30 percent.

But those -- that was automatic or more proactive reinstatement closer to the time of their next donation. So the question is these long-term deferred anti-core donors, what would be our success of getting those back?

But, again, as I said, if we don't do this for anti-core, then we might as well not do reentry for other -- any other marker, because this is clearly the highest marker of why we defer donors for test results. And we hear from donors -- I alone hear from donors every single day about, how can they be reentered for HIV, HCV, HBV? So it's definitely

something that the community or these committed donors want.

ACTING CHAIRMAN ALLEN: Dr. Nelson.

DR. NELSON: Yes. It seems to me that beyond the numbers of donors that could be captured or reentered, the benefit -- there would be a benefit to an individual person to know that, in fact, the test was false positive, the initial screening test, and that he doesn't have a chronic infection -- an infection with a chronic infection, viral infection. So I would think there would be some benefit to that.

And probably if the blood bank didn't do it, if a person is asymptomatic, probably nobody else would. I doubt his physician would do that. So I can see some benefit to the individual person who was repeated reactive on the core antibody alone. And it seems to me there would be some individual donor benefit from that.

ACTING CHAIRMAN ALLEN: I certainly agree. And, in fact, the availability of more specific tests probably could allow a total reexamination of the testing scheme, which tests are used, and in what sequence, quite apart from the reentry issue.

Dr. Fitzpatrick, do you want to introduce yourself formally, please?

DR. FITZPATRICK: Mike Fitzpatrick from America's Blood Centers. Two things. One is it's very hard to measure the impact of reentry of anything. And it's hard to measure the impact of the deferred donors on other donors who have been deferred for some reason.

But the thing that I would suggest is that this would be the first step toward the next step, which is a supplemental algorithm to core testing that would allow us to use a battery of tests, whether it's NAT or a more specific -- more sensitive core, to evaluate those initial repeat reactive tests and not have that initial deferral.

And so if we don't have the initial deferral on a new donor, we don't have to worry about reentry, and we're not deferring donors who are eligible to donate. So I would see this as a first step in the progression toward accumulating more information about the tests available and the results available, so that we can come up with an algorithm for supplemental testing that will allow us to not defer those individuals who shouldn't be deferred.

ACTING CHAIRMAN ALLEN: Thank you.

Any questions or comments on that?

DR. SAYERS: My name is Merlin Sayers, and

I'm CEO at Carter Bloodcare, which is the community independent blood program for Dallas/Ft. Worth.

Dr. Allen, this will be an acronym-free statement.

(Laughter.)

I'd like to applaud the FDA for taking this approach to donor reentry. And without wanting to downplay the importance of the question that prompted these comments from the floor, it's not just the yield of a reentry program that's important.

One of the issues that we are dealing with is increasing incredulity on the part of donors who perceive that their donor deferral flies in the face of their own self-assessment of good health. And this is particularly true with regards to core antibody deferral.

These donors who dispute why they might have been deferred, if we cannot confirm to them that the reason they are deferred is because there is genuine risk to their health, these individuals essentially have become disincentives to others in the community when the deferred donors, particularly these core antibody deferred donors relate their experience to friends, family members, and neighbors, saying that they have indeed been deferred.

But during the deferral process, we do not have anything really beyond the precautionary principle to invoke to explain to them what the dual core deferral might mean to their good health. Reentry of these individuals, even individuals who might have been deferred 10 or 15 years ago, is going, to a significant extent, enable us to restore credibility in the minds of deferred donors, and I hope reduce the likelihood that their experience is going to act as a disincentive to other would-be donors.

So we do applaud the possibility of reentry for this particular group of deferred individuals.

ACTING CHAIRMAN ALLEN: Thank you. I think the point that you make is extremely important, and along with that -- with the comment from Dr. Fitzpatrick, about perhaps with the right evaluation, with new tests, we may never need to send out that letter of deferral initially, or the letter of -- "There is something that's not quite right in your testing mechanism. Please come back and donate again, so we can retest you."

You know, the point being that if you take healthy people with tests that have less than 100

percent sensitivity, and less than 100 percent specificity, you are going to have some inaccurate test results. The specificity being -- for a perfectly health donor being the most important measure there.

And our donors are supposed to be a totally healthy population. So it -- it really is an important issue in terms of the message that is given to them about their health.

Dr. Kleinman.

DR. KLEINMAN: Yes. One other aspect moving forward -- and that follows up on these comments -- is that when we do go to a new anti-core test, hopefully it will be more specific and the people that we defer really will have anti-core. But we still may have some false positives, even on a new test. So I think if we have a reentry algorithm in place, it permits our donor notification message to make more sense.

We can say to people, "We don't know" -- you have these results, and if you want to check them out further, if you're concerned, you can come back and potentially" -- and this is for the newly-identified people, you can potentially be reentered.

Now, we may not be able to reenter many of

those, but we could get a bad lot of reagent, for example, and wind up deferring persons for an anti-core test result that doesn't reproduce in the future. So I think just moving forward it gives us the ability to notify donors, and that issue that always comes up, "Well, if I'm okay, why can't I donate?"

And we never have an answer for it, and it creates cognitive dissidence in people's minds. "They are telling me I'm okay, but they're telling me I can't donate. That means they don't really think I'm okay. They just sort of think I'm kind of okay."

And by at least offering people reentry you can say, "We have a way of knowing -- we have a way of coming back to you and giving you further information." And I think that's valuable in the notification message also, psychologically valuable for donors, that we've gone the extra step.

ACTING CHAIRMAN ALLEN: Yes.

DR. SCHREIBER: I guess I'm a little bit confused, because when we're talking about reentry, we're only talking about reentry of the people who were deferred, and, therefore, had two subsequent repeat reactive tests and different donations.

80 percent of those, as Sue indicated, will -- that have the first repeat reactive will never

show up again in the door. So they will never be deferred for hepatitis B tests. So whatever you put out, they will never get a message letting them know what their true status of infectivity is, because they will never come back to the blood center, unless you go through some process to rerecruit those.

Sue's data showed that of those 20 percent of first-time donors that came in, 88 percent were -- were repeatedly reactive a second time. So those are the people who are deferred. And then --

DR. KLEINMAN: And that's with the current test, because you had --

DR. SCHREIBER: Right, right. But still -- you're still going to have a significant number of people, unless you really drive that false positivity rate way down that are not going to be told to come back and be retested.

I guess the other question I had is that when we talk about this reentry algorithm, and it seems that it's been deferred because there is not a more sensitive core antibody test, what about all of those people that are still out there?

And if we really are talking about a reentry algorithm, why wouldn't we institute it now, because that test is really dependent on the

subsequent NAT and other test followup. And the chance that they would get a second or a third -- a third core antibody test would probably be really, really small at this point.

ACTING CHAIRMAN ALLEN: Dr. Kleinman, are you responding to --

DR. KLEINMAN: Yes. I just wanted to clarify that. Maybe we didn't state it strongly enough. For reentry to really work, you have to have a licensed, more specific test that the blood center switches to. And without trying to promote or detract from any companies, we do believe that the Abbott PRISM assay is that more specific test, we do think that most Abbott users will switch to that test once it's licensed. And we do believe that the false positivity rate will go down.

But, I mean if you had a reentry algorithm and you're using the same test you used before, it's quite -- I mean, it wouldn't make sense, because you're still going to be repeatedly reactive on that non-specific test. Most people who are repeatedly reactive are repeatedly reactive to that test over time. They don't -- it doesn't go away. It's not a one-time thing.

So, really, we need that new test, and,

you know, it's been under consideration at FDA for quite a while, and hopefully it will be licensed at some point. And then, at that point, once test centers are using it, then reentry hopefully, if we validated the NAT assays and have the claims, then it might be a practical thing to do. But without that new assay, it's unlikely that reentry would have as good a yield as we would hope.

I don't know if that clarifies it a little bit.

ACTING CHAIRMAN ALLEN: Dr. Epstein.

DR. EPSTEIN: Yes. You know, I think what would be helpful to the FDA is if the committee members would comment specifically on the elements of the proposed algorithm and their scientific validity. And they are posted here on the slide that Dr. Kaplan put up, but just to highlight it it's the idea of an eight-week delay. That's to allow, you know, full-blown development of markers.

It's the idea of an offline test. In other words, you don't collect a unit that might be at risk before you've resolved the status. It's the idea of hepatitis B done on the individual sample -- in other words, ID NAT -- but with a sensitivity of at least 10 copies per mL. And I think you've heard that

that's hard to reach, but it is feasible.

And that, you know, some data appeared to have been -- generated less sensitive assays that, you know, could be debated. And that we have dropped the idea of looking at an anti-HBS as part of the reentry algorithm on account of the vaccine issue, and retaining the concept that you must demonstrate at least some negative anti-core test, whether it's the same assay or a different assay.

It's been explained that it's highly desirable to switch the assay, because if you simply use the same assay over again the likelihood it will be reactive again is very, very high.

So I think it would help us if, you know, there were specific comments on the elements of the algorithm.

ACTING CHAIRMAN ALLEN: Thank you. We will get to that.

Dr. Kuehnert.

DR. KUEHNERT: Yes. I just had a question about how this would work practically. So someone comes in, they're repeat reactive the first time, under this algorithm they would not get any kind of a letter or indication that they have a positive test. Is that right? Or are they going to have some

indication that they have a positive test the first time but can donate?

