It's an experimental process, but it shows that it is possible to get higher levels of removal or inactivation if, in fact, the product you get out of this is still useful, and I haven't heard too much about that yet.

Yes.

This to show you what this data looked like from our laboratory because I have this data in hand, the way we do our clearance calculations is the challenge itself we performed in duplicate. These are the dilutions for each one of these experiments. These are the various experiments right here.

Wherever you have a big S, the animal develops scrapie over the course of the incubation. Where you have a dot, that means there was an animal there, but it never developed disease.

And so we have down here our challenge inoculum. The titers were very consistent between the two challenges and gave us a great deal of confidence in our titrations.

And here's the full filtrate, the filtrate followed by ion exchange; the ion exchange itself over here, and here's the UHT sterilization.

Next.

This is summarized in this table right

here, and I'll just make a couple more points, and then we'll finish up.

We started with ten to the ninth ID-50 per mL, and these were the titers we obtained and the various pooled fractions, but these had to be multiplied by a volume because we had a large volume involved in all of these things.

And this is the step-wise clearance that was calculated. This was diluted one to 1,000, by the way, and so that's the comparison you're making here, and this is the step-wise clearance for each one of these steps. And this would be the cumulative clearance.

And what I've shown you here is that there wasn't a big difference between the filtrate itself and the filtrate followed by ion exchange. Therefore, you cannot really look at these separately. You have to look at them together in this process.

Provided you can combine the removal that you get from this process with the removal you got from the sterilization, UHT sterilization, you'd have around six logs. This may not be valid, and the reason for that is that if this material is being selectively removed here was already selectively removed during the degreasing step, for example, it

wouldn't be valid to include it at this stage.

But what we can say is at the level of the purification stage of the experiment, we're getting somewhere between four and six logs of removal.

Next.

Let's see. I think I've just made these point, except for this one right here. I was surprised at the results of the filtration and the ion exchange steps because we've done this type of removal experiment for a number of other clients and different validation studies, and especially these depth filtrations are often quite effective and remove, you know, more typically four or five logs of infectivity, and it's a warning that the context in which you do these things is important, but we don't know what's important, whether it's the matrix that's different, the actual materials that we use for the filtration, and actually this was a pooled result from several different matrices; whether it's the apparatus, the geometry of the apparatus and the configuration that's different; or whether it's the gelatin itself. Gelatin is often used as a carrier to protect biologicals, and maybe it's serving that purpose here as well.

Next.

Finally, UHT result, just a couple of points. It's consistent with the earlier kinetic experiments that I had done earlier. I think we have to be careful here because it may be sensitive to small variations in time at 140 degrees, and it would be nice to know just how robust this process is, and we should get some evidence for that once the mouse experiment has developed.

And as it looks now, and it's still in development, it's certainly within a log of what we're seeing here, what we saw with the hamster.

Next.

This was just to make those points, and I think I made them verbally. So we don't need the diagram.

Finally, to conclude, it looks from the preliminary data that we have right now that we can expect from the crude gelatin extraction process to remove three to four logs of infectivity, but I think an important aspect of this is that the degreasing step may be the most effective step, which is not what we had originally expected necessarily.

The purification steps remove an additional four to six logs, and the filtration and ion exchange seem to remove the same type of

infectivity, and the UHT provides a potentially secure inactivation step.

This gives us a cumulative removal somewhere in the range of six to seven logs, and an inactivation of somewhere in the range of five to six logs.

Next.

I think that's it.

CHAIRMAN BOLTON: Thank you, Bob.

Questions? I'm sure there will be some. Peter.

DR. LURIE: You mentioned a number of alternative processes to the liming, the acid process, the .3 normal sodium hydroxide process, a number of others, which actually seem to be more effective in reducing infected material than liming.

And my question is: what is the impact of those alternative processes upon the quality of the material in terms of its marketability?

DR. ROHWER: Well, that's a very valid question, and I can't answer it. But I think there are people here who could. Does someone from GME want to take that question? Yeah.

CHAIRMAN BOLTON: Please step to the

microphone.

MR. GROBBEN: Could the question please be repeated because I could hear it very poorly here?

CHAIRMAN BOLTON: The question basically is: is the product that's produced in the experimental .3 normal sodium hydroxide treatment or the other procedure was the autoclaving at 133 degrees C., 20 minutes, followed by direct extraction; do those produce an equivalent gelatin product or useful gelatin product?

MR. GROBBEN: Well, the gelatin produced by the acid process, including sodium hydroxide short treatment is the same kind of gelatin as the gelatin produced by the acid process without this step.

The gelatin produced by the heat and pressure process is a gelatin which its main use is in the food industry, but will also be used in capsule manufacturing as an addition to the limed gelatin. In general, the gelatin is gelatin with a lower gel strength, a gelatin produced by a heat and pressure process.

CHAIRMAN BOLTON: So from the bioengineering standpoint, these are not necessarily direct substitutes for existing gelatin products, but might be additions to or used in conjunction with the

existing products?

MR. GROBBEN: Well, the gelatin produced by the acid process, including the sodium hydroxide step, can directly replace acid bone gelatin. The other gelatin is a different kind of gelatin indeed.

CHAIRMAN BOLTON: Other questions?

I have a few actually, Bob. Let me see if I can decipher my own handwriting.

Well, the first question I guess I have is I didn't see an experiment, or maybe I missed it, that begins with a spiking at bone chips and takes it all the way through the filtration, the purification steps. It seems to me that --

DR. ROHWER: No, there was. The very first two experiments go the whole way.

