

performed? And how should these studies be designed, and what challenge rate should be examined? And we're back to the theme of the challenge. So sort of rather like motherhood and apple pie. Should active and passive studies be done? Who would like to comment on that?

Yes, Sam? Thank you.

DR. KATZ: As one, again, with apologies, not working with plague, it seems to me the important question is in what context do you envision a vaccine being used? Are you going to give a vaccine to the whole population to protect them against an aerosol? You'll get a response like you got with smallpox vaccine, where only the Department of Defense members were immunized and a very tiny portion of the civilian population.

If I understood what I've seen, and Karen commented on this a little, the kinetics of immune response to these vaccines, the incubation period of the disease is much too rapid. So you're not going to give vaccine post exposure. And we can do this with rabies. We can do it with measles. We

can do it with smallpox because you have long incubation periods, and the vaccines work much more rapidly than the evolution of the pathogenesis of the natural infection. So this isn't going to be a post-exposure vaccine.

Fortunately, you have antibiotics, so you can treat recognized or known infections. So how are you going to use this vaccine from the point of view of active immunization other than selecting out populations you think might be exposed? And how you define them other than armed forces, military groups, I'm unable to say.

Passive immunization might be fine for the individual who's exposed and for some reason or other you're not going to give antibiotics. I was impressed with the data you showed on transfer of either antibodies developed in animals and mice or antibodies developed in other species. They work fine. But again, the pragmatism of how you would use passive immunization escapes me in the context of what you're worried about, which is a bioterrorism event.

DR. McINNES: Sam, thank you.

I think when we come to question 4, there's actually a somewhat pointed question. Is there any role for post exposure? And I think we want to bring this back up again in the context of how the vaccine might be implemented and what the indication might actually be.

In the context of studies to develop the correlates of immunity for plague vaccines in humans and looking at the animal studies and animal to human studies, what is the role of active and passive immunization studies, and should they be undertaken? I think we saw some very nice data on the passive protection studies.

I would like to pose whether there are any alternative thoughts on how these studies should be designed and, once again, to the challenge route, and if people have specific comments on active and passive immunization studies in the animals or between humans--vaccine stimulated antibodies and passive transfer to animals?

DR. SNOW: Hi. This is Doris Snow. I'm

from DVC.

And I actually have more of a general question. The panel's been focused really on pneumonic versus bubonic plague. But as a sponsor, we're going to have to have a very specific indication, and our Animal Rule studies are going to have to be designed to, you know, justify the use of our product for that indication.

And I want to get an idea from the panel, do you assume that a pneumonic indication would actually be effective to use in a population of people that are being exposed to a threat which may not be pneumonic plague? It may be aerosol intoxication or a bomb or an event of some sort. So does pneumonic plague indication equal aerosol exposure indication? Because I think that decision will really depend on which models we choose.

And then I think Dr. Pitt's discussion of the aerosol model is really the appropriate model. Because from our perspective, as a sponsor, if our indication is protection against an event, a biowarfare event, then the aerosol model is really

the appropriate choice. And that will lead to how we design and discuss challenge routes and model, you know, for those pivotal studies.

I think from an R&D perspective, you can have a lot of different models to screen potential candidates. And you have a lab set up, and you can screen and down-select candidates with a lot of different measures. But for really those pivotal studies, does pneumonic indication equal aerosol event?

DR. McINNES: Anyone on the panel wish to comment on that? Louise, do you want to comment?

DR. PITT: Well, I agree with Doris. I think if the indication is pneumonic plague, then aerosol is definitely the route for challenge.

In terms of the passive studies, I think passive transfer as correlates is extremely important. It's one of the ways where you can bridge species. You can get antibodies from active immunization in animals and humans, put them in a single animal model and start comparing activities, getting an idea of what the animal model versus the

human looks like. So I think they're extremely important.

In terms of the challenge route for those studies, I think that could be debated depending on the question you're asking of those passive transfer studies. And depending on that question, I could see potentially both a parenteral route and an aerosol route could give you different pieces of information and could both be important.

DR. McINNES: Yes?

DR. SMILEY: Steve Smiley, Trudeau Institute.

So in terms of this passive transfer, it's been well established in the mouse that that can protect. But in the primates, they seem to have high-titer antibodies, and yet they fail to be protected from pneumonic plague in some situations. So I think an interesting question is whether humoral immunity will suffice in the primates? And what I guess challenge people to do--I don't think it's been done--is can you passively protect primates?

If someone could show that, then we would know that antibodies would suffice in primates, and then we wouldn't have to necessarily develop assays for CMI in those primates. We'd be comfortable with assays for humoral immunity. I don't know if anybody from USAMRIID has tried that type of experiment. I know it's a difficult experiment.

DR. PITT: We would love to and hope to do that sometime in the future.

DR. FERRIERI: May I comment more on the passive immunity? I'm very enthusiastic about studies, doing passive transfer of antibody to understand a number of features here and what might be translated then to the human situation eventually.

And I can envision a situation where you might want to stockpile a plasma, for example, with antibody to the relevant virulence factors as we've defined them today because perhaps there would be situations where individuals receive different doses through a bioterrorist event. And they may have received, depending on your proximity to the

release of it, you might have had a smaller dose. It's conceivable that you might be a candidate, that children might be a candidate as well for such passive treatment with the equivalent of using intravenous immunoglobulin.

I was a big proponent of this on the IOM panel that I served on on anthrax, as were other members. And we had a very hard time convincing certain members of our wider community, public health community of the merits of doing passive experiments in animals, for example. And that has caught on now, and those studies are being done, as I understand it. But I think there is a role potentially for this. Not just for the sake of doing it, but because there may be a role eventually in application.

DR. McINNES: Thank you, Pat. Yes?

DR. LOCKMAN: Hank Lockman, Battelle Memorial Institute.

To build on that last comment, passive transfer may also serve to reduce the lethality post exposure, which I believe is based mostly on

the failure of antibiotic treatment. If antibiotics are not--if antibiotic treatment is not begun early enough, the disease is uniformly fatal. But passive transfer may rescue--may provide some rescue therapy that's not provided by antibiotics.

DR. McINNES: Thank you. Yes, sir?

