In The Matter Of:

Paul Halderson, et al., v.
Star Blends, et al.,

Lewis G. Sheffield
May 9, 2014
Volume 2

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STATE OF WISCONSIN CIRCUIT COURT TREMPEALEAU COUNTY

Paul Halderson and Lyn M. Halderson, N17388 County Road T Galesville, Wisconsin 54630 and Arctic View Farms, LLC N17388 County Road T Galesville, Wisconsin 54630, Plaintiffs, vs. Star Blends LLC 1919 Riley Rd. Sparta, Wisconsin 54656 and ABC Insurance Company, a fictitious company and Northern States Power Company d/b/a Xcel Energy Services Inc. 1414 W. Hamlin Avenue Eau Claire, WI 54702, Defendants.

Case No. 12-CV-74
Code Nos: 30303 & 30201

STATE OF MINNESOTA COUNTY OF CASS

IN DISTRICT COURT
NINTH JUDICIAL DISTRICT
11-CV-12-1670

Randall and Peggy Norman, Plaintiffs, vs. Crow Wing Cooperative Power & Light Company, Defendant.

VOLUME II

Continuing Deposition of LEWIS G. SHEFFIELD, PhD, taken pursuant to Notice of Taking Deposition, and taken before John T. Kirby, a Notary Public in and for the County of Dakota, State of Minnesota, on the 9th day of May, 2014, at 1 South Pinckney Street, Madison, Wisconsin, commencing at approximately 9:22 a.m.

APPEARANCES:

Scott Lawrence, Esquire, of the LAWRENCE LAW OFFICE, S.C., 403 South Fourth Avenue, P.O. Box 117, Saint Nazianz, Wisconsin 54232-0117, 920-773-2811, ATTORNEYS@LDLAWSTN.COM, appeared representing the Plaintiffs, both captions.

Timothy R. Thornton, Esquire, of the firm of BRIGGS & MORGAN, 2400 IDS Center, Minneapolis, Minnesota 55402, 612-977-8400, tthornton@briggs.com, appeared representing Defendant NSP/Xcel Energy.

Paul F. Carlson, Esquire, of the firm of KENNEDY, CARLSON & VAN BRUGGEN, LLP, 116 Ash Avenue NW, P.O. Box 647, 218-631-2505, pcarlson@kcvblaw.com, Wadena, Minnesota 56482-0647, appeared representing Crow Wing Cooperative Power & Light Company.

ALSO PRESENT:

Theresa A. Peterson, DVM.

VIDEOGRAPHER:

Mark C. Haskins, HASKINS MEDIA SERVICES, 1071 Whitney Drive, Apple Valley, Minnesota 55124, 952-997-6455, mark@haskinsmediaservices.com.

EXHIBIT INDEX

EXAMINATIONS

* * * *

WHEREUPON, the following proceedings were duly had:

* * * *

MR. HASKINS: Today is Friday, May 9, 2014. The time is approximately 9:22 a.m. This is Volume II, Tape Number 1, of the continuing video deposition of Dr. Lewis G. Sheffield, taken by Defendant Northern States Power Company in the matter of Paul Halderson, et al, versus Star Blends LLC, et al, State of Wisconsin, Circuit Court, Trempealeau County, Case Number 12-CV-74. This deposition is being held at the Law Firm of Boardman and Clark, Madison, Wisconsin.

My name is Mark C. Haskins, I'm the video technician of Haskins Media Services, Apple Valley, Minnesota 55124.

Will counsel please note their appearances, after which the court reporter will swear in the witness.

MR. LAWRENCE: The Plaintiffs appear by attorney Scott Lawrence. Also present is their consultant, Dr. Theresa Peterson.

MR. THORNTON: Tim Thornton for NSP.

MR. LAWRENCE: Then, I think you ought to put the Minnesota case into the record also, please.

MR. HASKINS: Sure. Again, this matter has also been noticed in the matter of Randall and Peggy Norman versus Crow Wing Cooperative Power and Light Company. Court File Number 11-CV-12-1670, in the State of Minnesota, County of Cass. I think that covers it then?

MR. LAWRENCE: Sure. And the Plaintiffs in that case, at least for the time being, appear by Scott Lawrence, and also Charles Bird may appear by phone later this morning.

MR. CARLSON: Paul Carlson representing Crow Wing Power.

MR. HASKINS: Okay.

LEWIS G. SHEFFIELD, PhD,
an expert witness in the above matter,
after having been first duly sworn,
testified under oath as follows:

CONTINUING CROSS EXAMINATION

BY MR. LAWRENCE:

Q Good morning, Dr. Sheffield.
A Good morning.
Q Good to see you again. I am going to, in a few moments anyway, continue asking you questions about exhibit 254, that long spreadsheet we've talked about the last time,

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and the other documents you have in front of you which I will get to in a minute.

Before I do that, with respect to the second study, the later one in time that we were discussing in the first round of your deposition that involved measurements of involving Messenger RNA, could you describe how that study came about and how it was funded, please?

A You're asking some very long ago - -

Q Understood. And if you don't remember, you don't remember.

A Well, I can sort of, I think, give a general gist of this. I had been part of an earlier study with Douglas Reinemann, in which we did a lot of functional measurements. And, I am sorry, I am not going to remember years from this, but there was a request for proposals through the College of Agriculture, I believe that came about from a line in the state budget. But this is, again, very old memories.

I responded to that and this project was selected for that. And it was designed to be, in some respects, a follow-up to the previous Reinemann study looking at some broader ideas.

Q So the funding basically came out of the Wisconsin state government?

A I believe that is correct.

Q And if I understood what you just said correctly, you believe it came through the Department of Agriculture, is that right?

A Well, it came through the - it was funneled to the UW

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through the College of Agriculture.

Q Oh, the College of Agriculture. Excuse me.

A Yes. I don't know about state departments that would be involved. I may have known that ten years ago, but I don't recall that.

Q Sure. All right. And can you describe with any more specificity than that, than what you just did, what the object was in follow-up to the earlier study?

A Yes. Yes. The objective was to first develop some tools to assess gene expression in cattle. At that time, this technology was rather poorly developed, particularly in cattle, it was beginning to be developed in humans and model species, like mice.

Our objective was to try to get as broad a spectrum as we could of things that might be relevant to immune function. And, of course, along the way, include some things that might be either general of some genes that might be part of the immune function, but part of bigger things as well. And as you might notice in here some things that we used as controls that you wouldn't expect to see in immune function cells, and then use this to determine whether exposure to very low voltages in dairy cattle affected any of these potential measures.

Q And we discussed before, a little bit anyway, about exhibit 254, which is a four-page spreadsheet, and I'll represent to you that that spreadsheet was printed off of materials that were subpoenaed through the University and provided in April of 2008. They were copied by a copy shop here in Madison directly from the University Counsel's office and provided to us. And that spread-

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sheet appeared on a disk that was - probably you have never seen this disk cover before, but it's got your name on it and it talks about SV ARRAY. And there's all kinds of files on there, one of which contains that spreadsheet.

Do you recall seeing that spreadsheet in the past, back when you were doing - after you were doing - -

A I don't recall whether these - all the last rows of the means were in the spreadsheet or not, but it looks very much like this, and I do seem to, my suspicion - well, let me say, I didn't realize that it would have also been sorted by treatment group, whether they have treatments at random. But it does look very much like the spreadsheet that I would have generated. And I don't know - I don't recall whether those last rows were something I had generated in there or not. The numbers in them look like the numbers that I would have generated, had I done it. So.

Q Okay. And there very well may be a copy of this - -

A I'm not saying it was on the original, I'm just saying I don't recall that it looked exactly like that. But the data looks right.

Q There may be a copy of that spreadsheet on that disk without the means and so forth, the last four rows also, I'm not sure. But that one does appear - -

A This does look like something I would have generated. Like I said, I don't recall that being on there, but I'm not saying it wasn't. It was like ten years ago that I generated this.

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Q Sure. And on the disk, as we received it from UW, there was the - well, the data can be opened in EXCEL, what seems to take a few steps to do that with software that's available these days. Can you describe for me what software you were using back then?

A I believe that was done in the program called Minitab.

Q Can you spell that for John?

A M-i-n-i-t-a-b.

Q All right. Go ahead.

A That is a fairly standard statistics program. It's commercially available, and reasonably widely used.

Q And I take it from your earlier testimony, you do not recall asking the program to calculate the means or P values in this case, is that correct?

A Oh, I would have done that.

Q Okay.

A I just don't recall putting them on this spreadsheet. I certainly would have asked the program to calculate means, standard errors and P values.

Q What statistical test would you have used initially to do that on this particular data?

A The initial test would have been a t-test. We have two treatment groups, and so there would have been a - what's often called a Student's t-test done on each piece.

Q Then, is there such a thing as a two sample t-test?

A That's what the Student's t-test is. That's what I would have done.

Q Just so that we can get the mathematics straight, let me mark another exhibit, please. This will be 277.

MR. THORNTON: I thought we were on 276.

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Q That's the disk.
MR. THORNTON: Oh, okay.
Q I'll just give you a moment to look at 277. I believe this just reflects the basic statistical computation that would be done to accomplish a two sample treatment and control t-test in these circumstances. Would you take a look at that and tell me if you agree?
A That looks like the t-test, yes.
Q All right. So, in this particular case, given that we have ten treatment cows and ten control cows, I believe we'd be looking at a t-test with - or a pool t-test with 18 degrees of freedom, is that correct?
A I think that's correct.
Q And so, assuming the mathematics to be correct as at the bottom of exhibit 254, let's just talk about what those numbers are. We will take the first variable, ACK2 as an example. The C mean would be what?
A That would be the mean of the control group. That is the cows that were not treated with voltage.
Q Sure. And that's just a simple average - -
A That's just an arithmetic average in this case, yes.
Q And the T Mean would be what?
A That would be the arithmetic average of the animals, the treated animals, that is, exposed to the current.
Q And the Fold (T/C) is what?
A A common way of expressing gene expression is to look at how much it changed as a relative. So, basically, that is taking the T Mean divided by the C Mean.
Q And then the P value at the bottom is what?
A The last row is what is called a P value. And that is

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taken from the t-test, as described here, comparing the treatment and control mean. And the smaller the number, the higher the level of significance we put on it.
Q So, mechanically, to arrive at the P value, one would take the data, compute the means, compute the T value in accordance with exhibit 277 in front of you, and then go to either tables, or there are many online calculators that will do this for you these days also, and the tables or the calculator will give you the P value from the T number, correct?
A Essentially. Most standard statistics programs, including Minitab, will compute that. I don't know the algorithm it uses, but that's correct.
Q Back in the days before software did this for us, we probably looked it up in a table at the back of the book that had many pages in it.
A That is how I learned to do it many years ago.
Q And essentially, the P value shows you how far out you are on the bell shape curve away from the mean, correct?
A Essentially, yes. Actually, let me correct that slightly.
Q Sure. Please do.
A What it actually shows you is how far away from zero your effect of your treatment gets. And the smaller the number, the further away from no treatment effect you are.
Q Then, I've also marked as an exhibit this morning, exhibit 275, and I believe you had at least 10 or maybe 20 minutes to look at that before we got going this morning, is that correct?

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A That's correct.
Q And I will represent to you what that document is, in the interest of getting through this testimony today in less than three day's time, and in the interest of not driving Mr. Kirby too nuts with long words. I asked Dr. Chris Chase, who is a veterinarian and a professor at South Dakota State University, and a past president of the American Association of Veterinary Immunologists, to look at a copy of exhibit 254 and summarize his understanding of what those variables mean, where he could. I think there's a couple where he couldn't come up with the meaning, and I'm going to ask you about that. All right.
I also asked him to indicate his opinion on whether or not a change in each variable is indicative of an immune system specific effect, as he characterizes it, or, in other words, if there's a change in that variable, is that more likely than not indicative of a change in immune function as opposed to something else. And he has given his opinion on that.
I want to ask you those questions, too. And you may not agree with him on all of them, obviously. But this is going to permit us to do this in shorthand.
MR. THORNTON: I object. Exhibit 275 is hearsay, and is the work and opinion of an expert that was not timely identified in this litigation.
Q With that in mind, Professor Sheffield, let's go back to ACK2, which we talked about a little bit in the last deposition. But in shorthand, would you agree with Professor Chase's characterization of that variable on exhibit 275?

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A Yes, I'll say that's a - in all of these cases, obviously there's far more one could say about it, but that's a fair assessment.
Q All right. And if you agree with his comment in the immune system specific effect column that he says, yes, and in particular, it has an effect on mast cells?
A Yes, it does.
Q You told me in the first portion of your deposition that there were variables in this study that you did not expect to see an effect on. Would this be one of them or not?
A This one you might see an effect on or might not. It depends on what effect you have.
Q Well, certainly wasn't one of the variables you put in here where you did not expect to see an effect?
A I would have thought that neither result would have surprised me very much.
Q I'm contemplating asking you which of these variables you did not expect to see an effect on and which variables you should not have seen in the cells at all, because you told me that was true of some of them. And I take it there probably are relatively few of those in this study, is that correct?
A There are not many.
Q Why don't we do that first then, and I won't have to ask you that about each individual one. Okay? First of all, those that you did not expect to see an effect on. If you want to just scan the list and tell us which ones those are as we go.
A Yes. Before that, may I define what I mean by expect?

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Q Please.
A Because I think this is an important thing. Let's assume, if the voltage exposure was having some measurable effect on the immune function, would you expect to see these changed? So, I'm not - when I say expect, I'm not trying to imply that I expected voltage to either have or not have an effect. It's if it had an effect, would this be something that it might have affected? Is that clear and fair?
Q I think so. Mr. Thornton, any problems with that?
MR. THORNTON: Can you try it again?
A I'll try it. Okay. When you say expect, there's two aspects. Am I expecting that voltage has an effect, or am I expecting that if something has an effect, would it affect that measurement? So, whatever we use as our treatment, if it affected immune function, would this be something you might expect it to see changing. As opposed to a different question, which is, did I expect going in, because I try to approach things scientifically as I don't know what the answer is before, in terms of whether my treatment was having an effect. It's a different aspect. Am I expecting the treatment effect or am I expecting this measurement to reflect any possible treatment effect?
Q So, perhaps stated another way, if we find the relatively few variables where you did not expect to see an effect, that's another way of saying there's simply not - those variables aren't at play with the change in immune function. Would that be fair?
A I think that's fair.

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Q Okay. Which ones are those then?
A Okay. The ones that I would expect not too see an effect?
Q Correct. Before you even start.
A Adenyl cyclase. ATP Synthase. The CaATPase; calcium ATPase. Casein Kinase. ClevagePolyA. I guess I was unsure about CREP1 and CREP2. GAP. The column labeled Glu TransV, which is Roman numeral 5. Stands for Glucose Transporter 5.
Q I'm sorry. Let me see if I can find that one.
MR. THORNTON: Bottom of page 2.
Q Or 254, would that be the one on the far right column or has a Roman I, not V.
A Oh, yes. That is an error on exhibit 275. That should be Glu Trans IV, not V. So, the spreadsheet here is Glucose Transporter 5. On this - get my numbers right - exhibit 275, it is typed as GluTransV. That actually should be an IV in there.
Q And on 254 it says GluTransI, I think there should be a V.
A That's Glucose Transporter 4. That is the most common of the Glucose Transporter proteins in those.
Q So that would be one of these variables?
A Yes. And that's one I would not expect to change in this situation.
Q Okay. Please continue.
A Hexo Kin 1. Which stands for Hexokinase 1. I would be unsure about IGF1Rb, insulin-like growth factor 1 receptor beta form. And similarly IGR1R. I would be unsure about Leptin.