CHAIRMAN BOLTON: All the way to your steps of filtration?

DR. ROHWER: Yeah, yeah.

CHAIRMAN BOLTON: And those results are not back yet then.

DR. ROHWER: But we don't have the results at the -- we don't have the results from the purification steps.

CHAIRMAN BOLTON: Right.

DR. ROHWER: Those are still on

titration. They're on titration, but they haven't developed far enough to report those yet.

CHAIRMAN BOLTON: Okay, and the next question is -- and this is difficult. Believe me, I appreciate how hard it is to do this -- the initial spiking is in adding this macerate to the bone. How do you determine the titer of the spike? Is that done just from a sample of the macerate?

DR. ROHWER: Again, I did not do this part of the experiment, and Mr. Grobben did, but I know that what he was doing was he weighed everything before and after, and determined how much tissue actually ended up on the bone by weight difference.

MR. GROBBEN: Mr. Chairman, can I make an observation on that?

CHAIRMAN BOLTON: Yes.

MR. GROBBEN: Well, what I actually did was that I made a liquid of the mouse brain tissue by milling it very fine and injected this into the spinal cord of a piece of cattle spinal cord, and that was later then cut to sizes of one and a half centimeters, and part of the material in the syringe was smeared over industrial crushed bones, which was mixed with that piece of cut spinal cord to have very realistic model of infective starting material.

The syringe was filled with mouse brain, with fine milled mouse brain which was that thin that it could be injected, and the syringe was rated before injection and after injection.

CHAIRMAN BOLTON: The question really is: how was the sample taken to determine the titer of the agent that was used to spike? Was it --

MR. GROBBEN: The sample, that was a sample of that mouse brain that was in the syringe.

CHAIRMAN BOLTON: Okay. So no attempt was made to actually take some of the bone with the material and crush it and titer that. So this is my next question.

DR. ROHWER: No, it was the macerate that was titered.

CHAIRMAN BOLTON: Right, okay. What was the first step that was bioassayed downstream from that?

DR. ROHWER: It was the extracted gelatin.

CHAIRMAN BOLTON: Oh, so no intermediate steps were bioassayed. Okay.

I ask this question because in validation studies that I have done, it's always nice to be able to compare an early step with the spike to insure that you actually had that material present at some early

stage.

I realize that in this case it's difficult because you're really handling bone for much of the early process.

Yes?

MR. GROBBEN: Well, Mr. Chairman, on itself the bone would not be a very big problem because you would be able to powder that bone very fine, even taking all precautions necessary because it's highly effective, but the problem is making dilutions with solid material. That is the big problem.

CHAIRMAN BOLTON: Yes. No, I appreciate the difficulty in doing that.

Other questions? I don't want to monopolize. Yes.

DR. BELAY: Bob, is there any way to estimate the concentration or the titer of the agent that may be associated with natural infection in the bones? In other words, is there any way that you could estimate the titer in a natural infection that could be associated with, let's say, the skull bones or the vertebrae?

And if so, how would that compare with the concentration you used in the spiking material or in

the spiking experiment?

DR. ROHWER: Yeah, well, there is an ongoing titration of BSE brain in cattle in the U.K., and the problem with that titration is we have no idea what the incubation time of the disease is at limiting dilution in the cow. So you don't know when the titration is going to be over. It's out five or six years now. I don't think it's even six years, is it? Maybe. Maybe it is out six years, and I know it's up around ten to the seventh or ten to the eighth per gram of the inoculated tissue, or at least that's what I've heard, but it could go higher as the incubation progresses.

If anybody has more recent information on that titration, that would be useful.

MR. GROBBEN: Well, what I heard from my contacts with David Taylor, he says, well, if you take a value of about ten to the seventh as the infectivity for BSE cattle brain, that will be in the right order. It could be a bit higher.

Further, if you make, say, a kind of risk calculation, then you will see that the infectivity in bone will be in the most thinkable worst case not higher than about ten to the 1.8 per gram, while in these experiments we were using an infectivity

somewhere in the order of ten to the six to ten to the eight ID-50 per gram of bone.

So we are something like 10,000 to a million times higher than what could ever happen in reality.

DR. ROHWER: I'd just like to emphasize that that titration is not over, the cow titration, and the numbers I heard were larger than that.

CHAIRMAN BOLTON: Additional questions? Is there one from the floor?

DR. SCHOENTJES: Yes. Dr. Rohwer very interestingly put the question about the robustness of the UHT sterilization process, quoting the data of four seconds and 138 degrees. I just want to remember these values of four seconds, 138 degrees is actually a commitment value for UHT treatment from the GME members dating back in '94, and we didn't know the efficiency.

We expected, as Dr. Rohwer said, from his experiments, amongst others, that it would do something. Now, typically, talking about robustness, typically, because it's a continuous process, we are operating in most of the member -- and maybe they can contradict me -- at 140 or slightly higher and for between seven and ten seconds.

So that's just because of the dispersion of the residence time and dispersion of the temperature control we operate at high, more severe conditions, but the four and 138 is a commitment.

Thank you.

DR. ROHWER: That's extremely reassuring, and my expectation would be ten seconds could be enormously greater effect than four seconds.

CHAIRMAN BOLTON: Bruce?

DR. ROHWER: I apologize. I have a hard time hearing you because there's so much ambient noise from this ventilator.

DR. EWENSTEIN: Okay. I'll try to speak up. Just two sort of experimental questions because I don't do this kind of work. Is it possible to input enough infectivity at the top of the process that you can follow it all the way through both steps?

