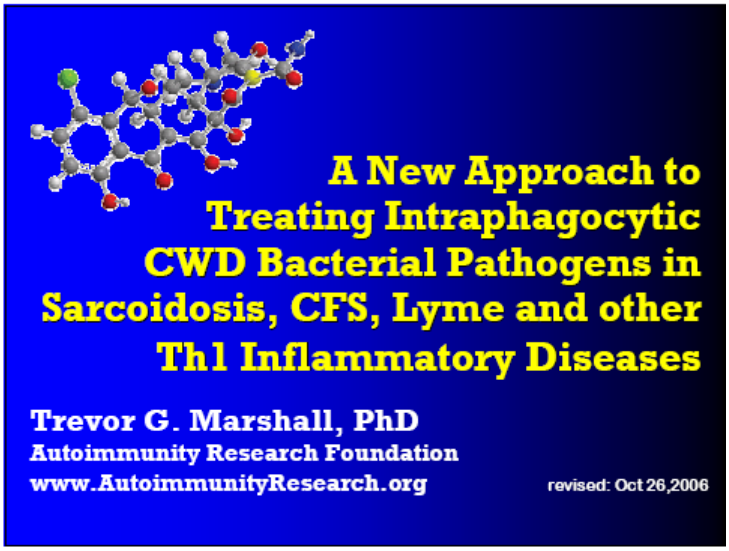
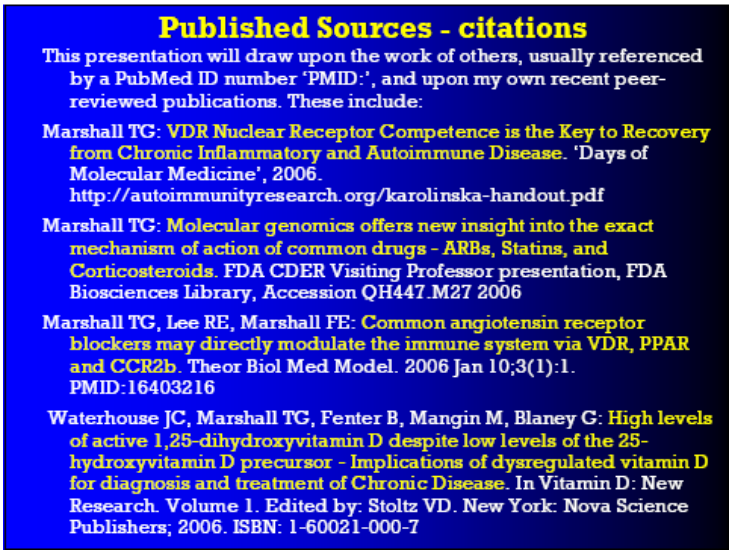
"The Brain and the Environment"
Immunity, Infection, Inflammation & Toxins







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TRANSCRIPT OF 27 OCT 2006 PRESENTATION

Dr. Trevor Marshall

Title Slide
0.07 / 1:23/02
Introduction by John L. Wilson, MD
AAEM CME Committee Chairman

Our next speaker is Dr Trevor Marshall. He, Dr Marshall, graduated in 1985 from the University of Western Australia with a PhD in Bioengineering. His thesis research included a non-surgical method to cure cryptorchidism, effective male and female infertility interventions and insulin infusion technology. He is currently director of Autoimmunity Research Foundation in California. He will be discussing, among other things, Olmesartan, Minocycline, Clindamycin, Azythromycin and FDA designations for Sarcoidosis. Also will be discussing generalized Th1 inflammation.

His topic. I have heard about for years about the Marshall treatment plan for treating intraphagocytic Cell Wall Deficient (CWD) organisms and I think it is a real privilege for us to be able to have at our academy someone, the person who really developed the Marshall Plan (This is not the same plan that ended WWII BTW... chuckles).

He will be discussing a new approach for treating intraphagocytic CWD bacterial pathogens in Sarcoidosis, Chronic Fatigue Syndrome, Lyme disease and other inflammatory diseases. Dr Marshall. [Clapping].

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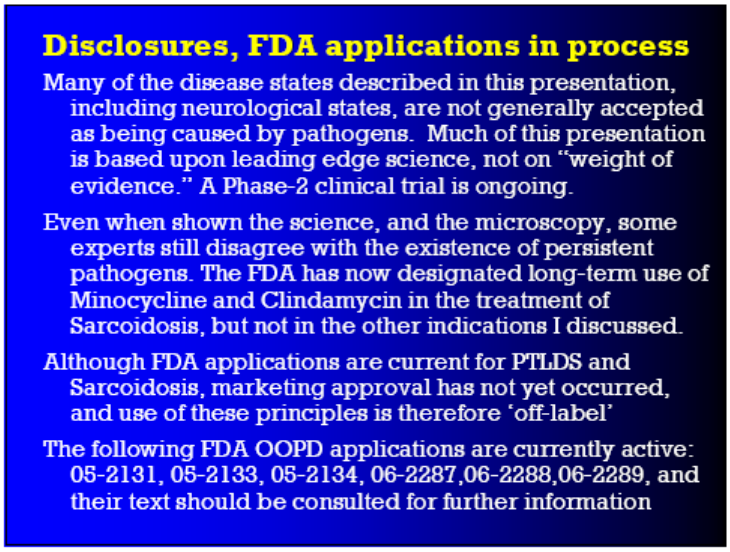
SLIDES: 1:51 / 1:23/02
Published Sources - Citations 2 slides

Thank you John, and thank you to the academy for inviting me to come.

Well the first two slides are just links to some of the papers that we've published on these topics. They are there for your references.

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SLIDES: **Disclosures, FDA applications in process**
2:19 / 1:23/02

Our FDA coordinator also told me to make sure that I point out that there are FDA applications current for both Sarcoidosis, designations in the drugs in Sarcoidosis, and also in PTLDS or Post Treatment Lyme Disease Syndrome, what most of you would know as chronic Lyme. Active, and the numbers are all at the bottom and you can find a copies of all of those applications on the Internet if you want to look in more detail at the regulatory issues.

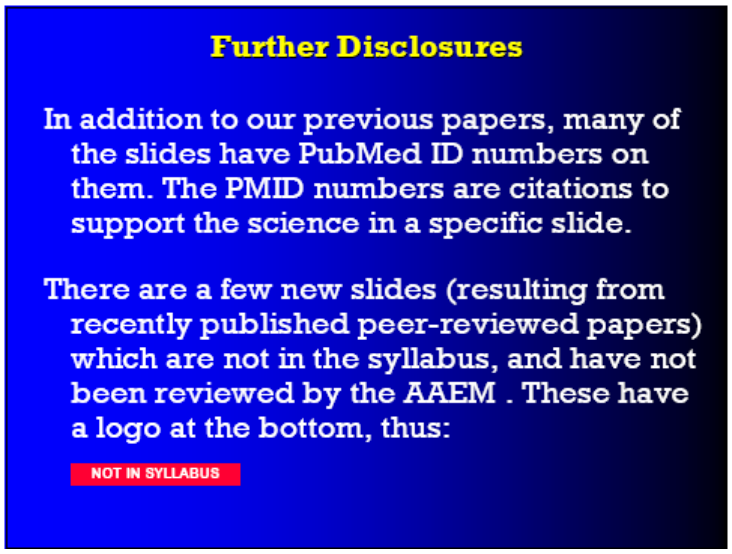
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SLIDES: 2:38 / 1:23/02

The disclaimers. Many of the disease states that I am going to cover in this presentation, including neurological states are not generally accepted as being caused by pathogens. Much of this presentation is based on leading edge science and not on weight of evidence. The Phase 2 clinical trial is ongoing.

Even when shown the science and the microscopy, some experts still disagree to the existence of persistent pathogens.

The FDA has now designated trials for long term use of Minocycline and Clindamycin in the treatment of Sarcoidosis but not in the other indications I am going to discuss. We are going to Phase 3 trials over the next year or two.

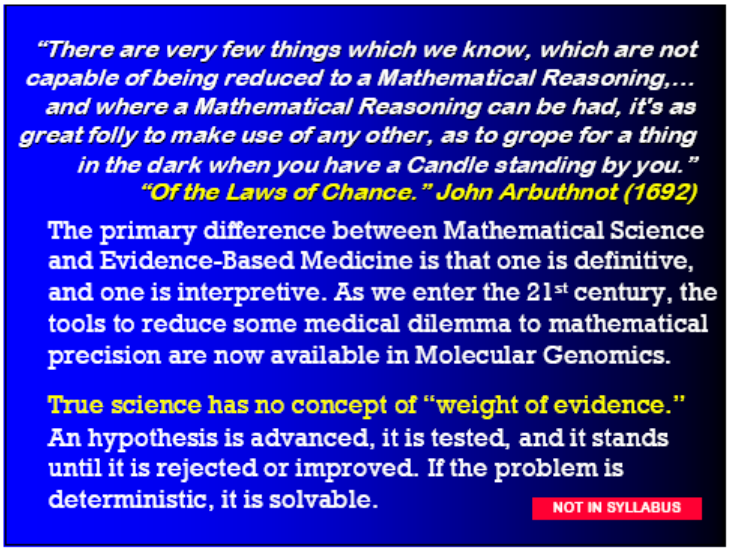


Now in addition to our previous papers, the list of which I've just gone through, many of the slides have Pub Med ID numbers on them. Those are citations to support the science in any specific slide. So if there is something on a specific slide that you feel I'm reaching a bit with, make sure you look at the Pub Med ID citations first.

And there are also a few new slides resulting from recently published peer review papers which are not in the syllabus and which have not been reviewed by AAEM. These have that little red logo at the bottom saying "NOT IN SYLLABUS."

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SLIDES: **John Arbuthnot Quote (1692)**
4:04 / 1:23/02



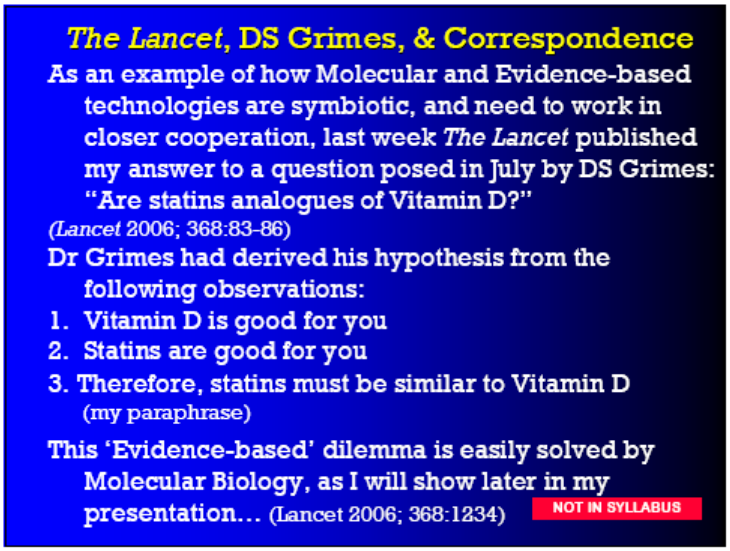
So here is the first one, "Not in Syllabus." Well, actually, the top part is in the syllabus but I changed the text on the bottom, as you will see on the next slide.