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MR. THORNTON: You say what?
A Leptin.
Q What page are you on?
A Bottom, near the last on Page 6. The various collagenases, mmp1, 3 and 9, I think would be unlikely. I wouldn't say it's impossible, but it's, I would say unlikely. PLC and PLCa, although I would expect activity of those to go up, I'm not sure I would expect the expression level to change.
I would say the same thing about the next several genes, PKACAlpha 1, PKACAlpha, PKACDelta, PKCalpha, PKG1beta, RASGAP, RhoGDI. GDK is an interesting one. Classically, we don't think of this as even being present in things other than in the illo cells.
So, at first glance, you don't expect to see very much of it, if any, in this. Although it turns out there are a few studies that have shown TEK to be involved in lymphocyte proliferation and activity. And we did see this gene in this study. We didn't see a change in it, but we did see it expressed at the above basil levels.
Q Sure. You came up with a P value of about .13, it looks like, from the spreadsheet.
A I don't remember that. I would have to look at the spreadsheet to see.
Q Sure.
A But I'll take your word for that for now.
Q Okay. So that would be a question mark, perhaps?
A That was one I - there's not that much known about it in

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lymphocytes, but it's kind of - was unexpected to me to even find any substantial amounts of it.
Q So, I guess it's neither a yes or a no, but it's interesting?
A It's interesting, and that's how I would describe that. On Page 11, Casein we wouldn't expect to see at all. That's only present in mammary tissue. The Klebella, that is actually a gene from a bacterium, (negative batgerium), Klebella pneumonia. I don't remember exactly which gene, off the top of my head. It's one of the lymphocyto (ph) genes of the bacteria. And that is so different in the lymphocyto RNA genes, if you carry out, you would expect to see a signal there. And we didn't.
Q Sure. And I'm sorry, plus I take it would be one you shouldn't even see it - -
A Shouldn't even see it. You'd be watching an empty well, or very close to it. That's what we call a specificity control. Same thing with pGEM. This is a plasmin that is present in certain bacteria, not in the geriots (ph). And GAPDH is another one that is - is very commonly used as what's called a housekeeping gene. It's something that you rarely see changing.
And then the Empty, that is his assumption here that nothing was added to that well, that's correct for that.
Q So, because you jumped around a little bit there, I would like to just summarize then. The items that you should not have seen in the cells at all, I believe would be - well, on exhibit 254, it would be on the last page, right and the end, I think, if I understood you correctly?

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A That's correct.
Q And they would include casein, Klebella, pGEM, and Empty, is that correct?
A That's correct.
Q Did I miss any? Are there any others that you shouldn't have seen at all?
A I think that's correct.
Q And then starting on, let's stick to 254 for a moment. You described PLC as being one that you did not expect to see a change, at least in gene expression, is that correct?
A That's correct.
Q And then the same comment, this is where you jumped around a little bit, so I would just like to clarify it for the record, if we may. The same would be true of PLCa, next to it, is that - -
A That's correct. PLCa is what we call an isoform. It has essentially the same activity, but it's a slightly different sequence of the gene.
Q If my notes are correct, the next one going to the right on 254 would be PKACAlpha. I'm not sure I got that right though.
A PKA - I may have skipped over one. There's a column here, says, PKAbCat. That I wouldn't expect to change either.
Q And, again, - -
A PKCAlpha, PKARIIb.
Q I think the next one in that category was RASGap, GAD, is that correct?
A Actually, I'm not sure - I don't think I would have

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expected PKG1Alpha, and PKG1Beta. And then we go to the RASGAP.
Q That, also, you did not expect a change, correct?
A I didn't expect that to change.
Q And the RhoGDI also, is that correct?
A I didn't expect to see it, no.
Q The rest of the sheet going to the right on the third page of exhibit 254, none of those fall in the category of did not expect a change, correct?
A All of them fall into the category of, you may see a change.
Q On the last page, just as the first one we covered at length, was interesting. After that, the first one I noted as you did not expect a change was PKCDelta, about halfway across the sheet. Did I miss any or did we miss any?
A There is another one in here called PKAR2Alpha. I may not have mentioned that one. But that is one I wouldn't have expected.
Q I think you did say that before, at least I wrote it down anyway.
A Is that - - there's a - - these abbreviations start looking very similar. The PKA stands for protein kinase 8. And the C and R stands for catalytic or regulatory sub-unit, and then we have 1 and 2 alpha and beta for each of those. So, it's easy to let them run together for a while. But that is one I would not expect to see changing. I may have mentioned it earlier. I think that's correct.
Q All right. And then I think that is the last one that

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falls into the category of did not expect to change until we get to casein, and we covered those last several a few moments ago.
A I think that's correct.
Q All right. I think we probably have them all then. I would then like to go through, I think we will go through the ones that you didn't name as falling in that category, and I will ask you for each of those variables, whether you would agree with Dr. Chase's description of the variable, and his conclusion regarding whether that variable is associated with an immune system specific effect.

I think the first of those that we - well, go ahead.

MR. THORNTON: I'm going to object again based upon an opinion of an expert that hasn't been identified in a timely fashion or hasn't produced a report.

Q We've talked about ACK2 already, I believe. I don't think we need to cover that again.

The next one is cFos. Do you agree with Dr. Chase's characterization of that item, and whether or not there is an immune system specific effect?

MR. THORNTON: I don't want to continue to interrupt you, but can I have a continuing objection?
Q Certainly.

MR. THORNTON: All right. Go ahead.

A I will agree with his assessment. I do not know exactly what is meant by immune system specific effect. There are two possibilities that come to my mind. I am

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assuming what he means here is that the effect it has is very specific to the immune system, it is not something you see throughout the body. That's what I assumed when I first saw that column.
Q I think that's a fair characterization.
A Okay. I want to make sure that what I'm saying lines up with what everyone is. I think the question I'm answering is the question that is intended to be answered.

There is another aspect, and cFos is a very good chain to describe this with. CFos and cJun and some of the others on here are very common genes of the body. Almost every cell in the body has cFos in it. It's normally expressed at a very low level, and certain activators of the cell change the expression level of cFos.

For example, if you take a cell and add a growth promoting agent to it, one of the very early effects is that the expression level of cFos goes very high. So, things that would affect the immune system might well affect cFos even though it is not an immune specific event, which is why I put it in the category of maybe you would see an effect on it. I would say many of the effects on cFos are very transient, it goes up and it comes back down very quickly.
Q The next one in the list is cJun. Same question for each one. Do you agree with Dr. Chase's description as set forth on exhibit 275 and his conclusion about immune system specific effect?
A I will agree with that, and I will say the same thing I

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did about cFos. It's another of these genes that is activated by a lot of different things, and present in most cells in the body.
Q For each one we come to, I'm just going to ask the same question so we don't make a lot of transcript here. The next one - -
MR. CARLSON: To the extent that Mr. Thornton objects, I'm going to join in those objections rather than making similar objections. So, both cases, okay?
MR. LAWRENCE: Understood. I don't have any concern about opinion objections.
MR. CARLSON: All right. But to the extent he makes an objection, I don't want to waste time voicing the same objection because I'm here on a different case.
MR. LAWRENCE: Understood.
MR. CARLSON: Okay.
Q The next one that was not a yes to, did not expect to see an effect, is CasKin1. At least that's the abbreviation on the spreadsheet.
A Yes. His assessment of this is correct. It's one you might see. Because this is a very general enzyme in cells, you might expect to see some increase, but not seeing an increase would also not be too surprising.
Q You can skip CasKin2 and go to CD14.
A I will agree with his assessment of CD14.
Q And he does assess that as to be something to the immune system, and you would agree with that?
A Yes, definitely. Same with CD23. Is that okay, if I just go down the list of these?

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Q Please.
A Same with CD23. Same with CD8. Same with CD3. These are all very common proteins on the immune system cells and they have major roles in the immune function.
Q Cdk1?
A Cdk1, his assessment of it is correct. Cycline dependent kinase is expressed during what we call the cell cycle. That is, cells are proliferated - or stimulated to proliferate, so it is present in most cells. So we wouldn't expect it specific to the immune system, unlike the CD genes, but if cells are being stimulated to proliferate, you might see it increased.
Q I think we can skip ClevPolyA, and go - well, we probably should cover CREB1 and CREB2, because you were unsure, shall we say, on those with respect to - -
A Yes. Correct. His description of what they do is, I will agree with. These are cyclic A and P. To put this in some context, is a very common substance we call a second Messenger. It has a wide variety of effects on activity of many enzymes and expression of many genes. And it's used as a second Messenger in many cells in the body, including some in the immune system.
CREB1 is a protein that is activated by cycline A and P. So I actually would say it probably wouldn't see the expression change a lot, but there are some cases when you do see changes in CREB1, and the same thing with KREB2, expression levels. So that's why I said I was unsure about it. It is not something that's specific to the immune system, I would say a very, very common protein in many cells in the body.

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Q Both KREB1 and 2?
A KREB1 and 2, yes.
Q Then I think the next item - the next variable is CuZnSOD.
A That's right. I will agree with his assessment of that.
Q And can you tell us what the next variable is, Desmogelin?
A Yes.
Q I think Dr. Chase couldn't make it out, so he doesn't have any restriction.
A As near as I can tell, that is a rather bad typographical error on there. I believe that that is a protein called Desmogelin.
Q Can you spell that for John?
A Yes. I expected to spell that. And I am a terrible speller, so I'm going to get it close. If I misspell it, please forgive me on that.
Q We will.
A It is spelled, D-e-s-m-o-g-e-l-i-n. I think that is correct.
Q Okay. What is that protein and what's its function?
A This is one that, at first glance, you would not expect to see at all, if you look at the classic definition of what it is and where it's found. Desmosomes, D-e-s-m-o-s-o-m-e-s, are what we call junctions. They are very common in epidemial cells and sometimes found in other cells as well.
They basically hold cells together. If you've got a sheet of cells, something has to hold them together. There are several things that do this, and

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desmosomes are among them. So we see this very commonly in epithelial cells.
I was surprised to find very much of it in these cells at all. It turns out, however, that there are some lymphocytes that do seem to express this, and it seems to be involved in their ability to invade tissues. Lymphocytes sometimes need to attach to tissues and epithelial tissues, the lining of the vascular system, and lead to vascular system. It's been studied in a few disease processes where this occurs.
Q Well, at the time - -
A At the time I was kind of surprised to even see it there.
Q So, I take it this perhaps falls into one that you didn't expect to see an effect or - -
A I would not have expected to see any effect of that. I actually would have expected to see very low levels of it.
Q I take it from that, that if there were - if it were detected and if there were a change, you wouldn't consider that likely to be immune specific, is that correct?
A That would have been my initial reaction to it.
Q As you sit here today, would that have changed?
A I would be maybe a little more qualified. There is not a lot known about it in the lymphocytes. It is not well studied, because it, as I said, it has been traditionally thought to be in epithelial tissues. That's where almost all the work on it has been done.
Q And the knowledge of the expression of lymphocytes is something that has come about since this study was

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performed, I take it?
A I think there were a couple studies before this study was performed. I may be mistaken on that, but I believe there were a couple that showed that it might be present in lymphocytes.
Q Well, I guess, obviously, in general, the human knowledge of gene expression in human or cattle cells is a subject that is just exploding over the last decade or two, is that correct?
A In the last ten years, it has exploded, yes. There's a lot we know now that was not known when this was done.
Q Would it be fair to say the same is true with respect to immunological function of humans and animals, such as cows?
A I would say that's true, yes.
Q The next item on the spreadsheet, and I'm going from left to right on exhibit 254, but I think you can do the same thing on 275. The next one is FAS. Same question.
A I will agree with his assessment here of what is FAS, as well as FASLigand. This, possibly you could see some changes in FAS. I think changes in FASLigand, would be much more likely to see if you're seeing immune system effects.
Q And, Dr. Chase in the immune specific - excuse me, immune system specific effect column, had a no for FAS and a yes for FASLigand. Go ahead. I'm sorry. Sounds like you are sort of in the same ballpark?
A I am. I am. FAS is a receptor. It's present on any cells. It induces cell death. So, you're going to see effects, very wide spread effects in distribution of FAS.

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FASLigand is the Ligand for that receptor.
Q GAP is one you didn't expect to see change. So let's proceed to GlutPerox.
A Yes. Yes. I will agree with what he says about that.
Q Again, a change in that item would be likely immune specific, correct?
A Yes. I'll agree to that.
Q And then, going to the right, the next is GluTrans - it should be Roman numeral IV?
A It should be Roman numeral IV, not V.
MR. THORNTON: It's Roman IV on 254, but it's Roman V on 275?
A Yes.
Q It's actually Roman I on 254, but - -
A 254 is sort of IV, quantity 4.
Q That's right.
MR. THORNTON: No, it's 4.
A IV, four.
MR. CARLSON: I think you might have said 4 before - you might have said 1 before, but it's actually 4.
A Whoever typed this just left the I out. It should be Glucose Transporter 4.
MR. THORNTON: Roman IV.
A Roman IV, yes.
Q Assuming that we don't know that Dr. Chase had that in mind, do you agree with his comments or is - -
A What he actually says, Glucose Transporter IV in the typed section. It is just in the column, for the abbreviation he has the V.

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MR. THORNTON: But it's Arabic 4 in the - -
A He has Arabic 4. You will see it written both ways. And I will agree with what he says about that.
Q The next item on 254, is the first one on the lefthand side of exhibit 254, is GMCSF. Same question.
A I will agree with what he says.
Q Next is HexoKinase1. Excuse me. I'm sorry. That's one you did not agree with.
A That's one I wouldn't expect to see a change.
Q And the next one then is HSP70. Same question.
A I will agree with his assessment of what it does. Because this is a very common protein to see expressed during stress situations, you might see it expressed with inducing some kind of stress here. It would not be specific to the immune system, that we can say almost universal protein.
Q The next one is IGF1Rb. You were, I'm sure whether you would have expected to see an effect on that one. So same question with respect to what Dr. Chase has summarized.
A His description I will agree with. This is the receptor for IGF1, insulin-like growth factor 1. It's very widely distributed. I was not sure if I would have seen a response or not. If I had to choose a side, I would have said less likely than likely.
Q Would the same be true of the next item, IGF1R?
A Yes.
Q You agree with Dr. Chase's characterization?
A His characterization, yes, I'll agree with that.
Q And then we have IgG1HC. Same question.