I understand why in the first cut you would sort of break it up into components, partly you know maybe because different labs have different expertise or just, you know, it makes sense, but because we keep hearing about the issue of subpopulations and could you be removing the same kind of infectivity in the first step as the last step?

DR. ROHWER: That is the ideal way to do

any of these experiments. You want to always condition the spike by the preceding steps in the process before you test it, and so, yes, I would endorse that entirely.

The issue here was especially because the degreasing step is so effective in terms of removing tissue mass, and basically you can see the stuff you put on disappear in that step. We weren't really sure there would be anything left by the time you got to the extraction step, and so this was done in that way.

And also we were expecting that, well, maybe you'd have a little bit left, but you wouldn't see the full potential of those steps. Quite frankly, I was expecting to see a lot more removal than we saw at least from the filtration step and perhaps from the ion exchange step, and for example, if we had been down to a couple of logs, we wouldn't have seen the UHT result.

So in the best of all possible worlds, you do both things. You take and run the process from beginning to end. You tested each step of the process, and you also run step-wise validations for the various process steps to take care of this contingency that you might miss something because you run out of infectivity somewhere along the line.

In fact, that's the way we've been able to do some of our blood validation studies, but it gets to be quite pricey.

DR. EWENSTEIN: But now that you, you know, put in ten to ninth, I think it was --

DR. ROHWER: Yeah.

DR. EWENSTEIN: -- and now that you only see the first piece at four logs, was it, or so, four to five logs, is there enough coming out then as it turns out to run it all the way through and see the next four logs?

DR. ROHWER: Oh, definitely, but what I was pointing out to Dave is that's already occurring.

DR. EWENSTEIN: Oh, okay.

DR. ROHWER: You know, these protocols, the alkaline protocol with BSE and the acid protocol with BSE have both been carried all the way through to the end. So we are looking right now at this intermediate titer, and in a few months we'll have a readout on the final titer after that intermediate stage has been carried through.

That removal, it didn't remove all of the infectivity. So definitely there is infectivity challenging those filters and those ion exchange columns, but we're not looking at the intermediate

steps there. Only the final step is going to be looked at. So we're not going to know how, for example, the infectivity conditioned by the earlier process behaves and the filtration. We won't be able to answer this reservation I have about the filtration, for example, from those experiments.

CHAIRMAN BOLTON: Yes, Pedro.

DR. PICCARDO: How are these kind of experiments being done using material of things from cattle with BSE and titrating back into cattle? So the cattle with BSE, you produce gelatin, and then that is challenged against cattle or transgenics, meaning human, with the human gene.

DR. ROHWER: That would be an excellent experiment, and the technical problem there is just working with cattle, but the transgenics may actually give us an opportunity to get around that issue.

I see that Stan has left already or has he left or is he just down -- I don't know, but can you tell us? Because my impression is that there's actually a BSE titration in your TG animals that has been completed or is underway, and it was quite successful, right?

DR. DeARMOND: Yeah. That's been worked out to some extent. The BSE in the transgenic BOPRP

mouse, and I don't know the titers, but it's in the order of ten to the ninth in brain. I think the issue here is we don't have a good feel for what titer is in skin or in bone without central nervous system contamination.

DR. ROHWER: But I think Pedro's point is that you could do this experiment starting with bovine bone from a BSE affected animal, titrating it into your mouse and look directly at the cow, you know, instead of doing a spike with a mouse derived material spiking bovine bones. Just start with a bovine spinal cord or a bovine skull, and use your transgenic.

DR. PICCARDO: Actually transgenic is in Dr. Prusiner's lab and DeArmond's lab is with the bovine gene, also would be to do it transgenic with the bovine gene or challenge it transgenic with the human gene. That would be the other way.

DR. ROHWER: Well, yeah, you could do that, but it seems to me that the question you're really asking is whether you're removing the indigenous bovine infectivity that comes in with the bovine bones, and for that you'd want a bovine assay.

DR. PICCARDO: Yeah, right. I guess the one question refers to the removal of the infectivity and the other goes into --

DR. ROHWER: Into risk, yeah.

DR. PICCARDO: -- the experiment into the human.

CHAIRMAN BOLTON: Stan, would you like to comment?

DR. PRUSINER: Yeah. I mean, I'm sorry I missed the beginning of this discussion, but just a technical comment. There's really not much point to stick BSE into humanized mice because it doesn't go into these animals, but the bovinized mice would be perfect to do what you want done, and I think it's very appropriate because I was thinking when you're in a slaughterhouse and you watch them remove the skin, the hide, whatever you want to call it. I wonder how much nervous system tissue is really being pulled away as you do that.

Of course, the skin is highly innervated. There are lots of nerves in there, and how many ganglia are getting pulled? My guess is not too much, but there's still a lot of nerves, and I have no idea what the titer in these peripheral nerves that innervate the skin is.

CHAIRMAN BOLTON: Let me get a comment from the floor.

MR. GROBBEN: I should like to make short

observation on your question, sir. Well, of course, we have had a very brief look at the beginning of the experiments at different --

DR. PICCARDO: Sorry. Can you speak up, please?

MR. GROBBEN: Yes.

DR. PICCARDO: It's hard to hear.

MR. GROBBEN: We had a brief look at different possible models at the beginning of the study, and while doing the study with actual bovine infective material was also shortly mentioned, but then looking at how we should have to titrate it and the enormous amount of cattle involved for that, well, it would make it impossible.