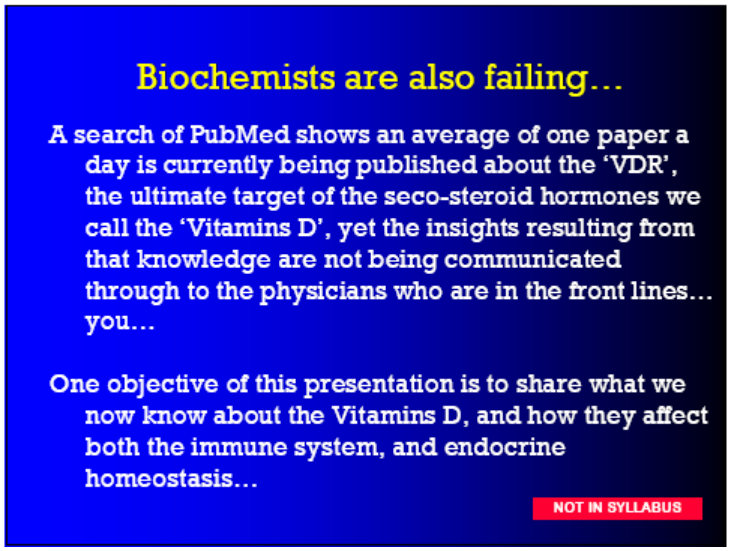
The quotation at the top, from a gentleman called John Arbuthnot, from Sixteen Hundred and Ninety two. Believe it or not, this quotation really forms the basis of evidence-based medicine. This is the gentleman that wrote the first book on statistics of the laws of chance in 1692 and as you would all know, without statistics and without the concepts of statistics, we wouldn't have the clinical evidence-base that we have at the moment.

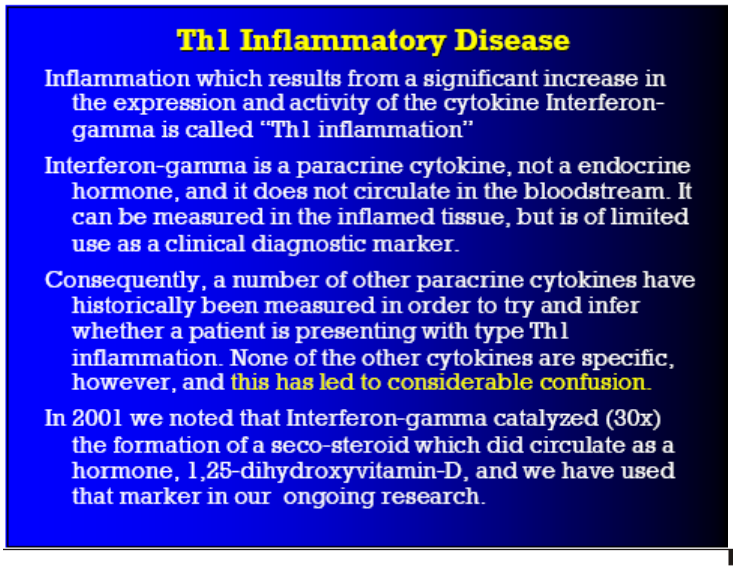
What John said [read quote on slide].

The primary difference between mathematical science, which is primarily what I'm going to deal with in this presentation, and evidence-based medicine is that one is definitive and one is interpretive. Mathematical science tries to define relationships between metabolites. Tries to define what's going on. Evidence-based medicine tries to interpret the results of observations in a reliable manner.

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As we enter the 21st century, the tools to reduce some important medical dilemma to mathematical precision are now available in Molecular Genomics. Now one thing I want to point out is that true science has no concept of weight of evidence. It doesn't matter how many times you add up the balance in your checking account, you can start at the bottom of the page and work up, you can start at the top of the page and work down, the numbers you are going to get are exactly the same and any body who has done high school arithmetic can sit down and check your calculations for you. So therefore, there is no need for weight of evidence.

As a hypothesis is advanced, it can be tested and it stands until it is rejected or improved. E=MC2 has been improved many times over the years, but the basic E=MC2 is still there, still in place.

If the problem is deterministic, then there is a solution.

.....

SLIDES: **The Lancet, DS Grimes, & Correspondence**
6:49 / 1:23/02

This is a new slide, and as an example of how molecular and evidence-based knowledge are symbiotic and the need for both fields to work in closer cooperation, last week The Lancet published my answer to a question which had been posed in July (2006) by Dr DS Grimes. Dr Grimes posed the hypothesis, "Are statins analogues of Vitamin D?"

[Read slide 1-3]

Now that's my paraphrase and I'm being a little bit cruel but I wanted to give you a quick encapsulation of what the hypothesis is about. This is a hypothesis which cannot be solved by evidence-based medicine. It cannot be solved because the variables that are involved, the number of variables that are involved is too high and they are not appreciated by the investigator.

When you have too many degrees of freedom, your statistical analyses of the epidemiology just break down. However, the evidence-based dilemma is easily solved by Molecular Biology and I'll show you this later in the presentation. I'm going to be talking about statins. That was actually in the syllabus.

.....

SLIDES: **Biochemists are also failing...**
8:27 / 1:23/02

But I wanted to point out that biochemists are also failing. [Read slide to 'Vitamins D']. The VDR stands for Vitamin D Receptor. [Continue reading slide to knowledge] Yet the insights that are resulting from science that the biochemists are producing [continue slide].

.....

SLIDES: **Th1 Inflammatory Disease**
9:16 / 1:23/02

First I need to say little bit about Th1 inflammatory disease. [Read slide first Paragraph]

Unfortunately, interferon-gamma is a paracrine cytokine. It exists only in the tissues and it's not an endocrine hormone. It doesn't circulate through the bloodstream. [Read 2nd Paragraph to tissue] if you want to do a biopsy [finish 2nd Paragraph].

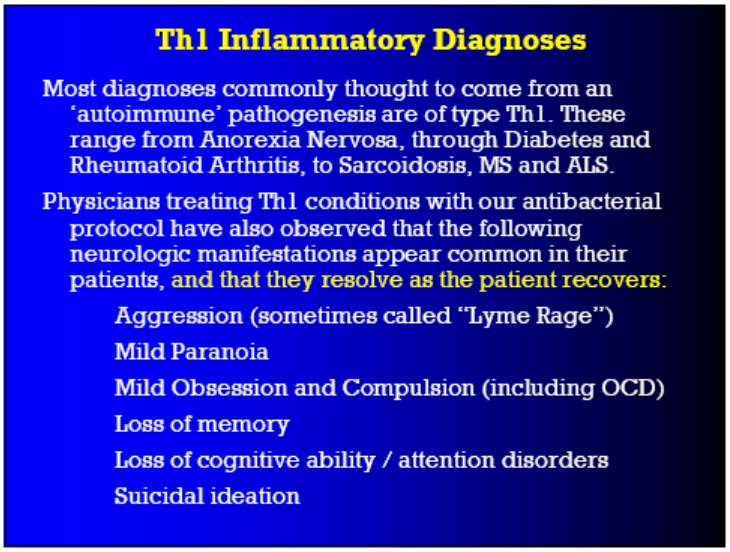
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Consequently, a number of other paracrine cytokine have historically been measured in order to try and infer whether a patient is presenting with a type Th1 inflammation, or a type Th2 inflammation. A type Th1 inflammation is typically those which result from infection, such as Tuberculosis, Leprosy, Syphilis. All those are Th1 or they cause Th1 immune responses. Th2 is more typically those which result from what we call allergies. However, none of these other cytokine are specific and this has led to considerable confusion. People think in terms of Th1 and Th2, but basically all of the autoimmune diseases are Th1.

[read final paragraph from slide]

Louis Pasteur once noted that in science, chance favors the prepared mind. There are 2 factors: Chance, it's all based on luck (chuckle), being in the right place at the right time. But the prepared mind is important. You have to be able to observe what you see, when you are there. We were lucky, we were able to break that particular nexus and realized that all of these diseases we were dealing with were in fact, Th1 diseases.

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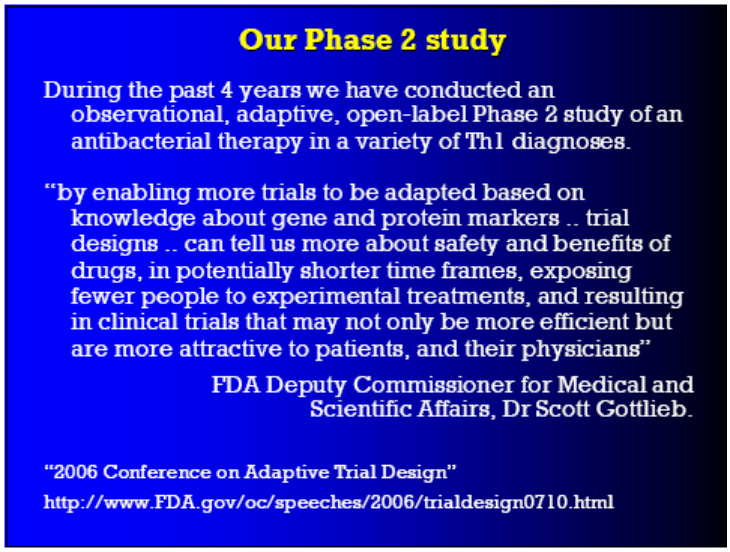


SLIDES: **Th1 Inflammatory Diagnoses**
11:48 / 1:23/02

[read slide]

... Suicidal ideation, this is not something I've heard anybody speak about, so far at the conference. It's a big problem with us. As the immunopathology takes over, the cytokine storm hits. Suicidal ideation occurs in a significant number of these people and it's treatable. As a matter of fact, it's avoidable.

.....



SLIDES: **Our Phase 2 study**
13:06 / 1:23/02

[read first paragraph]

Now those of you that know about the Marshall Protocol and have been following what we have been doing over the last five or six years would know that we've conducted this on the Internet as an open study. Any physician such as yourselves can register and get full access to the database of all the reporting from the patients in the study going right back to day one, right back to about 2001.

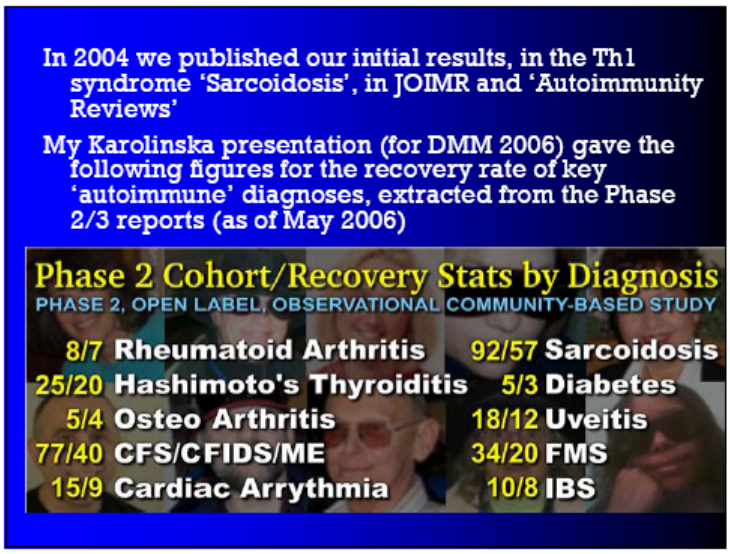
A year ago, this was heresy. Everybody said you can't do an open study. You can't do an adaptive study. You can't change the conditions as you are going along. You can't do anything on the Internet.

Well, things change. The drug pipeline is now nearly empty and the FDA is getting quite concerned that not only is the drug pipeline nearly empty, but the drugs that are there don't work properly anyway.

And so I put a quote in there now which is given by the FDA Deputy Commissioner for Medical and Scientific Affairs, who is actually in charge of clinical trial evaluation at the FDA, Dr Scott Gottlieb, from an actual conference that was held earlier this year on adaptive trial design. And basically, I'll save time by saying that he says [paraphrase] "we've got to figure out better ways of doing these things, guys." And whenever I read the stuff he writes, it makes me think of the things we are doing.

.....

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SLIDES: **Karolinska**
14:52 / 1:23/02

Now we published our initial results in 2004 in the Th1 syndrome called Sarcoidosis. Sarcoidosis is a rare disease and it's a disease that we chose to investigate first. We were lucky to choose this one, it happens to be a prototypical case for all of the Th1 diseases. And there are a number of things in Sarcoidosis which makes the analysis of the disease state very much easier than it is in some of the more complex conditions.