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A And I agree with what he says.
Q His response would likely be immune specific, correct?
A Yes. This is an immunoglobulin. Immunoglobulins are antibodies. So, yes, that would be a very important immune system response.
Q I sometimes have been asking you follow-up questions about the righthand column on Dr. Chase's spreadsheet. I'll just assume the general question includes that also, okay?
A Okay.
Q If you would also. Thank you. The next one is IgG2HC. Same question.
A Same answer. I agree with what he says.
Q Then, IgJ.
A And I agree with what he says about that.
Q Why did you choose that one in particular?
A The IgGJ?
Q IgJ, yes.
A We wanted to include as many of the immunoglobulins as we could. Circulating in blood, there are four major immunoglobulins. There's IgG1, IgG2, which are very similar, but slightly different. IgG immunoglobulin A, and immunoglobulin M, IgM, we didn't get a good probe for IgM, unfortunately.
Q I'm sorry, You say you did not get a good what?
A Probe. Assay for IgM. Immunoglobulin AG, or not, Immunoglobulin G is the dominant immunoglobulin circulating in blood. The most common antibody in the body, however you total up, is probably not IgG, it's actually IgA. This antibody is involved in what we call

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mucosal immunity. It's actually secreted in mucosal tissue, such as the GI tract, the lining of the lungs and the mammary duct. So, it is a very important antibody.

And this is a piece of that antibody, it's sometimes called secretory piece, that is necessary for it to be secreted into those foods.

Q So, what consequences would a decrease in IgJ following treatment have for the immune function of a cow?

A There's a couple of things that could be going on here. One is, if the animal is producing less immunoglobulin G, then that could result in a lowered mucosal immune response. As you imagine, the tissues I mentioned, the lining of your lungs is actually outside your body. It is exposed, potentially exposed to all kinds of pathogens, as is the lining of the GI tract, for instance. And this mucosal immunity plays a key role in that protection.

The limitation to this study is, we looked at what circulates in the blood, not what's in the mucosal secretions. It's also possible that, what's happening is, the cells producing the IgA are leaving the circulation, even though they are still there producing it at a different cycle. Does that make sense?

Q Well, I think it does. If a change in IgJ is found, what, if any, conclusions can be drawn about changes in the immune function of the cow?

A (No response).

Q I think you kind of answered that, but I am just asking you to expound a little more.

A Yes. I think that I probably answered it in a rather

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round about way. In this particular study, we are looking at expression of IgJ protein in cells that are circulating in the blood. That's where our cells came from. So you can make - there's two possibilities here, two major possibilities, anyway. One is that the IgJ production has actually gone down, which would impair immune function. The other is that the cells producing the IgJ gene - or IgJ unit, are not circulating. That would suggest they have been recruited to somewhere else, which would mean a change, but not necessarily an inhibition of the mucosal immunity.

Q So, to summarize, I think what you just told me is that decrease in IgJ in the blood can indicate either an impairment of immune function if its overall production has gone down, or it could simply be indicating that there's an immune response going on in the animal somewhere?

A Yes.

Q Somewhere, taking it out - -

A Yes, I think I'll agree with that.

Q Now, in that particular item, and according to the specifics or the run that summarizes the spreadsheet, the P value arising from the t-test was 8.21, if I understand the notation correctly, times ten to the minus fifth, is that correct?

A That's what this says. And I do recall that that was different. I didn't - I don't recall - didn't recall the exact number. But I do recall that IgJ and IgA were both lower in the treatment groups.

Q And to be a little more specific, 8.21 times 10 to the

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minus 5th would be .0000821, is that correct? Four zeros and 8121?

A Four zeros and 821, that's correct.

Q Four zeros to the right of the decimal point. And that is about - well, it's almost a hundred fold less than the .05 usually considered as statistically significant, correct?

A I'm not sure you can do that kind of calculation with P values, but it is much lower than your typical .05.

Q And that's just mathematically.

A Yes. It is - it is - that would be considered a significant response. You get no dispute on the significance of a P value that small.

Q It's fairly extraordinary to see a P value that low in any experiment, is it not?

A I don't know. Oftentimes in experiments, we're looking for things that haven't been discovered before, so we're often looking for things where the response is fairly subtle.

Q But it's certainly one of the larger responses that you would see?

A It's a highly significant statistical.

Q If you recall, did you or anyone also, to your knowledge, do any further statistical analysis related in particular to IgJ beyond what is summarized on the spreadsheet?

A Not to my knowledge.

Q Would that be of any of these variables on the spreadsheet, was any further statistical analysis done beyond what's on this spreadsheet, to your knowledge, by you or anyone else?

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A Not to my knowledge.

Q With respect to the results of this experiment, the one involved in Messenger RNA expressions we've been discussing, generally speaking, whose job was it to do the statistical work? Was that yours or somebody else's?

A I did the statistical analysis on this particular study.

Q And I take it, nobody else did any further analysis of any kind that you're aware of?

A Not that I'm aware of.

Q Proceeding to the next variable is IgAHC, is that correct?

A That's correct.

Q And my rudimentary understanding is, HC means heavy chain?

A Heavy chain, correct.

Q Same question that we were going at some time ago about Dr. Chase's summary on exhibit 276?

A His summary I will agree with.

Q And again, there we have a P value based on the t-test of 0.003211, correct?

A That looks correct.

Q And with respect to the immunological function of the cow, what does that tell us, if anything?

A The same thing the IgJ would. One actually would expect those to change more or less in parallel, although IgJ is involved in another antibody immunoglobulin M that it's a major part of the IgA molecule. So you might expect those to change in about the same way.

Q And again, the P value for IgAHC is less than .05, correct?

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A Correct.
Q Dr. Sheffield, Mr. Thornton discussed with you last time Bonferroni adjustments or computations and we talked about that generally a little bit, about the controversy and statistics surrounding that sort of adjustment.
Did you, in the course of your analysis of the statistics related to this study summarized on 254, consider making adjustments of that general nature, given that you studied many variables?
A I didn't do that. I do think it probably would be a reasonable thing to do. But I did not do that.
Q And this, I'm going to ask you this question and maybe one that you can't answer on the fly very well, and if so, then so be it. But given that there were quite a few variables here that you did not expect to change when the experiment was designed, how would you apply that sort of adjustments to these circumstances? Could you, either generally or specifically, to the extent you can address that?
A That is one of the problems with the Bonferroni. If you have maybe, let's just use as an example, ten measurements that you would expect to change and then ten that you don't expect to change, but maybe they will change, do you do the Bonferroni correction based on the ten or the 20? I can't answer how I would do that. I would probably call a statistician I know and ask him how to do that.
Q If I understood the answer or series of answers you gave Mr. Thornton the last time we were together, I take it your experience is that if you call ten statisticians,

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you might get ten different answers, is that about right?
A I don't know if I would go that far, but you will see discrepancies from time to time about things like, what is the best way to do adjustments for something like this.
Q And given that there were, I think it's a little less, but approximately a hundred variables studied in this experiment, and let's assume it's a hundred, that's not quite right, but it's close, okay? If one were to do a very simple Bonferroni adjustment based on 100 variables, the procedure as I understand it is to reduce your P value for significance by dividing the number of variables into .05, is that correct?
A It's something like that. I don't know if that's exactly right. I would have to look up the Bonferroni correction. But it's along those lines anyway.
Q And with respect to a number like the one for IgJ, which is reported on the spreadsheet as 8.21 times 10 to the minus 5th, if you make that simplistic adjustment, based on 100, you would compare that to a P value for significance under these assumptions of .05 divided by 100, correct?
A If that is correct, that's what you would - let me - say that again.
Q Sure. Could you read it back, John? I think I said it right, and I don't think I can say it better. So, I'll have him read it back.

(The last question was read aloud by the court reporter).

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A I believe so.
Q And .05 divided by 100 would be, I believe, 5 times 10 to the minus 4, is that correct?
A If I can do multiplication in my head, that's right.
Q Well, .05 is 5 times 10 to the minus 2, correct?
A Yes. 10 to the minus 2. That's correct.
Q So, even doing what - well, what one could call a full Bonferroni adjustment for all approximately 100 variables, the change in IgJ still appears as statistically significant after a fine Bonferroni in that manner, if one were to do that, is that correct?
A That seems right at the moment, yes.
Q I tell you what, we've been going for an hour and a half. I told Mr. Bird I'd call him and see if we can include him at about this time. Shall we take a short morning break? Perhaps you could use a break, doctor?
A Sure.

(At this time a recess was taken - 10:37 to 10:54.

Q Dr. Sheffield, we'll get back to the spreadsheet in just a moment, but Mr. Bird on the phone over the break reminded me to ask you, I believe that my office sent you a copy of the transcript of your first deposition in this matter after it was taken. Did you receive that?
A I don't recall. I would have to look to see if I did. I might well have.
Q I take it you haven't read the transcript then?
A I think I would remember if I had actually read it.

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Q Sure. Well, even without having read it, can you recall anything today that you said the first time around that, upon reflection, you would like to correct or change?
MR. THORNTON: I object. That's way too broad.
Q It is broad.
A I would want to read it before giving a definitive answer to that. But off the top of my head, I'm not - you're asking me to recall something that was over a month ago, and it might be something - I'd have to read the transcript.
Q You are correct on all of those accounts, but if there's anything that came to mind, I just wanted to know about it. Thank you.
Going back to the spreadsheet, exhibit 254, and Dr. Chase's summary, exhibit 275, the next item on the list is IL1a, where I think it's frequently referred to as IL1Alpha, is that correct?
A Alpha is what it's usually called.
Q And it's probably true of all the small subscript A's throughout the spreadsheet, is that correct?
A I think that's - at least most of them, yes.
Q And do you agree with Dr. Chase's summary in exhibit 276 (sic) about the function of IL1a, or alpha?
MR. THORNTON: 275?
Q 275. Excuse me. You're right.
A Yes.
Q And the next item is IL1b, or beta, correct?
A Correct.
Q I think you discussed with counsel the distinction

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between them, which is minimal, is that correct?
A They are different, but there's - they're very, very similar.
Q And in the case of IL1alpha and beta, the P values, as calculated and indicated on the spreadsheet, are, in the case of alpha, 8.74 times 10 to the minus 6; in the case of beta, 2.55 times 10 to the minus 6, is that correct?
A That is what I'm reading here, yes.
Q And which are even smaller than the IgJ P value of 8.21 times 10 to the minus 5th, correct?
A That is correct, yes.
Q So, if one were to perform a very simplistic Bonferroni adjustment to the P value, as we discussed before, these would still be statistically significant if P equals 5 times 10 to the minus 4, correct?
A If that correction is the way it's done, that would be correct, yes.
Q I'm not implying that one way or the other. But if one were to do it that way, that would still be true?
A Yes. Correct.
Q All right. Then, the next item is IL1. Could you tell me how that word is pronounced?
A It's an abbreviation for antagonist, or inhibitor.
Q But pronounced - -
A Pronounced antagonist.
Q Do you agree with Dr. Chase's summary relating to it?
A Yes.
Q The next item is IL2, is that correct?
A Correct.
Q And do you agree with Dr. Chase's summary with respect to

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it?
A Yes.
Q The P value there is also - well, it's in the same general ballpark as IL1alpha and IL1beta, correct?
A Looks like these are a little out of register, so I have to make sure I'm looking at the right column. That's correct.
Q Specifically, it's 4.98 times 10 to the minus 6, correct?
A That's correct. Yes.
Q The next item, could you tell us what it is and whether you - just how you say the full name, in other words, whether you agree with Dr. Chase?
A Yes, this is the IL2 receptor. And that is what he thought it was, and what he says about it is correct.
Q Moving on to the right, next is IL3. Same question.
A And I agree.
Q Next item is IL4. Same question.
A And I agree with what he says of that.
Q Next is IL6. Same question.
A And I agree.
Q Next item is IL8. Same question.
A Looks like IL8, yes.
Q Oh, I'm sorry, did I say IL6? I'm sorry.
A No. No. I just said, I was meaning to imply that that looks like an accurate description of IL8.
Q Okay. Thank you.
MR. THORNTON: You didn't say IL8.
A I'm sorry.
Q The next item is IL10. Same question.
A And I agree with that.

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Q If I understand correctly, IL1 is a pro-inflammatory cytokine, and IL10 is an anti-inflammatory cytokine. Is that correct?
A That is the sort of the general one word description of them. Like a lot of these, they have many functions, but that's a fair summary.
Q And by the measurements you made in the experiment, the fold for IL1 alpha and beta were around 1.65 for one, and 1.78 for the other, correct?
A Correct.
Q In other words, the serum levels of those cytokines went up in the treatment counts, correct?
A That's correct. Yes.
Q And in the case of IL10, the serum levels halved, essentially, is that correct?
A That's about right. Yes. IL10 was lower.
Q And I think you discussed with Mr. Thornton the last time that the immune responses seen in this study were smaller than you would typically see in an acute disease outbreak of some form in a cow?
A Yes, you would see if an animal has an acute infection, you would see much bigger changes in IL1, for instance.
Q And perhaps in some of the others?
A And some of the others as well, yes.
Q By the way, I don't think that TNFalpha was on your list here, is that correct?
A For some reason, I thought it was.
Q Maybe I missed it.
A Yes, it is.
Q Okay. Where is it?

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A It's on the spreadsheet on page 4.
MR. THORNTON: Chase's deal, it's - -
A About a third of the way in. And - -
MR. THORNTON: Page 11 on 275.
Q Okay. Very good.
A Yes, it is - it is on this.
Q Then we will get to that. I'd forgotten. I'm sorry. Is there a concept in biology called cytokine induced symptoms behavior?
A Probably. I'm not all that familiar with that specific term.
MR. THORNTON: Objection. Foundation.
Q And I take it you either haven't reviewed, or if you have, you don't recall much of the specifics of any literature about that subject?
A I don't recall reviewing any literature on that - anything that was called that.
MR. CARLSON: Let me just clarify. Are you referring to the cytokine storm as well? Is that the same thing, called it a cytokine storm?
Q I guess I wouldn't choose to say they mean the same thing or not. I don't know.
A I am familiar with the cytokine storm. But I haven't kept up with it very much, but I do know something of that. And I was assuming you meant something different with this.
Q What is a cytokine storm, in your understanding?
A During inflammation, you get a - frequently get a massive release of a variety of pro-inflammatory cytokines. The cytokines are actually important in inducing responses to

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an infection, protective responses, but they can also damage normal tissue. And in many cases, it's the production of massive amounts of these inflammatory cytokines, they cause some of the deleterious effects during infection and inflammatory responses.
Q All right. And if things are going well with the body, and with this massive cytokine release, one would hope that the inflammation serves its purpose and passes relatively quickly, is that correct?
A That's what one would hope.
Q Doesn't always happen that way though?
A No, it doesn't.
Q Are you familiar with any studies in any type of organism of the impact, if any, of a long-term elevation of cytokines at the levels of approximately two-fold or a little less than we're seeing in this study? In other words, I'm asking if there's been anything studied about - -
A There has been.
Q - - the consequences of that is a chronic process rather than an acute process?
A There have been.
Q Can you describe what you know about that, in general, please?
A This is not an area I have reviewed recently.
MR. THORNTON: Objection. Speculation. No foundation.
Q Please continue.
A So, I was familiar with this some years ago. I haven't looked at the most recent literature, except in

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preparation for teaching, which is a little bit lower level than what one would do if I were going to be doing research on this area.
Q Do you recall who - -I'm sorry. Go ahead.
A What I was going to refer to is, two points. Certain diseases, cardiovascular diseases, are felt to be caused by, or related to - maybe caused is not the right word - related to low level chronic inflammations. Some of these, not the interleukins, the TMFAlpha that you alluded to earlier, for example, is produced in adipose tissue. And this is actually where my familiarity years ago with this came from, that it's thought that some of the adverse effects of obesity on things like cardio-vascular health might be mediated by this long-term sub-acute inflammation, in other words, TMFI is usually implicated in that, in my understanding of that, rather than the interleukins, I don't know about the IL1 and its implication in that.
Q All right. Do you recall who any of the folks who you would consider to be the leading researchers are or were in that area?
A Not off the top of my head. If I were going to look at that again, it would be fairly easy to find it on health.ed. But I don't recall the names, off the top of my head.
Q What would you search for on health-ed? What kind of key words?
A Probably - -
MR. THORNTON: Objection. Speculation.
A The word that's often used is the adipokine.