MR. : So a comment and a question regarding the role of antibiotics. Just a comment. In terms of what to prepare for, I was thinking that we should assume that the aerosol strain would be resistant to all of the antibiotics that plague is susceptible to. Because the engineering of antibiotic resistance is so easy to do in a laboratory to all of the antibiotics we use.

But the question has to do with what study should we do for immune correlates? I've heard a lot about the acute response to vaccines. I've not heard anything about the long-term response and duration of response.

Now with other antibody-driven vaccines that are successful like hepatitis B, we know that

there's a certain titer that's protected. But you can fall below that, no problem, and you're still protected because you're going to boost. We don't know if that's the case for plague. We have no idea whether an anamnestic response plays any role whatsoever in protection from plague, and I just wonder if the panel can help us with that?

DR. McINNES: Yes, Bob?

DR. PERRY: Let me comment on aerosol delivery and multiple antibiotic resistant strains. You know, at a threat assessment meeting we were at several, maybe a year ago now, Luther? I mean, surprisingly, you know, everybody sort of decided that engineered strains were not going to be extremely likely. It was going to be using a natural strain in most cases.

Because while it's true that you can easily genetically engineer the bug if you've worked with for years and stuff, we're not talking about state-sponsored programs, and you're more likely to get somebody going someplace and picking up a dead prairie dog and isolating the organism

and distributing it. So we ended up thinking that highly engineered strains were a little less likely than I think all of us assumed when we first started talking about it.

And maybe the emphasis here is a little too much on an aerosol delivery. That was really sort of thought of as maybe the number-one ideal delivery route. But again, you might use more primitive methods. And so, there might be other methods that aren't that far down the list from aerosol as a delivery method.

Now at least I think that, you know, if you have something that's going to protect you from an aerosol delivery, it's going to protect you from any other route of delivery as well. So it's not maybe a huge mistake to focus a lot on aerosol. But I think we also need to do some testing along the way with other routes of delivery to make sure the vaccines that are developed work.

So like I say, I'm pretty sure that anything that protects against an aerosol is going to protect against any other form of

administration.

DR. MIZEL: Steve Mizel, Wake Forest.

One of the things we haven't talked about in this in terms of correlates is, are you correlating it with mortality? But what about correlating things with morbidity? So that you may be able to get protection where someone gets sick, but they have reduced morbidity. And so, we may throw the baby out with the wash if you ignore things that deal with morbidity as opposed to mortality.

So I'd be interested in the comments of the panel on that.

DR. MCINNES: Thank you. Karen?

DR. ELKINS: Karen Elkins, CBER.

Bob, I want you to go back to that statement you just made about things that protect against pneumonic exposure would protect against other routes of exposure. Could the plagueologists, which are assembled here, comment a bit more about the data that speaks to that point and how strong it is?

DR. PERRY: Well, I think it's basically from the aspect that the pneumonic route is much more rapid, much more highly fatal, and there may be differences, especially with the lung pathology, in the two diseases as to what we see. But, you know, if you have a nonpneumonic route, you know, a lot of people survive on their own. The disease takes longer to develop.

And so, I think if you have something that's going to protect against this rapidly developing, fatal, you know, rapid bacteremia that progresses, it's likely to protect against the other types of disease as well. Not on any firm data on histopathology or things like that, just, you know, the level of lethality, fatality, and the time to death and incubation periods.

DR. LYONS: Yes, I think most of the data on that are sort of anecdotal experience with particularly two veterinarians that got pneumonic who were vaccinated with one that particularly protected them--would have theoretically protected them against bubonic, from historical data. But

they died from pulmonic. So it's pretty anecdotal.

DR. LU: For the passive immunization, I think there may be potential it has utility there as well pointed by several members. But I think to use passive immunization or antibody through IV, or whatever, as an immune or vaccine surrogate marker, I think we have to be careful.

This is basic immunology knowledge. Just think about that. When you have a pathogen come in, you have a passive antibody. A passive antibody cannot go higher. It just keep going down. But you have a memory response from active immunization, you will expand it quickly. It continues fighting, stimulate that. So I think that part is very important.

So if we use passive immunization, we can only use a secondary standard. Again, I'm only thinking about vaccine licensure or move from Phase 1 to Phase 3 or from animal to human study. So I think that we have to be cautious on that.

DR. McINNES: Yes, Pat?

DR. FERRIERI: Well, I'd like to respond

to that. I didn't mean to imply that the priority should be on passive. I view that as a secondary priority, and in no way do I view it as conferring long-lasting protection in any way. I view it as an emergency.

DR. LU: Oh, no, no. That part I agree 100 percent. I don't think there's a difference. But I think in the context of this discussion here, there are two definitions. One is as a surrogate and one as immune correlates, establishing the immune protection as a vaccine. That part I say we can use that. Actually, I think it's a great idea to use that. But it's not the same value as the active immunization.

DR. McINNES: And you're also making a plea for understanding kinetics of durability of antibody response and anamnestic response with waning antibody and exposure to antigen sometime remote from vaccination.

DR. LYONS: Pam? Just on active, you know, I notice something that's, to me, missing badly here. But--and I don't want to complicate

things. But it seems like we're basing all the information on one adjuvant that we probably know is not the best adjuvant in the world anymore. And you wouldn't want to miss the opportunity to incorporate studies with new adjuvants that-- particularly for as we get into post exposure.

I mean, there are adjuvants that probably may enhance the immune response well enough to maybe benefit there. Right now we seem to be focused on alum. So or at least all the data I've seen has been using aluminum hydroxide of some base. And I know it complicates things, but you'd hate to miss the opportunity to take a look at that along the way. It just seems like such a ripe opportunity.

DR. McINNES: No. I agree. Certainly, I think in any R&D venture it is a ripe opportunity to look at alternative adjuvants. I think when one is--the counterpoint to that is trying to drive hard to a product--

DR. LYONS: Oh, absolutely.

DR. McINNES: --that can't be licensed in

interim, and you couple that with a novel adjuvant approach.

DR. LYONS: Sure.

DR. McINNES: But absolutely, we should use this opportunity to drive it.

Yes, Sam?