ACTING CHAIRMAN ALLEN: Dr. Kaplan, do you want to address that question?

DR. KAPLAN: Yes. So, basically, a person will be repeat reactive on any test, and then it will be deferred about eight weeks, and then it will be asked to -- a new sample will be collected. And so it will be again tested for surface anti-core and NAT.

DR. KUEHNERT: So they would be asked to come -- I guess I'm a little confused. So they'd be -- they'd get a letter saying that they have a positive test, and they need to come back for testing, or it would be when they come back to donate?

DR. KAPLAN: Well, this would have to be a testing at eight weeks, because they were reactive twice and --

DR. NELSON: The person has already been tested twice and found to be anti-core positive. So under the current algorithm, they are permanently deferred.

DR. KUEHNERT: Right.

DR. NELSON: And I guess what this is, it's a -- it's to try to reenter those people are permanently deferred because the feeling is that a lot

of permanently deferred --

DR. KUEHNERT: I understand that. I'm just saying, subsequent now to this, not thinking about people who have already been deferred, but people who in the future then are repeat reactive to anti-hep B core --

DR. KAPLAN: Okay. Basically, we are not right now talking about in the future, what will happen with a future license test. And that's something we are developing and we are thinking about it. However --

DR. KUEHNERT: But that will apply. If you change this algorithm, that will apply to them as well, won't it? What am I not understanding?

DR. KAPLAN: Yes. Robin wants to comment on this.

DR. BISWAS: Robin Biswas, FDA. This after somebody is repeat reactive on the initial -- initially. They wouldn't be deferred at that point. You know, the unit, of course, would not be used. The person could return and donate. I think that the blood organizations said that they do get a letter, in fact.

ACTING CHAIRMAN ALLEN: The Red Cross does send out a letter --

DR. BISWAS: Right.

ACTING CHAIRMAN ALLEN: -- after the first repeat reactive that invites the donor to come back in.

Dr. Stramer, do you want to clarify that?

DR. STRAMER: First-time core reactives are not deferred. It's up to the blood center -- well, clearly, the policies of two times deferral are up to the blood center. As Steve mentioned earlier, some blood centers will defer after the first time core reactive, because the yield on the second time is so low.

Some blood centers do the two times deferral policy and don't notify after the first time core deferral -- I mean, the first time core reactive and just let the donors come back and only notify them based on the second deferral.

We do notify after the first-time deferral for the reasons that these donors truly could be hepatitis B, in fact, and we believe it's the right thing to do. But they still can come back a second time.

So did I help clarify that?

DR. KUEHNERT: Sort of. I mean, what I'm asking, if this gets put into place now --

DR. STRAMER: Would that change?

DR. KUEHNERT: Yes, right. That's what I'm asking.

DR. STRAMER: Okay. I don't think the question on the table is whether the two-time core deferral policy will change. I think the question just is for those donors who have been core reactive two times, can they be reentered using the algorithm that the FDA proposes.

DR. KUEHNERT: That is correct. I know other people -- it will just be explained to them that this was a false positive test, and that now they can donate?

DR. KAPLAN: Well, the following slide -- it's basically at this point people will be told that they can donate, come back, they're negative, and then they will be tested again on the donation. And if it's negative, everything is fine. They can donate if everything is negative.

So I don't know if you can put the next slide --

DR. NELSON: Do you have an estimate of what proportion of the people who were twice positive will qualify under this?

DR. KAPLAN: No. Because we don't have to

-- we haven't seen the data yet with the PRISM, and that's what Sue Stramer's data was -- would fill that blank, and then let us know how -- fully validate this algorithm.

ACTING CHAIRMAN ALLEN: We've got several hands up. Dr. Strong, and then Dr. Doppelt.

DR. STRONG: We don't have good data on that. However, if you go back to the -- several years ago when the PRISM clinical trials were being done, comparing current license tests with the clinical trial results, there was about a 10-fold improvement in specificity. So it could -- that's what got everybody excited about the possibility that these donors could be recovered.

ACTING CHAIRMAN ALLEN: Mr. Doppelt.

DR. DOPPELT: I was just going to say I -- I thought I had it straight. Now I'm -- perhaps I'm confused. But in terms of fairness to the potential donor, it seems to be there's a difference between whether you say come back a third time and we're going to retest you, to see if you can be a donor, versus come back a third time and we're going to do some different tests and try and sort out whether or not you really have an -- you're really infectious or not.

So I'm a little bit confused as to what

information is going to be specifically relayed to the potential donors.

ACTING CHAIRMAN ALLEN: I think that's an important question to answer. And my guess is based on what we've already heard from different blood collection centers today that since there isn't any single way of handling that first time, that this is an issue that needs to either be clarified by the FDA or to allow the marketplace to sort it out on its own as currently has happened.

DR. DOPPELT: I mean, it just seems to me that you are far more likely to get patients to return if you tell them there is some additional testing that may be done to help sort this out versus come on back and try it again and let's see what happens.

DR. KLEIN: In the proposed FDA algorithm, they are going to do a NAT test that's quite sensitive. So they are, in fact, going to be doing something different.

ACTING CHAIRMAN ALLEN: Right. And one might, you know, raise the question -- and I'm sure there aren't data there to answer that today, but one might raise the question, if you've got a donor who is repeatedly reactive the first time, if you want to give them the best possible information, maybe you

ought to take, you know -- take the samples from that first donation and subject them to these additional tests instead of asking them to come back in eight weeks.

And I understand the reason for -- you know, for the eight-week or longer delay in terms of trying to find out if there's progression of markers over time that might indicate real infection, but if you want -- you know, if you want to give the most reassuring message back to the donor the first time, you would do that before you contact them the first time.

So, you know, I think there's a lot of permutations here that aren't really on the table and haven't been discussed.

Dr. Klein.

DR. KLEIN: Yes, I -- just to move this a little forward I hope, I'd like to support the concepts of the FDA algorithm. I think it's a good one, and I certainly support reentry for those people who have been deferred because of a two-time causative core antibody test.

At the same time, I'd also certainly like to encourage you to do whatever is possible to get a more specific core antibody test available, and also

keep an open mind to the possibility of having an algorithm that will prevent us having to defer such donors in the first place.

ACTING CHAIRMAN ALLEN: Thank you for that statement.

Dr. Strong.

DR. STRONG: Actually, I was going to support his proposal. I think we -- we have lots of -- we have lots of donors who get that message by letter. The letters are constructed in different ways, so the recipient may get a different message than was intended. But many of them at least will be told that they should see a physician, and they go to a physician and they get tested and it's negative, because they're using different tests.

So I think also to accelerate the discussion that we've probably heard enough, and I would certainly support it.

ACTING CHAIRMAN ALLEN: Yes. Dr. Hollinger.

DR. HOLLINGER: Again, I support this statement. I think it's a good statement. You might ask the question, you know, even when you look up, there will -- why even have to do -- I mean, if you have a test, a new test, let's say, that's licensed,

that is completely specific, then why do you even have to do HBV NAT in these patients?

And it goes back to the other issue is -- what has been taught by the blood bank a lot, and others here, is that it's important to tell those donors who do not have an infection, or do not have any evidence of exposure, that they're clear of this disease. I mean, there's nothing really there.

But I -- I also want to say that the blood bank does the clinicians a great service in this regard, because it also tells those who may be infected that this is an issue. So we've talked so much about the ones who may be falsely positive, and what it's going to do by telling them and talking to them about this issue.

It's just as equally important to talk to those who have a positive result of whether they might be infected, and that's where the HBV NAT comes in. And by looking at all of the things -- one of the things I like about getting information from the blood bank is that they are willing to send to clinicians all the information -- the cutoff levels, which I like to look at, the ALT levels, which are often -- when they were doing them, which is also -- can be elevated, and yet the patient -- the donor can

contribute. It could be a variety of things.

But all of that information is really helpful when you then can sit down with a person, and certainly by looking at this, then I could say to that person, "Look, I see no evidence that you have an active infection here. Even if it's a very specific positive anti-HBC test that's highly -- that has a high titer, or high concentration of the antibody," and the HBV NAT is negative.

Then I might be able to tell them that and reassure them that I see nothing in these studies that would have me concerned. I don't mind telling them that they're not infected. It's not a real issue at that point. And if they're positive, then one can deal with that issue also and reassure them about their risks, and so on, for transmission and other things which are usually very small.

ACTING CHAIRMAN ALLEN: Thank you.

Dr. Bianco, first of all, if you'll introduce yourself, and then Dr. Biswas.

DR. BIANCO: Celso Bianco, America's Blood Centers. I want to make two quick comments. One is the reason why only old people remember that plain -- the reason why we do it twice in terms of only deferring on the second time is from day one we

recognized that this is a lousy test, in terms of specificity. And we were hoping that we could recover some donors. Hopefully, a new test, more specific, we will be able to do it only once and make a conclusion about that. We don't need to wait for two.

The second point that I'd like to make is addressing one of the questions that Dr. Kaplan has asked us, and Dr. Epstein, is the question of offline testing. I think I'd like very much to discuss that. It appears that it's a note of caution that you will call back the donor, collect only a sample, and then, after getting the results on that sample, you will allow the collecting of blood units.

Donors, unless they are very angry with us, and they come together with a bill from their doctor for all the tests that they did because of our core positive doctor, and they want the blood center to reimburse them for that, they don't come back just for a test. They come back for a blood donation. That's what attracts them is their sense of altruism, sense of trying to help.