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Q Can you spell that, please.
A A-d-i-p-o-k-i-n-e. Or adipokine hypothesis, which is this idea that adipose tissue produces hormones, and that is considered a subset of that hypothesis. So, you could look for words like TNFAlpha and adipose tissue.
Q Now back to IL10 for a moment. I think you've answered my basic question about that. The P value that is indicated in your summary is 2.93 times 10 to the minus 5th, correct?
A Correct.
Q And again, just grossly, if you were to make that very simplistic Bonferroni adjustment that we went through earlier, not saying it's right or wrong, that number is still less than 5 times 10 to the minus 4, and then this item would be - would still be considered statistically significant, even if one made such a Bonferroni adjust-ment, correct?
A That looks right, yes.
Q Next item on the spreadsheet is IL12Alpha, I believe, is that correct?
A Correct.
Q Put the same basic question relating to 275, Dr. Chase's summary.
A I'll agree with that.
Q Next item is IL13. Same question.
A I will agree with that.
Q Next one is IL15. Same question.
A Yes, I'll agree with that.
Q Then, IL16. Same question.
A And I'll agree with that.

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Q Next item is labeled INTb1. What's the full name of that item, and then do you agree with Dr. Chase or not?
A The full name is interferon, i-n-t-e-r-f-e-r-o-n, beta 1. And I'll agree with what is said there.
Q The next item is Int2? Could you give us the full name and whether you agree with the summary on 275?
A That is interferon 2, and I will agree with that.
Q Next item, which is the lefthand column on page 3 of exhibit 254, could you pronounce the full name for us and answer the same basic question.
A The full name is lactose peroxidase. Do you want me to spell that?
THE REPORTER: I can look it up.
Q It's in the exhibit, so I think we're fine.
A And let me find it again.
MR. THORNTON: On Page 6.
A Yes. I'm just looking to make sure. He's asking me if I agree, and I wanted to make sure I know what I'm agreeing to.
Q Please.
A Yes, I'll agree with that.
Q Next item is Leptin. Same question.
A Yes, I'll agree with that.
Q Next item is mmp1, and is spelled out in full in exhibit 275, so the same basic question.
A Yes, I'll agree with that.
Q Next item is mmp3. Same question.
A I'll agree with that one.
Q And we have mmp9. Same question.
A And I'll agree with that.

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Q Next item is PLC. Same question.
A I'll agree with that.
Q The next item is PLCa, and in Dr. Chase's summary, he says, "Not sure what this is." Can you expound on that one in some length, please?
A PLCa stands for phospholipase C alpha. And it's one of many forms of phospholipase C, which is an enzyme that is involved in a wide variety of cell signaling pathways, in many cells in the body, not specific to the immune system.
Q And so in terms of whether a change would probably relate to immunological function, that would be a no - it could, but not likely?
A It could, but it's the sort of enzyme that you might expect activity to change more than expression.
Q Next item is PGDSynth, S-y-n-t-h, or Prostaglandin D synthase, is that correct?
A That's correct.
Q Do you agree with Dr. Chase's summary there?
A Yes.
Q Next item is labeled PGSH2, and Dr. Chase asks us to double check with you as to just what this is. Could you tell us, please?
A Okay. It stands for Prostaglandin synthase H2. It's an enzyme involved in Prostaglandin synthase, much like the PGD synthase is. These enzymes go by a wide variety of names. An alternative name for this one is Cyclooxgenase, or COX-2.
Q Is that the item we had - is that one of those items that one of the drugs for humans that was inhibiting that was

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pulled off the market a while back?
A Exactly.
Q All right. Thank you. At least I got that right. And is this an item, a change in which you would expect ordinarily to relate to immunological function?
A Yes. It's involved in inflammation.
Q And it's pretty specific to inflammation?
A There are other places and times when it does occur, but it is not exclusively, but it is very often associated with inflammation.
Q Next item is pim1, and the same basic question about Dr. Chase's comments on 275.
A I would agree with those assessments.
MR. THORNTON: Excuse me. You didn't ask him specifically whether he agreed or disagreed with Chase's assumptions on PGSH2.
A He didn't make any assumptions on PGSH2 because he wasn't sure what it was.
MR. THORNTON: Well, but there's a description here.
A That is actually referring to the PGD, which is also called COX-1, is the way I interpreted that. I may be misinterpreting it.
MR. THORNTON: I don't think so. Well. It's unclear.
Q Well, Dr. Chase makes no summary about COX-2, which is what the PGSH2 is, or another name for it.
A I think I want to make sure this is clarified here.
Q Please.
A Or should. COX-1 is the same thing as PGDSynthase. So,

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what is written beside the PGSH2, referring to COX-1, I will agree with, if we assume that that's referring to the PGDSynthase, or COX-1. The PGSH2 is also called COX-2. Its roles are fairly similar to COX-1, it's involved in synthases across the glandins, and it has an immune specific effect in inflammation. It does have some other roles, but its main role is mediating inflammation.
Q That's why I didn't ask, because I don't think it's clear. There is a comment there.
MR. THORNTON: The comment is sort of screwed up. Usually, when you've got the wikipedia reference, that's the end of the comment. But the comment for PCGDSynth appears to go down all the way to pim1 1, and there's two wikipedia references.
A Without having the Web site here, I don't know what that wikipedia reference says.
Q Sure. And that's why I didn't ask the question. It is not clear what that is referred to, and Dr. Chase, obviously, was having trouble identifying what PGSH2 was, which is why he asked to double-check it with Dr. Sheffield. Okay. Yeah, I think you answered the pim1.
The next item is PKAbCAT.
A Okay. These next few that start with PKA are various sub-units and forms of an enzyme called protein kinase A, and his description I will agree with on this.
Q I think on these, on all of these except maybe PKCalpha, you indicated you really didn't expect to see a change in any of those, is that right?
A I wouldn't have expected changes.

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Q Would that include PKCalpha?
A That is a rather different enzyme. And that is another one that one doesn't always see changes in expression level, but often sees changes in activity level.
Q And by expression level, you mean the expression that would show up in MRNA?
A Correct. Correct.
Q Okay. After the PKs, the various PKs, the next item is PRASG. Again, that's one you didn't expect a change, correct?
A Correct.
Q The same with the next one, RhoGDI, correct?
A Yes.
Q Then we have a series of five or six STATS, is that correct?
A That's correct.
Q And, generally speaking, maybe it's fair to ask these together, maybe we need to take them one at a time, but I suspect you're going to agree with Dr. Chase that changes in all of those would tend to be associated with immune function, is that correct?
A Let me just look through the list to make absolutely sure.
Q Sure.
A Yes.
Q Proceeding on to page 4 of exhibit 254, and which is near top of page 10 on exhibit 275, the next item is TEK, and I think you described that one as interesting before.
A Yes.
Q And I guess I'll ask you to respond to the standard

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question, do you agree with Dr. Chase's summary?
A What he says is correct. I would add that the statement is expressed almost exclusively, and that is correct. But he almost doesn't indicate that there are a few other places it can get expressed.
Q Such as?
A There are some, as I discussed earlier, some, a few studies that suggest it might be expressed in leukocytes that have roles in them.
Q Any particular type of leukocytes?
A I'm not recalling that off the top of my head for that one. I would have to look that up.
Q And do you agree with Dr. Chase, that changes in PK would not be particularly immune specific?
A No, they wouldn't.
Q Next item is TGFb1. Same question.
A I agree with what he says about that.
Q Next is TGFBP as it's shown on my spreadsheet, which is different on 275.
MR. CARLSON: Looks like EB on the spreadsheet.
Q I guess, let me ask you more broadly then, Dr. Sheffield. Can you tell us what that is?
A I believe that refers to a transforming growth factor binding protein, not P2. Now, what he says, it is the same, TGFB2, that's the same function as B1, that is correct. But there is a trans - for many factors like this, there are proteins that are called binding proteins that are secreted, bind to the growth factor, and modified its function. Sometimes they stimulate its

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function, sometimes they inhibit it.
These are best known for insulin-like growth factors, but they exist for other things as well. And I am almost positive that this is referring to transforming growth factor binding protein, not the B2 protein.
Q Would a change in that item tend to be indicative of - -
A That's a possibility.
Q Let me finish the question. Would such a change tend to reflect an immune system specific effect?
A It's not specific. These things would be present in a lot of places in the body, just like TGFb1 would be. But it is important in the immune system. It's one of these things that covers a lot of ground. TGFTransforming growth factor proteins are involved in a huge variety of processes. They're involved in the immune system, but they're involved in things like limb pattern formation in embryonic development, to give you an example of how involved their actions are.
Q The next item is - well, let me ask you, tell me what the spreadsheet says. It appears to be Tie1 or maybe Roman 1 or I, I'm not sure what it says, frankly.
A Yes. This is Tie 1 and 2.
Q All right. Dr. Chase does comment on those. Standard question.
A Let me look at his comment a little more carefully.
MR. THORNTON: He's got a different - -
A He has it listed as TiaI. And I want to make sure if he is referring to the same thing that I am here. I'm not sure he is referring to the same thing that I am

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thinking.
Q Okay. Could you please tell me what your item is on exhibit 254, please?
A What I believe those are, are two very closely related proteins that are involved in cell death processes. I'm not sure if they are the same as Tia that he is stating here, but they are involved in what is called program cell death.
Q And would a change in either of those tend to be something that would be immune system specific effect?
A It would not be specific to the immune system. These are fairly - they're more often associated with the immune system's, if I recall, distribution, but they are found in other places as well. But you would expect to - a change in them, in the immune system cells, might be seen in alterations of the immune function. They could be seen in other things as well.
Q The next item is TIMP3. Same question. Standard question, I should say. Bottom of page 10 on 275.
A Yes, I'll agree with that.
Q Then we have TNFalpha or tumor necrosis factor alpha. Standard question.
A Yes, and I'll agree with his assessment there.
Q Next item is TNFRec, tumor necrosis factor receptor, is that correct?
A That's correct.
Q Same question.
A Yes, I'll agree with that.
Q Next item is TPA. Same question.
A I'll agree with that.

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Q Next item is Urokinase. Same question.
A I'll agree with that.
Q Then we have IL1Rec or interleukin 1 receptor. Same question.
A I'll agree with that.
Q Then we have a series of variables that start with PK, I guess, I believe it's the next six. I'll ask you the same question for each. If you prefer to deal with them collectively, that's fine.
A I would make one correction, and that is, the one that says PKARI, Roman numeral I, alpha, that one is not an isomer of PKC, it's an isomer to PKA.
Q I see. With respect to those six variables, do you agree that those that Dr. Chase has in the column on immune system specific effect?
A Yes. These are the sorts of enzymes you find in most cells of the body in varying amounts.
Q Then we have INFalpha. What is the full name of that item?
A Interferon alpha.
Q And same question, the standard question.
A I'll agree with his statement.
Q Then we have those last four items, which I think we have already covered in detail when we got started?
A Correct.
Q I see, for example, with Casein, Dr. Chase says, "Good control," like that of some of those others. Okay.
MR. THORNTON: Before you leave that, could you explain what Empty is?
A Yes. We didn't put anything there. There's no DNA probe

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at all. So you'd expect - that's not background. You'd expect it to be close to zero. That's sort of a 'no signal at all' control.
Q We still get a number, but they're much, much smaller?
A Yeah. You will always - you will always get a - like if you're trying to detect for radiation, there's always a background radiation that you have to correct for, and that's what that means.
Q All it means is, there's something in that well generating the light that the lab machine is detecting?
A Right. Right. It's another of those controls for how specific things are.
Q Oh, before I forget this one, you didn't review the transcript, but there's, I think two - approximately two occasions in the transcript of your first deposition, you were describing that machine detecting the light signals, and the area of the intensity and so forth, where Mr. Kirby got the word protons, and I think you meant to say or said - I think you said photons?
A Photon is what it should have been.
Q Resisting the urge to ask you whether light is a wave or a particle.
A Yes.
Q We will pass on that one. Oh, by the way, Dr. Chase got his PhD in veterinary science, veterinary medicine here at Madison in 1990. Do you recall knowing him at all?
A I do not recall knowing him. I may know the person he studied with. But I don't recall that name.
Q You never had any professional interaction with him, I take it?

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A I don't recall any. I may have met him at a seminar or something at some point, but I don't remember.
Q Sure. For example, I happened to attend a seminar given by him most of the day in Wisconsin Veterinary Medical Association in the fall of 2009 over - it was over farming methods and vaccinating dairy cows and that sort of thing. He did that commonly. Have you ever heard of one of those?
A I know that they're there, but I have never been to one.
Q In with the materials that were provided by the University back in 2008, and following in late 2007, there are a number of memos, either from you or to you, usually between yourself and Steven LeMire, the gentleman who is doing the statistics on that first study, about various alternative information and alternative statistical tests, and I'll get some of them out and talk about them here in a moment. But do you recall anything about your asking for alternative statistical analyses of the data from the first study, the one that was submitted to the Minnesota State Government, after the study was published in the year 1999?
A That was a long time ago. I don't recall anything specific. I think we did have some discussions about how to analyze the data. And I don't recall the time-frame of any of those, whether that was before or after various events occurred.
Q Well, I would like to show you some of those documents and talk to you about them a bit. I understand it was a long time ago, and they're not - fortunately, they're not lengthy documents, though some of them have tables and

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numbers and that kind of thing which are in a lot of information. So, in doing so, I will certainly try to give you plenty of time to look at them. In fact, if you would like, I could stack them up and we can take a lunch break and you can take a look at them while we're on break and take them up afterwards, and I'll provide those to counsel, too. Does that sound reasonable?
A (No response).
Q It might go a little quicker than if I simply hand them to you and give you the opportunity to read them and spawn through them. But if you've got - -
A It's up to you. It depends how long they are and how many there are. Might be able to respond quickly or might need to look at them.
Q Well, let's give the first one a try.
A Why don't we try that.
Q Sure. About 8 or 9 of them, I think, total. Actually, I have one here and the others are back here. I think I'll try to do them in chronological order, that might be easier. The first one doesn't involve you, I don't think, but start with that.
Exhibit 278 is a series of documents that were produced by Professor Reinemann in response to the subpoena, and appears to be a memo from Steven LeMire to Doug Reinemann, dated June 28, '99, just a couple days before the first paper was sent to the Minnesota State Government.
Do you recall ever seeing this document before, particularly the cover sheet?
A I don't recall seeing it. I recall seeing the informa-