DR. KATZ: I think one aspect that relates to the question that the gentleman at the microphone asked previously is I don't think we have any vaccines that protect against infection. They protect against illness. And the shading between morbidity and mortality I think is in individual human response to a pathogen. But a successful vaccine will present--protect against morbidity as well as mortality.

However, what I haven't heard discussed, and maybe I missed it because I came a little late, is kinetics and duration. A, if antibody is the answer, how quickly can you detect effective antibody? And, B, how long does it persist? I agree with Dr. Frothingham that we've shown with some antigens, hepatitis B being the most cogent,

that you may have undetectable levels, but challenged, you will have an anamnestic response.

Are you going to need boosters of this vaccine? Your primary immunization series, whatever it is, one, two, or three doses, 5 years later, 10 years later or what? Are you going to have to provide boosters if you really believe you're going to provide protection? And those sort of studies in animals could go on for years before you ever had a licensed vaccine.

DR. MCINNES: Thank you, Sam. Yes?

DR. MIZEL: Steve Mizel, Wake Forest.

For those of us who stayed up last night and watched the debate, one of the pieces of data that we heard, in 2008, that the baby boomers will start to retire. And nowhere in our discussions yesterday or today have we talked about aging and the immune response in the aged, which is quite different. We know that very dramatically from the flu vaccines.

So one of the issues in these models and in these correlates is, at some point, we're going

to be looking at a population that, by and large, is not young monkeys or mice. It's old people. And we haven't discussed that. So somewhere in here we have to get to that issue of aged models and because that's a sizable part of the American population we should be thinking about protecting.

So I think that somewhere in our discussions this morning, that ought to come into play.

DR. McINNES: Thank you.

I think one would hope that if you go through the process of having a vaccine, going through your animal studies, developing an assay that's characterized, that's correlated with the functional assay, that some of that readout, in fact, would come from human clinical trials and looking at comparative immunogenicity. I think it's an interesting question on whether you need to have the equivalent counterpart in the animal world. I mean, traditionally, we have done that in human populations.

I'm going to move on because we're going

to loop back, with this particular question, to some of the issues we have already touched on, which is how can a correlate of protection in animals be translated to a correlate of protection in humans? And once again, the question about what functional assays need to be established and validated.

And I think if--I'm not sure if we've beaten that one as far as we're going to go on the functional assay. Drusilla, did you want some more on that one? Are you all right, Karen? So we could focus on the first part of this question, which is how can the correlate of protection in animals be translated to a correlate of protection in humans?

And I'm not picking on you, but I wondered whether someone from DSTL would like to comment about how they see this path of moving forward and the bridging that they would propose to be able to show? Sorry to do that to you, but did somebody want to--Di, thanks.

DR. WILLIAMSON: Well, I think the thing

is that one needs to be very certain of the assays of immune response in the animal models and then translate those assays to man, to the equivalent sera, peripheral blood mononuclear cells in man, and determine whether we're seeing the same kind of readouts. Simply that.

Also I think passive transfer of antibody from human to animal models is going to be vital. That's it. I mean, Karen's presentation this morning really summed up very nicely how to bridge from the animal models to human, I thought.

DR. MCINNES: I agree. I thought it was very, very helpful. Are there any comments from the panel about this in terms of the strategy for bridging from positions of certainty? All right.

So any comments from anybody else from the floor, have any issues they want to share? All right. Yes? Yes, sir.

MR. HEATH: I just wanted to point out in the first--I'm Dave Heath from USAMRIID.

I just wanted to point out in the first presentation, there was a gentleman who gave a

presentation on the vaccines, the older vaccines. Jerry Andrews at USAMRIID looked at the Greer vaccine and found that it had plenty of F1, but it didn't have V. And hence, the protection against bubonic, but not pneumonic. So that's just an old historical perspective I wanted to throw in.

The other thing is about F1, on the actual F1 and V together, when you have F1 by itself, it does delay the time to death. So why is that important? From a clinical microbiologist's perspective, if you have, say, a person who was exposed and they're in the hospital. And you're giving that clinical microbiologist or the physicians a couple of days extra to discover the organism, to isolate it, to characterize it, that's really important.

And it really becomes more important when you see the variability in the V antigen. So F1 even becomes more important there. So I would posit that F1 is a very important aspect of the vaccine. So that's just all I wanted to comment on. Thank you.

DR. McINNES: Thank you very much.

DR. FERRIERI: May I ask a question about-

DR. McINNES: Yes, Pat.

DR. FERRIERI: Dr. Lyons, do you understand why people with bacteremia following a bubo, who become quite ill but die infrequently, versus the rapid pulmonary death in someone with pneumonic, and my question is what's happening within the lung, within the macrophages?

Is this the key that unlocks the difference for the fulminant downhill course from a pulmonary point of view? The cytotoxicity that everything's up-regulated within the macrophages, and they're not turned on in peripheral blood, or is there some other very simple explanation?

DR. LYONS: I doubt if there's a simple explanation. I don't know. That's why I think--I mean, that hopefully will fall out over the next few years as we study it more.

I think it happens with--I mean, we see it with a lot of--and Jon can jump in here, too. But

staph infections are the same way. I mean, when staph goes bacteremic from the lung, it's a horrible situation. And so, I think there's some damage to the lung that probably interferes with oxygenation, which we know is a big problem. And mechanical ventilation does not overcome that.

So not only now are you faced with classic sepsis, which, at least in our models, the sepsis developed by plague is different than the sepsis developed by classic Klebsiella, things like that. It's not as--once it starts, it's a real bad situation. But it doesn't kick in until--at least what we've seen, it doesn't kick in until the numbers are extraordinarily high, extraordinarily.

And so, I think what's happened is you get some growth in the lung going, causing a lot of damage. And then, so now you have lung damage on top of sepsis, which is a real bad situation. But that's not the whole story, I'm sure. Jon?

DR. MCINNES: Yes. Thank you.

DR. GOGUEN: I can offer--I don't know if it's correct, but I can offer a simple explanation.

And that is that in the bubonic case, there's a longer time for the host to develop a specific immune response. And so, they have this--you're looking at a race between the development of specific immunity and the increase in the bacterial population to the point where the host is not able to recover. In the pneumonic case, this happens much more quickly.