And so the chances, I believe, of releasing a unit inappropriately because of a test result are very small. Usually units are released inappropriately when it happens. Because of other

issues, they are not so much dependent on the interaction, direct interface between testing machines, computers, and the computers that release the unit of blood.

So I would ask that these requirements be dropped.

Thank you.

ACTING CHAIRMAN ALLEN: Dr. Epstein first.

DR. EPSTEIN: Well, I think the other side of that argument needs to be heard, which is that there is a finite risk of inadvertent/inappropriate release of a unit, and the risk of consequences goes up if that is, in fact, an at-risk unit.

So that's why in our reentry algorithms we have always wanted to requalify the donor offline, because otherwise you've drawn an at-risk unit, you don't know its status yet, it might turn out to be true positive, and there's a finite risk unrelated to the testing that it might get out.

ACTING CHAIRMAN ALLEN: Dr. Klein.

DR. KLEIN: I'm not sure that those data are correct, Dr. Bianco. We've never had any difficulty getting people back for our studies just to be tested. And, in fact, we've found that people are more reluctant to give a unit that they think might be

discarded, so I don't know of any data to say that they won't come back to be tested. Our experience is exactly the opposite.

ACTING CHAIRMAN ALLEN: Dr. Kuehnert, and then Dr. Biswas.

DR. KUEHNERT: I just wanted to say that I'm supportive of the statement. I think, you know, this is probably going to apply to a very small number of people, at least the way the tests are currently applied. And I do have concerns about sort of donor counseling and health, but that's not at issue here.

I think it could be a good topic for another government committee, but, you know, just with that in mind I think donor health is important. And I think there is some work to be done here. There's probably going to be more confusion when the new algorithm is applied initially concerning counseling, but overall I think this is a good statement.

ACTING CHAIRMAN ALLEN: Dr. Biswas.

DR. BISWAS: I just wanted to say, you know, there's been a lot of talk, which I agree with you, about the specificity, lack of it, and, you know, the importance of having a specific anti-core test. But remember that the reason we are bringing this issue to you at this time is because of the

improvement or the development of NAT technology, and that really was the driving force here. I just wanted to say that.

ACTING CHAIRMAN ALLEN: Dr. Busch.

DR. BUSCH: Yes, I want to address the issue of the NAT sensitivity requirement. The only donors who will be reinstatable will have to be negative on test of records -- surface antigen anti-core assays. In fact, these donors, if they came in for the first time today, they wouldn't be screened out at all. They were the historical false positive anti-cores.

We know in true anti-core positives that are persistently anti-core reactive on all the assays, if you progressively increase the sensitivity of your NAT test, you will cull in a little bit more, you'll detect a smaller incremental fraction of very low viremic donors.

So the studies that we've done and Sue described where if you go from 100 copy to 50 copy to 10 copy sensitivity, you pick up a small fraction of additional low viremic carriage, but this is in people who are fully seroreactive and would be reactive on any anti-core test.

And, you know, the requirement for 10

copy, 95 percent hit rate is making all the companies have to do multiple replicates -- would make the NAT screening cumbersome for the blood centers even if these tests can achieve that level of sensitivity.

And I just don't understand where that number came from and why FDA is pushing such an extremely stringent low sensitivity threshold when, again, these donors would be eligible today. And in the absence of any NAT screening right now, and certainly once NAT is in place, it will be likely done on small pools, and certainly not with assays that have this level of sensitivity.

ACTING CHAIRMAN ALLEN: Would anybody like to respond to Dr. Busch's comment?

DR. BISWAS: Mike, I think you were asking -- I didn't hear everything you said, but your questioning the -- the requirement for 10 copies and less, that was it. I should say that -- actually, Dr. Kaplan did say it earlier -- that we are sort of flexible on that point. It will depend on some of the results -- on the results that we get back using the less sensitive test that Sue Stramer is doing with NGI.

And it -- but I should say that although it is stringent, it does seem to be as though it is

possible. But as I said, we would be flexible and will take into account the clinical trials -- the results of the trials that are being done under IND, and that we are flexible on that.

ACTING CHAIRMAN ALLEN: Dr. Kaplan, did you want to respond?

DR. KAPLAN: Yes. You know, someone was deferred -- gave twice anti-core, it was repeat reactive anti-core, so this -- there's some flag there. It could be -- you know, it could be a false positive measurement, but there's a flag there. So there's some rationale there to try to increase the sensitivity of the NAT. And as we heard from Roche, that's achievable.

And then, so what -- what you don't want to do is someone that's -- you know, has a very low core and a very -- a very low DNA, but it could be an infectious unit to -- to reenter it. So there's some rationale for asking the state-of-the-art sensitivity, maximum sensitivity achievable.

ACTING CHAIRMAN ALLEN: Dr. Lew.

DR. LEW: I think I heard pretty well that, you know, for those who are -- and someone can correct me -- anti-core antibody positive alone, but if you keep on going down, you know, might detect

real, real low levels, and for our immunocompetent patient it's quite possible these patients will not become infected.

But since a lot of our blood products go to immunocompromised patients, and they get a lot of blood products, I'm not hearing that potentially, you know, these -- if these patients do get it, that they will become positive. They will get infection. So I'm a little concerned about those patients and how this algorithm works.

ACTING CHAIRMAN ALLEN: I think that's a very important and interesting comment.

Dr. Hollinger.

DR. HOLLINGER: I think part of it comes back to we need to know how many of the -- these new licensed tests might be negative when the other test is positive, and that we need to know the false negative rate, if any, of the -- of HBV DNA in those samples.

If not, then it -- then I don't think that's -- if it's not there, and I suspect it may not be, then they're not at risk. I don't think they would be at risk in a -- with a good assay that's very specific. And if it's given to an immunocompromised individual, if there's no virus in the blood, or

detectable virus, then those patients are not going to get --

DR. KAPLAN: Can I add something? It's that this algorithm that you have the donor tested at least three times, you know, it was -- all markers negative, but it was repeat reactive, then you bring it -- bring them back at eight weeks, and then you test them with all the battery again. And then, if they are negative, they are -- then you have a donation.

So, you know, if it's low levels, you should -- you should be able to detect it at that time. I think that's a pretty well functional algorithm.

ACTING CHAIRMAN ALLEN: Dr. Lew, and then Dr. Strong.

DR. LEW: No, I don't really have a problem with this algorithm. I'm just talking -- hearing the conversation of other methods of how to decide. And as far as I'm aware, I don't know if there's any lab test that I know of that's 100 percent sensitive-specific. You know, I'd keel over if there ever was one.

(Laughter.)

So --

DR. STRONG: But I think the point is that it won't be worse than it is now, because the new tests are actually both more specific and more sensitive. So, if anything, we're going to catch more.

ACTING CHAIRMAN ALLEN: Dr. Kleinman.

DR. KLEINMAN: Yes. I just wanted to emphasize, I think the issue of reentering somebody is dependent on the sensitivity of the tests you use to reenter. And remember, the reentry scheme includes two tests, three tests actually -- surface antigen as well. But it includes another anti-core test.

So if you are positive on that anti-core test by a second manufacturer, you're out. It doesn't matter what your HBV NAT test shows. So if that's an equally sensitive test, I mean, if you -- you're negative on that anti-core test, and that anti-core test is equally or more sensitive than the one you're using, you don't really have anti-core. And, therefore, you wouldn't need any NAT testing.

I mean, it would just be the same as if that person came in today and had never been deferred in the past, and was screened with the new test. They'd be anti-core negative, and you could say, "Well, gee, we might be missing somebody. Maybe we

should do HPB NAT testing at 10 copies per mL on every donor in order to increase the safety of the blood supply."

Well, obviously, you're not going to do that. So I think we need to see the clinical trial data that validates the reentry algorithm, and then we'll know if there are discrepancies between -- if somebody is negative on the new test, and actually has HBV DNA in their serum. If we find somebody like that, how sensitive in that test did we need to do to find that person? Did we need to do a 10 copy mL test? Or was a 50 copy mL test enough? And then we would be able to test -- to maybe set our sensitivity levels.

Now, I don't know if we could do a big enough clinical trial, because I don't expect we'll have many people who are NAT positive failing the second reentry test. So it does become kind of arbitrary. But I think maybe that's why the FDA is saying there is some flexibility; we just don't have the data to know yet.

But I think, really, to answer the question that's on the floor we -- we do have protections in place with this algorithm, and even if we were to increase the -- or I guess decrease the

sensitivity of the NAT, increase the copy number of detection, even if we were to do that, I think our patients would be protected from getting unsafe units because of the second core test.

ACTING CHAIRMAN ALLEN: Dr. Stramer.

DR. STRAMER: Yes. Two points, commenting along the same lines that Dr. Strong brought up. As Blaine mentioned in his presentation with the use of reductant in this new, more specific test, it's the use of this chemical treatment that eliminates the false positive non-specific early IGM antibodies that cause interferences or false positivity in the test.

But because of this concern, the test before the FDA has gone through extremely robust validation. And as has been pointed out before by Mike, the test is not only more specific but is actually more sensitive because of the disassembly of all these false positive, non-specific antibodies, so that it's more specific for low-level true antibodies.

So the test is more sensitive, it's more specific, and what FDA is proposing is actually a very robust reentry algorithm. You would use the more sensitive and more specific anti-core test. The donor would not only have to be negative on followup, but then again negative at donation.