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tion that's in it, but I don't recall seeing the particular document.
Q On it are - there is a - obviously a portion of it is typewritten and then there's a whole bunch of hand written notations. Do you know whose handwriting is on that document?
A No, I do not.
Q And - -
A It does not appear to be mine, I can tell you that, but I have no idea whose it is.
Q As an example, Table 1 on the cover sheet, for the variable interleukin 1, micrograms per milliliter. We have a P value shown for serum of 0.071, the typewritten number, that is, over in the righthand column. Does that appear to be correct?
A That's what I see here, yes.
Q And up at the top of Table 1, it says, "Table 1. Blood file names and different responses in natural logs. Sample size is 12 per group." Did I read that correctly?
A Yes.
Q And we were talking about a blocked experimental design with multiple replicas the last time we got together?
A Yes.
Q Does it appear from this table that Mr. LeMire is analyzing the treatments and controls as the treatment group of 12 and control group of 12?
A I can't tell for certain from - just from this table. I don't see any indication on here of any blocked correction.
Q I guess we have to run the numbers to really know that,

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is that correct?
A Or get a printout of where those numbers came from, the program that was used to do it.
Q It appears that the following pages, which were all pretty much sequential in Professor Reinemann's materials relate to the SAS, S-A-S, program that was used for analysis. Is it possible that by examining those pages we could tell?
A Assuming those relate to this table, it appears that you might be able to.
Q Could you take a look and tell us whether you can tell whether a block design is accounted for in this analysis or not?
A I do not see anything that would suggest it is. It looks like it was not.
Q And, for example, I'll refer to Bates number pages down in the lower righthand corner where it says Reinemann with a number, numbers in the 2600 plus range. For example, on page 2656, the last variable on that page is iL1 serum, apparently, is that correct?
A That's what that would mean.
Q And it appears that the input to the SAS program is 12 controls and 12 treatments, correct?
A Correct.
Q And if this were a blocker, a replica analysis we would see something a little different, is that correct?
A Not necessarily. Let me put where this is, what this is. This is simply a calculation - I believe this is simply a calculation of the means, not the actual analysis, not an actual analysis of variance. If you've done an analysis

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of variance, it would look different than that.
So, let me look at the means. I believe that is the printout. Yes, that is a simple P test. That does not have blocks in it and you would see something different if it did.
Q So, I take it, there's no indication in these documents that a blocked analysis was performed?
A That's correct.
Q And you told us the last time, that you were not involved at all in the statistical analysis of the data from this first study, correct?
A Other than reviewing what was done and sometimes discussing what might be done next or in addition with Steve, I didn't do any of the actual computations.
Q The number, the typewritten number for iL1 serum on exhibit 278 of .071, is, I believe, the same number that appears for the P value for iL1 serum in the final publication, which we marked as exhibit 250 the last time. We can check that. Exhibit 250, page 9, Table 2, shows a P value for iL1 serum of 0.071, the same number that is on exhibit 278, correct?
A Yes.
Q Do you have any understanding of why an analysis of the experiment accounting for the three different blocks that was performed and was not done in this data?
A If you're referring to this table only (278), I don't. I don't know why Steve decided to run the initial analysis like this.
Q I think you told us the last time that it would be appropriate to do the block analysis where the study was

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done in block, correct?
A There Actually - it could be. There's another caveat in there, but I'm sure we will get to that in a bit.
Q All right. Well, this is as good a time as any. What is that caveat?
A Well, if I recall correctly, one of the analyses that were done was to use the initial values before treatment was applied as a covariant, and do an analysis of co-variants. That controls for a lot of the block to block variability, although possibly not all of it. And so it still might be appropriate to run a block, include a block of that in that analysis.
Q Do you know if Steve LeMire did, in fact, do a covariant analysis on this data?
A I believe so.
Q Again, not something you've ever run the numbers on?
A I haven't.
Q Do you have any understanding one way or the other as to whether the numbers published in the report that was sent to Minnesota, which is exhibit 250 here, utilized a covariant analysis? Feel free to look at this document, if it helps.
A That may help.
Q Sure.
A It appears it was not.
Q And you are referring in particular to the paragraph labeled Immune Function Responses at the bottom of page 8, just before Table 2?
A Yes. What it appears was done here in generating this table was to take the difference from base line level -

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yes, this is a - as I understand this analysis, and since I didn't do it, I could be mistaken, is that what Steve did was take the control group and look at it before and after change, and then do the same thing with the treatment group; and then look at the difference between those changes and do a t-test on the difference between those changes.
Q And apparently the t-test was run on all 12 control cows and all 12 treatment cows as one control group and one treatment group, correct?
A Correct.
Q In these circumstances, would that be the most appropriate statistical test or would the block analysis be more appropriate?
A I am not a statistician. In my opinion, as a non-statistician, this is not the best way to analyze these data, and some kind of analysis or variance which might include block effects would be more appropriate.
Q And we discussed that a little bit the last time around, in general. And I take it, that's a subject you've never had occasion to discuss with Steve LeMire or Douglas Reinemann, is that correct, in relation to this study?
A I don't recall those in any discussion like that.
Q It's about the lunch hour. I'm going to mark another series of 8 or 9 memos, and you're welcome to have copies of them before lunch, including you, Professor Sheffield. I'll ask you about those the first thing after lunch, okay?
A Okay.
Q Why don't we go off the record and we'll mark those

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items.

(At this time the noon recess was taken - 11:59 - 1:03).

Q Just briefly, Dr. Sheffield, before I get back to those memos. In the first study, the one, the output of which went to Minnesota State Government, did the workers in the barn, who were with the cows daily, keep notes in that study, do you recall?
A I don't recall any.
Q There are what appear to be some barn notes in the records.
A That could be. I just don't recall what they were or seeing them.
Q We will get back to that, perhaps. The memos in front of us, we have covered, I believe , 278, which is the analysis with the handwriting. Exhibit 279, and I believe - excuse me. Exhibit 279 has a cover sheet upon which it says Analysis of Part III requested by Dr. Lewis Sheffield, 10/6 of '99. Did you have some opportunity to look at this over the lunch hour?
A Yes, I have.
Q First of all, do you think that's accurate that you were requesting some further analysis?
A I probably did. I don't recall how detailed I would have requested it.
Q Do you recall why you were requesting it?
A Yes. Basically, I had some questions about whether the way it would have been analyzed in this initial 278, whether that was the best way of analyzing the data.

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Q And there are a number of memos relating to further analysis of the data that followed that, which we have marked as exhibits. Have you had a chance to look at all of those exhibits?
A I glanced at them, and we can go through those.
Q Okay. On the cover sheet of 279, in someone's handwriting, it appears to be written, "See iga serum." Is that your handwriting or somebody else's?
A I don't think it's mine. It doesn't look like mine. But someone did write that on there.
Q Okay. All right. And tell me what it was you were requesting upon this occasion as reflected in 279, and what the conclusions are from the re-analysis, if any?
A Well, this analysis, these two methods of analyzing the data, one is called general linear models, which is a fairly standard analysis. The problem with the general linear models analysis is, it assumes that your observations are all independent of each other. Or how one should analyze data that, where that isn't true, for example, when you take one animal and measure it sequentially as a very long history in statistics, it's not all that simple to figure out how to correctly analyze that, because the methods that ignore the potential for correlated error terms, if the errors are correlated can give erroneous results.
I don't recall the history of this, but the SAS, Statistical Analysis System, released sometime in the 1990s, and I don't recall when, a method called Proc, P-r-o-c, mixed. I, at the time, was not very familiar with that, I'm still not very familiar with the ins and

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outs of how Proc Mix works.
MR. THORNTON: Objection. Foundation.
Q Go ahead, doctor.
A But the procedure, as I understand it, accounts for the correlation that could exist from one animal to another, - or not from one animal to another, but one sample to another within the same animal. How it does that, I don't know.
So this was analyzed using Proc Mixed, and Proc GLM, taking into account the effects that individual cows have on the results.
Q Are the results of that analysis tabulated anywhere within the document?
A Yes. In fact, most of the document is the results of that. For example, page 15 begins with The Models, is the title on it. The first variable analyzed here is called CHEM, which stands for chemiluminescence. The first analysis here is the mixed procedure, mixed model for chemiluminescence. The results of this of the SAS output is shown here.
Q Now on page 16?
A Page 15, and continuing to page 16. At the top of page 16 you'll see a table that says, Tests of Fixed Effects. The mixed, in Proc Mixed, refers to what we call a mixed model. In statistics, we think of - often think of things as either being fixed effects or random effects. Fixed effects are things you decided on, is the easiest way to describe it, like which treatment you apply. We decided the treatments.
Random are things that are selected at

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random, like the cows were selected at random, they were randomly assigned to the treatment, so the cows would be a random effect.
We usually, in these types of experiments, are interested in the fixed effects. The random effects are things you want to control for. And controlling for them can reduce error variance, but we're not really interested in comparing this cow with this cow, we are interested in comparing the two treatments.
In this table you will see - -
MR. THORNTON: You're at the top of page 16?
A The top of page 16. You'll see source, degrees of freedom, type 2, which stands for type 2 sums of squares, and then the statistical test, which is Pr greater than F, that is the P value for treatment effect, day effect, and treatment by date interaction.
Q So, in this case the treatment effect by this computation came in with a P value of 0.07?
A That's correct. Well, 0.08 - .7958.
Q Oh.
MR. THORNTON: It's zero point, not zero point zero.
A Correct. Zero point seven-nine-five-eight.
Q Okay. I'm sorry. I misunderstood. I was getting some post-it notes when you were giving part of that explanation. I apologize. Let's just run through the columns again to make sure I've got it straight. I apologize.
The F means what?

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A I'm not sure.
Q DDF, degrees of freedom?
A NDF would be degrees of freedom. Yes, because there's one treatment, 5 day effects, 5 treatment by day. DDF, I'm not sure what that stands for.
Q Okay. The third column is what?
A That is the sums of squares, from an analysis of errors. SAS uses something called a type 3 sums of squares. And the final column is the P value.
Q Do you see a low P value in the day row?
A Day row.
Q What's the significance, if any, of that?
A It fluctuates from day-to-day, and I don't know what the significance of that would be.
Q Then I take it there are similar analyses for the other variables?
A Correct. It follows with the glm model for chemiluminescence, which is a different analysis. And as we follow through this, on 15, we have the beginning of the analysis of variance table on 16. And then on 17, that continues.

About the middle of page 17, it appears to be expecting mean squares. Least square means for the two treatment variables. I'm looking through here to find a least squares means by date for these. Here's what I was looking for. At the top of page 17, we have the analysis, the variance table giving treatment, cow within treatment, effect, the effect of the individual cows, the day effect, and a treatment by day interaction. And again, in the final column you have the P values for

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these.
Q And again, the P value for treatment in this case is 0.7958, which I think is exactly the same as was noted in the prior table we spoke of a moment ago?
A Correct. If you notice, the treatment effect here, - -

MR. THORNTON: Where are you. Top of page 17?

A I'm on the top of page 17. We have a treatment effect, and there's a table here in which it shows that P value as .5066. Now this is something about statistics programs. When you do an analysis of variance, everything you include in the model is a model effects. Everything that's left over is assumed to be your error.

That's not always correct. That's where this expected means squares table comes in. What we really want to use is the error term, not the residuals what's left over, but the cow within treatment effect.
Q And how is that reflected in these tables?
A That is what this last line, which is a test of hypotheses using the type 3 means square in this, means square, for cow (treatment) as an error term.
Q So that is the row that has in the last column P value you're looking for?
A Correct. That was included as a line within the commands given to the SAS Program to use that as the error term for testing treatment.
Q Which happens to be exactly the same P number - -
A It should be similar. It should have been similar.
Q As the four digits that appears, it was the same as under the mix model that you discussed on 16?

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A Yes.
Q Then, - -
A And then it repeats for all the other variance.
Q Is there any one of these models, either the mixed model or glm model, that you consider most appropriate for these circumstances as set forth in these calculations?
A Most people today, I believe, would use the Proc Mixed procedure.
Q And many of these results as expressed in the P value are different than what was reported in the original publication provided to Minnesota?
A Well, it is a different way of computing the P values, and that's not too surprising, but there were a couple that were - -
Q Different by quite a bit?
A - - were different by quite a bit. And the IgA in particular, is one.
Q And it looks as if interleukin 1 is serum?
A Interleukin 1, I think 2, also. Let's see.
Q Let's go through those results - -
A We will have to go through those.
Q - - a little bit?
A I think IgA was the first one of those where it looked very different.
Q And that's, I believe those are the results that are found on about pages 36 and 37?
A 36, yes, page 36 and 37.
Q Under the mixed model, for IgA, what's the P value?
A Under the mixed value, it says .0003.
Q That's at the top of page 37?

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A Top of page 37, yes.
Q As originally reported in exhibit 250, the P value under the two tailed independent test, 12 treatments and 12 controls that we spoke about earlier, was 0.796, is that correct?
A Yes.
Q And that is an established - -
A That is very different. And when you look at the raw data, you begin to see why that is. I'm going to go - let me find it here. Beginning on page 11, we have a table of needs for control and treatment groups for each day. And if we come down on page 11, where it says, Observation, which is row 37 in this table, IgA serum, the 3 in the day means this is taken on day 3. 7 means day 7. Control and treatment. We see here the control and the treatment start off very different. And the same on day 7. Now, if I recall correctly, day 3 and 7 no treatment had been applied yet. So, for reasons that I can't even guess, for whatever reason the serum IgA was different at the very start of the treatment. So, when we did a before and after study or analysis, as was done on this table in exhibit 250, it didn't show much effect.

This particular calculation could have all of the days in there, whether they're just looking at after the treatment had been done. And so it's including a difference that was at the very start before any treatment was applied. And that's why, I think, it is so radically different.
Q Then, with respect to interleukin serum, I think the results of that under the mixed model, the next few

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pages, I think I'm seeing them on 39 and 40?
A Yes. That's correct. And again, I think this is because of the differences in this. When you look - this particular analysis, basically, didn't recognize - the first two points in there were not actually points of where the treatment had been applied. And when you take that into account, you get that the effect is bigger than what it appeared in this analysis.
Q So, would it be fair to say, in summary form with respect to exhibit 279, although the method of analysis you've tested it, that was attempted to be applied, might be more appropriate than the original paper, it wasn't applied correctly to this data set?
A I would say that's correct.
Q So, then there are some more follow-up analyses that you requested, starting with exhibit exhibit 280 a couple weeks later, is that correct?
A I don't remember requesting 280, but that is a follow-up analysis, and in looking at that, I'm not entirely sure what that analysis was. It appears to be a multi-variable analysis of some sort. But there's not enough of it there for me to see what - it appears that it's being applied to IgA serum, but I don't see the actual SAS code for it, so I am not a hundred percent sure what was actually done with that.
Q Fair enough. Then 281. This goes several months later, into February of 2000, according to the date on the document. Do you recall requesting this re-analysis?
A I recall discussing with Steve using those days when no treatment was applied as a co-variant barrier to the

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analysis.
Q For some reason, it looks like we don't actually have the numbers - -
A All I see is what looks like a cover page for a report that Steven has sent me.
Q Then, 282 is a memo directed from Steve LeMire to Doug Reinemann. It indicates that you had requested the summary statistics that are on the second and third page. Do you recall requesting this information?
A I am pretty sure I would have requested that information. I don't recall the specific request, but it's certainly something I would have requested.
Q And why is that?
A Well, these are just the basic characteristics of the cows that we were using in the study to just - to document what the animals were like, what their milk production was, how old they were, which lactation they were in, those kinds of things.
Q And then, for each of those types of statistics or variables, there's a mean, a standard variation given in the table, correct?
A Correct.
Q From that data, or will that data reflect at all upon how, shall we say, successful the randomization process was?
A I don't know if it would reflect on how successful randomization was.
Q Or how random it was, I guess is what I was looking for.
A It would basically reflect on whether the two groups were really similar. As I recall, these were randomized. I

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believe Steve actually did the randomization, probably using a computer program that is essentially a random number generated to assign the cows to treatment. I believe that's - I could be mistaken on that.
Q Well, does it appear that the two groups of cows were reasonably similar?
A From the types of gross variables, like age of the cows, milk yield, lactation number, that they looked fairly similar. That's fairly typical of what you see with a well-randomized experiment.
Q And then, exhibit 283 appears to be the result of another request for a different type of statistical analysis from yourself, is that correct?
A Actually, just a different variable. One of the things that we had recorded, and had not included in the initial analysis, was body temperature, morning temperature of the cow's body temperature. And that appears to be what's being analyzed here.
Q On the third page of that document, there is a table 2, it has both typewritten and handwritten numbers. Any idea what the handwriting is all about?
A Let's see. The very first here, over where it says treatment and treatment by time, it's just to indicate that those are the P values in those columns. I do not know what that split plot refers to. The check marks are just apparently checking with the computer printouts to make sure that the numbers were typed correctly in the table. The Concanavalin A blastoenesis that has a square around it indicates that before the .413 should actually be point .4013.