Second, at the time we developed the study, the 31V NDV mice (phonetic) was the closest model for BSE. Using actual BSE with, say, cattle adapted or transgenic mice was not possible yet because those mice were not there yet.

CHAIRMAN BOLTON: Right. Steve.

DR. PETTEWAY: Yeah, just a comment on the issue with the potential for resistant or refractile populations of prion or infectivity during processing not being removed. We've looked at that with several processing steps and processes, say, for

immunoglobulin, and for step one, either prion or infectivity that wasn't removed, in other words, wasn't partitioned away, we then took that infectivity in prion and we spiked it into the next step, step two, and we found that still there was some prion or infectivity that wasn't removed at the second step.

However, when that prion or infectivity was spiked into step three, all prion or infectivity that could be measured was removed. So at least in some cases over a series of process steps, it's possible that all input prions through either one step or another that's mechanistically independent could be removed. It doesn't mean it will happen with all processes, but at least some it will happen.

CHAIRMAN BOLTON: Yeah, and it's comforting to know as I misunderstood the first time that this process actually has been tested from beginning to end and is under test and will produce results by the fall or, I guess, early 2002.

DR. ROHWER: We put all our titrations on at the same time. So we'll have a year on these animals late September, and we'll feel pretty confident with the 301D model.

Actually this is the first experiment we did in that model. So I'm not sure -- first

titrations we've done in that model. So I don't know what the incubation time is really, but we're still getting animals coming down at a fairly regular clip right now, and once we get to a point where we haven't seen any new infections for several months, I'll feel pretty confident that we can draw our conclusions from the experiment. We're just not at that point.

But we'll have a year on it by late September, and on all of the arms of our experiment, the hamster experiment is essentially over. We haven't seen an infection there in a long time, and the animals are starting to die of old age.

This was not by our choice. This was an EC decision to hold these animals this long, but in any case -- oh, the other point though is that the Edinburgh experiment was staggered. The inoculations were staggered. So some of them are quite far behind the others, and there will be a longer lag for some of that data, but it's all in animals.

CHAIRMAN BOLTON: Very good. Any other questions, comments from the committee?

One more from the floor, yes.

MR. GROBBEN: I should like to make a few observations about things Dr. Rohwer probably did not understand while he was, well, talking with us about

this presentation.

He speaks about that the results, final results of the concentrated gelatin in Edinburgh will be available in August, but I'm afraid that will be a few months later. In August the titrations are finished, and after that the pathology of the animals has to be done, which is also taking quite a while because we are talking about a few thousand mice.

Then the heat and pressure process is not only an experimental process, but this process is, indeed, already actually used in a factory in the Netherlands, and at the moment still the major use for this gelatin is for the licorice industry because it seems to improve the taste of licorice enormously.

CHAIRMAN BOLTON: Good. I like licorice. I can't wait to try it.

(Laughter.)

DR. ROHWER: We know alcohol doesn't do anything.

CHAIRMAN BOLTON: I usually don't mix alcohol and licorice.

DR. ROHWER: Right.

CHAIRMAN BOLTON: I think what we'll do now is take our break, come back in 15 minutes. That would be 10:15 for the open public hearing, and we'll

see you back then.

(Whereupon, the foregoing matter went off the record at 10:56 a.m. and went back on the record at 11:15 a.m.)

DR. FREAS: Could I ask committee members to take their seats and the audience take their seats, please?

CHAIRMAN BOLTON: Welcome back. We're entering the home stretch. I would now like to open the topic number three to the -- have the open public hearing portion and welcome comments from the floor, from the public.

MR. GOOSSENS: My name is Patrick Goossens. I am President of GME.

And I would like to come back to two points which have been raised this morning. The first one is after the presentation of Dr. Schoentjes, I was a bit amazed by the number of questions that have been asked about European raw materials, about what is happening to European bones.

I think it's quite confusing, and therefore, I would like to emphasize again that, in fact, today that is not the issue because European raw materials, or at least bovine raw materials, today are not used by GME members to produce gelatin that is

imported in the States for pharmaceutical or for food applications.

We have some European bone gelatin produced, and that goes to photo applications, but none of it is going to pharma or to photo -- to food applications.

Now, where do the bones come from? Well, as you will remember from Dr. Schoentjes' presentation, a quite substantial amount of European gelatin is produced in Europe based on American bones.

Now, the question is where do the other bones come from. Well, again, I reemphasize they are not coming from Europe. We are importing bones also from Africa and from Asia.

Now, the question, of course, is that if you're importing from Africa and Asia, how can you be sure that everything is okay because no matter how you look at it those countries might have a certain reputation. Well, I can say we only import bones from Africa and from Asia from countries which have submitted a file to the Scientific Steering Committee in Europe to be -- to get a classification, a GBR classification.

Now, submitting a file means that you must have a system in place in the country itself for

looking for BSE, for reporting BSE, and then the whole system around it. We're only importing bones from those countries.

Furthermore, the suppliers where we are taking bones from in Africa and in Asia, they are audited by the local authorities and also by the gelatin producers themselves.

So once again I would like to reemphasize today we are not using European bones to make bovine bone gelatin to import in the States. In other words, all the gelatin that we import is FDA compliant, is compliant with the FDA guidance to the industry.

The sourcing of raw material is, in fact, the major reason -- the major -- how do you say that? -- the major ground for us for guaranteeing the safety of gelatin.

The guarantee of safety that we give starts with the raw material. The study that we're doing on the production process today, in fact, we only look at it as a kind of back-up if something would go wrong, in the very unlikely case something would go wrong with the raw materials that we use. Then we prove that -- with the process, that the process is removing the prions also.