And we're kind of quibbling -- is 10 copy, 50 copy -- certainly, if you want to increase the catchment, a more sensitive test is what you should do, or to really identify those people who are circulating DNA, from a public health perspective and from a reentry perspective, and there are tests that are -- you know, that can be achieved.

Whether it's 10 copies or 30 copies, I don't think that makes a difference. I mean, we were -- when Red Cross presented data to FDA, FDA actually said 30 copies was inadequate. So we moved it down to 10 copies quite arbitrarily, just because if we thought 30 wasn't enough, well, what would be sensitive enough? So we just chose 10 as something that we thought even the FDA wouldn't reject.

So that's kind of the derivation of the 10 copies per mL. But robustness has ben built in the algorithm, and robustness has been built into the tests that are before the FDA for licensure.

ACTING CHAIRMAN ALLEN: I would like -- okay. Dr. Lew.

DR. LEW: If I could just say that -- just for -- I think everyone agrees it's a given, but for clarification, that this statement seems suitable for most people -- given the caveat, we're talking about

a more sensitive hepatitis C core antibody test. I mean, just for that clarification.

ACTING CHAIRMAN ALLEN: Dr. Epstein.

DR. EPSTEIN: I just want to comment on what we know and what we don't. What we know, and these are Sue Stramer's data from the histogram that you showed of viral load, there are indeed samples that have a viral load of 10 copies per mL or less in individuals who have a repeatedly reactive anti-core test, and who -- 65 percent of whom have a negative HBsAg test.

What we don't know is whether those same samples would be found if a third independent test were negative for anti-core. And we couldn't show you those data because we don't have those data. But no one should think that there are no samples with low-level viremia in individuals with so-called anti-core only by current testing. There are such people.

So the problem here is, you know, can we really place our faith that there won't be any such low-level viremias in those in whom another EIA is negative. We just don't know until we have those data.

So I just don't see -- but the, you know, proposal that we use assays as sensitive, that is

based on the observation that some of the true positives do have DNA at that low level. What we don't know is whether the ones with the further negative EIA would have DNA at that level.

ACTING CHAIRMAN ALLEN: Thank you.

Let me come back to the proposed FDA statement here and just -- we've heard some -- actually, all of the committee members that have spoken have spoken in favor of the basic algorithm. What I'd like to do is just ask the committee for any additional comments that anyone might have about the FDA proposed algorithm and issues directly related to that.

DR. QUIROLO: Well, it would seem to me that if this is such a sensitive core test that you wouldn't really need to come back twice to have two -- a repeat reactive and then a third test, it sounds like, before you did the NAT and the surface antigen again before donation.

The other thing is if this is such a sensitive test, what's the possibility that somebody would come back for their second core test, being reactive the first time, and being negative the second time? And if you're using the same core over and over again, isn't that the same dilemma you're in now where

you're -- if you're reactive once, you're more likely to be reactive over and over again?

DR. KAPLAN: Well, this algorithm basically solves a present problem. And so, you know, Mrs. Stramer -- through the number of a million people that they could be -- reenter with this.

I think that the -- the other issue that you are raising is: what is the performance of this new test that has not been approved? And how will that fill into the -- this algorithm or the deferral -- deferred algorithms for repeat reactives in core? It's something we have to see at the moment of approval of that new test. I think that's on the table at this point.

However, we are -- we agree with -- I personally agree with what you said is that, yes, that's a very important point that we have to retain -- or reevaluate when we have this more specific, more sensitive test available. Yes.

ACTING CHAIRMAN ALLEN: In actual fact, if the belief is that most of these people are not truly infected but are false reactives, then, in fact, the issue is not -- it really doesn't matter how much more sensitive the test is. The question really is needing a test of increased specificity, and the new

generation tests coming on the market should meet that.

DR. QUIROLO: Will those be the initial tests, though, for the new donors once the test is available? Will that be -- will the more sensitive test be the initial test? Or will there be two tests, a less specific and then another second test after you fail the first one?

DR. KAPLAN: We don't know how many people will adopt that. We don't even know when this will be licensed at this point. And I think that's a market force -- speculation at this moment.

ACTING CHAIRMAN ALLEN: Yes. As you were asking your question, I saw heads nodding around the room from blood collection people.

Dr. Strong, do you want to comment on --

DR. STRONG: Yes. Once we have a new test that has greater sensitivity and specificity, the old test goes away. We'll only be using one test. And the donor is likely to be deferred, because we fully expect that we're going to be doing DNA as well.

ACTING CHAIRMAN ALLEN: Other specific comments from the committee members on the FDA proposed reentry algorithm? Anybody have major heartburn over it? We basically heard comments in

support with a few questions on technical details.

Okay. I would like to add my support to the basic proposal, and, you know, would encourage, based on all that I've heard today, I would encourage the FDA to continue working to get this completed as rapidly as possible.

Does the committee -- and this is a straw vote -- does the committee believe that this needs to come back to the committee again once as a formal question, or have we given sufficient direction to the FDA and would like to encourage them to move forward as rapidly as possible to implement?

DR. EPSTEIN: Jim, if I could just clarify, that, you know, the committee serves to advise us on the science. You know, FDA takes unto itself the responsibility of determining the policy. So in phrasing your question, really, is -- the question is: are there other scientific issues that need to be brought back to the committee?

ACTING CHAIRMAN ALLEN: Thank you. Yes, that was what I meant.

Dr. Kuehnert.

DR. KUEHNERT: I just wanted to ask if there -- there was some discussion about the sensitivity level required, if that's a -- still an

open question, or has FDA gotten enough guidance on that issue?

DR. EPSTEIN: Well, our position is that we want to see the data that emerge from combining historic, you know, twice anti-core repeat reactives with negative results of a more sensitive and specific screen, and then see: a) if there are any DNA positives, and b) what their levels are. But we think that those studies need to be done with the most sensitive available assay, otherwise we'll never get a meaningful answer.

So I can't answer the question on point. I can only answer it by saying this is why we want to see the studies that we describe.

DR. KUEHNERT: And I think, you know, the discussions we've had in previous meetings about mini-pool NAT screening in general, you know, the consensus was for more sensitive screening. So I think this all works towards that.

ACTING CHAIRMAN ALLEN: All right. Any other comments or questions? Does the FDA want further discussion from the committee, or have you achieved what -- okay.

It's approximately 12:15. The official game clock here says it's 12:21. That's a little

faster than my watch, which tends to be one or two minutes fast most of the time. We will adjourn for lunch. Let's plan to have -- have people back -- we will reconvene at 1:20 by the game clock here.

(Whereupon, at 12:15 p.m., the proceedings in the foregoing matter recessed for lunch.)

DR. SMALLWOOD: We're ready to reconvene. May I ask all Committee members present to please take your seats, and may I have the attention of the audience?

Before we start, I just wanted to make an announcement. Dr. Martin Ruta this morning mentioned a draft guidance that was expected to be published, and I just wanted to announce publicly that on the FDA web site guidance for industry, use of nucleic acid tests on pooled and individual samples from donors of whole blood and blood components, including source plasma and source leukocytes to adequately and appropriately reduce the risk of transmission of HIV I and HCV has been posted as of this morning. So it is on the FDA web site. Dr. Allen, were you ready? We're ready to reconvene.

ACTING CHAIRMAN ALLEN: Good afternoon. We're ready to continue our discussion with Topic 2,

the potential risk of Simian Foamy Virus transmission by blood transfusion. Our first presentation will be the introduction and background by Dr. Tabor.

DR. TABOR: Good afternoon. I'd like to begin by thanking the other speakers who will be participating in this session. Dr. Kahn from Sieber but also people who came from a great distance -- Dr. Heneine, Dr. Brooks, Dr. Peter Gantz and Dr. Lerka -- all of whom you'll be hearing from soon.

The potential risk of transmission of Simian Foamy Virus by blood transfusion is being brought to be BPAC at this time because of a report in the Lancet that this retrovirus is being transmitted under so-called natural conditions from non-human primates to the human population in Cameroon. This not only places a renewed focus on Simian Foamy Virus transmission but also represents a mechanism by which Simian Foamy Virus and other non-human primate retroviruses might enter the human population and ultimately the blood supply.

This issue is made more urgent by recent research developments related to the possible transmission of Simian Foamy Virus to non-human primates by blood transfusion, and this information will be presented by the other speakers at this

Advisory Committee meeting today. Can I have the next slide, please?

Transmission of Simian Foamy Virus to humans due to occupational exposure to infected non-human primates has been reported to occur in two to five percent of persons working with non-human primates in research institutions and zoos. Most of the infected persons had histories of scratch or bite injuries caused by the non-human primates.

Because of these reports, the topic of Simian Foamy Virus was discussed at the December 13, 2001 BPAC and the consensus of BPAC in 2001 was that more data were needed to determine whether Simian Foamy Virus presented any risk to the safety of blood transfusions. Next slide, please.

This year, in the March 20, 2004 issue of the Lancet, transmission of Simian Foamy Virus to humans by so-called non-occupational contact with non-human primates was reported by Wolfe et al. and was accompanied by a commentary by Peters et al. In fact, the exposure was only somewhat non-occupational in the generally accepted use of the term, "occupational," since the authors felt that the transmission probably occurred as a result of a hunting preparation and consumption of food made from tissues of non-human

primates, sometimes referred to as bush meat. Next slide, please.