So it's simply that the host in the bubonic case has more time to develop an adequate response and has a chance of recovery. In an untreated bubonic plague, I think the recovery rate is something like 50 percent.

DR. LYONS: Right.

DR. GOGUEN: So you're right at the limit there. Pneumonic, it goes a little faster, and so there's--you just push it to the point where there's essentially no chance to develop an adequate specific immune response.

DR. McINNES: Sue?

DR. STRALEY: I'd just like to comment that we focus a great deal on the macrophage, and

it certainly is important because it's the cell on the spot. But I don't--I think we ought to not overlook PMNs because they're very numerous, and they make all of this--you know, a lot of cytokines that are important.

DR. McINNES: Thank you.

Just some very practical issues now regarding the strain that should be used in animal challenge studies. How should those strains be produced, characterized, and monitored for stability and virulence over time?

I think one of the rate-limiting steps that we really have identified in the rPA, and even in the MVA vaccine development efforts, has been challenge capacity, challenge dose, source of the challenge, characterization of the challenge, potency and stability testing on the challenge material, facilities in which to conduct the challenge. So I think we sort of added the second part to the question because I think it may not be terribly interesting, but it's terribly important in our being able to move forward.

And in terms of what I believe the two strains that I saw reported yesterday, the Colorado 92 and then the C12, which is F minus. Correct? Are there some--from people who have used those strains, are there some distinct pros and cons and specific utilities? Are there some things that should we be looking at alternatives? And then what are some comments from people who have been trying to go through challenge experiments in terms of accessed and what--should we be producing a standardized reference pool, for example, that has actually a stability and potency program established with it?

I'd like to hear some thoughts about that. So go ahead.

DR. PITT: Can I start off? The challenge strain with *Yersinia pestis*, I am fully aware of the problems that have been occurring with the anthrax rPA program. That one's extremely simple compared to *Yersinia pestis* because it's a vegetative bacteria. So every time you do a challenge you have to grow it up. You can't just

pull it out of the fridge, as you can a spore, and do your experiment.

So you have many, many more steps in where there is going to be variability. There will be much more variability. You can standardize your procedures. You can standardize your media. You can standardize everything, but you're going to have to change your lots of media at some point. You're going to have to change. So that's just something to keep in mind in terms of--

DR. McINNES: So you would, in terms of having I hate to say even an SOP process, but in terms of having some buy-in about how to produce each time, the media issues, you would see value to that to the community as a whole?

DR. PITT: I see great value in there being a standardized SOP because we have been through some very painful experiences where we've thought we had a certain concentration and we don't. Some of them are not as viable as you would want because they don't stand up to the aerosol procedure based on how long they've been incubated.

So standardizing the procedure is very, very important and sticking to that. The temperature the organisms are held at once they are prepared is extremely important. So, yes, a standardized procedure is invaluable. And a standardized stock that you then take out and just grow up for your challenge is incredibly important.

DR. McINNES: Correct. Any other comments?

Yes, Dr. Perry?

DR. PERRY: Louise is right. I mean, you have to standardize things. And it is going to be difficult from the standpoint of, you know, some of us have been trying to grow the organisms as we thought they might be from a natural infection or from a natural aerosol. And if you're worried about an artificial bioterrorism event, are these people going to grow them at 26 or 37, you're going to get very different profiles of what is made, the metabolism of the organism, and that. So that's going to be very different. There's really I don't think any way to predict that.

Another problem is if you want to store your strain, *Yersinia pestis* grows at refrigeration temperatures very slowly. So, once again, if you've grown the cells at 37, and now you stick them in the refrigerator over night, when you take them out the next morning, you know, they're not going to be 37-degree grown cells, they're going to have replicated a little bit or at least have adapted now to the cold. So you've got that problem.

With the issue of specific strains, if you look in the literature, you can see primarily three different strains have been used. The C12 strain is really a derivative. So I would say, you know, it's an isogenic strain of Colorado 92. So there's no big issue here. So Colorado 92 has been used extensively at USAMRIID and other places. We and others have used KIM.

So there's three biotypes of plague, and Colorado 92 is the *orientalis*, and KIM is the *medievalis*, and I don't know that an *antiqua* biotype has been used much in virulence testing or

animal tested at all. But if you go back to the old literature, there doesn't seem to be much difference in the level of virulence among the three biotypes.

I think DSTL had been using a strain called GB for a while. I'm not sure what biotype that is. So that's the third strain that's been used in some virulence testing, and I'm not sure there's a big difference in which strain you pick. I'm not sure there is a real issue of, well, we need to test more than one strain. I'm not sure you'll see a big difference.

The first two strains that were sequenced were Colorado 92 and the KIM strain. And you do see differences. The KIM strain has been in the laboratory longer, and so it's not clear whether there are some differences that have accumulated from growth in the laboratory. However, if you look at the degree of virulence, there's really not any significant difference between KIM and Colorado 92.

So I think we can pick one strain that can

be used. I'm not sure that we need to worry about different strains that we're looking at. But really, so the issue is how you're going to grow the strains, how you do this.

For most of our studies, when we have wanted to mimic bubonic plague, we have deliberately grown the bacteria at 26 degrees and have done it in the presence of either iron or hemin, since the flea is going to be probably a relatively iron-rich environment. So that's been our standpoint. But that's not really relevant for what we're considering today.

DR. McINNES: Thank you, Bob. Pat?

DR. FERRIERI: Well, I don't work with this bug, but I would make a case for great standardization and that you know the lineage of it and that everything in one lab. You're able to correlate with what is done in another lab.

And an anecdote. Years ago, one of my fellows wanted to work with HiB, haemophilus influenzae type B. So I called Arnie Smith, who gave me the strain that was used by Haddy Alexander

years ago. And so, we felt that we could talk turkey with everyone else in the field of HiB because that strain was not passage once a week, and you knew exactly where it came from and how it had been treated all these years.

So I mean, do you have something that's been lyophilized and it's been shared? It gets very complicated. And I don't understand. It's great that the genome has been established, I guess, for Y. pestis. But do we understand its stability and the virulence factors, their stability? And what should we be doing so that--I mean, you're working with this bug. Well, we have a recombinant fusion protein vaccine, but we still need to have great standardization of the bug to do these critical in vivo animal studies and in vitro assays.