But once again, the sourcing of the raw

material is for us the major point for guaranteeing the safety of the gelatin. This is also why today we are not asking to the FDA or to the TSE Advisory Committee to approve European bones.

We hope that once the final results of our study are available, that then putting together all the measures in place in Europe, all the legislation, all the selection of the bones, and also the guarantee that or the proof from the study that the production process is removing the prion, at that time we will try to come back as GME and say, "Look. If you put everything together, then we think that the final product, even when it's based on European bones, is a safe product."

But this is not the issue yet because today the study is not final, but that will probably happen, I hope, for the next TSE Advisory Committee.

CHAIRMAN BOLTON: Thank you.

Questions? Stan.

DR. PRUSINER: Yes. We had a rather content -- I don't know whether you were hear yesterday, but we had a rather contentious discussion last --

MR. GOOSSENS: I wasn't here yesterday.

DR. PRUSINER: -- late into the evening,

when everyone, at least I was exhausted. And we were talking then about plasma fractionation, and this concept of fractionating European plasma one week and American plasma the next week in a particular plant, wherever it is.

Now, is that happening in GME producers where you make gelatin from European bones one week and then the next week you use Asian bones and the next week you use African bones and the next week you use American bones?

MR. GOOSSENS: Well, most of the production facilities for bone gelatin in Europe, they are using bones from different sources. That might be American bones, sometimes European bones if it's for photographic --

DR. PRUSINER: So one production facility uses bones from different places?

MR. GOOSSENS: Yes, indeed. But we must also say that in most of the plants between the different production processes for different raw materials, there are cleaning procedures in place.

Now, you can discuss about those cleaning procedures, and that would take us a long way. On the other hand, there is also another issue, and that was covered this morning also.

When we produce gelatin from whatever raw material, we always apply the most stringent conditions, and the reason is that if a certain customer asks you a certain quality of gelatin, the gelatin production process is not such that you can turn a few buttons in the beginning of the process and you get then the final product asked by the customer.

In the end, the final product for the customer is a blend of different operations of gelatin. Of course, if that customer requests gelatin only from, for instance, American bone, and most of your American customers do so, then of course the operations that are used are only for American bones.

But again, final products of gelatin are usually blends of different operations, and that's why it's very difficult to limit your production process in certain production plants to one source of raw material or one production system even.

Is that an answer to your question?

DR. PRUSINER: Yes.

MR. GOOSSENS: I had one second remark, and that was on the acid bone and the possible sodium hydroxide treatment which was presented by Dr. Rohwer. Now, it has been proven that the sodium hydroxide treatment helps a lot in removing the potential TSE

effectivity.

The question then was asked, well, acid bone or even lime bone, if you would applied the sodium hydroxide treatment, would that give a comparable quality of gelatin?

The answer is yes, but there is another practical question, and that is very often the gelatin today when it goes through pharmaceutical applications is used for registered products, and that means that even if the gelatin industry is ready today to apply sodium hydroxide treatment to acid bone or to lime bone gelatin, our customers, they want some more time because they have a registered product.

And if we change a step in the production process the registration has to be changed. There are tests on shelf life which have to be done. It's not something that can be done from one day or another. It takes several months.

But we must say that, in fact, the gelatin industry is ready to apply the step if the market is ready for it.

CHAIRMAN BOLTON: Very good. Thank you.

Other questions or comments from the public? Questions from the committee?

(No response.)

CHAIRMAN BOLTON: Well, very good. I think we'll move on then to a presentation by Dr. John Bailey, who will give the FDA summary on topic number three.

Dr. Bailey.

DR. BAILEY: Okay. Thanks.

I'm going to provide just a very quick, brief summary or transition into the panel discussion that will close out today's meeting just by noting that the agency has been considering the safety of the products that we regulate with regard to TSEs for a number of years, probably will continue to do it for many more years.

Included have been a number of what we call processed ingredients that are produced from bovine raw materials, and certainly gelatin falls within that category as a processed ingredient.

The committee first took up gelatin in 1997, and shortly after that we issued our guidance document, which remains as the current FDA position and policy on the safe use of gelatin.

Clearly an important part of our consideration relative to the safety of gelatin have been the studies that have been conducted over the years as part of the assessment of the manufacturing

process and reduction of infectivity, an infectious agent at the various steps.

And the industry has, I think, shown a good commitment to undertaking these studies. As you can see they're very complex, very costly I'm sure, but I think have been -- the commitment that we've seen from them has been very reassuring on our part.

Gelatin is on the committee agenda today primarily to allow for the report of progress of these ongoing studies that are being conducted by the industry and the results that have been obtained to date. And I think we've heard excellent presentations by both GME and also Dr. Rohwer of the progress that has been made and what's planned for the future.

It's an opportunity I think also for the committee to consider to have available experts so that if you have questions or want further clarification or to even make suggestions about studies or directions this is a good time to do it. We have the folks here who are knowledgeable and can answer these questions for you.

However, as Dr. Rohwer, I think, pointed out very clearly, this is work in progress and is subject to change, and the time course for these additional studies will certainly run out throughout

the remainder of this year and probably into next year.

So we are very interested in following these and want very much to continue to receive updates, and especially if any of the information that's generated may have an impact on our decision making.

Finally, I would point out that FDA does intend to continue to monitor this issue very closely and may take up the issue of the safety of gelatin at subsequent meetings.

At this point, I don't think we've actually scheduled anything on the agenda, but certainly we remain very interested and will follow this very closely.