Wolfe et al. studied 1,099 residents of the tropical forest area of Cameroon who had regular contact with blood or body fluids of non-human primates. Antibodies to Simian Foamy Virus were found in ten persons. Among these ten, Simian Foamy Virus itself was found by RTPCR in the peripheral blood lymphocytes of three. These three apparently had acquired Simian Foamy Virus in three distinctly separate transmissions from non-human primates. The individuals were each from different villages and the nucleotide sequence of each isolate of Simian Foamy Virus showed that the Simian Foamy Virus in each person was from a different primate species, each consistent with that person's individual hunting and food preparation history, enveloping gorilla, mandrill and cercopithecus species, respectively. These observations are consistent with the known fact that the different Simian Foamy Virus strains are each highly specific for their host species. Next slide, please.

Simian Foamy Virus is quite prevalent in the non-human primate populations, ranging from 31 to 61 percent among non-human primates in the wild and

from 70 to 90 percent among those in captivity. In contrast, the prevalence of Simian Immunodeficiency Virus among non-human primates in the wild, in the same geographic areas, is about 16 percent, and the prevalence of Simian T-Lymphotropic Virus is about 11 percent.

What is the risk of Simian Foamy Virus becoming widespread among humans once it has entered the human populations. Recent reports by Heneine et al. and Switzer et al. provide strong evidence of persistent Simian Foamy Virus infections in humans, with infections lasting as long as 19 and 26 years or longer. However, human-to-human spread of Simian Foamy Virus has not yet been shown to occur. We do know that the study of a small number of wives evaluated in studies of persons with occupationally acquired Simian Foamy Virus infection has not revealed any instances of spousal transmission. And the low percentage of infected persons working in primate facilities suggests that human-to-human transmission, if it ever occurs, must be very inefficient.

Nevertheless, there is clearly primate-to-primate transmission among non-human primates in the wild. Transmission between primates is believed to occur by means of saliva since Simian Foamy Virus can

be isolated easily from the saliva of infected non-human primates. Transmission by bites theoretically could be one mechanism of transmission from captive, non-human primates to their handlers.

In support of this theory, a gorilla strain of Simian Foamy Virus was detected in two Cameroonian hunters who had had multiple bite injuries during separate fights with gorillas. As we evaluate additional human cases, particularly those occurring in areas where careful observation is possible, we may have to revisit the issues of human-to-human transmission and the potential infectivity of human saliva if any evidence is found contrary to the concept that human-to-human spread does not occur. Next slide, please.

There are few clinical studies to evaluate the transmission of Simian Foamy Virus by blood transfusion. In one small lookback study, reported by Boneva et al., summarized in this slide, no evidence of transmission of Simian Foamy Virus by human-to-human blood transfusion was found. Although this study may provide a basis for optimism, its small size and the absence of information about viral load in the blood donor preclude any firm conclusions. And, in addition, no ideologic association between Simian

Foamy Virus and any human disease has been established so far. Next slide, please.

Our concerns today are twofold. We wish to discuss the possibility that Simian Foamy Virus could be transmitted by blood transfusion, and we wish to discuss the risks suggested by Simian Foamy Virus as a model of cross-species transmission of a retrovirus. We know that two Simian Immunodeficiency Virus strains emerged to form HIV Types I and II. And two strains of another Simian retrovirus, Simian T-Lymphotropic Virus, emerged to form HTLV times one and two.

Human diseases associated with infections with these emerging retroviruses were not recognized for many years, and in the case of HTLV I this delay was due in part to the fact that fewer than five percent of persons infected with the virus develop a disease. Even though no human disease has been linked to Simian Foamy Virus infection, these theoretical concerns I've just described may leave many people to urge taking precautionary measures. However, such precautionary regulatory measures require careful consideration of risk level and of the impact on the availability of needed blood products.

Handlers of non-human primates in the

laboratory setting are not the only people with close contact with non-human primates. Other groups include zoo workers, people who have non-human primates as pets -- there are about 15,000 households with such pets in the United States -- and bench laboratory scientists and technicians who conduct testing of primate serum and tissues. Any of these people could be at various levels of risk for acquiring Simian Foamy Virus infection. The risk for scientists conducting behavioral studies on non-human primates theoretically could be lower than that for scientists conducting other types of studies. It would be a challenge to define precisely which individuals would pose a risk as blood donors.

And now I'd like to show the questions we have for the Committee. Next slide, please. The first question: "In the absence of any known disease association, should FDA be concerned about the potential for transfusion transmission of Simian Foamy Virus?" Next slide.

The second question is, "Do the recent evidence of Simian Foamy Virus infections in humans and the evidence of transmissibility of Simian Foamy Virus by blood in animal studies heighten concern that known and unknown pathogenic viruses of non-human

primates could enter the human blood supply?" Next slide.

And the third question, "Do the available scientific data warrant possible consideration of donor exclusion criteria for exposure to non-human primates?" And we would like the Committee to please discuss the factors that we should consider if you recommend this.

Thank you. I'll now take any questions and after that we'll move on to the other presentations. Are there any questions? Dr. Lew.

DR. LEW: Can you just remind me, did you tell us if it causes any disease in the monkeys?

DR. TABOR: I believe it does not, but the next speaker is a sufficient expert that I'll defer to him. And with that, I'll introduce Dr. Walid Heneine from the Centers for Disease Control and Prevention.

DR. NELSON: I was a visitor on one of the other FDA committees a few years ago, namely the -- I forgot the name of the committee but it dealt with transplanted organs and tissues. And my recollection of that meeting is that currently tissues from non-human primates are not acceptable or permitted by FDA to be transplanted into humans. Is that correct or am I wrong on that?

DR. TABOR: I'd like to defer that question to Dr. Epstein, if I may.

DR. EPSTEIN: The FDA doesn't regulate organ transplantation.

DR. NELSON: Oh. Well, somehow that committee was discussing it and it was an FDA committee. Maybe I was in the wrong room.

(Laughter.)

DR. NELSON: Xenotransplants.

DR. EPSTEIN: Oh, xenotransplants. Oh, okay. Sorry. Well, there was a moratorium on xenotransplantation from non-human primates on account of various viruses and primates. But I believe the moratorium is now lifted, and I can't comment on the current status.

DR. HOLLINGER: This may be discussed later and if it is, Ed, just let me know. When we talk about donors who have been infected from like '81 to 2000, this is a Boneva study, what do we mean by infected from -- were there bloods available then in which they found nucleic acid and so on?

DR. TABOR: Well, let me first ask, is Dr. Boneva in the audience by any chance? No. Okay. I've read the paper and I can describe it to you, but perhaps I can ask Dr. Heneine to answer that one too

since he's from CDC. The question was how the blood donor samples were collected after the donor was identified in the Dr. Boneva study.

DR. HENEINE: I'll get to it probably in my talk, but let me follow up on the answer of Dr. Epstein regarding the xenotransplantation guidelines. I think the moratorium on using non-human primates as sources of tissues and organs is still in place, and the only species now we think is useful is pigs as sources of xenographs because of the issues of xenogeneic infections and xenogeneic viruses.

DR. EPSTEIN: I just wanted to clarify, I thought what Dr. Nelson was asking was whether there's a policy to exclude human organ donation from humans exposed to primates, and that would not be something that's FDA regulated.

ACTING CHAIRMAN ALLEN: We will have a chance for discussion later, so what I would suggest is that we get through our presentations, asking questions just of the presenters just for clarification, and then we'll get into the broader discussion later. And I suspect most of the -- or all of the presenters will still be here and can answer at any time. Dr. Heneine?

DR. HENEINE: Thank you again for giving

me the opportunity to present our data. I begin with the first slide. I'll be giving a summary of our data thus far. How can I move the -- move it for me. Next slide, please.

But I'd like to reiterate the fact about our experiences or the lessons we've learned from pathogenic retroviruses we're aware of that have resulted from cross-species transmission. And of course like you've heard from Dr. Tabor, HIV-1, HIV-2 are primary examples that originated from transmission of Simian Immunodeficiency Viruses from chimpanzees and sutimangabees, respectively. But we have also additional examples, human T-Lymphotropic Virus Type 1 or HTLV-1 that resulted from STLV-1; Gibbon Ape Leukemia Virus resulted from a strain of Murine Leukemia Virus and Feline Leukemia Virus that also resulted from transmission from Murine Leukemia Viruses. So these are not dead end zoonotic transmissions, but very successful cross-species infections that have become established and endemic in the new host end-cause disease. So this really highlights the ability of retroviruses to cross species that persist and then spread into the new host and cause disease in many instances. So next.

So what about transmission of Simian

retroviruses, what are the mechanisms that could be in fold in this transmission? Or course, hunting non-human primates in the wild, butchering, preparation consumption, keeping non-human primates as pets and of course occupational exposures in zoos and primate centers. Next.

So although we know that HIV-1 and HIV-2 as well as HTLV-1 resulted from these cross-species infections, we still don't know if SIV or other Simian retroviruses continue to cross species to human and what are the public health sequences of these events. We were aware almost ten years ago of isolated cases of transmission of SIV, something our lab has done, and Germans reported a couple of cases of Simian Foamy Virus transmission in occupationally exposed person. And, of course, these reports have raised concerns about the magnitude of these events. So back in 1995 we decided to establish a link study for volunteer testing for simian retroviruses in exposed laboratory workers and primate handlers. Next.