And in particular, by the time the inflammation has advanced to the stage where it forms self-perpetuating granuloma, there are very few antibodies being formed. So there is no longer the red herring of the antibody formation and perhaps the antibodies making people sick. We knew the antibodies weren't making people sick because there weren't any. We knew there was something else that was making people sick, probably a cytokine storm. Where was it coming from?

That was the other Pasteur piece of luck. We happened to be studying the correct disease that turned out to be the prototype for many, many other Th1 diseases.

So at Karolinska institutet in Stokholm, at DMM 2006, I gave the following figures for the recovery rate in several key autoimmune diagnoses. These were extracted from the Phase 2 and 3 reports. These are people that have mainly been on the protocol for twelve months or so on average.

As you can see, Rheumatoid Arthritis has very good success. Eight have a primary diagnosis of Rheumatoid Arthritis, seven of them are reporting improvement in their condition.

With Hashimoto's Thyroiditis it is a lot easier. Either they need the supplements or they don't. There the ratio is twenty out of twenty-five are improving.

Osteo Arthritis is an interesting one. I was surprised how many people with Osteo Arthritis we had in the cohort.

And then the ones that we are known for: CFS/CFIDS/Myalgia Encephalmyelitis. Let me stick with Chronic Fatigue Syndrome. Seventy-seven with about forty reporting ongoing resolution.

Cardiac Arrhythmia is an interesting one. This came up in a case study yesterday. And I would like to have an opportunity to join this group again in about five years time when we have more data and a lot more history under our belts. And talking again about cardiac arrhythmia, this is a very classic symptom of late stage Th1 disease.

Sarcoidosis of course, ninety-two with fifty seven recovering.

Diabetes, five and three.

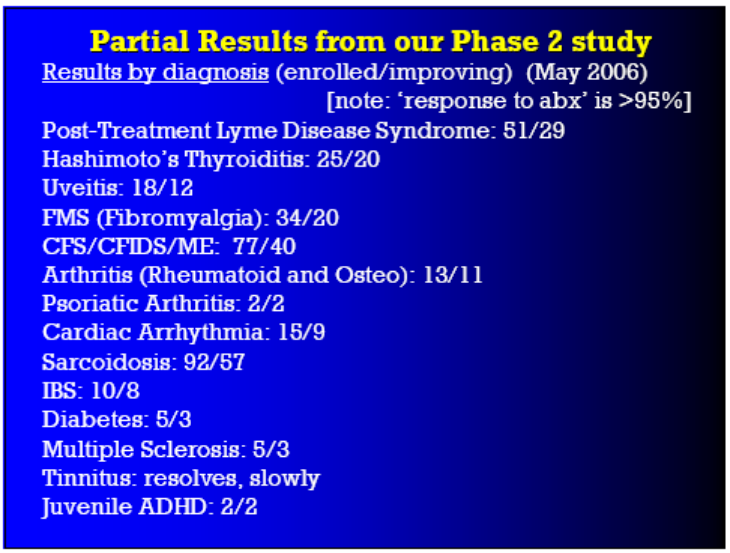
Uveitis, eighteen and twelve.

34/20 in Fibromyalgia and 10/8 in Irritable bowel syndrome.

It is an open study, we don't put any preselection conditions on the patients that want to join, and that's why we have accumulated such a wide range of diagnoses. There are about 500 patients at the moment that are regularly reporting and about ten times that number that we are aware of... of course, that don't report.

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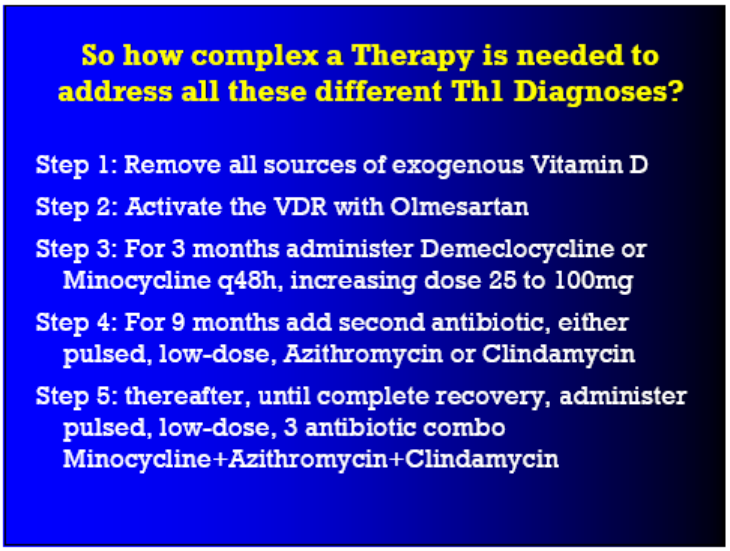
SLIDES: **Partial Results from our Phase 2 study**
18:39 / 1:23/02

There are a few more diagnoses on this slide. It is in your syllabus so I won't go over them in too much detail. Point out Psoriatic Arthritis is a new one, also Multiple Sclerosis. We have quite a few Multiple Sclerosis patients now, at least it is getting up to statistical significance. Most of those are being looked after by Dr Greg Blaney in Canada.

Tinnitus resolves. Usually acute Tinnitus resolves quite quickly and then takes quite a lot longer to resolve fully, several years to resolve fully. Like peripheral neuropathy, Tinnitus is one of the slow symptoms to resolve.

Juvenile ADHD, I've got two out of two and I've already chatted with the autism, the people that are interested in autism. That was quite a surprise to us. Both of those patients had primary diagnosis in other areas. One had a Lyme disease diagnosis, the other had Sarcoidosis and Uveitis diagnosis. The uveitis being key there. The patient was going blind. All of the problems have resolved. As the primary inflammatory diagnosis resolved, the attention and other problems, psychological problems, resolved as well. Resolved spectacularly. Both are back at school and doing very well.

.....



SLIDES: **So how complex a Therapy is needed to address all these different Th1 Diagnoses?**
20:19 / 1:23/02

Well, five steps basically. [Read to end of step 2]

Olmesartan is licensed for sale throughout the world as an angiotensin-receptor blocker. But it has significant activity in other receptors. One of the receptors it is very active in is the VDR. It activates the VDR.

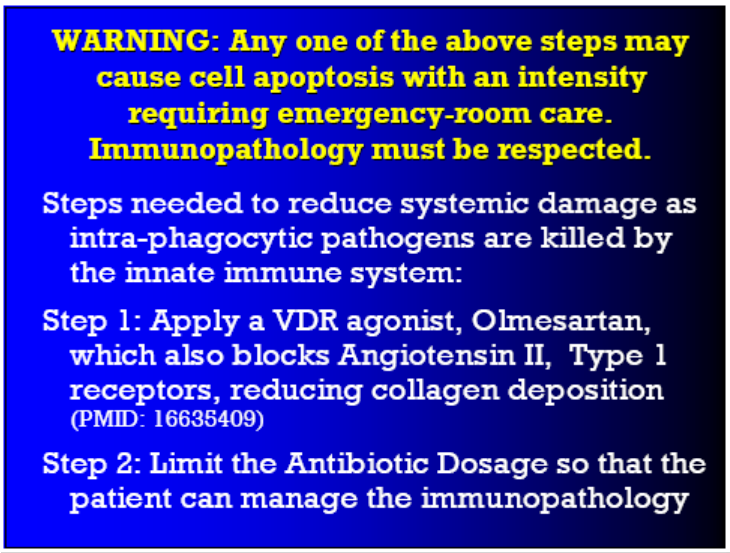
[Read step 3, to "25"] And when the patients start with 25mg every 48 hours it is usually all they can handle. And then increasing to 100mg over about three months.

[Read step 4] The choice is made based on suicidal ideation. Clindamycin tends to exacerbate any suicidal ideation or depression that is present and we avoid it in the early stages of the protocol with people who have that tendency.

[Read step 5 to 3 antibiotic combo] "Low dose?", well Azythromycin, some of them can handle 1/8 of a tablet every ten days when they start. They eventually work up to about half of an Azythromycin tablet as ultimately the bacterial load is totally removed, and then they can just guzzle the antibiotic like candy, like a healthy person can.

But initially, when the immune system is working, those antibiotics do an incredible job of killing the bacteria.

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SLIDES: **WARNING**
22:20 / 1:23/02

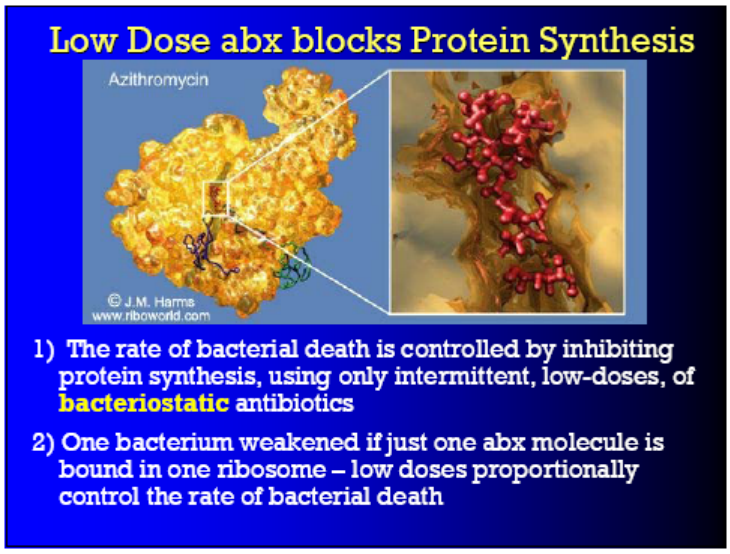
So, this was a warning that my FDA liaison — contact — said "please put in."

[Read warning to apoptosis] programmed cell death [continue warning].

Immunopathology. That is the word that is being used instead of

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"Jarisch-Herxheimer." Jarisch-Herxheimer typically refers to the cytokine storm that occurs in acute episodes. "Immunopathology," I first heard Rolf Zinkernagle use it at Karolinska and it is becoming more and more commonly used. It is basically what happens to the body when the immune system is doing its job. It includes apoptosis, cytokene storm, all of the other effects on the body from the immune system doing its job. Immunopathology.



[Read Steps needed].

The moment pathogens start being recognized and start being killed, your white and red blood cell counts are going to go all over the place.

[Read Step 1 to receptors], which is rather handy to reduces fibriotic tissue deposition.

[Finish and read final phrase]

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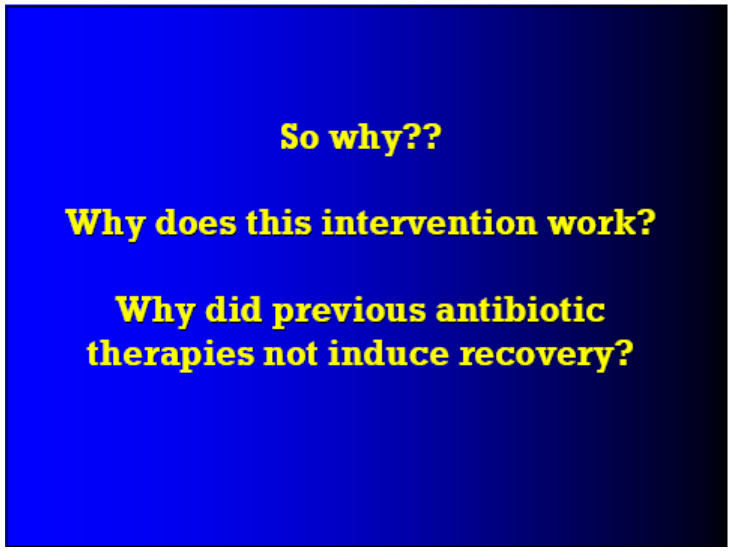
SLIDES: **Low Dose abx blocks Protein Synthesis**
24:07 / 1:23/02

Now we use special antibiotics. You might not think they are special, but they are special in their interaction. The antibiotics have a direct action on a part of the bacterial genome which is called the ribosome. The ribosome, if you cast your mind back to molecular biology, is where the messenger RNA from gene transcription, are turned into protein.