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Q Do you know whose handwriting it is on this page?
A I am not entirely sure. It looks a little like mine, at least in places. So, I may have written that. The split plot does not look like my handwriting. My handwriting is not that neat. But the scribble at the bottom does look like my handwriting.
Q Up at the top it talks about - -
A Yes, the P, I'm pretty sure I wrote that P. That looks like the way I would write Ps, at least the first one.
Q And on table 2 at the end of the typewritten introduction there, the third line, it says, "n equals 12 treated and 12 control cows," correct?
A Where?
Q Top of the third page, third line.
A Top of the third page, third line. Yes, that's correct.
Q Would that again indicate that this was run as a simple two tailed t-test and not with a block design or analysis?
A No. This was run, as I recall, as an analysis of co-variance design, where the initial values were included. I do not believe that a block was included in it. With that much of a block effect, it's included in the co-variant effect. Not always, but it is possible that much of it would be.
Q So it's a different way of getting at some of the concerns about multiple groups, is that correct?
A It's - no, it's a way of getting at the concern I mentioned earlier about the IgA, that the cow started off with a different IgA. That's what it's really trying to get at.

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Q And as I think you indicated earlier, the original analysis in the published report, published in the sense that it went to the Minnesota government, the variable that was analyzed was the difference in cows from the first week test when no treatment was being applied to the later test, is that correct, with later assays, I should say?
A Yes.
Q And if one did that and one also took into account the block design of the experiment, that would yield an appropriate statistical analysis, would it not? That is, using the differences, the variable of interest in analyzing pursuant to replication of blocked statistics.
A It might. I would run that by a statistician.
Q Fair enough. 284. Another memo from Steve LeMire to Doug Reinemann, copy to yourself, indicating it was another analysis that you had requested, dated August 8, 2000, correct?
A That's what it says.
Q Do you recall requesting this one?
A No, I don't. I do know what it's about, so it's quite possible that I requested it, but I don't recall making the specific request for it.
Q Okay. And what is it about?
A A question came up. If this is the one I'm thinking it is, a question came up of a positive control. If we were to take cows and do something to them that we knew affected immune function, would we be able to detect it with the assays we were using? I do recall some discussions about that. What are we going to do? And

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the decision was made. I don't remember who actually came up with this suggestion, but it was suggested that a classic immune suppressant treatment is high dose glucocorticoids.
Q Doctor, the first two words were high dose?
A High dose, yes.
Q And that was, such a control test was run in the original Part III study using dexamethasone, is that correct?
A That's correct.
Q Go ahead.
A Yes. It was actually after we had finished the results we had been discussing up to this point. We did a short study with a fairly small number of animals where we injected dexamethasone. I think it was a three or four days of treatment, probably says in here - yes, four days treatment, and made our various measurements on them. And this was just an analysis of those results to see if, in fact, we did effect immune function.
Q Sure. And they were also analyzed in a different statistical manner in the original paper, correct?
A Were they recorded in the original paper here or not?
Q Page 11 on exhibit 250.
A We're looking at 250. I didn't recall those even being in this paper.
Q Isn't that page 11 reports in exhibit 250?
THE REPORTER: I'm sorry, I'm not hearing you.
A It says positive control. Let me read this. It makes the statement that they were suppressed, but I'm trying to find the table that actually shows the data. So I do

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not know how it was analyzed at this particular study.
Q I don't know that we have the data, the table with the data, but Table III on page 11 gives the results of the statistical analysis, I believe. Looks very similar to table 2, which has the electrical treatment.
A Yes, I believe that is correct. The analysis here is a different analysis, a different way of analyzing it. But you're correct on that.
Q Would this again be a t-test, basically?
A This is a t-test, this Table III on page 11 is a t-test.
Q And there's no issue with respect to the blocked or blocking issue because there's only one replicate?
A Correct.
Q And with a known immune depressant and dexamethasone for the 13 values reported were less than .05, correct?
A That's correct.
Q Okay. Then exhibit 285 appears to be another memo from Steve LeMire to Doug Reinemann, copy to you, about some more analysis you had requested, according to the document. Do you recall requesting this analysis?
A This appears to be the analysis co-variants I was referring to.
Q With respect to the positive control study?
A Let me look at that. I don't think so. Must have been. There's not enough cows there.
Q Under Introduction, it says, "It covers the positive control blood data"?
A Okay. Yes. That's what that says.
Q All right. In the course of all this analysis and re-analysis, did Steve LeMire, or anybody else associated

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with this experiment, that is, the Minnesota funded one resulting in the Part III paper, exhibit 250, ever do an analysis of the statistics of the difference between the after treatment and before treatment levels using a block design?
A Not that I'm aware.
Q Was that ever discussed among yourself and Mr. LeMire and Professor Reinemann or others associated with the experiment?
A Not that I recall.
Q So nobody ever expressed reluctance to do it that way, I take it?
A Not that I recall.
Q And it was Mr. LeMire who was primarily in charge of that part of the work, correct?
A Correct.
Q And the person overseeing - who is the person overall in charge of the whole experiment? Was that Professor Reinemann?
A That would have been Professor Reinemann.
Q Then, with respect to the second experiment involving the Messenger RNA, the gene expression, who would you describe as the person in overall charge of that experiment, if there was one?
A That would have been me.
Q In the materials from the University, there were - the one graph of that I found in those materials came out of Professor Reinemann's file. Maybe I can find that. (Exhibit 286). I printed off the sheets from Professor Reinemann's disk right after that.

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How was it that this document came to be drafted? Did you write it? Did somebody else write it? Did you have assistants or somebody else? How did that work?
A This appears to be - this appears to be something that I wrote, and would have shared with Dr. Reinemann, who was involved in this, particularly in figuring out how to deliver the current to the animals.
Q This particular item, again, which is off a disk supplied by Professor Reinemann, has some typed in red, appears that editing was going on by someone. Do you know what that was all about?
A I do not know who would have made those edits. They may have been changes that I made in the document, they may have been things that someone else suggested. I wouldn't know.
Q But your last - at the first portion of your deposition in March, another version of this document was marked as, I believe it's exhibit 251. I believe the text is the same. We can check that, but.
A It would look very similar.
Q It didn't have the red type and so forth, but I think it was the same text in the first four pages plus the references anyway.
Would you turn to the third page of that document, and I think the text will be the same on the other one. Under Results and Discussion, in the second paragraph, the third sentence, it says, "There was a tendency for IL1a, or alpha, and IL1b beta mRNA, to increase slightly, P less than 0.10, but it did not reach

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significance at P less than 0.05." Did I read that correctly?
A That's what that says, yes.
Q And yet, in the tables of data and analysis that we went through this morning, exhibit 254, those P values were on the order of approximately 10 to the minus 5th, or 10 to the minus 6th, something like that. Do you recall that?
A Yes. And I do not recall - that's why that maybe in looking at these columns, I don't know that I generated those numbers in that table. I don't know where those numbers came from.
MR. THORNTON: The last four lines?
A The last four lines of this (254). I'm not sure. Because this does not look like a version of this that I actually had. It's got a lot of the numbers in it. So, I don't know where those numbers came from. I don't recall putting in those particular numbers, so I can't vouch for the veracity of those last four lines of this table.
Q Okay. You supplied documents to Kathleen Erwin, the University's counsel, in response to the subpoena that was served on the University back in late 2007 and through early 2008, correct?
A That's correct.
Q That those documents were subsequently copied when in document form and provided to myself and others who were interested in obtaining those documents.
Did you ever send that documentation back or did it stay with the University?
A As far as I know, it is still with the University. I

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don't have it.
Q Well, I will represent to you that it was provided to us on a disk whose cover was, as marked on exhibit 276, and on that disk - mark some of these if we need to later, but I would just like to work through it before we do. There are files named "503 cow data sheets." And then, "Array 503 gene. Array 503 layout. Array 503 statistics. And collated Array 503," as it appears on the disk produced by the University.
Do you have any idea what the 503 number refers to?
A Just a number we gave the file.
Q And that particular spreadsheet, including the bottom four lines, appears in the file labeled "Array 503 statistics." I'll also represent to you that Professor Frank Martin, who is a retired statistics professor at the University of Minnesota, with a long time appointment at the veterinary school, has reread those numbers and verified them.
A Okay.
Q And again, it would just be a matter of - -
MR. THORNTON: I'm going to object to that. You are testifying, number one. Number 2, frank Martin hasn't been identified as an expert in this case and hasn't made any expert disclosures in this case.
Q In any event, coming up with that analysis is just a matter of running the math, either by computer or however, correct?
A Correct.
Q Where did the .05 to .10 P values come from?

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A I don't recall where those would have come from. They could just be things that I entered incorrectly in this tape. I may have inadvertently got my decimal point off and meant .01. I honestly don't recall.
Q And by table, you are referring to the table 1, which is part of the exhibit - -
A Table 1, yes.
Q It appears that there are no P values in that table, is that correct?
A That's correct.
Q However, the fold number, or a fold number anyway, is in that table, correct?
A Yes.
Q Did you ever have occasion to discuss the P values from the gene expression study, the second study that is, with Professor Reinemann?
A I don't recall any such discussion.
Q Dr. Sheffield, I would like to ask you to assume, hypothetically, that the P values from this second study are, as set forth in exhibit 254, pursuant to a t-test as we discussed this morning, and that the P values for IgJ of 8.21, times 10 to the minus 5th; interleukin 1 alpha of 8.74 times 10 to the minus 6th; IL1 beta of 2.55 times 10 to the minus 6th; IL2 of 4.98 times 10 to the minus 6th; and IL10 of 2.93 times 10 to the minus 5th, do reflect an accurate two tailed P analysis of the experimental data.
Under that hypothetical assumption, would the Results and Discussion section of the paper need to be redrafted, in your estimate, if it were to be

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submitted for publication?
A Well, it would certainly, when it refers to IL1 alpha and beta, would need to be modified from slightly to significantly, with a PS to .01, which is how very low P values are usually reported, and he did not reach significance would be struck. The significance of the IL2 and IL10 is not changed, just the P value changes. And the same thing with .04, the IgA, heavy chain, and secretory piece.
Does that answer it?
Q Sure. And again, as we have already discussed, an increase of interleukin 1, either alpha or beta, that is somewhat under two-fold, is certainly not indicative of a change associated with an acute infection, correct?
A Correct.
Q The main objective of a commercial dairy herd, certainly one of them, is to produce high levels of milk production and good components, correct?
A Yes. That is a major objective.
Q Do you have any opinion one way or the other as to what the likely consequence of chronically elevated interleukin 1 alpha and beta between 1.5 and two-fold, what would be there without electrical exposure, would have to the productivity of a commercial dairy herd?
MR. THORNTON: Objection. Foundation.
A I am not familiar with anything that would let me assess that reliably.
Q Is it true, as I've heard some people express, that elevated interleukin 1 is one of the principal factors determining how we feel lousy when we have a cold or

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other infection?
MR. THORNTON: Objection. Relevancy.
A I've heard that.
MR. THORNTON: Objection. Hearsay.
Q Do you have any professional opinion as to whether or not its true or do you know?
A I don't know.
Q Do you know of any studies or publications indicating that one of the symptoms of chronically elevated inflammatory cytokines, such as interleukin 1, use some degree of inappetence?
A Excuse me?
Q Some degree of inappetence, not getting hungry?
A I'm not aware of that. But I haven't read the literature on this in a while.
Q I think you would probably agree that when dairy cows get some degree of inappetence, that's a big deal to a commercial dairy, fair statement?
A Yes, feed intake is a big issue in dairy production.
Q I have way too much paper here. What, if any, are the consequences of a decrease in IgA, either in serum or in tissue, to local immunity of the utter of a cow?
A In tissue, it's fairly important. Circulating, I'm less certain about how important that would be.
Q And why are IgA levels in the utter important to local immunity of the utter?
A Well, IgA in many mucosal tissues is the major anti-body that is secreted into a secretion. It turns out in cows, they are a little bit different in their mammary glands than most other species, in that cows' milk actually has

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more IgG in it than most other species do. Humans, for example, the major anti-body in milk is IgA. Cows have significant amounts of IgA in their milk, but they also have large amounts of IgG. But IgG is still an important part of immunity in these tissues that are exposed to the environment, such as the utter. If you think about it, the inside of the utter is outside the body.
Q Explain that a little bit more, please. That last comment, I mean.
A Well, the mammary gland is lined with an epithelium. Milk is produced in small structures called alveoli and is transported through a system of ducts to the outside. So, it is just like the lining of your GI tract, it's actually outside the body. That means it is very easy for bacteria to enter through the teat, what we call the the streak canal, and infects the utter. And the utter has, as you'd expect, a lot of defense systems, among them, various anti-bodies in milk.
Q Unfortunately, the next document I want to refer to is sitting down in the trunk of my car. I can get it if I need to.
MR. THORNTON: Do you want to take a short break?
Q Yeah, we could do that. Maybe I can ask you if you can recall anything about Professor Reinemann making a presentation at a seminar relating to - or entitled Stray Voltage, something like that, in Campbelltown, Pennsylvania, in the spring of 2003, in which his references included a reference to a paper that was reported in the document in press, being submitted in The Journal of

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Dairy Science, in which you were a co-author at that time.
Does this make any sense to you?
A Possibly. I think there was a paper that we were drafting. I don't recall it ever being actually accepted for publication. I assume that is what he is referring to.
Q I believe in 2003. Would that be approximately the likely - -
A That would sound like about the right time-frame. I'm assuming that's the paper he's referring to. It was never published.
Q Is that the same paper that was marked in your earlier deposition as exhibit 249?
A Let me see exhibit 249.
Q Here we go.
A Yes. That's the one I'm assuming he's referring to there.
Q To your knowledge, was that paper put out for peer review?
A I don't recall.
Q You never saw any feedback from any peer review - -
A I don't recall anything like that. It could have been. That was a long time ago. I don't recall that.
Q Were the results of the second study with the MRNA gene expression ever put into a form that was intended to be submitted for publication?
A No, they weren't.
Q And you personally have no results of statistical analysis of that data from the gene expression study at

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all, I take it, unless they are in what you brought to the first deposition? I don't believe there's any - -
A I don't believe I do.
Q Back in the late '90s and the very early 2000s as this work was going on, did you know that Dr. Reinemann had been testifying for utilities in stray voltage litigation since approximately the early 1990s?
A I know now that he had been. I honestly can't remember if I knew at that time if he had been or not.
Q Not a subject that ever came up at the time?
A I don't recall any discussions with him about it. I may have known it, but I may not. I just don't remember what I knew when.
Q Other than responding to the subpoena here today and the last time we got together, you have never been involved in litigation - -
A I have never been involved in any such litigation.
Q Sure. You've never been involved in any litigation as an expert witness, is that correct?
A No, I have not.
Q At least until whatever we've done here. I don't think I'll bother going down to the car. I think we have covered it.
I've got copies of these, but may not be very significant. Let me just ask you some questions regarding them.
Exhibit 287 is what I understand to be some notes taken by folks working in the first research project, the Part III paper project, taken off the materials provided by Dr. Reinemann in response to the

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University subpoena.
Does that refresh your recollection at all as to whether there were any barn notes taken?
A Well, that is what this appears to be. I don't recall seeing these specific notes before, but I knew that things like water consumption and temperatures were being recorded. I didn't see the raw records, but that's what this appears to be.
MR. THORNTON: Object to that document. Foundation.
Q The document, the Bates number from the University materials, the documents start with Reinemann 1595, for the record, and runs through - well, there's a number of pages there, The last one of which is Reinemann 1611.
And at the bottom of the sheet where the water and temperature and so forth are recorded, there's a Comments section, is that correct?
A That's what it seems to be.
Q And what was the purpose, if you know, of having that Comments section on the form?
MR. THORNTON: Objection. Foundation.
A I would only be able to speculate, since I did not - -
MR. THORNTON: Objection. Speculation.
Q Since you did not, what?
A Since I did not design the form or have any input into it.
Q Well, were you not the person in charge of experimental design as it related to things having to do with the cows themselves?
A No.