So what today I will do is summarize the data that we have generated from these studies. I will be talking about the prevalence of Simian retroviruses among North American primate handlers, prevalence of the Simian Foamy Viruses in Central

Africa, what we have learned from these studies about human-to-human transmissibility and disease and also comment on the risks from other Simian retroviruses, the topic that Dr. Tabor highlighted earlier. Next.

So a little bit of background about those viruses. SIV, we know it's prevalent in African non-human primates, shows a lot of diversity, at least nine different lineages have been described, and the list keeps growing as more studies are done. They're generally benign and natural hosts and only cause disease when they sometimes spread to different hosts. The Simian T-Cell Lymphotropic Viruses, again, they're prevalent in African and Asian primates, there are three different viral species here and can cause disease. Simian Retrovirus Type D or D Type retroviruses are prevalent in Asian Macaques, can cause a pathogenic international host and cause AIDS-like illnesses. And, finally, the Simian Lymphoma viruses, they're ubiquitous in almost all primate species, show species-specific clades and like SIV they appear to be benign in their natural hosts. Next.

So what is the study design on the surveillance in North America that we've been doing? We have a protocol where we invite primate research

centers who wish to participate on a voluntary basis in a linked study for testing for these viruses. Participants fill out the questionnaire and provide a serum sample that gets tested serologically first for all those viruses, and we've developed assays that are not commercially available. The active samples are then identified, and those persons are contacted again to provide an additional sample where serology is repeated and DNA is obtained from peripheral blood lymphocytes to do PCR sequence analysis and in many case virus isolation. And we've developed diagnostics to do those tests. And once the individual is found to be infected, the participant will be interviewed closely. Next.

So this is an update of the results we have so far. We have 20 institutions that have joined the study, a total of 3,000 samples collected. Institutions have two choices: To only get tested for SIV or get tested for all four retroviruses. But 441 persons so far have elected to be tested for the four retroviruses. They're from 13 research institutions and four zoos. And here are the results. We saw only two cases were positive for SIV, and these are older cases that we've reported in the past. No STLV infections have been identified. Two cases were

seropositive for Simian retroviruses, and I will comment on this later. However, the big surprise was with the prevalence with the Simian Foamy Virus where 15 cases have been identified so far, giving a prevalence of 3.4 percent. One of the SRV-positive cases is also Simian Foamy seropositive. Next.

This gives you an idea about the distribution in research centers versus zoos; again, 3.2 percent in centers versus 4.5 percent. And here are the papers. Just to clarify, the first four cases were published back in 1998, ten additional cases were published or reported earlier this year, and the 15th is a recently identified case from a new institution that enrolled. Next.

Again, we saw similar results from a unlinked survey of zoo workers that was led by Paul Sandstrom and he was with us at CDC where the prevalence of Simian Foamy Virus was found to be about three percent as well in this population versus zero percent rise in workers that had no contact with non-human primates. Next.

Again, as I mentioned, these viruses have cross-speciated with their natural host and therefore follow genetic trees that are similar to the hosts, so, therefore, when we see virus and analyzed it and

we can look with which viral species or cluster, you can identify the species origin of these infections. Here, for example, is the ape group, here's the monkey group from baboon, African Queen monkey, macaque and so forth. The samples we were able to evaluate thus far show us that there are eight cases that have chimp-like SIV, four that have baboon-like, one macaque and one African Queen monkey-like. So large variety of different SIV clades that have been identified. And this makes sense because these are the species that are usually commonly used in primate centers and in zoos. Next.

From the interviews, we collect a lot of case histories and histories of exposures, and we also obtain archive samples in these institutions which gives us an idea about the duration of seropositivity in these individuals. In general, all the cases usually report working with the primate species that was responsible for the infection, and many but not all report receiving bites or injuries from that species. The duration of seropositivity, again, shows recent as well as long-standing infections. Next.

So what about the key issues here, about disease and human-to-human transmissibility. Here's some basic background on those cases. Thirteen cases

were males and two were females; in fact, they were from the zoos, the females. Seropositivity is a mean of 17 -- or, no, I think it's 19 years, six to 28 years documented from archive samples. Now, what about disease cessation? Those cases report being in generally good health, but, again, this is -- I'd like to stop at this point because we receive a lot of questions on this issue. This is a limitation of surveying health persons. We enroll full-time employees that are health, and we identify some of them to be infected, and then we ask them, "How do you feel," and the result is, "We feel fine." So in fact this is not the best design to identify disease cessation. So keep that in mind, that limitation of the study in mind as we discuss the implications of the data.

However, we can tell something about sexual transmission. So six wives of men that reported regular sexual activity, unprotected sexual activity, remain uninfected despite mean documented exposure of 14.5 years, suggesting that probably male-to-female transmission, sexual transmission is not very easy. But, again, we don't have a lot of power in these numbers, and we cannot exclude transmission after longer exposures, similar to the case scenario

with HTLV-1 or HTLV-2. We require longer times for sexual transmission.

However, we have a second protocol where we invite infected cases to participate for a long-term follow-up study where we can follow them up clinically and immunologically and virologically for five years, and seven cases have opted to participate in this study. Next.

Of course, the main issue here is transmissibility by donated blood. This is important because we document persistent peripheral blood lymphocyte associated viremia in all cases. We can easily amplify the viral sequences from lymphocytes and also isolate virus from those cases. We're a bit surprised that 11 of the workers we identified were blood donors and six were confirmed to be positive. This is confirmation retrospectively at the time of donation. So if I understood the question earlier about the case from the lookback study which is here, this is a case that was identified retrospectively to have donated blood, and the lookback study here that we worked with with Dr. Boneva really targeted the recipients of that components from this case, in particular two recipients of red cells, one of filter red cells and one of platelets and all tested

negative. Again, very little data, not a lot, but we need additional data to have conclusive information. Next.

So the bigger question now is what about transmission in the natural setting? Are these infections only get transmitted to humans in special type of occupational exposures or they also occur in the natural setting? As I mentioned, hunting, butchering and keeping pets is a primary mechanism for this transmission and keeping in fact the estimates about the bush meat trade mainly in Central Africa, central and deforested areas in West Africa. But one to five million tons annually has been estimated to be traded. So there's a lot of contact that occurs in that region. Next.

So we collaborated with Rubin Hopkins to answer the question of prevalence of SFV in samples that were collected from rural villages and persons that reported direct contact with non-human primates. The progress we saw was 0.9 percent. They were all in lowland forests. This is the forested areas where hunting takes place, is prevalent. There were seven men, three women from different villages, and three were PCR-positive. Next.

These are the sites of data collection and

these were the areas where the seropositive cases were identified, and the ones in red were those that were also PCR-positive. Again, this area where prevalent practice of hunting and butchering takes place. Next.

Analysis indicated that we have one case infected with a gorilla, one with mandrill and one with a cercopithecus species, more precisely dubrozakeenan. Next.

These are the examples, the pictures of these non-human primates. Next.

And, again, more recent better to confirm our findings from the Pasteur Institute in Paris where the lab reported those in the fourth international Foamy Virus meeting in July in Germany where he screened also southern Cameroonian villagers for Foamy and found 11 out of 720 to be positive, three were PCR-positive and two were hunters, 60 and 67 years old, that have gorilla-type SIV and reported injuries, like you've heard earlier, from gorillas. There was a third person who was a woman that did not -- has no history of -- did not report hunting but contact with bush meat. And she has a chimpanzee-type Simian Foamy Virus infection. Next.

So the bigger question right now is what is the scope of these infections? Is it an infection

that is only limited to people that have direct contact with primates or are we dealing with a global dissemination of this infection that has gone so far unrecognized? And this is one scenario, how can a global dissemination or emergence of this virus can happen, first, from exposed injured individuals that some of them do get infected but not associated with secondary transmission. However, very few can lead to secondary human-to-human transmission for maybe one or two generations, but then maybe this local epidemic can die out or it can have -- one of those can really adapt this virus and be able to disseminate and spread among humans. So we really do not know where we are in the scheme of things. Are we here or are we already here? However, only expanded screening would probably tell us where we are. Next.

So how widespread is SIV? Is it -- first, you can think of it in West Central African countries. What about the situation in Congo, Gabon, Equatorial Guinea, Central Africa public, DRC or Nigeria where all practices of non-human primates hunting and consumption and butchering takes place? Do we have a situation where endemicity has already occurred, and this is sustained by human-to-human spread? We are very interested in looking at this scenario.

We started in fact looking at samples that are collected from different groups, different populations. This is unpublished data that we have recently generated by looking at recently collected Cameroonian blood donors from Yaounde, the capital of Cameroon. We have screened 180 samples and one found SIV-positive individual, giving a prevalence of 0.5 percent. The virus has a mandrill-type SIV, which is consistent with the common hunting of mandrills in Cameroon. More interestingly is that this blood donor was also HIV-1 infected, so we are now dealing with a co-infection situation with HIV-1.

The second population of samples which were already available to us at CDC was a collection of samples from sex workers from the Democratic Republic of Congo, from Kinshasa. Those were collected back in 1985 and we screened those, and one was positive, giving a prevalence of 0.72 percent. Again, the sex workers was also HIV-1 co-infected. Next.