So with that I will turn it over to the committee if you have any further discussion, questions, anything else.

CHAIRMAN BOLTON: Thank you, Dr. Bailey.

Well, my agenda says "committee discussion, committee discussion." So I suppose that means we should discuss things twice.

(Laughter.)

CHAIRMAN BOLTON: We are open for discussion on topic three, and I would just point out

as Dr. Bailey, I think, just emphasized, now is a good time for us to contemplate what information we would like to have probably six months or a year from now when we are back contemplating some recommendation on policy that the FDA may ask us for on this issue.

We're not asked to give those recommendations now, but I'm sure we will be. So the industry representatives are here. We have people who are expert in and already conducting the validation studies. If there are any items, particular items of concern about the design of the validation studies or things that you think were left out, this is a good time to bring time up.

Other questions of policy that could be possibly resolved by anticipating factual information that would be needed, this is the time to bring those up and discuss them.

Don.

DR. BURKE: Just a point of clarification. There was one of the papers that was provided in our kit that we get from Nature on amplification of misfolded prion proteins. Does that have any implication for detection in this kind of assay?

It may not have a perfect sensitivity, but it might have enough to be used in this kind of -- and

I just -- there's enough experts here. I'd like to ask you: does that have any implications for doing this kind of work?

CHAIRMAN BOLTON: Stan, do you want to talk about that?

I think it has potential but it's unproven at this point.

DR. PRUSINER: Yeah, I think we don't know whether the paper is right. It could be like in a -- Nature papers fall into two main -- three main groups. One group is you have no idea why it's in Nature.

The second group is it's clear it's in Nature because it's exciting, but never is reproduced.

And the third group is that it's exciting and it's reproduced.

And we don't -- of the latter two, because we know this is an exciting concept, we don't know whether it's reproducible or not.

And I think we have to wait and see whether this is really reproducible in other people's hands and then it would have application.

CHAIRMAN BOLTON: And there's always a concern if it works as advertised, so to speak, is the amplification really a measure of infectivity in the sample or is it a measure of some abnormally folded

protein, PrP, that may not be necessarily the infectious component?

And so a lot of work needs to be done to sort that out I think.

DR. DeARMOND: There's a further issue, too, that they didn't clarify. Their controls are a little weak. One of them is the possibility that the amount of material of scrapie PrP that they spike, whether with each cycle, they don't denature it just a little bit so that their detection system now sees more of it because the protein tends to be aggregated.

So they haven't resolved that issue either. So we don't know whether it's amplification or just better detection with each cycle.

CHAIRMAN BOLTON: Yes, Don.

DR. BURKE: It at least raised one other possibility that there might be <u>in vitro</u> amplification during other chemical handling of nervous tissue, and although it seems unlikely, it made me wonder about whether or not in the handling of processed tissues that you might inadvertently get amplification as well.

CHAIRMAN BOLTON: I think that's a good point, and possibly a scary point to contemplate at this time.

Other questions? Steve?

DR. DeARMOND: It seems to me the -- I guess I don't know, unless, Stan, you've heard anything, but I don't think we know in the worse case of BSE what the infectivity titer of hide and bone is. I don't think that's been resolved. We certainly don't know it for skeletal muscle, although there's some issue that skeletal muscle itself in some animal models can be infective.

So that's certainly one piece of information, I think, that has to be derived.

The other to me has to do with something that Stan kind of alluded to, is the uniformity at which the hide is dissected and the bones are removed, because there's always a chance of contamination in those processes. Certainly the bones that are close to the spinal column really become a problem.

And there may be a very precise process in some rendering plants, but I'm not sure it's precise everywhere. We should know something about the uniformity in which that's done.

So the two issues, we need to know something about infectivity in the worse case of these tissues, of these organs, and we need to know how uniform the process of removal is.

CHAIRMAN BOLTON: I guess the question then becomes whose obligation is it to obtain that information?

Do we put the onus on industry to provide that or is there some mechanism to encourage independent investigators to determine that?

I'm not sure how. Stan?

DR. PRUSINER: Yeah, I was sitting and we were talking and we were whatever. Dr. McCullough and I were speaking about the fact that probably most of the people at this table have never been at a slaughterhouse. I used to spend a lot of time there collecting the cord plexus out of brains of cows, and I think somehow it would probably really instructional for this committee, and maybe the FDA can arrange this, either to go through a slaughterhouse and look at it first hand or have some really graphic films here about from the moment the bolt hits the head of the cow what goes on and all of these issues.

Because I just think that the committee doesn't really have an appreciation of just what happens to these animals and how violent and horrible this is, and these are the conditions under which these animals are slaughtered all over the world, and it's not exactly the most humane kind of thing.

That's really irrelevant, but the process is so messy and the blood is dripping everywhere and it's very hard to contain this, and then what about the next animal and the next animal?

So I think it would be probably very useful if you want to have a discussion of gelatin or other products from cows, now, down the road, it seems to me there ought to be some education about this.

CHAIRMAN BOLTON: Bill, what's the closest slaughterhouse to Bethesda?

No, I think that's a reasonable recommendation. I actually have participated in the slaughtering of cattle, and it is a messy business. And would you like to?

DR. CRAWFORD: I would like to second that motion as one who grew up in a slaughterhouse. The second thing, though, is the prospect of this committee --

DR. PRUSINER: How tall are you? Stand up.

(Laughter.)

DR. CRAWFORD: I was six feet, four.

(Laughter.)