In bacteria, they only have one ribosome. The human body has two ribosomes. Bacteria only have one, called the 70S ribosome, and the antibiotics that we use block the action of the 70S ribosome. They block the ability of the bacteria to produce protein. Particularly to use the protein that they use to evade the immune system. Because clearly, the bacteria wouldn't be capable of living within the phagocytes unless they had a mechanism evolved probably over centuries that allows them to evade phagocytosis.

So the rate of bacterial death is controlled by inhibiting the protein synthesis, using only intermittent low doses of bacteriostatic antibiotics.

The other advantage of going after the ribosome is a small amount of antibiotics kills a small number of bacteria. In fact, one molecule binds into one ribosome. So it is not quite linear, but it is a proportional response.



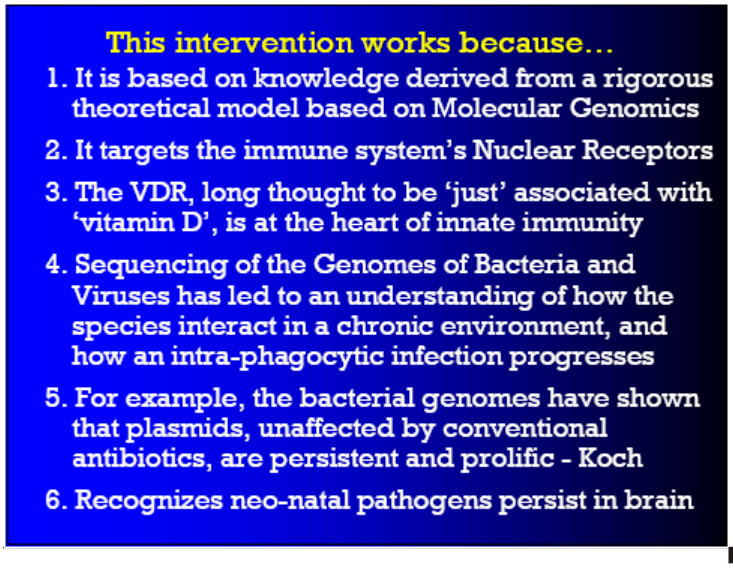
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SLIDES: **So why??**
25:53 / 1:23/02

So why? Why does this intervention work when all the previous antibiotic therapies used for these diseases didn't induce recovery?

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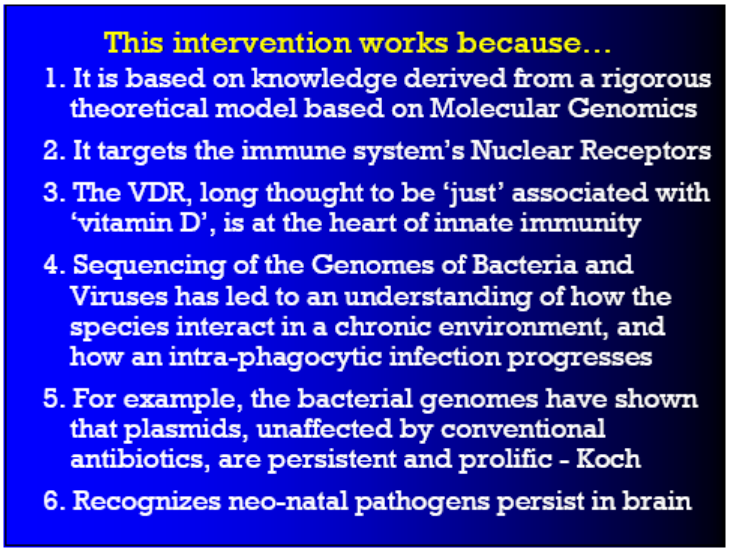
SLIDES: **This intervention works because...**
26:40 / 1:23/02



[Read 1]

In other words, there is a model. We can fit the disease that we observe to a model. So when I say that five MS patients is a good sized sample, its because they are behaving exactly as the model predicts. So we have the extra statistical guidance from the model as well as from the data.

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It works because [Read 2].

[Read 3] The VDR, which was long thought to be 'just' associated with 'vitamin D', is now known to be at the heart of innate immunity. And I'll talk a LOT more about this as we go forward. [emphasis repeat] Vitamin D is at the heart of innate immunity.

[Read 4]. That is part of the model. We have to know how the people get sick, and how the sickness progresses over the years.

[Reference 5]. For example, sequencing the bacterial genomes have shown that many of the common bacteria, even Staph aurious, has got plasmids. Plasmids are little self-replicating loops or sticks of DNA that can exist and replicate on their own without the main chromosome. And many bacteria have these plasmids. The plasmids can persist and if they are not destroyed by the immune system, they can interact with other species of bacteria.

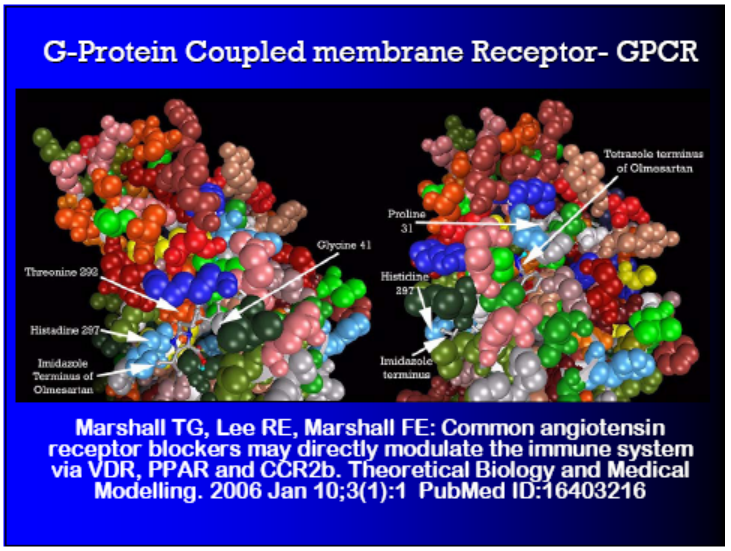
Toward the end, I will show you exactly how that happens with a bacterium species called antrhax.

So the plasmids are persistent and prolific. So when you have a chronic disease which is accumulating over a lifetime, certainly over many years, you're not going to have just one species that is active. You are going to have a number of species that are active. You are going to have a number of plasmids that are active. You are going to have a number of viruses that are active.

The ubiquitous EBV. I think I can find a paper for just about any disease you like name which blames EBV as the cause of that disease. No. But EBV is very hard to kill off. Its plasmids is just two genomes long; something like a 160,000 base pairs long; it's very, very small; very, very hard for the immune system to recognize and kill; and it persists. And it interacts with any other species of pathogens in exactly the same way as it interacts with the human genome. And it will cause mutations. The bacteria cause mutations.

And the last reason the intervention works is [reference 6] because the MODEL recognizes that neo-natal pathogens persist in the brain. [Repeat intended] Neo-natal pathogens persist in the brain.

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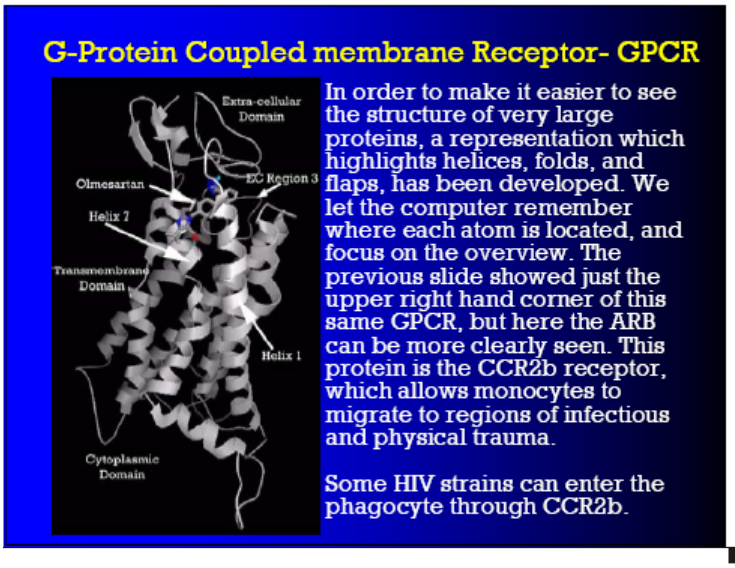


SLIDES: **G-Protein Coupled membrane Receptor-GPCR (balls)**
29:34 / 1:23/02

Well, now we get into some pretty pictures.

These are pictures of a receptor, it is a G-Protein Coupled Receptor that goes through a membrane. And each of the little colored balls are atoms. Each of the similarly colored atoms form part of an amino acid, which together make the protein which is folded into the receptor.

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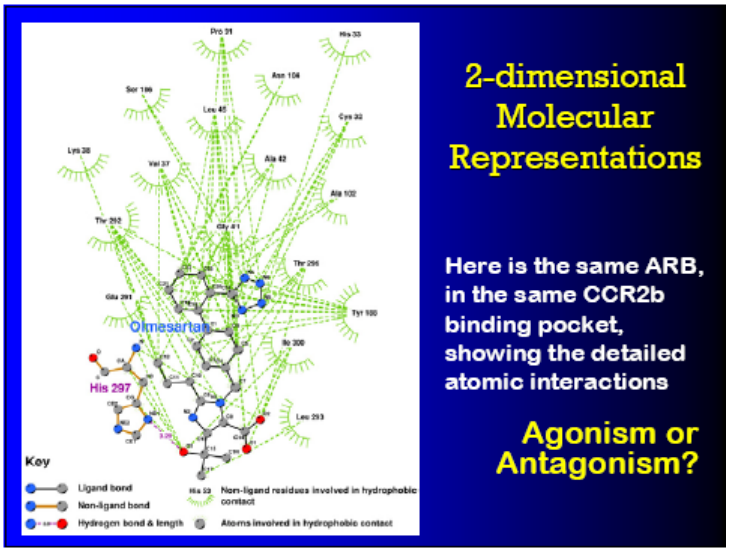


SLIDES: **G Protein Coupled membrane Receptor-GPCR (spirals)**
30:04 / 1:23/02

Well it's not very easy analyzing that sort of diagram, so biochemists tend to draw everything in terms of spirals and loops. Each one of these spirals is a cascade of amino acids. It is shown diagramatically as a spiral so that we can see more easily what it is actually doing.

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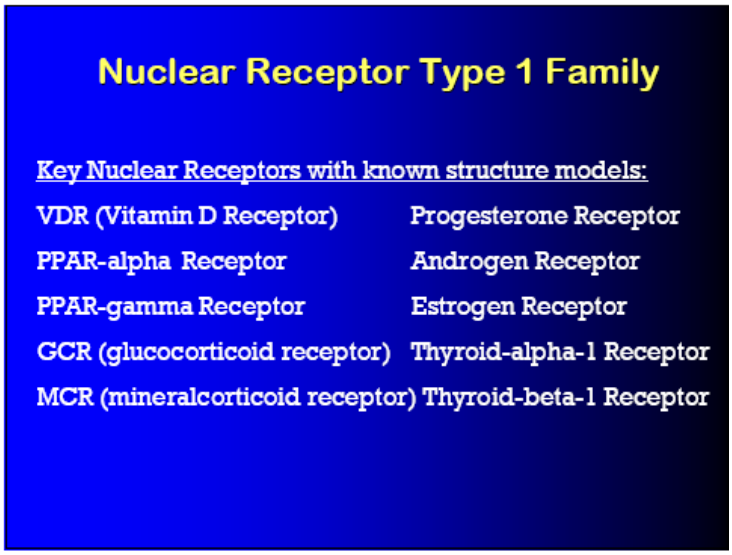


SLIDES: **2-dimensional Molecular Representations**
30:25 / 1:23/02

We also have a more precise 2-dimensional representation that you will find in some of my papers. And here we have the actual amino acid of the protein (see lower right "Leu 293" highlighted in video) and it shows which are the atoms or ligand of the chemical. In this case it is olmesartan but it could be any of the vitamins D.