Now, back to this part of the world, again, how widespread SIV is here? Is it only in population exposed to primates or it already has spread out? We do not know. We know cases have been identified in Canada and the U.S. We know in Europe,

at least Germany, we have cases identified. We don't know the rest of the other countries. And there's no reason why primate and zoo workers in those countries should not also show evidence of infection. So, again, we need expanded surveillance to answer the question of whether this virus has already spread outside populations exposed to primates. Next.

So I'd like to end up with a summary and conclusions. We've identified substantial SIV infection in U.S., Canadian and Cameroon persons exposed to non-human primates -- you'll probably hear more from the Canadian speaker later about the Canadian data -- demonstrated infection with multiple SIV clades, at least seven so far, in both men and women; demonstrated old and recently acquired SIV, at least we can date back 28 or three decades where those infections have been occurring.

So all together it implies that Simian retroviruses are actively crossing into human populations. We used to think that probably HIV-1 or HIV-2 the estimates were probably 70 to 60 or 50 to 70 has crossed into human population and caused the pandemic and that active transmission has stopped since then. Probably those data are a reminder that active transmission has not stopped; it is still

ongoing. However, we need to better define disease and spread of Simian Foamy among humans. We have little data on that. Next.

The issue of disease, very important. We think our data can suggest that it is largely still undefined, especially if you consider that you might have disease like in HTLV-1, low incidence, five percent less, after long incubation periods. So we cannot exclude at this point from the available data the incidence of disease. There's many issues that may surround the question of disease. Disease cannot be clade-dependent, like we have seen with HIV. Only two lineages cause disease in humans: The chimpanzee and the sutamangabees, and there's at least seven other lineages that so far appear to be non-transmissible and non-pathogenic to humans. So is this going to be the case for Foamy? We do not know. Definitely opportunities for human-to-human spread will lead to evolution of pathogenicity, and this is something we've documented that we've learned from other virus systems such as SIV.

The recent data on co-infection with HIV in the two cases we've identified are a little bit surprising to us because it begs the question on the impact of co-infection of HIV on disease incidence for

SIV as well as for HIV and, again, the issue of increased transmissibility, human transmissibility of SIV. The fact that one of those cases was a blood donor and one is the sex workers of course has implications for blood-borne transmissibility and sexual transmissibility.

But we're becoming increasingly -- because of the reasons I mentioned, we've become increasingly convinced that we probably need different study designs to identify disease cessation, not from surveying healthy people but probably identify endemic populations and then screen different sick populations just to see where we have some disease cessations. But we are following up the infected case to see if we can identify any incident disease. Next.

So the topic again and the question to the Committee that Dr. Tabor highlighted is what about emergence of other viruses, and what do these data tell us about that? In fact, SIV could be a good surrogate market of xenotransmission of other viruses, including Simian retroviruses, such as SIV or Simian-type D or STLV. And in a sense, it's the center for other possibly more pathogenic viruses. You can think of it this way. We have ongoing screening of our Cameroonian samples for SIV and STLV. Next.

But, in fact, we do have data that tell us that other retroviruses are also crossing, and this is what I alluded to before from our domestic surveillance where we identified two cases that are SIV-infected and two other ones that are seropositive for Simian retrovirus infection, and this is a joint study with Dr. Lerker who will be the other speaker where two cases were identified to be serologically positive. This is the Western Blot here, utilization antibody-positive but virus isolation-negative and PCR-negative. So no evidence of viremia in them but evidence of seropositivity.

This was observed over a two-year period in one person, suggesting probably an infection in this one. However, Case 1 here is the one that is also Simian Foamy-positive. So, again, it's a reminder that the question of other viruses or other Simian retroviruses that may be also crossing is not a hypothetical. Next.

So I will end up with some questions on the emergence of SIV and its implications for the blood supply. What do we consider are the criteria for a new virus to process for the blood supply? Of course, you can think of the infected donors to be asymptomatic, but the viruses causes persistent

viremia, so it can transmit. The virus is able to spread among humans and of course can cause -- has the potential to cause some disease. For SIV, I think the available data show that this is true, this is true, because you have at least PBL associated viremia. This is still unclear at this point, and this is still unclear at this point. But at least some criteria have been met so far.

And my last slide is a big thank you to the people that have been contributing to this work. Many names have already in or out because many have gone to other places, but a large number of people have contributed to it. A special thanks to Bill Switzer, who's the PI of the domestic surveillance, and Nicholas Lab for the Simian Type D serology, our collaborators from Johns Hopkins and Cameroon for the Cameroonian studies. Thank you.

ACTING CHAIRMAN ALLEN: Thank you very much. That was a very good overview. Comments or questions for Dr. Hemeine? Dr. Strong?

DR. STRONG: Within a given species, since you have substantial sequence data, do you see this virus being more or less mutagenic as compared to other viruses?

DR. HENEINE: The virus causes cytopathic

effect in-vitro, so it's not an oncogenic virus like HTLV, STLV and others, so it's a cytopathic in-vitro, it causes cell death. In-vivo, in the natural host, it seems to -- the host seems to control it well and we don't know of any disease that is associated with it. If I think I understood your question is do we see any evidence of adaptive events or mutations after cross-species? We're very interested in this, and actually we've been screening sequences we have already; this is ongoing work. We don't have any data at this point.

DR. KHAN: If I can just comment on that question. In my lab, we have looked at various naturally occurring viruses from Rhesus macaque and from pigtail macaques, and we have analyzed the sequences as well as studied the biological properties of the viruses in-vitro, and we have found that within any one group -- within any one species there is a diversity in terms of the sequences as well as in terms of the biological properties in-vitro, namely replication properties. So we have not found any two viruses that are identical.

ACTING CHAIRMAN ALLEN: Would you identify yourself to the reporter, please?

DR. KHAN: I'm sorry. Arifa Khan from

Sieber.

ACTING CHAIRMAN ALLEN: And do the immune responses cross-react with those differences? In other words, do the antibodies that are formed in the animals also recognize the same?

DR. KHAN: In general, the antibodies against the highly conserved proteins are cross-reactive and can pick up the various different viruses.

ACTING CHAIRMAN ALLEN: Dr. Lew?

DR. LEW: There was a question earlier about viral load. I noticed in your slides you did have a PCR, it looked like, for the Simian Foamy Virus. And so you had the group of husbands and wives. Did anyone try to look at viral load in those husbands and look at viral load over time? Do you have any sense of what the viral load is?

DR. HENEINE: We know it's a cell-associated infection predominantly, so most of the viruses in the peripheral blood lymphocytes, again like HTVL or other HIV. We only analyzed samples from four or five cases where we looked at cell-free viremia by RTPCR and we were -- all the samples were negative except one time from one case, the seropositive for both type D and Foamy.

Regarding your question on the pro-viral loads, we're beginning to look at that, and we have data from the chimp-infected SIV where we're trying to compare the pro-virus load in naturally infected chimpanzees to the human cases. So far they look similar. We don't have big differences. And they're detectable easily from the peripheral. So if you think of comparing to what we know from HTLV, HIV and Foamy, HTLV has the higher pro-viral loads, Foamy is next and then HIV, and the asymptomatic stage is lowered. So that's the trend we're seeing thus far.

ACTING CHAIRMAN ALLEN: Just picking up on that question, do you have any evidence in the humans that you know are infected, any variability over time in terms of viral load?

DR. HENEINE: The long-term follow-up study where we enrolled those seven cases is going to provide us that information. And we're collecting samples over six months to 12 months, so we'll be able to answer that. I think at this point we don't have any information.

ACTING CHAIRMAN ALLEN: I suspect you don't have any known infected humans that have died that gives you an opportunity for looking at other tissues for evidence of pathology or infection. I'm

thinking in particular if you've got peripheral blood lymphocytes, are there other -- you know, what's happening in lymph nodes, in the spleen and so on? What about some of the non-human primates that are infected, what does pathology show there?

DR. HENEINE: Probably I should have put one slide from the long-term follow up where we looked at distribution. It's not a lot of cases but we were able to amplify viral sequences from semen, from cell palates from saliva and from cell palates from urine. So it looks like -- of course in addition to peripheral blood lymphocytes. So it does look like there's a wide distribution in the biological fluids of virus-infected cells.

And in some instances, we were able to isolate virus from the throat swabs or cells from saliva. Virus titers, we don't have idea about it, but it does seem at least that the scenario is similar maybe to the natural host in terms of tissue distribution. But, again, very preliminary data on very few cases.

ACTING CHAIRMAN ALLEN: Dr. Lew again and then Dr. Klein.

DR. LEW: Yes. And you mentioned cytopathic effects. Which cell lines are affected and

what type of cytopathic effects are you seeing?

DR. HENEINE: The Foamy Virus are notorious to have a wide cytotropism, wide host cytotropism, and they're cytopathic to many different cell types. We routinely grow them in human cell lines, IG cells, whatever, canine cell lines, dark cell lines, I mean they grow very easily in different cell lines from different primates -- very disparate primate species.

DR. KLEIN: Do you have any indication of the percentage of circulating lymphocytes infected in the animals and any quantification of the nodes or other lymphoid tissues?

DR. HENEINE: No, we don't, but based on the pro-viral load data, the limited data that we have, we did some comparison with our experience with HTLV and asymptomatic HIV-1. It does seem it's in between. HTLV-1 is really at the higher end, Foamy is in between, and there was no differences between primates and humans.

DR. KLEIN: Is there any evidence on what happens to a newly infected animal in terms of spread of the virus?