DR. MCINNES: Anyone wish to comment on that?

MR. : I think standardization would be a critical thing to do, and probably C092 would be the best strain at this point to choose as

the standard, I would think. It's the more recent isolate of the sequent strains.

I would also comment that I think at the end of the day, not necessarily during the testing, but at the end of the day before--I think we want to know if the vaccine is broadly able to protect against a variety of plague that's out there. And at some point, I think we'd like some recent isolates from different parts of the world to run against the final product.

DR. McINNES: Very good point. Bob?

DR. PERRY: I can't remember whether it was Pat Worsham or somebody else brought up the EV76 strain, the vaccine strain. And you know, everyone who works with plague has EV76 stored in their freezer, and I'll bet no two of them look exactly alike.

And I think we're even starting to see that with Colorado 92 because as it's been disseminated now, we're getting, you know, differences coming up in the laboratories where they've been grown. So, you know, the

standardization probably is sort of important.

DR. McINNES: Yes? One last comment.

DR. FERRIERI: There's a flea in the room that I hope didn't come from some animal experimental station. It was here yesterday, and it's circulating around the table today.

DR. McINNES: Go ahead. Yes?

MR. SCHRIEFER: Marty Schriefer at CDC in Fort Collins.

We are currently making available a panel of *Y. pestis* strains, eight of them, which include all three biotypes. And Colorado 92 will be made available through a subcontractor of ATTC.

I agree that standardization of protocol for growth and maintenance of any of these strains is critical to standardization of vaccine or other animal protocols and would be happy to participate in that. But would just like to let everyone know that within a few months, these strains that I just referred to should be available through this subcontractor, which is, I believe, BI, subcontractor of ATTC.

DR. McINNES: Thank you to CDC colleagues for that information and setting that up.

We're back to the challenge dose, for completeness sake, on E. And I think we heard--we heard a strong preference for aerosol challenge from some colleagues. I think we heard a little bit more pragmatic approach that alternative delivery routes might be valid. And I don't know whether we want to pin anybody down any further on that.

I think that the challenge by aerosol route has been the most compelling to this point, and I wondered whether we wanted to have just again some pragmatic thoughts about intranasal and intratracheal delivery. And Rick, do you want to just give us a summary again on how you feel about this and where it may play in some of these--which will become very important in the pivotal study.

DR. LYONS: Yes, I guess I believe in the mouse, particularly. I mean, the mouse just simply because the aerosol route is not efficient, and getting high doses is going to be difficult. I

mean, it's going up very high, and that was brought up today by someone else, and trying to do that.

I mean, that may be difficult even in-- from practicality matters, even in the primate. But I couldn't say that with authority. But getting multiplicity of infections, moving logarithmically or however you want to do it, you know, that would be more straightforward to do by either intranasal or intratracheal routes.

And again, I just haven't seen any data to suggest that the behavior is different, no matter how it gets to the lung--yet. You know?

DR. McINNES: Thank you.

I wanted to go in terms of to the actual dose that we had some discussion about that, I believe yesterday it started, about what would actually constitute challenge dose. And I also want to pick up on here this issue of the readout of the feet-up and--the feet-up readout that we're currently dealing with, which is the mortality endpoint and the sort of theme that has been percolating around about some intermediate endpoint

that might measure disease conditions, some biomarker readout, some histopathological readout, some count readout.

And to toss out to people in the discussion with what should the challenge dose be, is something like lung infection for pneumonic plague a feasible endpoint readout? And I toss that out to--is it even feasible?

DR. PITT: Can I just comment on the nonhuman primates? I have never seen a nonhuman primate that gets pneumonic plague survive. If they get pneumonia, they die.

DR. McINNES: I think the point is can you, at an earlier point, instead of waiting for that, is there some readout at an earlier stage that you could think about, that you could--I'm looking at this puzzled look, and I'm thinking I'm not communicating properly.

DR. LYONS: I don't think I understood the question. I guess I agree with Louise. I don't think we have a correlate that we could rely on right now to say we should do this, if I'm

understanding your question. So like a morbid timepoint as opposed to--

DR. McINNES: Correct. A morbid timepoint as opposed to--

DR. LYONS: I mean, I think in mice anyway, if they get sick, they're going to die. I mean, that's pretty much what we see. But I think when you start looking at vaccines, I think Dr. Katz's point is well taken. I mean, we've seen some vaccine studies where they get very sick and they get better with time.

So you're really looking at a spectrum of illnesses when you look at vaccinated population versus in a pure naive population. So I think you want to be a little careful about calling your endpoint too short. That's all.

DR. PITT: Based on our IACUC animal requirements we already do, we do not allow our animals to go to death. So we are collecting some of those pieces of information as we do these studies. And I will say as soon as the animals have fluid on their lungs and they are audible

through your respiratory protection equipment that you are wearing, those animals are immediately euthanized because there is no way back from there.

DR. McINNES: Yes, Brad?

DR. LEISSA: Brad Leissa, Center for Drugs, FDA.

Since we're looking at correlates of protection through the Animal Rule, somewhat related to this. But interested if the panel, anyone here in the audience have thoughts about for the purposes of showing efficacy in correlates of protection, we're certainly--in the human trials that will be done, there will be women and men in that.

In the animal studies, do people have opinions on whether or not the nonhuman primates that will be tested, whether it should include both males and females or whether it really matters?

DR. PITT: We always use both. We mix them 50/50 whenever possible.

DR. McINNES: Thank you. Yes, Mark?

DR. ABDY: Hi. Mark Abdy with CBER.

I want to get back to the challenge dose. I think I heard a discussion that sort of the group seems to be happy with going with Colorado 92. We had a discussion earlier this morning where we talked about plague, and we sort of settled on a 200 LD 50, and the question came up "why?" I think--

DR. PITT: About anthrax.

DR. ABDY: I'm sorry, anthrax. Sorry, anthrax. We had a 200 LD 50 target. And I say a target.

I want to preface this by coming back to the Animal Rule. We need to remember as we work on these studies that we don't want to set the bar low. We have to set the bar high because we will never have the ability to test this in human beings. So we need to be pretty conservative in how we do that.