It shows which of the atoms interact with the ammino acid residues (see lower right O1 red atom highlighted in video), and it also shows the hydrogen bonds when they exist as well. Making it very easy to try and figure out whether we are dealing with an agonist, or an ANtagonist. Much harder to do that in 3-dimensions.

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SLIDES: **Nuclear Receptor Type 1 Family**
31:10 / 1:23/02

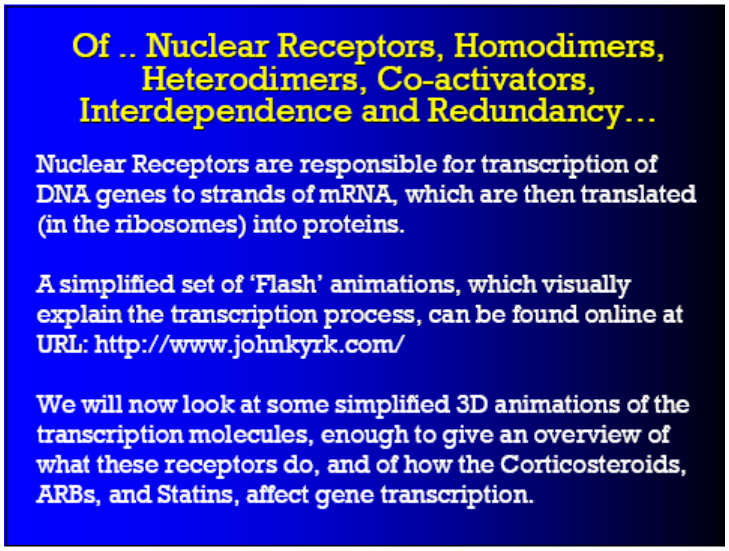
I want to talk about the nuclear receptor type one family because it turns out that this family is most closely related to the operation of the immune system — both the innate immune system, and the adaptive immune system.

The key receptors for which we have known structure models. In other words, we not only know that the bacterial genome looks like this but we also know in 3-dimensional space that x-ray spectroscopy has told us exactly where each atom is in the molecule. In this case, we're talking about the human genome. But it's the same theory, we've got the VDR (Vitamin D Receptor), the Progesterone Receptor, PPAR-alpha (which is one of the lipid and immune system mediators), PPAR-gamma (which is also lipid and immune system), Androgen Receptor, the Estrogen Receptor, glucocorticoid receptor, Thyroid-alpha-1, Thyroid-beta-1, and the mineralcorticoid receptor.

Now surprisingly, if you look at the pictures of each of these drawn as spirals (I guess maybe I should have done a slide of it for you), but they are all very, very similar. And that is why they are the type 1 nuclear receptors.

And you know molecules that will go into the VDR usually only go into PPAR-gamma and some of the other type 1 receptors as well. They affect more than one function of the body.

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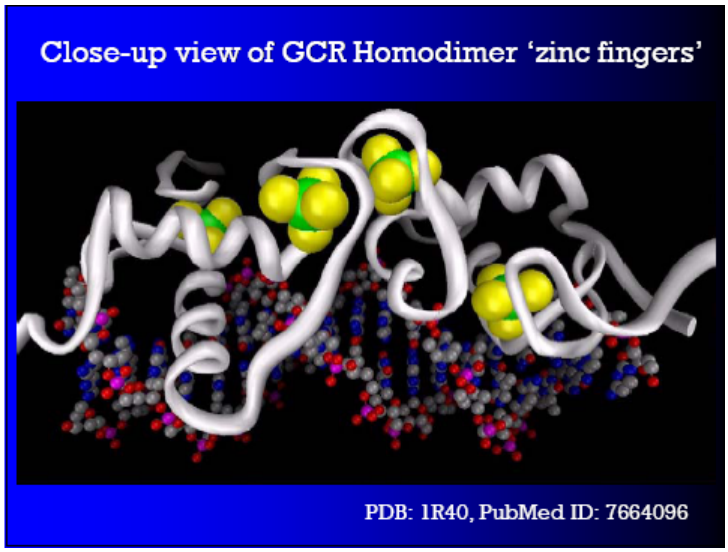
SLIDES: **Of .. Nuclear Receptors, Homodimers, Heterodimers, Co-activators, Interdependence and Redundancy...**
32:47 / 1:23/02

So nuclear receptors are responsible for the transcription of the DNA genes to strands of RNA, which are then translated into proteins in the ribosomes. And they are very important.

I've put a link there to a simplified set of flash animations for those of you that want a crash course on how this all works. It is a very simple crash course. I really recommend you have a look at it. (http://www.johnkyrk.com/)

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SLIDES: **Close-up view of GCR Homodimer 'zinc fingers'**
33:17 / 1:23/02

Now we're going to look at some simplified 3-D animations of the transcription molecules, just to give us an overview.

What we have here, we have a strand or double strand of DNA (highlight blue/grey/red/pink balls base right of diagram). These are the base pairs that run across (highlight on video shows vertical ball lines at bottom center of diagram) the two backbones of the DNA. Across the center you have the hydrogen bonds that hold the DNA together (highlight moves L-R across center of vertical ball lines). Hydrogen bonds that are peeled apart is when the RNA is produced. The RNA is basically ½ of the DNA double spiral.

So when the nuclear receptors do their job and transcribe a gene, what they are actually doing is breaking the hydrogen bonds and producing the RNA in a very complex and clever fashion which doesn't occur very fast because it requires a lot of things, the statistically drift around and come together in exactly the right way.

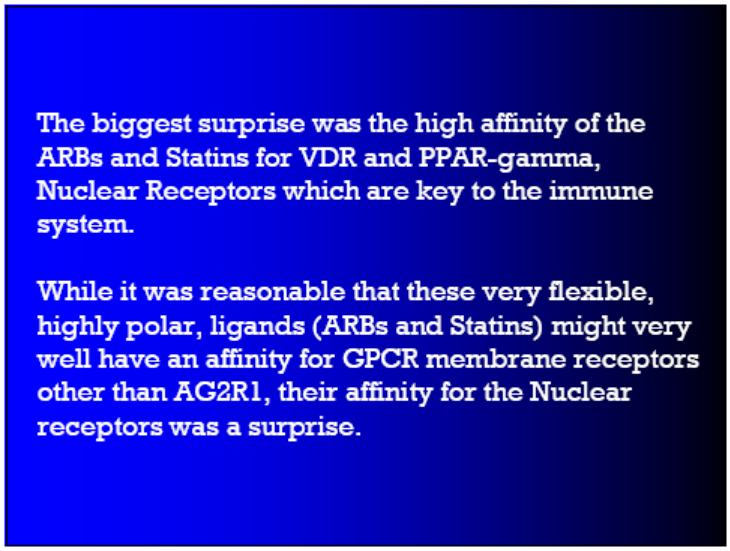
Nuclear receptors typically take days to effect their action.

Key to nuclear receptors are the zinc fingers here (video highlights upper left center large yellow/green balls). The green atoms are zinc, the yellow, sulpher-disulfides. Actually cystines to be honest. And they are a key part to positioning the helixes which go through the (video highlights lower left solid light grey spiral) DNA and find the exact chain that needs to be translated based on the exact mapping of attractive and repulsive forces and then do the DNA transcription.

It is an amazing system. I've shown here that there two receptors because usually one nuclear receptor doesn't do it by itself. It can form a homodimer but it usually forms a Heterodimer. So you have a VDR coupled with a retinoid-X receptor and then the two of them transcribe genes. There is also a cofacter that gets involved as well. Quite complex and we really don't know a lot about it yet.

I mean we now have the genes to transcribe, but we don't know exactly which co-factors go with which receptors.

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SLIDES: **The biggest surprise...**
35:30 / 1:23/02

The biggest surprise we got when we started doing this modeling, was the high affinity of the ARBs and Statins for VDR and PPAR-gamma, Nuclear Receptors which are both key to the immune system.

Now these are very flexible, highly polar ligands. And we thought they might have an affinity for other G-protein coupled receptor membrane receptors, in other words receptors that go through a membrane rather than exist in the nucleus of the atom.

But their affinity for the Nuclear receptors was a real surprise.

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Estimated Ki for ARBs and Statins into NRs

Drug est. Ki	VDR	PPARg	PPARa	GCR	MCR	PR	AT	BT
Candesartan	30	61	3	6	16	7	0.4	0.7
Irbesartan	10	6	0.9	0.8	47	4	6	0.5
Losartan	74	3	4	4	2	0.6	2	0.5
Olmesartan	10	12	3	1	4	0.3	28	2
Telmisartan	0.04	0.3	0.7	2	no	no	no	no
Valsartan	14	12	26	10	2	4	6	1
Atorvastatin	no	4	2	1	no	no	no	no
Fluvastatin	no	12	1	3	8	13	36	2
Lovastatin	10	0.2	19	15	2	0.5	2	0.3
Pravastatin	62	21	2	8	6	2	80	0.3
Rosuvastatin	no	24	18	7	3	14	no	0.6
Simvastatin	4	0.3	4	2	2	0.4	5	0.3

SLIDES: **Estimated Ki for ARBs and Statins into NRs**
36:09 / 1:23/02

Here's a big table which represents months of computing.

But what we have done here is used special software that computes the interaction between thousands of atoms and figures

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Estimated Ki for ARBs and Statins into NRs

Drug est. Ki	VDR	PPARg	PPARa	GCR	MCR	PR	AT	BT
Candesartan	30	61	3	6	16	7	0.4	0.7
Irbesartan	10	6	0.9	0.8	47	4	6	0.5
Losartan	74	3	4	4	2	0.6	2	0.5
Olmesartan	10	12	3	1	4	0.3	28	2
Telmisartan	0.04	0.3	0.7	2	no	no	no	no
Valsartan	14	12	26	10	2	4	6	1
Atorvastatin	no	4	2	1	no	no	no	no
Fluvastatin	no	12	1	3	8	13	36	2
Lovastatin	10	0.2	19	15	2	0.5	2	0.3
Pravastatin	62	21	2	8	6	2	80	0.3
Rosuvastatin	no	24	18	7	3	14	no	0.6
Simvastatin	4	0.3	4	2	2	0.4	5	0.3

out for each of the (video highlights drug names in the sartan group) ARBs and each of the Statins (video highlights drug names in the statin group), which receptors they are going to have an affinity for — if any — and which receptors they are going to switch on or switch off.

And if we just look at the statins first, thinking back to the question posed by Grimes in The Lancet: "Are statins analogues of vitamin D?" Well the first thing that pops out is that the VDR here (video highlights VDR column head), there is a huge difference between the statins.

Simvastatin (video highlights 4 at base of the VDR column) hs got quite a low number which means quite a high affinity, but Rosuvastatin, fluvastatin, and particularly Atorvastatin (Lipitor) don't have any affinity for the VDR at all. Pravastatin that is effectively no affinity. Lovastatin would have a little affinity if a low dose is used in vivo.

So there is a huge difference between the statins. Just thinking of the statins as a group is a concept that is dangerous clinically, let alone to try and extrapolate that to talking about the statins functioning as a hormone would.