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Q Who was in charge of that?
A Dr. Reinemann did most of the setup, designing the stalls and working with the barn crew. I was responsible for handling the samples after - the blood samples that were coming into the lab.
Q There's a person named Misty, I believe the last name is Davis, that appears in these forms. Are you familiar with her?
A She was someone that worked with Dr. Reinemann. I don't know if she was a graduate student or post doctorate or her exact status, but she was an employee with Dr. Reinemann in Ag. Engineer.
Q Well, I tell you what, why don't we take a short break. I'm close to done. And these gentlemen will want to ask you some follow-up questions, I'm sure, especially Mr. Carlson, who wasn't here before. Why don't we take ten minutes, shall we say, or your pleasure. It's your flight, so I'll make it shorter, if you want.
MR. THORNTON: That's fine.

(At this time a recess was taken - 2:15 - 2:26).

Q Dr. Sheffield, we've marked as exhibit 288 another packet of barn notes, if you will, the water meter, et cetera, measurements on them and comments, and I believe these are from Replica 2, and were taken in January, 1999.
Would you turn to the third page of those barn notes, Bates number 1663. Look through some of these quickly. I'm sorry. Actually, turn to the fourth page, if you would, 1669. There's a fairly long hand-

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written description about cow number 3861 having a bad day there. Do you see that?
A Yes.
Q The initials at the bottom I think, appears to be RK. Do you know who that is?
MR. THORNTON: Objection. Foundation.
A A watcher, and I think the last name was something like Kasper, if I recall. There was a Roger Kasper that was involved in this, but I can't say for sure if that's who that's referring to.
Q Roger was a one-time State Agriculture Department employee, I believe. Did you know that?
A I seem to recall that, yes.
Q And toward the middle of the page, it's indicated by the author of this note, - I'll point to where I am on the sheet. "Decided to share concern with Doug and then Lewis." Do you see that?
MR. THORNTON: About half way. The line that begins with "Jerry and - -
A Yes. Okay. I see that.
Q Then, toward the bottom of the page, it's indicated, "Lewis, Doug and Josie felt okay to stay in trial." Do you see that?
A Yes.
Q And up on top, it describes her having trouble getting up, struggling to get up and things of that nature. Do you see that?
A Yes.
Q Do you recall anything about that cow as you sit here today? Long time ago, I realize.

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A There was at least one instance. This may be this particular instance, where someone, and I don't remember who it was, possibly - probably Josie, since she worked with me, came and said, "We've got a cow that seems very bad. Can we come look at her?" By the time I got there, she did not seem to be having those problems. But that's really all I recall about that.

I do recall at least one instance where there was a cow they were concerned about in the morning, but by afternoon seemed to be doing okay. But I don't recall any more details about it.

Q Seems to be several pages into the document, about seven pages in is a typewritten memo.

MR. THORNTON: What's the Bates number?

Q The date on it is January 25, 1999.

MR. THORNTON: I'll just note the page in front of it is 1674, then we go to 1394.

Q It is. Taken out of a different section of Dr. Reinemann's materials.

In there it talks of cow 3861 having fallen, so forth. Do you see that?

A I see that.

Q And further down is a duplicated, an e-mail from Roger Kasper, to apparently Dr. Reinemann and others.

Are you among the recipients of that e-mail?

A I don't recall seeing this. I will look at the Cs on it to see if I'm on there. I don't seem to be on the CCs to that. My e-mail address doesn't appear on this, and I don't recall seeing this.

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Q Sure. In the second paragraph below the addresses, there's a line, indicates, "Doug, Lewis and Jerry discussed the situation, decided giving her a tube of Calcium Oral Gel would help boost her energy. Also some surface ointment was applied to the spot on rear hip where the fur has rubbed off from rubbing against the stall support." Do you see that?

A I see that.

Q And if we go through these notes, we will find notes relating to cow 3861 on January 22, 23, 27, 28 and 29. I don't necessarily want to go through all of them with you, - -

A I appreciate.

Q - - but just take a quick look and verify whether or not there are multiple days with notations about that cow?

A Okay.

Q Is there multiple entries about that cow?

A It does appear that way, yes.

Q On multiple occasions either down or having trouble getting up, that sort of thing?

A There seems to have been some concern about her mobility.

Q And from the treatment prescribed, it appears that the consensus was that she was hypocalcemic?

A I don't recall there ever being - I don't recall a discussion about that particular treatment. I mean, it's possible she was, but - let me. I would have to spend some time looking through where she was in lactation and all of that. But I don't remember a discussion about treating her with calcium.

Q But it is indicated in Roger Kasper's e-mail though, correct?

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A It is indicated that she was treated with that.

Q And if that were done, the only reason for that would be, the thought process was, she was hypocalcemic?

A That would be what you would use that for.

Q Now, according to the table in exhibit 282 that we went through earlier, I'll just show you my copy here, it indicates that that cow was, I believe 175 days in milk at the start of her participation in the study?

A That's what that says, yes.

Q Is it normal for a cow in mid-lactation, such as that, to be hypocalcemic?

A Not normally. That's usually something that occurs early in lactation.

Q I believe the Part III paper indicates that there were no noted differences between treatment and control cows in behavior, or makes that indication at some length, is that correct?

A I think it does.

Q Would it have been a reasonable thing to evaluate statistically the number of cows, treatment and control, through these three replicas that exhibited some unusual behavior or some health problems, such as hypocalcemia?

A Because the incidents of it is so low, I don't know how you could make any conclusion from that.

Q Well, that would certainly be true for any one condition?

A Or any one condition, yes.

Q How about, would it be reasonable to evaluate the incidents of any unusual behavior for health problems in the population of cows?

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A I'm assuming you mean by this, simply having a health problem, yes or no. I don't know, just because that seemed like a fairly vague thing to do. Most researchers would want to break it out into exactly which health problem you're talking about.

Q Understood. But the more you break it out, the more you run into the difficulty of small numbers of cows in - -

A Yes.

Q - - experiments that are practical, right?

A Yes.

Q In exhibit 286, the draft paper that was not published, there's a reference in perhaps more than one place, but one of them is at the bottom of page 1, the very last line, and on the top of page 2, about epidemiological study of over 15,000 Swedish cows. Do you see that?

A Yes. I remember reading this study.

Q And that had to do with herds that use or did not use electric cow trainers?

A That's correct.

Q I'm going to show you what we have marked as exhibit 289, which is out of an LC of your journal, titled Preventive Veterinary Medicine, and it appears to be a copy of an article entitled, "Associations between use of electric cow-trainers and clinical diseases, reproductive performance and culling Swedish dairy cattle," and the lead author is Pascal A - not even attempt to pronounce this, O-l-t-e-n-a-c-u. Is that correct?

A That's correct.

Q Does that appear to be the article that you were referring to in your draft?

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A That does appear to be the article. Let me double-check this. Yes, that appears to be the same article.
Q Then, on the fourth page of your paper, the page where the references start at the bottom. The draft paper says, starting on the fourth line, "An epidemiological study by Oltenacu - for want of a better pronunciation - et al, 1998, that found mastitis and reproductive problems were associated with the use of cow-trainers. This could suggest impacts of electrical exposure. But other explanations are also possible, including the herds with such problems may be more likely to use cow-trainers to solve them." Correct?
A That's what it says there, yes.
Q And do you recall that at least one of the herds studied, a larger herd utilized one group of its cows and in the before and after use of cow-trainers - -
A Yes, that was in some of the data in there, yes.
Q And even in that herd using the same cows as their own control, if you will, there was found to be an effect, is that correct?
A It has been a while since I've read this paper.
Q Well, and I don't want to make you sit here and read it.
A I think I do recall that.
Q Okay. Fair enough. We can all read the paper and find out.
Have you had any occasion to read exhibit 252, which is marked from the last time, which is a group of materials prepared in connection with other litigation, not this case, by Dr. Frank Martin, retired physics professor I have referred to?

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A I have read it. I haven't read it in incredible detail. By I have read through that, yes.
Q With respect to Dr. Martin's statistical methods applied to the data from the Part III study and/or the MRNA gene expression study, do you have any critiques or issues with the statistical methods he approached those with?
MR. THORNTON: Objection. Foundation. Objection. Identifying the work of a witness who has neither been designated as an expert or provided a report in this litigation.
A Okay. I was not sure exactly what he did in his re-analysis, but the idea of using the general type that he referred to would seem reasonable.
Q Counsel talked to you about the issue of blood samples collected from the cows in the second study, the gene expression study, which is referenced, I believe, in the middle of the second page of exhibit 286 or exhibit 251, whichever you prefer, and which indicates blood samples were collected via the tail vein immediately prior to applying current and at the end of a three week exposure period." And I think you told Mr. Thornton when we got together before that you didn't know where the data was from the initial blood draw?
A I don't think we ever analyzed those samples.
Q I see. Is there a particular reason why or why not?
A Running out of time and money, and the analysis is quite difficult, tedious and expensive to perform.
Q And then these gene expression assays are expensive and the use of sophisticated equipment, correct?
A Yes.

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Q I think you mentioned earlier that people had to share the University's equipment with other departments, it's not like it was available to you for as long as you wanted, I take it, is that correct?
A For some of it, that's true.
Q By the way, did you ever talk to Mr. Thornton before the first leg of the deposition in this matter?
A Only to reschedule the deposition.
Q Have you ever had occasion to discuss with anyone what you were going to be asked about in this deposition before it occurred, and I don't mean just today, but the first leg of it?
A No, other than to state your opinions on this work.
Q But have you had occasion to discuss something with Mr. Thornton just as we took a break awhile ago? Did it have anything to do with this case or just pleasantries?
A Just the contents, you know, about, I think the comment that I made had to do with I can be kind of frustrating at times because I will answer a question by saying this, but on the other hand.
Q In other words, lawyers like more defined answers than scientists often have?
A Yes, I think sometimes I could be frustrating because everyone seems to want me to give a yes or no answer, and I'm seeing nuances in things.
Q I have enjoyed listening to your nuances, Professor Sheffield, but that's all I have at least for now. Counsel may have some follow-ups.
MR. CARLSON: Do you have anything, Tim?
MR. THORNTON: Go ahead. You haven't had a

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chance to talk to him at all. I kind of get the feeling we're going to be coming back here.

RE-DIRECT EXAMINATION

BY MR. CARLSON:

Q Dr. Sheffield, correct?
A Correct.
Q I just want to kind of go over a few things. I apologize in advance if I cover any ground that's already been covered, and I'll try not to. And I have read the transcript of the first portion of your deposition.
Have you had any contact with attorneys Will Mahler or Charlie Bird or Jeremy Stevens, or anyone working on behalf of Randy and Peggy Norman?
A No.
Q Is it your understanding that your testimony or March 14 and today will be to render opinions regarding your work?
A Yes.
Q Now, the work of Frank Martin has been described to you a few times, and I'll represent to you that he has done some work in the case of mine, including the Randy Norman versus Crow Wing Power case, which is why I'm here. Dr. Martin testified that you've lost data, and he's referring to that initial blood test data.
I take it that you were referring to the exhibit 286 abstract research that was done.
MR. LAWRENCE: Object. Foundation. Go head.

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Q My question is, did you lose any data?
A Not that I know of.
Q And when you testified just a few moments ago that time and money were running out and you had to spend money, I guess more wisely, you didn't analyze the initial blood tests?
A The initial blood tests were not analyzed. It wasn't that the data weren't collected and lost. I don't believe we ever analyzed those.
Q And do you agree that it would be helpful - it would have been helpful to have analyzed that initial blood test data so that you could compare it to what happened later?
A I think more data is also better served. I would say it would have been better to have done it.
Q And when I say what happened later, what I should have more artfully said, compare it to the end of research blood test results?
A Yes.
Q Are you aware of any authoritative referenced table that establishes what are good or acceptable levels or ranges of levels for various substances found in the immune system, such as IL1, IL2, IL3, IL10, IgA and I think you've been calling it IgG, I have heard it called Ig3 sometimes. Is there any such authoritative reference table that you're aware of?
A There are accepted values for what is normal for some of those. I couldn't point you to a specific reference, off the top of my head. But for others, I don't think there are.
Q I take it, off the top of your head, you can't tell me

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which ones that there are basically no - -
A Well, - -
Q If I could finish. I take it that you're not aware, off the top of your head, of which one of these or any of the other - I'm calling them substances, I don't know if that's the best term, but better analyzed in exhibit 254 and described by Dr. Chase in exhibit 275. You couldn't, off of the top of your head, say that there's known values for these that are acceptable?
A Well, those are different than protein values. Those values are Messenger RNA values, and I think it would be very difficult to find standard values of what those would be.
Q What about on the various cytokines, the pro-inflammatory and anti-inflammatory, and I'm going to say compounds, if that's okay. I'm not in your field. But is there any table of known, accepted, you know, reasonable values for those?
A Not that I'm aware of for cattle.
Q And what I'm getting at is, I go to the doctor and he does a - runs a blood test on me. Gives me back a report and says, "You're good and bad cholesterol levels are this, these are the acceptable ranges. Your enzymes are in this range, and this is the acceptable or good range." So there's nothing like that for dairy cattle regarding their immune system?
A For immunoglobulin levels, there probably is. There are some numbers that you might think are high or low. But for the cytokines, I'm not aware of anything like that.
Q And IL1, 2, 3, 10, IgA, IgG, those are all cytokines?