DR. CRAWFORD: The prospect of this committee coming into any one's slaughterhouse, I

think, is enough to strike fear into the economic world. So that's got to be done very carefully, and probably in concert with USDA which has regulatory control, and Dr. Ferguson can set it up for us this afternoon, I'm sure.

DR. FERGUSON: Wrong agency. But actually in all seriousness, there are some large plants in Pennsylvania that have some impressive operations.

CHAIRMAN BOLTON: Road trip. Okay. Well, we'll leave that as a recommendation to the FDA.

Yes, Bruce.

DR. EWENSTEIN: Actually this is more sort of a question to the FDA. I mean, we heard from industry that they're prepared to incorporate what sounds like an improvement in the margin of safety in prion inactivation. Their fear is, you know, legitimately that the pharmaceutical industry, obviously, has to have the new gelatin approved for a whole variety of products.

And so I think now the question is: how difficult would that be? Is this really just a matter of a few months and reasonably few tests or is this a multi-year process that we're talking about?

CHAIRMAN BOLTON: Would somebody from the FDA want to comment on that?

DR. CHIU: Four pharmaceuticals use the gelatin. We do have drug master files to document the process of preparing the capsules and preparing the gelatins. A change like that to increase the assurance of safety could be reported in any report. And the data would need to be submitted in the annual report to show the characteristic of the gelatin has not been changed, and in our applications for drugs.

We will also need it to be documented, the shelf life. The product characteristic will not change through the shelf life. That could also be documenting in annual report.

So there's really no need to go through the approval process, no waiting for doing that.

CHAIRMAN BOLTON: I'd like to invite a comment from GME on that to respond. Is that -- what time frame do those requirements impose upon GME and what about expense?

Please.

MR. SCHRIEBER: To the best of our information we have received from the capsule industry and representing the pharmaceutical industry the time frame will be at least 12 months to run though the whole process, including shelf life tests, bioavailability, and so on.

CHAIRMAN BOLTON: Is this something that would -- assuming that there's no pressure put on the industry to do this, but that you could move at your pace, is this something that the industry would move on voluntarily?

MR. SCHRIEBER: It is in progress.

CHAIRMAN BOLTON: It is in progress. Very good.

Stan?

DR. PRUSINER: I'd like to just, since we're talking about what we'd like to have, would like to see, and now this is out -- so I declare a total conflict of interest here. And let me explain.

CHAIRMAN BOLTON: I know where you're going.

DR. PRUSINER: I have been trying for a long time now to get a large series of tissues, and I may be successful soon from all parts of the cow and to titer these out from bovine transgenic mice.

And any pressure the FDA can apply to the Minister of Agriculture in Britain will only help because that's where these tissues reside, and I've been working -- just to show you how hard it is, I worked through the Minister of Agriculture. Strike that from the public record.

But I think this is the kind of data that everybody would like to have here. Everybody would like to know what is the titer of BSE prions in all these different tissues. This comes up constantly in the discussion of a cow.

We don't know the answers to that. I think this is really important. I mean, it's just as -- obviously it's just as important to know what -- are there prions, as Hank Baron was really pushing? We don't know the answer in the blood of somebody with variant CJD.

But there are precious few studies -- there are no reliable studies in cows. All of the data in cows has been pushed through R3 mice, which we know are four logs less sensitive than the transgenic mice. So if you say there is nothing in a tissue in a R3 mouse assay, you have another four logs that you could detect with a transgenic mouse with a bovine PrP gene on a null background.

So I think this is, as I said, this is all self-interest because I really want to know this data. I'm pushing to carry out these studies, but I think this is really important information that any regulatory body, whether it's here or in Europe or in Asia or in Africa, needs to know.

DR. DeARMOND: Well, that's why I also am arguing for it. Because how can we judge what these tests that are run, the Rohwer type tests that are run through the process, whether -- what it means?

If you start out at ten to the ninth, you end up with probably ten to the square or ten to the third infectivity still at the bottom. And we know -- I was talking with Stan last night that the prion protein tends to aggregate in funny ways, and it may not be uniformly distributed. So one set of gelatin could theoretically have a high titer, much higher titer than an other.

But if we start out at ten to the third or ten to the forth titer at the worse scenario, in the worse case of BSE, then the process could reduce it significantly to a point where there'd be no effect. We'd have a -- at least I would have a higher confidence that there's nothing in the end product that's dangerous.

But we still need to begin with what are you putting into the system and we don't know the answer to that. And that's true of virtually everything we've talked about. We don't know what's going into the system, and we don't even know what's in the end product, whether it's blood or whatever

we're working with.

CHAIRMAN BOLTON: Clearly, additional data on tissue distribution of BSE assayed in transgenic mice, bovinized transgenic mice would be very valuable.

Bob Rohwer.

DR. ROHWER: I'd just like to address one point that Steve just made and that is when you're doing a validation study you cannot remove more than you put in. You can't claim removal of more than you put in. And I think it's an important point that when you reduce infectivity down to zero, that doesn't mean that in the real world when you're working with tons and tons of stuff that there might not still be infectivity in that product.

You can only claim to have reduced as much as you measured. And so going to zero is really not an advantage, and you see people abuse this by you don't put as much in and you can get to zero a lot easier.

And so I think it's important to always push it in the other direction. You know, you start with as much infectivity as you can so that you've covered that possibility.

DR. DeARMOND: No, I think your studies

are nice in that regard. If you start with a maximum essentially, a very big bolus, and you were able to reduce it by ten to the forth to ten to the sixth by the time you're finished, what if you start instead of ten to the ninth, you start at ten to the fifth or ten to the fourth?