But you can also see if you take Simvastatin, not only does it have a high affinity for the VDR, it's got 10x higher affinity for the PPAR-gamma, same for PPAR-alpha as the VDR. The Gluco-corticoid receptor -the home for cortisole- it has a high affinity for, also the MCR. The Progesterone receptor, the Alpha-Thyroid and the Beta-Thyroid. Simvastatin is a very, very active statin.

Whereas Atorvastatin for example, primarily hits PPAR-gamma and alpha and incidently hits the Gluco-cortisole axis for the GCR.

However, some of the ARBs are a little bit better behaved than that. Telmisartan is very poorly behaved. It totally shuts off the VDR the moment you give it to somebody. This is NOT good.

Olmesartan, here (video highlights 10 in the VDR column) has got a moderately low affinity for the VDR, and that is one of the reasons why we have to use a higher dose when we are using it to modify the VDR than when we are modifying the Angiotensen-receptor where it has a much higher affinity. But it is nicely controlled in the PPAR-gamma and -alpha (video highlight across Olmesartan Estimated Ki row) and GCR. It hits the progestrone receptor but it doesn't really hit Alpha-Thyroid.

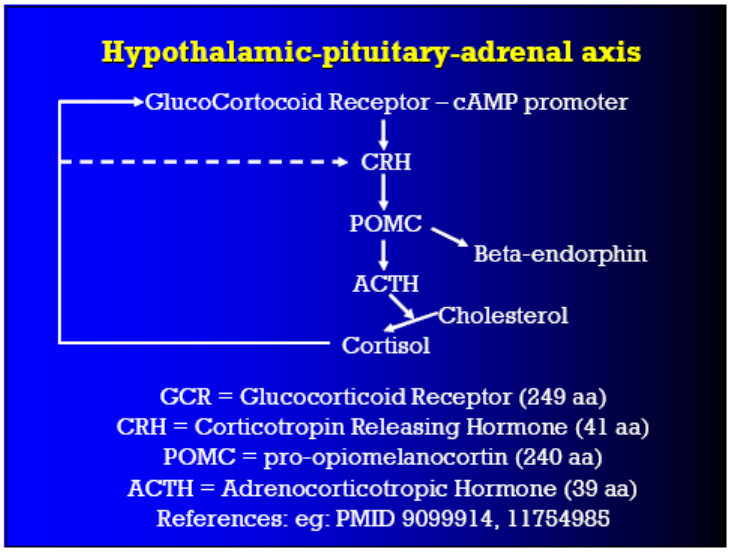
Beta-Thyroid it hits but we're not really sure what Beta-Thyroiod does yet. What we know about Beta Thyroid is that if you breed mice that don't have the beta thyroid receptor, then they are born deaf. Otherwise they seem to be fine. (Audience chuckles) So we're still trying to figure out what the Beta Thyroid does.

The ability to breed mice with gene knock-outs without these receptors is a PRIMARY new tool that has become available to the biochemist. It is invaluable. Unfortunately with the GCR, if you knock out the GCR the mice never survive past gestation. So we don't know a heck of a lot about the GCR either except it is at the root of the Cortisole axis of the adrenal.

But anyway, it is a big table and I have it printed on a sheet of paper and I have to refer to it all the time, so I'm not going to expect any of you to commit that to memory.

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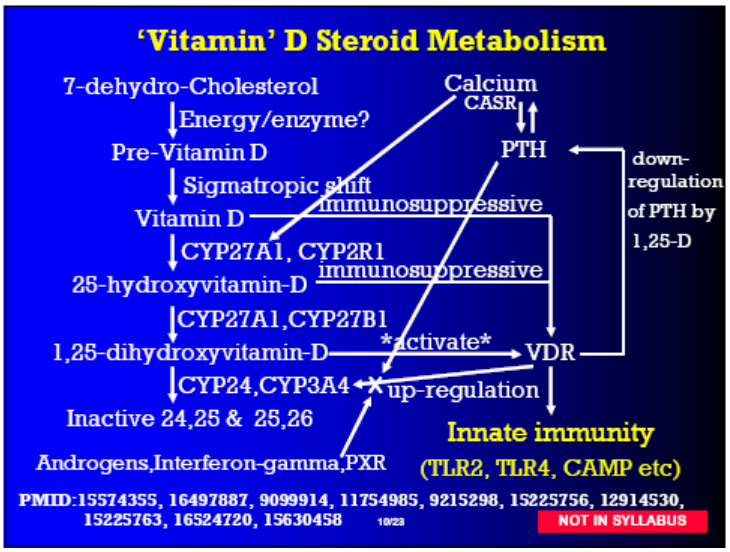
SLIDES: **Hypothalamic-pituitary-adrenal axis**
40:12 / 1:23/02

Now I'm going to look at a very simple hormonal system. The adrenal axis. And up here (video highlights top word "GlucoCortocoid Receptor"), I'm just looking at part of it and I'm looking at the GlucoCortocoid Receptor, the nuclear receptor, with a promotor which is "c" AMP (cAMP). Promotor binds to the receptors, and then the promotor and the receptor together bind to the DNA and transcribe the gene.

The GlucoCortocoid Receptor is responsible for producing Corticotrophin Releasing Hormone (CRH highlighted), and that in turn produces the Pro-Opeo-Milleno Cortin and when that is metabolized, you get Beta-endorphin and also an ACTH produced. ACTH of course you know, that catalyzes the conversion of Cholesterol into Cortisol. Then Cortisol goes up and docs into the GlucoCortocoid Receptor (highlight shows arrow line path left and up to GCR) and you have a feedback loop, which keeps the Cortisol levels of the body at roughly constant levels.

That is a very simple, straight forward hormone loop.

.....



SLIDES: **'Vitamin' D Steroid Metabolism**
41:24 / 1:23/02

Unfortunately, with 'vitamin' D, it is a little bit more complex. As you would expect for something that is at the heart of the immune system. It has adapted over many, many millennia.

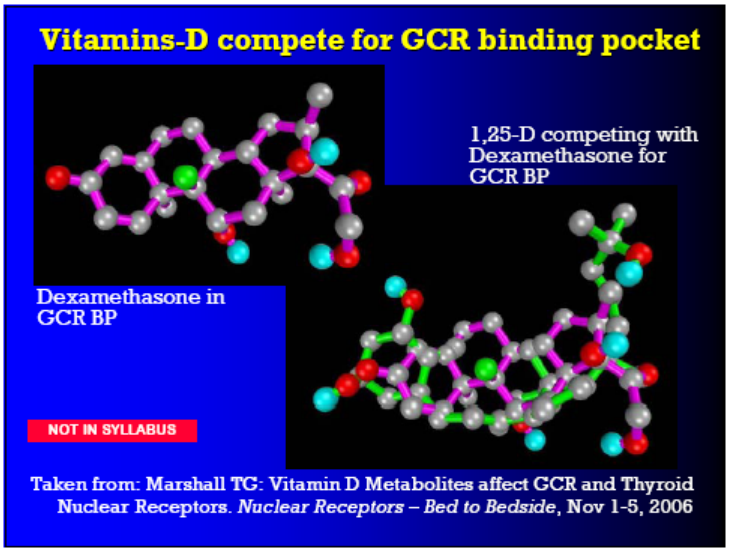
The "not in Syllabus" flag indicates some of the enzymes here have shifted from what is printed in your syllabus. Also, since that syllabus was published, there has been new studies showing that the Androgens (video highlights lower left corner word "Androgens"), particularly testosterone, reduce the breakdown of 1,25-D by the enzyme CYP24.

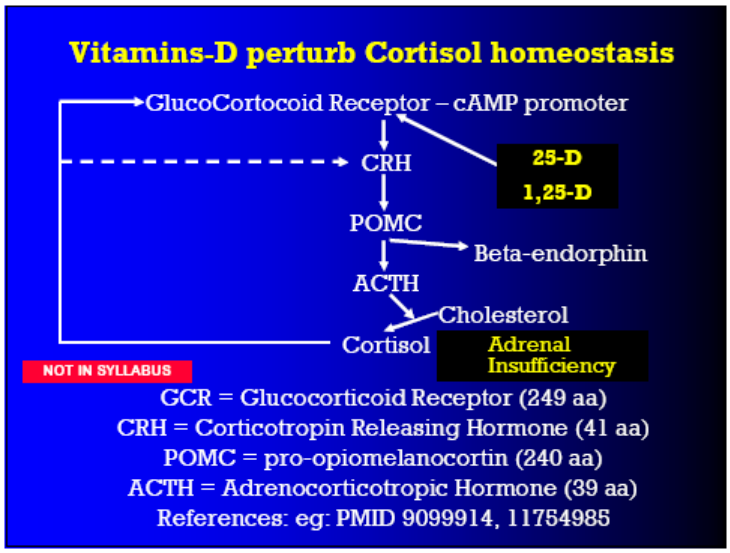
But basically, the body starts off with 7-dehydro-Cholesterol, which is part of the sterol subsystem. It is a cholesterol derivative (video highlights upper left 7-dehydro-Cholesterol), and that is converted to pre-Vitamin D by energy. Conventionally thought to be UVB, but the biochemists will easily tell you it doesn't have to be UVB and it can even be an enzyme. Enzyme at this point, we have no idea what that enzyme would be if it is an enzyme. But it certainly doesn't have to be UVB to convert to Pre-Vitamin D (Pre-Vitamin D highlighted). All you need is a contra-rotatory electrocyclic reaction which occurs in other substances and which is pretty well characterized.

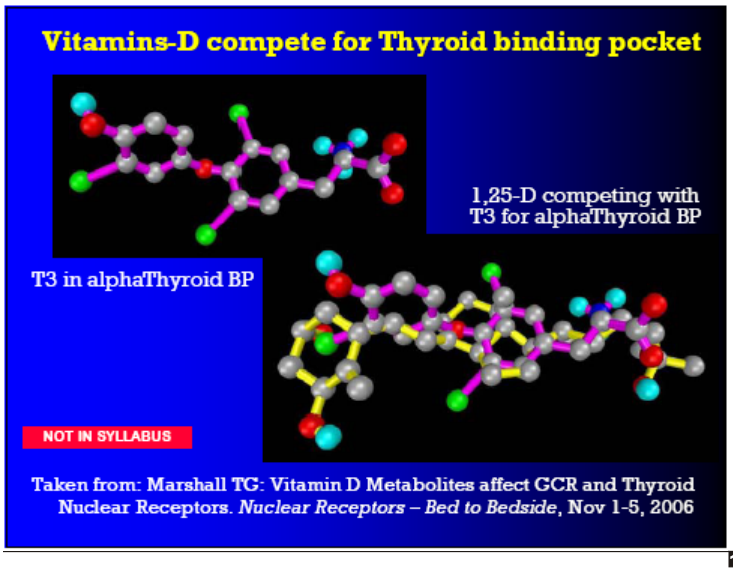
(Vitamin D highlighted) Then, Pre-Vitamin D is formed into Vitamin D by a Sigmatrophic shift. A very small shift of the electrons (????). Vitamin D is converted by the enzyme CYP27A1 -or- CYP2R1 and both of these are P450 digestive system enzymes into 25-hydroxyvitamin-D (highlight drops to 25-hydroxyvitamin-D) and this is the one that most of you are measuring when you measure somebody's 'vitamin' D levels, you are almost certainly measuring the 25-hydroxyvitamin-D.

(Highlight drops to 1,25-dihydroxyvitamin-D) That then is converted to 1,25-dihydroxyvitamin-D, hydroxillation by enzyme either CYP27A1 or CYP27B1, both of them will hydroxyllate. And that 1,25-dihydroxillate with activate (highlight moves to *activate*) the VDR and allow the VDR to do the gene transcription that is at the heart of innate immunity.