Knowing that, is there a recommendation or any discussion on what our target challenge dose should be for Colorado 92 in a mouse model and in a nonhuman primate model? Because we obviously have

to talk to sponsors, and we have to recommend a dose or a target to go with, and I'm not a plague expert. But I want--I'd like to be able to recommend something.

DR. PITT: I can only tell you how we chose the range of challenge that we use. It was based on the probability curve, the lethality curve. We wanted to get above an LD 99 because we wanted all our controls to die. And so, an LD 99 is around 50 to 100 LD 50s. So 100 LD 50 is usually the target that we use.

Having said that, we usually get anything between 50 and 200, 250.

DR. McINNES: Yes?

DR. ABDY: What about the mouse?

DR. McINNES: The question is, what about the mouse?

DR. PITT: The mouse, we have taken that you can use multiple groups of mice so you can do multiple challenge levels. So we have not really done that academic exercise as to what is the most appropriate challenge dose for a mouse. You can

give 100, 1,000--10, 100, 1,000 LD 50s.

DR. McINNES: Your comment, yes?

MR. : At least in the case of plague, an LD 99 is going to be a lot less than 100 LD 50s by the intranasal route. I can assure you of that.

DR. PITT: Pardon? Could you repeat that?

MR. : I was just saying that in plague, an LD 99 is going to be a lot less than 100 LD 50s, at least by the intranasal route. That's been our experience.

I asked about this problem several times. That is what's the aerosol dose that one would expect in a terrorist exposure or whatever? And I never seem to be able to get a good answer. Somebody told me yesterday that the battlefield dose that they plan for--and maybe that fellow is still here. I don't know who it was--is 150 LD 50s, which turns out to be about the same dose that the DSTL people have been using, and that's about 100,000 organisms.

And it seems a reasonable level to shoot

for, and that's--

DR. PITT: You're talking about a mouse now?

MR. : In the mouse model, yes. And that's about the same level that we've used in our--in some of our challenge studies. So that would seem a reasonable target. But it would be nice to know what is expected, and I have no idea what that number is.

DR. PITT: I think that's an extremely difficult question because it depends on the scenario. I mean, you can sit down in a room and come up with multiple scenarios, all of which will have different exposure levels.

So the question is, do you want to go for the worst-case immediately, or do you want to pick a reasonable dose that's somewhere in the middle to establish your parameters, understand your vaccine and your models, and then go in and see how high can we protect against?

MR. : But we also know at this point that we can get protection with some of the

current vaccines at that sort of dose, 100,000 organisms or about 150 LD 50s.

DR. PITT: In a mouse.

MR. : In a mouse. So that's not unreasonable. I had something else, but I'll stop.

DR. McINNES: So just to our colleague who has made an appeal several times for a morbidity readout, I think I have a somewhat equivocal interest in that expressed from people in terms of looking, may have a place in terms of a challenge post vaccination, where you may be looking at amelioration of disease and some sort of morbidity readout. And I think that sort of summarizes how-- correctly how people felt about that.

DR. FERRIERI: May I ask a question, Dr. McInnes?

DR. McINNES: Yes.

DR. FERRIERI: This is in response to Dr. McInnes's earlier point about earlier stages of morbidity. Can you attach, or maybe you do, a little oximetry device to a digit of the nonhuman primate so you know when they start to become

deoxygenated?

DR. PITT: By the time they're deoxygenated, they are really sick, and it's very obvious.

DR. FERRIERI: So you don't need that?

DR. PITT: No. The earliest--and that's why we use telemetry continuously in the nonhuman primates because that is the earliest notification that the animal is becoming sick. And the fever goes up before the animals show clinical signs. So--and I can tell you in our limited experience, if the animal's had a fever, the animal has died or has gone close to death.

DR. McINNES: Thank you.

Moving on, question 3 was regarding--I'm sorry. I'm going to have to continue. Oh, all right.

MR. : It's just an add-on comment for the small animal model for plague. Our experience with the mouse is that the only reliable parameter for morbidity is recumbence. When the animals become recumbent, they're going to die.

And we're talking about vaccinated animals. Every other parameter is not a useful indicator of outcome.

Now I'll also say that we did one study looking at hypothermia in the mouse, and that seemed to correlate with recumbence. But it's very difficult to do a large, you know, mouse study looking at temperature.

DR. McINNES: Very important. Thank you.

DR. PERRY: Can I make one comment? So in the subcutaneous mouse infections that we've done, if we look at--you know, we haven't done it real stringently. But we do see mouse that become moribund and nonresponsive, and a small fraction of those actually do recover, at least from a subcu infection.

Yes, you've seen the same thing? Yes, with a subcu. Now that's obviously different than aerosol. But you know, in looking at this, I think we want to be a little careful about assuming that everything's the same between aerosol and bubonic model.

DR. McINNES: Thank you.

Question 3. There are three more questions left, and I think they're sort of big picture, and we can deal with them in the time. We don't have a lot of time available.

The issues around the fact that there will be human safety and immunogenicity studies of candidate vaccines, and induced responses will be compared with those from the challenge experiments in animals to anticipate efficacy in humans. The question on the table is, in addition, should clinical field trials be considered to evaluate the efficacy against natural infection?

Sam, I'm going to, if you wouldn't mind, from your experience of thinking about the whole plethora of efficacy trials, the challenge of trying to do studies in endemic areas, the fact that being able to follow up on subjects and to do case ascertainment and to have medically appended illness in some of these endemic settings is really a challenge.

And seeing what you saw yesterday in terms

of the therapeutic intervention study and perhaps the site in Uganda and thinking about where you might have endemic disease, what are your thoughts on clinical field trials for vaccine efficacy in those settings and in those disease conditions? Yes? If you would be so kind?

DR. KATZ: I think that you have one advantage. That is that in the natural setting, you can treat with antibiotics if your vaccine fails. Whether you can organize a study of that sort in Uganda or Madagascar, where the disease is still occurring, I would ask Jacob Kool to answer that. He's been to these sites. I've been to Kampala, but I've not been out in the field in Uganda.