What would you predict with this system?

You should get --

DR. ROHWER: You should get zero.

DR. DeARMOND: Yes.

CHAIRMAN BOLTON: Well, correct me if I'm wrong, Steve, but I thought your point was that knowledge of the actual bio load, the prion load in native skin or bone, cattle skin or bone, would be important to know because then you know what the challenge of the actual manufacturing process is.

DR. DeARMOND: Exactly. Well, that's what I said, yes. But I didn't say virus. In fact, that's --

CHAIRMAN BOLTON: No. Did I say virus?

DR. DeARMOND: Yes. Didn't he say virus?

CHAIRMAN BOLTON: I would be -- I'm shocked that I said virus.

DR. PRUSINER: No, I said virus.

(Laughter.)

CHAIRMAN BOLTON: Did you say virus?

(Laughter.)

CHAIRMAN BOLTON: That would be even more shocking, Stan.

Additional questions and comments? Peter.

DR. LURIE: Just to follow up on what seems to be the good news that at least some gelatin manufacturers are moving to include the sodium hydroxide step, but my question is -- well, there's two parts.

One, what fraction of the GME members are actually doing this?

And secondly, since GME members account for only 45 percent of gelatin world production, what do we know about the non-GME members?

CHAIRMAN BOLTON: That may not be quite fair to ask the GME that comment on the non-GME members, but --

DR. LURIE: I'm sure they're very interested in what they're doing.

MR. SCHRIEBER: Let me answer the first question. Implementing this additional step is under consideration by all GME members, but again we have to verify this for all kinds and all grades of gelatin because when they're going to implement this, it will

be implemented as photographic gelatin, eatable gelatin, and mainly with regard to photographic gelatin we have to verify what is the potential effect on the final product in this respect as well, because very often the production process is partly becoming photographic gelatin, partly becoming pharmaceutical, and a part might become edible gelatin.

So we have to run though the whole thing. Up to now we have only studied pharmaceutical part, and we are confident that nothing's going to happen there. Edible is no problem, but photographic is still under progress.

So this, if you are going to decide to do it, it will be done by everybody. We're not going to segregate ourselves because all gelatin has to be safe, and therefore, of course, we would be happy if we would be able to convince the rest of the bone gelatin world outside GME to implement that steps as well.

I can't guarantee this, but of course, this will depend to a certain extent at the end of the day on regulation. So if it would be come a regulatory requirement then of course the rest of the world would have to follow what Europe is doing.

But I can't really cross my fingers and

asking about this, but I can't give any guarantees in the moment for non-members.

DR. LURIE: I guess that raises the question whether within FDA there's any consideration of providing just such a regulatory requirement.

CHAIRMAN BOLTON: Can we get a response?

DR. CHIU: Since we haven't seen the whole report and all the data, so right now, you know, I don't think we are in the position to tell the industry you must do this. However, once we have the final reports and we feel this is a really important step to assure the gelatin would be safer, then we will issue an additional policy to let the industry know.

CHAIRMAN BOLTON: Please introduce yourself.

MR. MASSON: My name's George Masson. I'm President of one of the members of the GMIA, which is the U.S. equivalent of GME, and I'm Vice President of GMIA.

Just to speak to the question of what is the rest of the gelatin world doing, you know that Europe, the GME members, produce something like 45 percent of global production. We in the States produce roughly 60,000 tons, which is about a quarter

of the global production.

So there you have between the two organizations, what is that? Seventy-plus percent.

Most of the gelatin made in the States, however, is actually porcine gelatin, probably about 60 percent of it. So again that's not an issue as far as TSEs are concerned.

The remainder is bovine between hide and actually born gelatin, and to the extent that bone gelatin is manufactured, a lot of that is actually made by Eastman Kodak. So again, not an issue as far as this committee is concerned.

The other bone manufacturer is a subsidiary of GME member and whatever GME is going to be implementing, I'm sure that will be implemented here in the States as well.

So that covers, I think, the situation for the U.S. industry.

CHAIRMAN BOLTON: Thank you.

Additional questions or comments from the committee?

DR. BELAY: This is just a minor comment. And that is estimation of what actually goes into the system in terms of the infectivity in an actual set-up should also take into account the pooling effect. In

other words, you know, the worst case scenario would be more than one animal that's potentially impacted could get into the system. In the type situation of just one animal or parts of the body, parts of the organs from one animal may not necessarily, you know, answer that question. So we need to take into account the pooling effect.

CHAIRMAN BOLTON: Exactly. Good point, very good point.

Others?

(No response.)

CHAIRMAN BOLTON: Well, I guess I would like to commend the GME for undertaking these studies. I don't think they were forced to do so, but I think that my impression is they had a forward looking approach and decided to look into this, and I think that was a wise and maybe even bold action.

I also have a recommendation that they return when these studies are completed to this committee and present the final data when that's available, and that we, again, consider that, and I'm sure the FDA will ask us to recommend something to them.

And at this time if there are no other discussions from the committee, I'd like to thank the

committee members, the consultants, industry representatives, and the general public for attending the meeting, and I think we can stand adjourned.

Thank you.

(Whereupon, at 11:50 a.m., the Advisory Committee meeting was adjourned.)

CERTIFICATE

This is to certify that the foregoing transcript in the matter of: TSE A/C MEETING

Before: FDA-CBER

Date: JUNE 29, 2001

Place: BETHESDA, MD

represents the full and complete proceedings of the aforementioned matter, as reported and reduced to typewriting.