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And then finally, the 1,25-D is inactivated (highlight opens on lower left "Inactive 24,25 & 25,26") by the VDR up regulating this CYP24 enzyme so that you don't end up with too much of it in the body.

And that is what goes wrong in Th1 disease. You end up with too much 1,25-D in the body because the VDR inactivation path — something goes wrong with it.

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SLIDES: **Vitamins-D compete for GCR binding pocket**
44:11 / 1:23/02

Well, OK. Here we have some pretty pictures. We have atoms and bonds. The grey atoms (highlight upper left picture, left grey atom selected highlight in video) are carbons, the red ones are oxygen, that green is a fluorine, That's a hydroxy OH there (highlight moving from green to lower center red/grey center bottom), the light blue is the H of the hydroxy. And this molecule is Dexamethasone in exactly the configuration where it binds into the Gluco-Corticoid Receptor (GCR binding pocket), you know the adrenal axis, the Gluco-corticoid receptor.

And here (video highlights lower right figure center), we see 1,25-Dihydroxyvitamin D competing with Dexamethasone for the binding pocket of the GlucoCortocoid receptor.

1,25-D has a very high affinity for the adrenal axis. This is new, it is not in the syllabus, it's taken from a paper which was accepted for the nuclear receptors conference next week, actually.

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SLIDES: **Vitamins-D perturb Cortisol homeostasis**
45:20 / 1:23/02

So what does that mean? If we go back to our nice simple adrenal axis, what does that mean?

Well it means that (highlight focuses on top black box 25-D and 1,25-D) 25-D and 1,25-D can both bind into the GlucoCortocoid Receptor and displace Cortisol from doing that.

When it does that, the amount of CRH and particularly ACTH is down regulated, and you end up with Adrenal Insufficiency (highlight on lower black box "Adrenal Insufficiency").

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SLIDES: **Vitamins-D compete for Thyroid binding pocket**
45:47 / 1:23/02

OK, the vitamins D also compete for the Thyroid binding pocket. I'm talking about the alpha thyroid here, the important one.

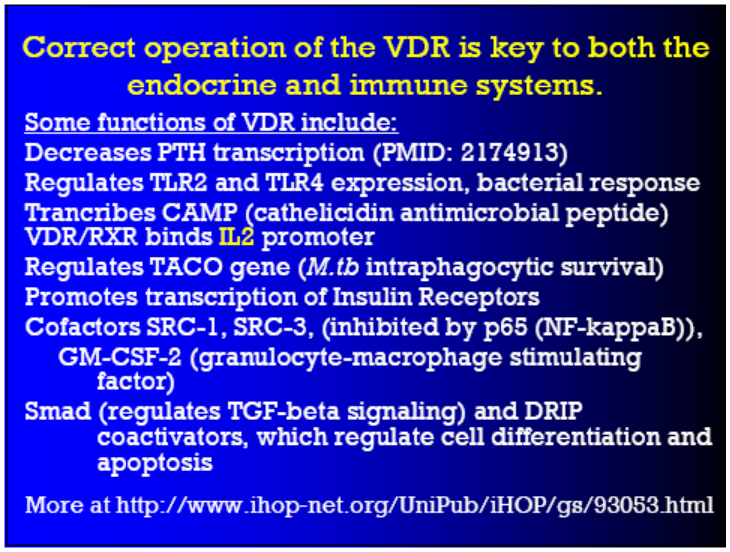
There we have T3 as it docs into the alpha thyroid to activate the alpha thyroid (highlight shows green end molecules on the top left illustration). These are the iodine molecules, three of them of course.

Here we have 1,25-D competing (highlight center of lower R figure) with T3 for the alpha thyroid Binding Pocket. It wants to dock in exactly the same binding pockets as the T3 would. And by doing that in high enough concentration, it will stop the T3 from binding. It will stop the alpha thyroid from working properly.

We end up with, among other things, hypothyroidism.

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SLIDES: **Correct operation of the VDR is key to both the endocrine and immune systems.**
46:37 / 1:23/02

So I've done a summary slide here of the operation of the VDR. It is key to both the endocrine and immune systems.

The VDR is responsible for decreasing parathyroid Hormone Transcription. [Reference slide to TLR4], and therefore the bacterial response.

Toll-like Receptor 4 is responsible for sensing lipopolysaccarhride. Toll-like Receptor 2 is responsible for sensing the presence of other bacteria protein. When the VDR is not working, because you have administered vitamin D and are experiencing the immuno suppressive action of the vitamin D, when the VDR is not working, you do not have a functioning TLR2 and TLR4 system.

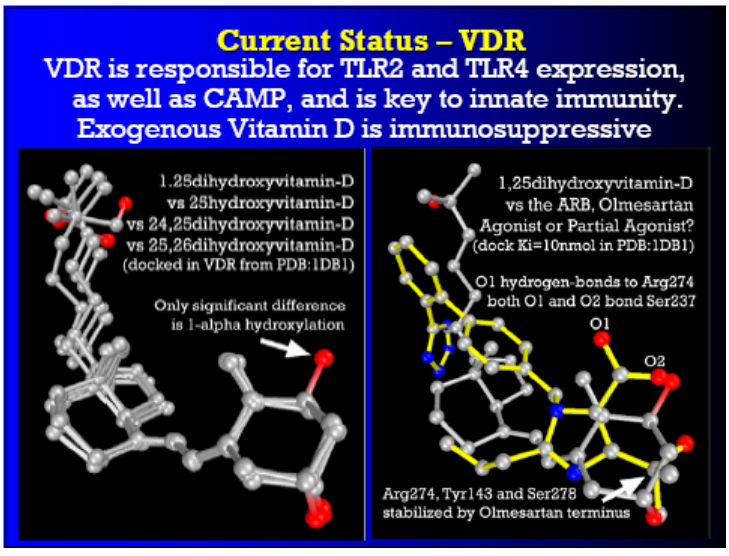
But you also lose the antimicrobial peptides. It transcribes the cathelicidin antimicrobial peptide (cAMP) and it's also responsible for beta defensins. The beta defensins are absolutely key to the proper operation of small intestine. The small intestine, in fact, the whole GI tract is critically dependent on the defensins and especially the beta defensins and the cathelicidins to work properly. When you knock out the VDR with immunosupression — either a steroid or 'vitamin' D — you lose that ability.

And finally, the VDR is responsible for binding the interluken 2 (IL2) promoter. Now you all probably know that you measure interluken 2 when you want to measure a Th1 reaction. Well, that is why, because the VDR actually produces the interluken 2, even if you can't measure it.

I've got some other functions there, I guess the main one there is it regulates the TACO gene, which is responsible for micobacterium tuburculosis intraphagocytic survival, and it promotes transcription of Insulin Receptors.

Let's skip the rest because we're running a little late bit short in time.

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SLIDES: **Current Status — VDR**
48: 53 / 1:23/02

The current status of the VDR is that the VDR is responsible for TLR2 and TLR4 expression, as well as cAMP.. hey, I just said that on the previous slide! Sorry.

What we've got here is two pictures and we have all the vitamin D's (video highlights left image) superimposed. We've got 1,25-Dihydroxyvitamin-D, that's the one that has the activating hydroxyl (highlight arrow and red atom in lower right of left image) here, the only one that can activate the VDR. Various other atoms can activate the GCR or de-activate the GCR and the thyroid, but the only one that will activate the VDR is this one (highlight again on arrow and red atom) on the 1,25-Dihydroxy. But you can see that all the vitamins D fit into the receptor in almost identical positions. Whether it is 24,25-D, 25,26-D or 1,25-D. Even vitamin D itself fits in there as well and they are all competitive.

If you're giving people 40mg, if you're giving people vitamin D such as such as to get the blood concentration at 25-D to 40 nanograms per ml, then you're sitting with 25-D in most of your VDRs and they are NOT activated, it is immuno-suppressive. The patient will feel better short term because the inflammation is

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suppressed. Long term, with the bacterial pathogens running right, it's a different situation.

The picture on the right hand side just shows Olmesartan binding into the VDR pocket and you can see if you look at it carefully, you can see that the Olmesartan hydroxyl here (video highlights upper right atoms O1 and O2 in lower right of right image) are actual capable of activating the VDR in the same way as 1,25-D does.



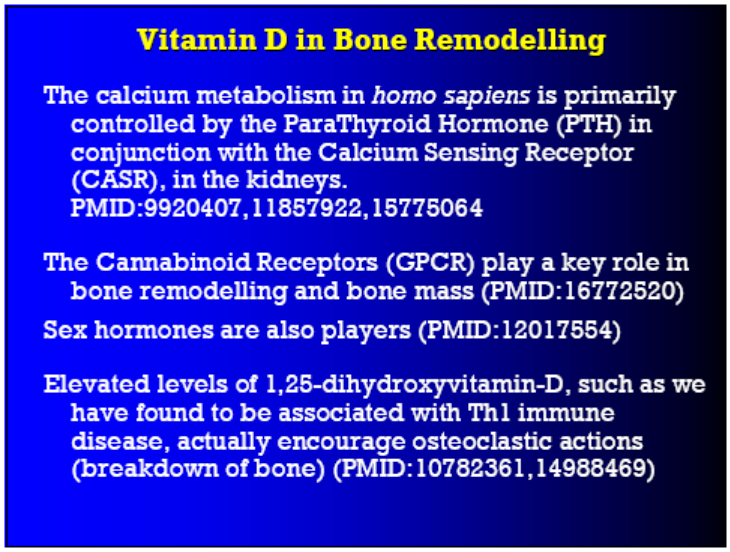
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SLIDES: **MDL video**
50:41 / 1:23/02

So, we've got some little pictures here. We have 2 molecules. What we have here is the steroid rings (video highlights right image, upper right rings of molecule) of prednisone. This is actually prednisone.

We have the steroid rings of prednisone, and over here (video highlights middle two rings on left figure) we have the steroid rings of "vitamin" D. What I wanted to show you was that you've got the steroid rings.... I want to show you the only difference between the secosteroid and the steroid is one bond. It's this carbon to carbon bond across here (highlight shows critical top left ring closure of right image, in 2nd ring from left). And it is missing across here (highlight now shows open ring in left image in the 2nd ring from left) in the seco-steroid. And that is why it is a secosteroid rather than a steroid.

It is a very subtle change, it actually gives the [vitamin D] [not VDR] more flexibility to move in a socket and that is one of the reasons why it binds into so many of the nuclear receptors. The structural similarity (video highlights cortisone bond again) between prednisone and vitamin D basically come down to that one carbon bond. And that is why it is a secosteroid and not a corticosteroid.



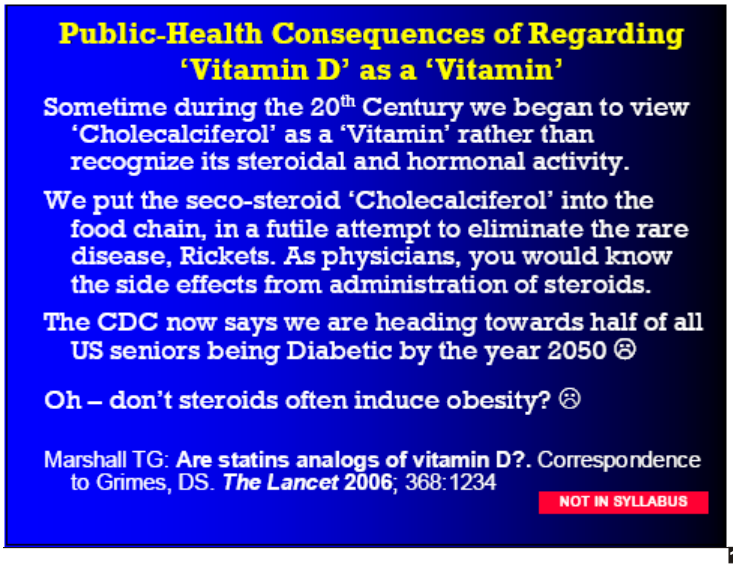
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SLIDES: **Vitamin D in Bone Remodeling**
52:29 / 1:23/02

So I have a slide here on Vitamin D in Bone Remodeling because I know all of you are going to say "well, If I withdraw vitamin D from my patients, their bones are going to get weaker." This gives you some up to date citations you can go away and look at, which show that actually "vitamin" D is not really responsible for calcium at all. It's the Calcium Sensing Receptor and the Parathyroid hormone that are responsible for Calcium homeostasis. And secondly, even the cannabinoid receptors play a key role in bone remodeling and bone mass. That paper is only a few weeks old.