But it would seem if you were going to organize a study, it would have to be extraordinarily carefully monitored to see that if there were potential vaccine failures, you were onsite to treat promptly. You know, life has become much more complicated. I think some of us forget that in July of 1796, Jenner gave James

Phipps a little virila--or a little cowpox, excuse me, from Sarah Nelms. And seven weeks later, he challenged him with smallpox, and he resisted challenge. And that was an N of 1, and that proved that it worked.

We're not in that era anymore, and the other issue, of course, that comes up is doing studies abroad, international studies in populations who are resource poor and who may feel that they're sometimes used as guinea pigs. And I think you have to be very careful in that respect.

I go back to our own experience, and it's not--it's apples and oranges. But we were pleaded with by people in sub-Saharan Africa to come and do measles vaccine studies because they had a 10 percent mortality from measles. But we did not do them until we had done them in the United States and the vaccine had been licensed. And then we felt it was no longer a fact of using these children as guinea pigs but showing that you could or you couldn't protect in a population that was at high risk.

But I'd turn this one over to Jacob.

DR. McINNES: Jacob, yes.

DR. KOOL: Yes, thank you. I think you said it all. The best way to do it is to go back to the old days, and you guys should just inject yourselves.

[Laughter.]

DR. KOOL: If you're not willing to do that, I think it would be doable, logistically, to do a clinical trial of vaccines. I have to admit I have no experience with vaccine trials. But I would think that it would be easier than a treatment trial.

For the treatment trial, you have to capture the patient on the first day before he's gotten any antibiotic yet. For vaccines, you'll be vaccinating the population, if I understand correctly. And then you'll be waiting for cases to appear. So you don't have to be there prospectively. All you need is good communications so that you can get samples and confirm cases, if necessary.

What I think might be a problem is what-- you mentioned that, too. You don't want to use these people as guinea pigs. And the only ethical way that you can do a vaccine trial is to be able to promise them that this will be in the advantage of the study population itself. So you have to be able to offer the vaccine afterwards to the country for an affordable price.

DR. PERRY: I guess there's one other question beyond the ethical issues, and that's the statistical issues. Since an N of 1 is no longer valid, how long is it going to take a study like this to accumulate enough data to be statistically reliable?

DR. KOOL: I can't tell you that. We are optimistic that we will get several hundred cases in our two-year study. Several hundred is all I can say. I will consider myself lucky if we get 400 cases in two countries in two years. But if we're not lucky--you know how it is with these plague seasons. Sometimes they have hardly any cases in a whole year. And other times, they have

huge numbers.

DR. McINNES: Right. Thank you, Jacob.

DR. LU: I think maybe FDA colleagues can provide more information on that. From my knowledge, including my present experience in the HIV field, even HIV is a very high incidence of disease.

Now the Phase 3 trial at a minimum is talking about 5,000 to 7,000 people, or several reasons the Phase 3 trial like from Merck, HPV, the requirement or the efficacy need 7,000, even go over 10,000 so for efficacy. So the vaccine trial requirement has become more and more complicated.

And also I agree with our colleague's comment that the ethical issue is not just providing vaccine. Later, when it becomes commercially available, actually now become ethical issue. You see, you have to go in with the public health education to reduce the incidence so they will not get infected.

So you cannot say I hope we have high incidence rate so we can see the efficacy of the

vaccine. You have to say public health education, including antibiotic treatment. So how do you pick a group treated, not treated becomes very complicated.

DR. McINNES: Thank you. Brad?

DR. KATZ: I think that one of the features that he's brought up that was tangential is at least Uganda, and I can't speak for Madagascar, does have a significant incidence of HIV infection. And whether efficacy of vaccine in an HIV-infected population can be extrapolated to a "normal healthy" population becomes another issue.

DR. McINNES: Right. Right.

DR. KATZ: And all the vaccines I think you're talking about are inactivated or nonreplicating. If you got into the business of an actively replicating organism or an attenuated live strain, I think that's out as far as any HIV population.

DR. McINNES: Valid point. Yes, Brad?

DR. LEISSA: Brad Leissa, CDER, FDA.

I assume that when we're talking about

natural infection, we're talking about bubonic plague?

DR. McINNES: Correct.

DR. LEISSA: Because naturally, pneumonic plague occurs as well, but from the statistical standpoint in deciphering primary from secondary pneumonic plague, you'd never do it. So to the bioterrorist threat, this wouldn't really suffice.

DR. McINNES: Just speaking from the perspective of really a lot of different field trials in some quite difficult diseases and in some really resource poor settings, the onus to be able to track and capture everybody involved in your study to thinking about the setting where you'd be implementing, the onus of, in fact, going through mapping your own trial site, knowing where everybody is, the fact that people are remote from health care settings, the fact that there will be deaths, the fact that you'll need to have some sorts of systems for validating cause of death, even if it's post mortem questionnaires, I think implementation of a prophylactic vaccine study in

such a setting is just an enormous challenge.

So that's outside of thinking about the numbers of cases that you would need to conduct a study. So I think one really cannot underestimate the infrastructure that would need to be established in order to conduct an efficacy trial in such a setting. I don't know if anybody wants to comment.

So moving on to really number 4, and I think, Sam, you had some discussion about this. Pat, you picked this up again in a sort of passive immunization from a therapeutic mode perhaps. I think we have all been talking about pre-exposure prophylactic use of the vaccine. And this is the situation that has come to really be an enormous challenge for us in terms of the anthrax rPA development program.

Are we thinking at all there will be any circumstances--knowing the disease, having the pathogenesis experts, all the animal model people here, are there circumstances under which vaccination should be considered in a post-exposure

situation of plague?

Yes. Dr. Perry?

DR. PERRY: I can't remember who gave the presentation, but there was one where they did a series of vaccinations, one, two, three, four, five, six days prior to infection. And it wasn't until you got out to six days before infection that there was any protection. And I think this is an aerosol model. It might be a little different for a bubonic model.

But I think that at least from what we see in mice, the disease is so rapid that you're not going to have time to develop much of an immune response, especially when we take into consideration not only is it post exposure, but you have to recognize there's been an exposure. So there's a couple more days after there's been the exposure. You've got your first people coming in sick.