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A No. The IL interleukins are cytokines.
Q Okay.
A The ones that start with Ig are immunoglobulins. Those you might find standard values for. I couldn't point you to a specific table, but there might be such.
Q Are you familiar with a method called, Repeated Measures Design?
THE REPORTER: Called what?
Q Repeated Measures Design.
A Yes.
Q And was that used at all in your work?
A That's what the Proc Mix was, is a type of repeated measures design.
Q And you use Proc Mix for some of them?
A For some of the analysis, yes.
Q And those are at least denoted in the exhibits, correct, which ones have been used for?
A I think so.
Q That's all I have for now.
A Okay.
Q Oh, wait. I'm sorry. I missed something here. I just want to read to you something.
Dr. Frank Martin has opined in the Randy Norman case that he will testify that in the 1999 Science Advisors, Part III experiment, 1 milliamp current had an effect on behavior and health of the treated cows at a highly statistically significant level. Do you agree with that statement?
A I'm not familiar with the behavior data that was collected on that, so I can't give an opinion whether I

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agree or disagree with that.
Q What about the effect of those levels on the health of the treated cows, did the 1 milliamp current have a statistically significant effect on their health?
A Not that I know of. But again, I don't have the specific data about health measures for that.
Q So, were your conclusions in the 1999 Science Advisors, Part III, he calls it an experiment, I believe it was research, so you didn't come to these conclusions that Dr. Martin comes to here, is that correct?
A We are referring here to - -
MR. THORNTON: 250.
A 250.
Q Yes.
A Okay. As I recall, that was a statement that was made based on the - I don't know what that statement was based on about behavior.
Q Are you talking about Frank Martin's statement?
A Yes. I don't recall - I don't know what that statement was based on. I am not aware of anything in that study that showed behavioral differences, but I'm not aware of data - specific data on behavior.
Q So did you, in exhibit 250, in that report, was a change in cow behavior one of the things that was being measured?
A That was not the major purpose of this. The major purpose was to measure the immune function.
Q Was there any data obtained from that research which would allow someone to make any type of - or draw any type of conclusion about changes in cow behavior during

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your 1 milliamp exposure testing?
A There were some things that one might call behavior.
Q What were those?
A Things like feed consumption and water in-take were measured. I don't know if you'd call those behaviors or not, but those were measured. They were analyzed. I don't recall the results of those. As I - well, I better not say, because I really don't recall the results of those particular analyses. I don't recall anyone ever discussing any usual changes in feed or water consumption.
I'm not aware of any other measures of behavior other than general comments that were made in the barn notes, which I had never looked at before today.
Q Such as the cow - -
A Such as - -
Q If I can finish. And that's what I was wondering. Was there any effort made during your research for exhibit 250 that would have recorded or in some way measured animal behavior, such as lapping at water, stomping, kicking, things along that - twitching, things along those lines?
A Not that I'm aware of.
Q And you were involved in that research from start to finish, correct?
A I did not do very much in the barn. Other people were involved in that. I was mostly involved in analyzing the laboratory bench-type analysis of the blood samples that were collected.
Q Did you see a copy of exhibit 250 before it was submitted

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to the State of Minnesota?
A I believe so.
Q And in you review of that, did you see any indications that cow behavior, other than like feed in-take and water in-take, the things I was talking about, the lapping at water, fidgeting, and that sort of thing, did you see any indication that any of those behaviors were ever measured or recorded?
A I don't recall them being recorded, and I don't recall any discussion of recording specifically like that for this study.
Q And if I'm correct, and I apologize if I'm rehashing. Did that report, the Part III report, conclude that 1 milliamp current had an effect on the health of treated cows at a highly statistically significant level?
A I don't recall that conclusion.
Q Have you seen Frank Martin's complete re-analysis of your work where he comes to that conclusion?
A No, I haven't.
Q So, I take it you're not able to opine as to whether Frank Martin's methods and conclusions are valid?
A Without seeing it, no, I can't.
Q That's all I have.

RE-DIRECT EXAMINATION

BY MR. THORNTON:

Q Dr. Sheffield, you did all the statistical work for the second study that you did?

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A I think so, yes.
Q And you're not a trained statistician?
A No, I am not.
Q And you said it would be best to adjust for the multiple variables that you were looking for, but you didn't do that?
A I did that initial analysis. No, I did not.
Q And you're not an electrical engineer or have no particular expertise in electricity, do you?
A No, I have no expertise in that area.
Q Who handled the administration of the electricity for the tests that ended up in table 254?
A Dr. Reinemann suggested an individual, whose name escapes me now, to design the device to administer the current.
Q And do you know - -
A So he designed the equipment to administer the current to the cows. Do you have another question?
Q I want to make sure you're done with your answer.
A I was the one that actually attached it to the cows.
Q Do you know anything about the credentials, skills or competence of the individual that designed the device?
A At the time I had looked at his credentials, but I don't recall what they were.
Q And you lack credentials to make sure that the device was properly attached to the cow, didn't you?
A Well, I can tell if it was properly attached. It had indicator lights on it to indicate that it was working. What I would lack is expertise in determining whether it was properly designed.
Q And why did nobody else participate in the second study

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that you did?
A Well, Dr. Reinemann did participate in assisting in designing the equipment and figuring out how to administer the electricity to the cows, the current to the cows. This was very heavily involved in looking at the immune function. We weren't measuring a lot of other things in it. And so, I was - for this part of that study, and the time, we had no need to have other people involved in there.
Q Well, in my experience, there is usually multiple authors of these types of reports. You're the only one who's indicated as an author on this study. Why is that?
A That's because I had prepared the report. And that's the reason I put my name on there. There are a few other people I probably could have put on there.
Q Had you had any kind of falling out with Dr. Reinemann?
A Not that I know. He may have felt so, but I don't recall anything that was a falling out with him.
Q We talked to some extent when we talked the last time that there's a difference between biological significance and statistical significance?
A Yes.
Q What would you describe the term biological significance to mean?
A Biological significance refers to affecting the overall function of the organism in some way.
Q And did you see any evidence of biological significance in either the first study or the second study?
A The second study, you really could not assess biological significance in, because we were simply measuring

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Messenger RNA levels, not any higher level functions. The first study we - the study was not big enough to measure things like disease incidents. So, if you're looking for that as a biological response, it would have been an inadequate study for that.

In general, I would say little effect. You might consider something like IgA to be a biological significance. I wouldn't. I would consider biological significance to mean something, such as in this case, mastitis incidents, milk production or other such things that would be of potential interest to a dairy farmer as opposed to serum antibody levels, that's of more interest to a researcher.

Q And you saw none of those symptoms?

A Well, we didn't report it, just because we knew the study would be too small to report them.

Q And is there any conclusions that you can draw to a reasonable degree of scientific certainty from either study other than more study should be done?

A Regarding possible biological significance or?

Q Anything that you can draw - any conclusions, any opinions that you can come to to a reasonable degree of scientific certainty based on either study, other than more study needs to be done?

MR. LAWRENCE: Object to form.

A All of the conclusions that I could come to reasonably would be about things like Messenger RNA levels, or levels of protein in the blood, and would be suggestive of overall health effects at best. Rather than saying health effects to a scientific certainty, if that's kind

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of what you are getting at, I guess I don't know quite what you're getting at in the question.

Q Well, I think you indicated and your paper indicated that there was some issues with assay collections and assay analyzation, is that correct?

A Issues?

Q Problems.

A Well, early on, yes, some of the - some of the assays were not as feasible as we thought they would be.

Q And problems with the assay collection and assay analyzation is going to effect the data integrity?

A If not corrected, they could, yes.

Q And explain this to me. You say in some cases the activity changed, but the expression level didn't, when you're talking about genes. What do you mean by that?

A In the second study we are measuring Messenger RNA levels. In the first study, we're measuring the levels of protein in the blood. There's several steps in between those. The first step in producing the protein is called transcription.

This is the production of the Messenger RNA using the DNA and the cells nucleus as a template. That Messenger RNA then under-goes a process called translation in which it's used as a template to produce a protein sequence. In the case of proteins that are in serum, that protein is then processed and excreted from the cells. And what you measured in the serum is a mixture of the rate at which that protein is secreted, the volume that it gets distributed in the body, and the rate at which it is destroyed by the body or excreted.

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So there are many steps between the Messenger RNA and the actual protein being detected. For some genes, it's possible to see a bigger change in one than the other. They often parallel each other, but they don't have to.

For example, for certain cytokines, there are reports where the level of Messenger RNA changes more than the level of protein in the blood does. And I believe that's what I was referring to there, was the fact that they're measuring very different things and they don't always - one does not always reflect the other one. This is because there are things affecting the protein in addition to the amount of Messenger RNA that is there.

Q So, Messenger RNA is associated with the electricity that's being administered, or can be?

A The Messenger RNA could be affected by that. The Messenger RNA - I'm trying to think how to explain this. It's the first step in making a biological response, or is often the first step in making the biological a response.

Q And the biological response is the creation of proteins to fight the antigens?

A That could be a biological response, yes.

Q But if I'm understanding you correct, some of the proteins that were being observed were not necessarily associated with the Messenger RNA?

A I think that what we're getting at here is two different ideas. The Messenger RNA is not perfectly correlated with the protein in terms of how much of it is there.

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That's the point I was trying to make. The Messenger RNA corresponds to, in a certain sense, the protein in terms of the sequence of the Messenger RNA determines the sequence of the protein. But the level of the Messenger RNA is one of several factors that determines how much of the protein is being made.

Q But there is a relationship between the production of antigen fighting protein and the Messenger RNA?

A Yes. There is a relationship. It is not a perfect relationship. That was my point.

Q Okay. Why did you decide that your second paper wasn't worth publishing?

A I never published it. It was a fairly - it's - although there's a lot of Messengers there, it was a fairly small study in terms of the sorts of things that were measured. I looked at it and I thought as soon as I tried to publish it, it's very likely that someone in the review process would say, "But have you actually seen the change in the protein levels," or other things that could have been measured. And I - I just did not think it would withstand peer review because of that possible criticism of it.

Q So, not only was there a singular author, there was never a peer review?

A No. That's correct.

Q Why was the study done the way it was?

A In terms of measuring the Messenger RNA?

Q Right.

A Well, that is the first step in producing these proteins. And it's allowed us to assess a fairly large number of

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possible responses; whereas, measuring proteins is much more tedious.
Q You talk at some length with Mr. Lawrence about block analysis. Could you please explain that in a way that somebody as dumb as me can understand it?
MR. LAWRENCE: Object to foundation.
A Yes. In this Part III, there were 12 cows total in each treatment group. But they weren't all used at the same time. We did the experiment once with a total of 8 cows for treatments in 4 controls. Collected all of that data at the same time from those 8 cows.
Sometime later, we did the same thing with 8 different cows. At some time later, the same thing with a final 8 cows, for a total of 24 or 12 per treatment.
A question comes up, are the conditions the same in the first group and the second group? For example, in dairy cattle research, this might be reflected in the temperature of the barn.
Q We know the environmental conditions - -
A The environmental, it might change. Could that change the results? And that is what is meant by the blocking effect. And it's the statistical technique to correct for the fact that the cows weren't - the three groups weren't all at the same time, there was a sequential factor to it.
Q So how do you correct for that?
A There's a fairly well known statistical technique that does correct for it, called, in this case, it would be a randomized block design. Off the top of my head, I could

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not describe the mathematics of it. But there is - it is a statistical technique that, in doing an analysis of variant, you have what is called a model, and then residual effects. You include the three replicates as part of the model.
Q But cow performance, stress on a cow, is going to be different in January and May and August, isn't it?
A Yes. Yes.
Q Now, in the first test, 250, you talked about co-variants. You did a comparison of the animals, what their condition was when they started, and what their condition was when you stopped the test in terms of protein production, correct?
A That's right. Yes.
Q And you didn't do that in the second test?
A That's correct.
Q And what do you mean by covariant?
A A covariant is a variable that might influence the results that you were trying to control for.
Q So, for example, when Mr. Lawrence pointed out to you that the P value on exhibit 278 for IgA serum was .7932, that was the beginning and end - that was based on data at the beginning and end of the treatment, correct?
A Let me get to that exhibit. You're referring to 278?
Q 278, it's about the fifth box from the bottom. Do you see there's handwritten in there, IgA serum?
A IgA serum.
Q And it shows a P value of .7932?
A Okay. Yes.
Q And that's not statistically significant because it's

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greater than .05, correct?
A That's correct.
Q And then, when you did the same analysis, as I understand it, and looking then to page exhibit 279, page 37, you got a P value of .0003, and that's based upon comparing the same protein between two different groups of animals?
A That's correct.
Q So, if we compared the control group with the treatment group, even before any treatment was administered, the difference in the IgA serum levels was statistically significant?
A Correct.
Q But when we compared the treatment group at the beginning and at the end, the difference between the IgA serum levels was not statistically significant?
A That's what this result indicates.
Q And the difference between the two is dramatic, statistically speaking, right?
A Yes.
Q So, when you only made the comparison between the control group and treatment group in the second study, not knowing what the difference was between the two groups when they started, that really calls into question any of your data, doesn't it?
A The fact that we randomized the assignment of cows to treatment should prevent that. I have no idea of why it didn't in this case. But it does - it is something I would concede as a possibility, yes.
Q Well, you thought the cows in the initial study were randomly selected.

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A Well, they were randomly selected or randomly assigned to treatments, and I do not know why there was a difference initially at the start in IgA.
Q But there definitely was?
A There was, yes.
Q And that would indicate, as to this criteria, these cows were not random?
A It just indicates that they were different. I don't know if that gets into a question of what random means. But we had - when we assigned them to the treatment, we had no idea what the IgA levels were. So. But it is something that was different in the two groups.
Q I gotta hit the airport. Let's go off the record.

(3:24 o'clock p.m.)

* * * *

READING AND SIGNING CERTIFICATE

I, LEWIS G. SHEFFIELD, do hereby certify that I have read the foregoing transcript of my deposition, recorded by John T. Kirby, of 5-9-14, and believe the same to be true and correct, (or except as follows, noting the page and line number of the change or addition and the reason why):

WRITING IN TRANSCRIPT WILL NOT BE ACCEPTED

_____ _____

DATE SIGNATURE

STATE OF MINNESOTA)
) ss.
COUNTY OF DAKOTA)

Be it known that I took the deposition of LEWIS G. SHEFFIELD, on the 9th day of May, 2014, at Madison, Wisconsin;

That I was then and there a notary public in and for the County of Dakota, State of Minnesota, and that by virtue thereof, I was duly authorized to administer an oath;

That the witness before testifying was by me first duly sworn to testify to the truth and nothing but the truth relative to said cause;

That the testimony of said witness was recorded in computerized Stenotype and thereafter transcribed by myself, and that the testimony is a true record of the testimony given by the witness to the best of my ability;

That I am not related to any of the parties hereto nor interested in the outcome of the matter;

That the reading and the signing has been executed as evidenced by the preceding page.

WITNESS MY HAND AND SEAL THIS 12TH DAY OF MAY, 2014.

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