DR. HENEINE: Well, maybe some data from the next speakers will tell us about the newly

infected animals.

ACTING CHAIRMAN ALLEN: Please identify yourself.

DR. SANDSTROM: Paul Sandstrom from the Public Health Agency of Canada. Dr. Brooks in his presentation that's going to be after Arifa's will present some data on viral load or at least quantitative data on viral load in comparison to cells as well as some indication of what goes on in the animals in the weeks after infection.

ACTING CHAIRMAN ALLEN: Dr. Hollinger?

DR. HOLLINGER: Just want to be clear about something. You find this only in the PBLs and not in the plasma at all; is that correct?

DR. HENEINE: Yes.

DR. HOLLINGER: Okay. The second this is you showed a slide which showed species specificity and the origin of some of these Simian viruses, the SFV. Do you find transmissibility then between gorillas and mandrills? I mean some of the studies have shown this to be transmitted apparently to humans, but I would think that you'd also see it transmitted to other non-human primates as well. Has that been shown?

DR. HENEINE: Yes. It's primarily species

-- the clades and their primate species have co-evolved over time and this is wide evidence. In fact, we could use Foamy Virus as the best model for host virus co-evolution in cross-speciation, the best model we have so far. However, there is also cross-species infections, and we think we've identified at least a couple of animals that have dual infections with their own clade and another clade. These came actually from captive animals that were in contact with other primate species. So primarily you see species-specific variance, but we also saw, though infrequently, cross-species infections among primates.

ACTING CHAIRMAN ALLEN: Dr. Cunningham, Ron Wilson and Dr. Epstein.

DR. CUNNINGHAM: I don't know if I missed this or not but when you have had a chance to look at samples over a period of time, is the genome stable or does it seem to have mutational alterations over some period of time?

DR. HENEINE: That's a good question. I think the genome of Foamy compared to HIV is more stable. We still don't have quantitative data when you compare it to, say, HTLV, which is very stable. But at this point we think it's more at the stable end rather than the high diversity end variable like HIV

is. But you could see evidence of quasi-species in the infected animal and in humans, and you could see some evolution over time, but it is slow compared to HIV.

MR. WILSON: Thank you, Walid. In the Boneva study, do you happen to know what the duration of storage of the blood components was before transfusion?

DR. HENEINE: I'm sorry, I didn't hear you, in what?

MR. WILSON: Yes. In the Boneva study?

DR. HENEINE: Yes.

MR. WILSON: Do we know how long the units were stored by the refrigerator at room temperature before transfusion? Because in the HTLV experience, we know that if units are stored more than two weeks, the rate of transmission falls off, presumably related to death of leukocytes. And I just wonder whether a similar phenomenon has gone on and whether we learned anything in that regard from the lookback study or, conversely, has it ever been examined in-vitro what is the storage stability at four degrees or room temperature of infected leukocytes?

DR. HENEINE: I don't recall the data if it's in the paper, but we can check the paper and get

back to the question of duration.

MR. WILSON: I think it's in Table 1.

DR. HENEINE: Stability in-vitro, we have not looked at it.

ACTING CHAIRMAN ALLEN: Dr. Strong.

DR. STRONG: SFV has also been considered to be an ideal gene vector for those that are doing molecular genetic engineering. Do you know to what extent it has penetrated that marketplace?

DR. HENEINE: Well, most of the vectors are actually non-replicating vectors, which is a good thing. There's also interest with some groups, including Dr. Folks in our branch, to use live vectors for gene delivery. It all depends on the incidence of disease and what these infections do. There is a lot of questions raised right now in the field as we're understanding that those infections are probably more prevalent than we previously thought than whether or not -- I guess it all depends on how the data come out. But the vectors have large number of advantages.

DR. STRONG: Thank you.

ACTING CHAIRMAN ALLEN: Other burning questions? If not, I think let's move on to our presentations and come back for the general discussion. Our next speaker is Dr. Kahn of Simian

Foamy Virus transmission studies.

DR. KHAN: Thank you. Next slide, please. As you have heard, SFV can be transmitted to humans by injuries involving infected non-human primates, most probably due to the saliva, as in the case of some animal handlers and zoo keepers. Additionally, recent data has shown that hunters in Africa can be infected due to exposure to blood tissues and meat consumption infected among human primates.

In all of these cases, the infection results in long-term persistence in the host, and in the next slide is indicated the reason why. Because the viral sequences in a retrovirus must integrate as a normal part of the host cell DNA as a critical part of the retrovirus life cycle. This results in lifelong infection of the host cell.

In the next slide, in general, in case of other retroviruses it's been demonstrated that retrovirus integration can in many cases result in the generation of pathogenic viruses or the virus insertion directly can result in mutagenesis by various mechanisms, such as activation of tumor genes or disruption of normal gene functions, such as tumor suppressor genes. Next slide, please.

Although to date there has not bee any

clear evidence that Simian Foamy Virus is associated with pathogenesis; however, this retrovirus is unique in its biological properties and therefore there are concerns regarding Simian Foamy Virus.

Number one, primarily it's the unrestricted host ranges of Simian Foamy Viruses. Regardless of the species of origin, most Simian Foamy Viruses have demonstrated a very broad host range. They can infect avian cells as well as a variety of mammalian cells, including human, as you have heard. They have a very broad tissue tropism as well as a very broad cell tropism, and the question came up earlier regarding how does the virus replicate or cause CPE in different cell types. My lab has done studies in a variety of different human cell lines, and we have found that the virus replicates highly efficiently in fibroblasts and then the replication varies depending upon the different cell types. In general, in epithelial cells, we found that the replication rate lags that of fibroblasts and in lymphoid cells the rate is also different. So the virus can infect all cell types of various species, however the replication of a particular virus is dependent upon the cell that it infects.

Additionally, there was a question earlier

regarding the different types -- or Foamy viruses of a certain species, and I wanted to just mention that, again, from one particular species in our experiences in macaques, we find that you can get viruses with different replication properties. They range from highly efficient to less efficient; however, in all cases we do get infection of the cells.

As I mentioned, we do see cytopathicity to various extents depending upon the cell type. Foamy Virus, in all cases you get infection that can result in latency, especially in humans, as you have heard. In one case, infectious virus was isolated 30 years post-infection from an infected human, and this was a CDC study. And it is this latency that is of concern in terms of its potential transmission in blood, because the virus can persist in a quiescent state and basically it can even go undetected, maybe asymptomatic. However, because it is a retrovirus it has the opportunity to generate into a pathogenic virus and then therefore result in serious consequences.

It is this concern of its persistence in human cells, especially PBMCs, that is the question that we are addressing today, whether there is a potential risk in terms of blood transfusion. The

next slide, please.

And this just states the question, and this question was also posed to the Committee in December 2001. The next slide. At that time, data was lacking in terms of blood transmission studies and this was the FDA study that I had proposed. Basically, we proposed that we would take whole blood from an SFV-infected Rhesus macaque and inject it into negative animals. The blood recipients will be monitored for SFV infection by a variety of parameters -- virological, serological, molecular as well as clinical analysis. And we will follow the inoculated animals or the transfused animals for at least one year to evaluate the infection by blood transfusion.

And in the next slide is outlined the blood transfusion study that we have now conducted in Rhesus macaque. We use a well characterized donor in which the Simian Foamy Virus has been isolated, the sequences determined in a limited extent, as well as the biological properties studied. And the recipient animals are retrovirus-negative. They were obtained from an FDA colony that's at Morgan Island, South Carolina, and the recipients were negative for other retroviruses, including SRV, SIV, STLV and of course for Simian Foamy Virus.

The animals were initially screened by antibody assays. At that point, while we were waiting for the results, they were housed individually. Prior to the actual initiation of the study, the negativity of the animals was confirmed by PCR and then further additionally confirmed by virus isolation, because we wanted to be absolutely sure that there was not any low-level infection in the animals that could eventually come up and confuse the results of our blood transfusion study. So we had confirmed negative animals for the study, and we additionally included a negative animal as a control in the study. And the study was done under approved animal protocol, of course, and the donor and the recipients were housed in different rooms and each was housed singly, of course.

And the next slide is the protocol that we followed. Blood was collected prior to transfusion to prepare controlled or pre-bleed samples or transfused samples for PBMC and plasma. And, additionally, the animals were tested by serum chemistry and hematology to evaluate their clinical status at the initiation of the study.

For blood transfer, 20 mls of whole blood was drawn in Heparin from a donor animal, and 10 mls

was injected into each of the recipients. And I should mention that we went with using Heparin as the anticoagulant because in SIV studies we know that blood collected in Heparin can transmit SIV into other naive animals -- monkeys -- and therefore we wanted to use a model that we know works for a retrovirus to initially evaluate the results. And the control monkey received 10 mls of just PBS.

In the next slide it's indicated how we monitored the animals for virus infection. We did antibody detection by initially Dot Blot and confirmation by Western Blot. Virus sequences were detected by PCR and nucleotide sequences determined for confirmation of identity. Virus isolation was done by using monkey PBMCs in cold-culture studies I will describe later, as well as the animals were monitored clinically by hematology and serum chemistry as well as by physical exam.

And I will not be able to present all of the data here, but I should indicate that initially during abut the first three months of this study the animals were very closely monitored initially on a weekly basis. All of these assays were conducted on samples, conducted weekly, and once we could see when the animal developed a positive result, then it was