And so, you know, you're really probably talking about three, four days, you know, after the event before it's going to be recognized enough, I

would think, and you'd be ready to give a vaccination. And for aerosol, certainly, it's already too late. You're already having deaths.

For another route, you're probably almost past the time when any sort of immune response is going to help before they reach that endpoint where they're going to get better or die.

DR. McINNES: Pat?

DR. KATZ: I tried to think hard, but the only post exposure I could imagine would be if you had a laboratory accident that you could time, and you then gave antibiotic, your immune globulin, and then also added after the immune globulin was catabolized, give your vaccine then.

DR. McINNES: Thank you. Pat?

DR. FERRIERI: Well, I think the priority in the vast majority of our resources for this whole project should be on pre-exposure. But I like this example that Dr. Katz gave. That laboratory person should have been vaccinated before, you know, in a perfect world. But there will be occasions when that hasn't happened.

And I thought I saw some kinetic data, and I can't find it, of course, that some antibody responses are as early as five to seven days? Would that be a true statement in the nonhuman primate, that you start to see a rise? This was-- it's too late for--that's too late for a big dose that you've aerosoled--aerosolized into the lung.

But maybe not if you were in a subway situation or in a train and you had a low dose. And what about if this bioweapon, in the attempt to make it antibiotic resistant, also defanged the organism slightly. So maybe the organism doesn't have the potency, virulence that it would have, and maybe the illness would be dragged out and would not be as virulent and fast.

So I think we have to think out of the box about how else do you manipulate the bug and to make it maybe less virulent as an accident. Maybe the vaccine then would be relevant post exposure.

DR. McINNES: Mark?

DR. ABDY: Mark Abdy from CBER.

I guess one scenario I'm trying to think

of, if it is a possible one with plague, I don't know what the antibiotic of choice would be and the duration of treatment. But if you wanted to shorten the duration of treatment because of compliance issues and then use a vaccine, a bit like what we're looking at for post exposure in anthrax. Is that a scenario that you could foresee in plague?

DR. POLEY: Gerald Poley from NIAID.

Post exposure for the plague vaccine, you're presuming just a single event. We have seen already that folks who want to do this may do it more than one time. So if one event does occur, that's your canary. And it will take quite a number of people, and it may be too late. But there presumably would be other folks who would demand protection and vaccination.

DR. McINNES: Thank you. Brad?

DR. PERRY: But I would say that's not post exposure for those people anymore. That's pre-exposure, you know? So--

DR. KATZ: It reminds me a little bit of

meningococcal disease, where with your initial exposure, vaccine isn't any good. You give your antibiotic, then you can give vaccine for subsequent exposure. But it's not going to do any good for that immediate exposure.

DR. LEISSA: Mark Abdy raised the parallel with regards to anthrax and post-exposure antibiotics, et cetera. And I think they're very different in terms of not having a spore, you know, the issue of 60 to 100 days of antibiotics. But in most settings, I think especially in a post-exposure setting for plague, most people are looking at seven days of duration. So I don't think it's an issue.

I also don't think, from an indication standpoint, that anyone would be comfortable with just a vaccine for plague, that they would be giving antibiotics and passive immunization as well.

DR. McINNES: Agreed. So I think what we-
-while I don't think we got a resounding 100 percent agreement that there is absolutely no

indication ever, I think there was, Pat, I think you did support that the priorities should be focused on pre-exposure and that the animal studies and the vaccine development program should be focused on pre-exposure at this point.

Does that--the panel is now looking for data.

[Laughter.]

DR. FERRIERI: You stated it as we presented it, Dr. McInnes.

DR. McINNES: Thank you, Dr. Ferrieri.

We are, I do apologize, seven minutes late. But I want to thank the panel very, very much for your very thoughtful input, and my sincere appreciation to everybody at this meeting who contributed to this discussion and put their two cents and two dollars' worth in. Because you have to be part of the path that's being moved forward, and I thank you very much.

Drusilla and Karen?

DR. MEYSICK: There's one more slide. And actually, it's probably the most--but if I can--

bear with me.

It's the most important slide. First off, I would like to thank, personally and for the entire committee, all the invited speakers, moderators, and panel members. I think by the high quality of the presentations and the discussions that have gone on today that it's a testament to all the hard work they have put into this workshop. And for that, I'm very appreciative for all of them. Thank you. Thank you. Thank you.

Secondly, to the program committee, a lot of you see me up here, I'm like a figurehead. But there is a program committee that came about and helped really cement this entire workshop, and those people are from NIAID, Judy Hewitt, Tony Macaluso, Ed Nuzum, and Vicki Pierson.

From Department of Homeland Security, Captain Lauren Iacono-Connors and Luther Lindler. From HHS, Jerry Donlon. And to HHS, we owe extreme thanks for their generous funding of the workshop. And then from CBER, Mark Abdy, Drusilla Burns, Karen Elkins, and myself.

To Rob Watson and the staff at SAIC, who was around, thank you so much for your logistics and meeting support and help and your patience with me. Also to the transcribers, thank you very much for your patience.

And to the guys at the Marriott, who I think did a really nice job setting everything up.

Finally, and I guess also very more important, thank you to all you guys out there. Because it is all of us coming together and putting everything on the table and discussing things which is the best way to get around and to really figure out what we're going to need to do to fulfill the Animal Rule.

It's obviously not an easy task. There's a lot of questions. There are still things we haven't even talked about. But I think this is a great starting place, where we as the FDA can go back and sit down and have the most informative and current data to make decisions. And for that, I appreciate everybody sticking around for so long.

And that's it. Thank you. And have a

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safe journey home.

[Applause.]

[Whereupon, at 12:10 p.m., the meeting was
concluded.]

C E R T I F I C A T E

I, **SUSAN A. HARRIS**, the Official Court Reporter for Miller Reporting Company, Inc., hereby certify that I recorded the foregoing proceedings; that the proceedings have been reduced to typewriting by me, or under my direction and that the foregoing transcript is a correct and accurate record of the proceedings to the best of my knowledge, ability and belief.

A handwritten signature in cursive script, reading "Susan A. Harris", written over a horizontal line.

SUSAN A. HARRIS