The sex hormones are also players in bone density, I think you know that one.

And that elevated levels of 1,25-dihydroxyvitamin-D, such as we have found to be associated with Th1 immune disease, actually encourage osteoclastic actions and breakdown of bone and deposition of bone into soft tissue. The last thing you want in these patients is high levels of 1,25-Dihydroxyvitamin-D.

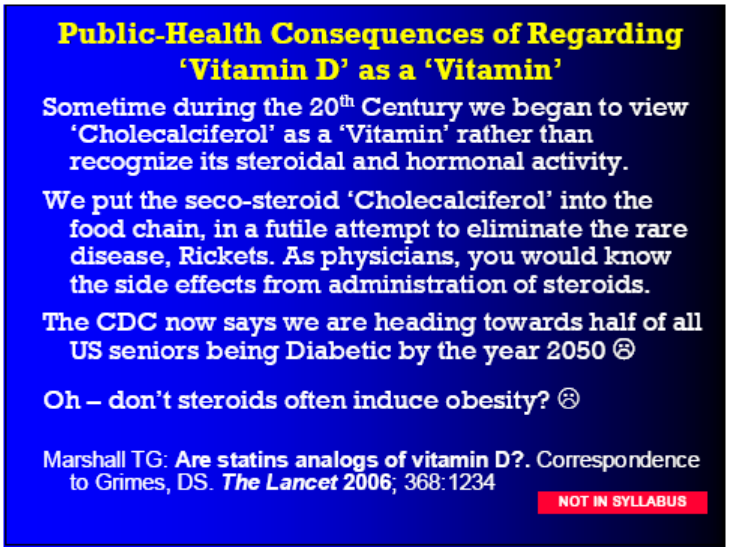


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SLIDES: **Public-Health Consequences of Regarding 'vitamin D' as a 'Vitamin'**
53:32 / 1:23/02

So this has got a "not in syllabus" picture because the text that I had in the syllabus was a lot more caustic. This is diluted.

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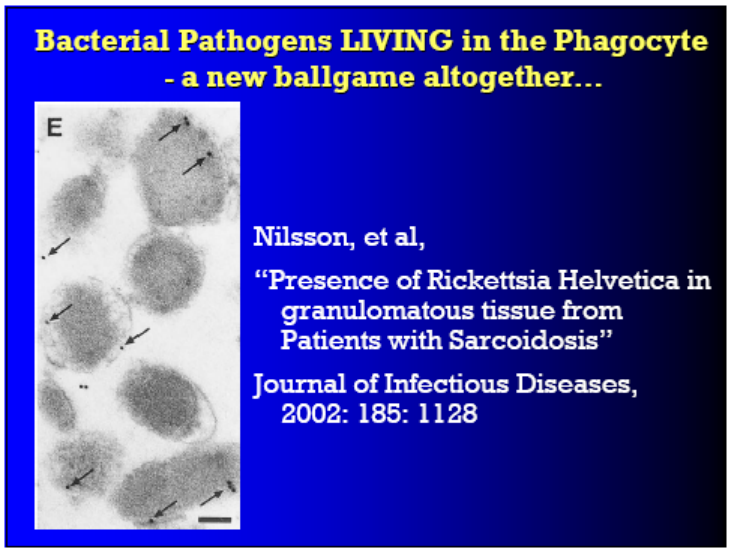


Public-Health Consequences of Regarding 'vitamin D' as a 'Vitamin.' Sometime during the 20th century, we began to view "Cholecarciferol' as a 'Vitamin' rather than recognizing its steroidal and hormonal activity. And we put the seco-steroid 'Cholecarciferol' into the food chain, in a futile attempt to eliminate the rare disease, Rickets. And as physicians, you would know the side effects from administration of steroids.

So the CDC now says we are heading towards half of all US seniors being Diabetic by the year 2050.

And, Oh, steroids often induce obesity. I'll leave you to connect the dots.

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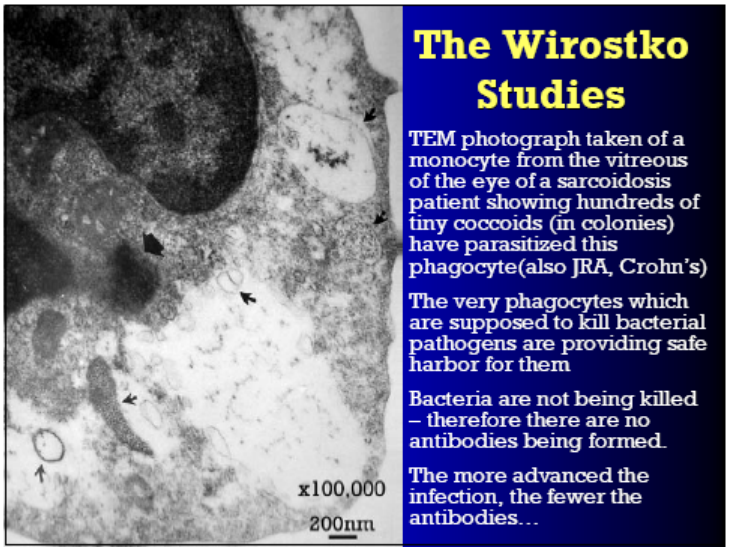


SLIDES: **Bacterial Pathogens LIVING in the Phagocyte — a new ball game altogether...**
54:39 / 1:23/02

I haven't said anything about bacteria yet. When I was trying to figure out how Sarcoidosis patients could be made sick in the absence of antibodies, this paper was published back in late 2001 on the Internet, and what it showed was Rickettsia Helvetica inside Phagocytes (video highlights upper right image E at two arrows). Persistent inside phagocytes, not broken down by phagocytosis but actually living in the phagocytes of Sarcoidosis patients.

That was the Eureka moment for me. I should have known it, there is plenty of other literature talking about micobacterium being Intraphagocytic, but that was the actual paper.

.....



SLIDES: **The Wirostko Studies**
55: 18 / 1:23/02

And then the real studies that you all will want to see if you are interested in the actual pathogens, were the Wirostko Studies from Columbia University in the late 80's. There are bout 30 or 40 electron transmisional microscopy photographs showing stained bacteria. Tiny, tiny colonies (video highlight shows lower left arrow circle of bacteria, then moves to second to left arrow colony, to middle arrow pointing to colony and then to upper right arrow) of stained bacteria in the phagocytes. This is a quarter of a macrophage.

And they were very thorough, they did macrophages, monocytes, lymphocytes and neutrophils. And they found that in the Sarcoidosis patient, they found the intraphagocytic bacteria persistent in all of them.

And in fact, the nucleus here (video highlights heavy bolded arrow left of center pointing to nucleus) is breaking away a little bit near this larger colony of bacteria (video highlights second lower left small arrow). That's why it has the big arrow on it.

Fascinating photographs. Very important if you want to understand the actual bacterial pathogens, the L-forms that cause these diseases (cell wall deficient bacteria called L-forms for the Lister institute where they were first identified). They are tiny pathogens. They are so small. They are hundredths the diameter of the macrophage. I've got 200 nanometers here (video highlights lower right slide legend). We're talking about cocciodes, tiny little dots that are about 10-15 nanometers in diameter.

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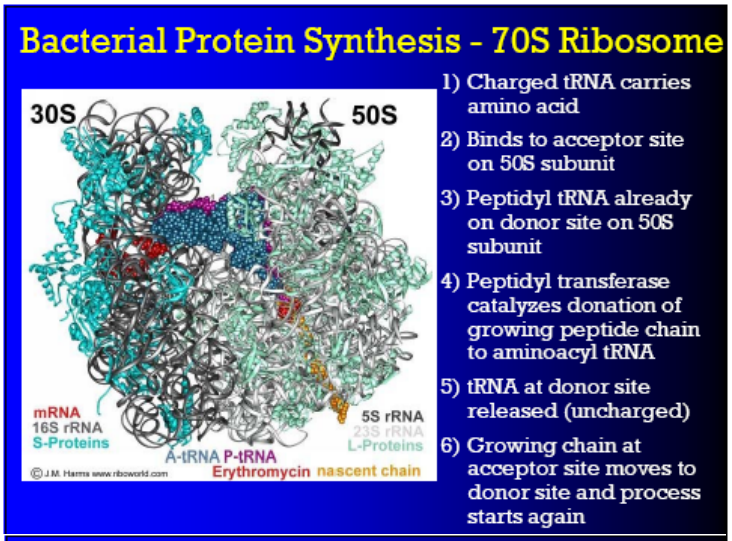
SLIDES: **Bacterial Survival in the Phagocyte**
56:49 / 1:23/02

These are pictures that show the diagrammatically show the survival of bacteria in the phagocyte. I've covered these in my 30th anniversary of Lyme meeting and with your permission, I'll skip over this. You can find it in any book. (because we're running late on time).

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SLIDES: **Phagocytosis**
SKIPPED / 1:23/02

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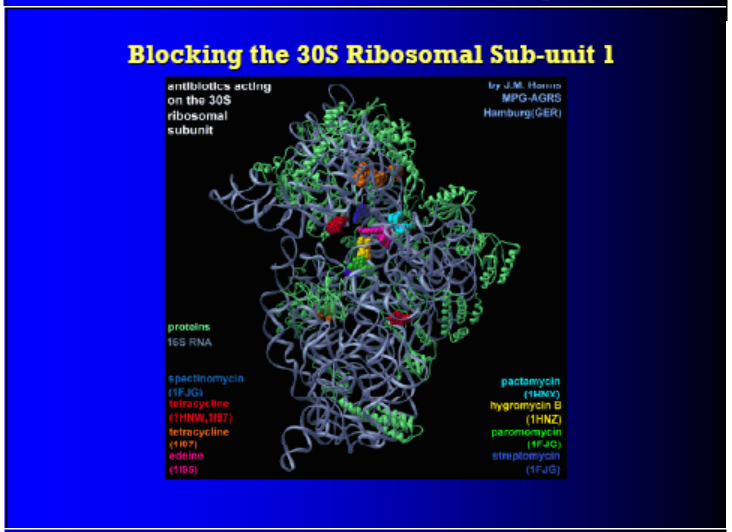
SLIDES: **Bacterial Protein Synthesis — 70S Ribosome**
57:06 / 1:23/02

Bacterial protein synthesis, and remember I talked about the 70S Ribosome, the bacterial Ribosome. Here we actually have the structure of the bacterial Ribosome, it is primarily made up of RNA, unlike the human ribosomes which are primarily protein based.

But the 16S RNA (video highlights the lower left 16S rRNA on figure), which you would all recognize as being the PCR, the substance which is used as the target for most PCR tests. 16S rRNA forms the 30S side of the ribosome (video highlights upper left 30S figure subhead). The top of this 30S we have a heliacal structure (video highlights top left ribbons) which is responsible for translate but actually for ... translating the mRNA into a protein.

The actual translation is done on the 50S side, you can see a protein (video highlights lower right quadrant) coming out the bottom here and it's in yellow. The protein transferase center (video highlights middle, slightly right) is right in the center which you see, and there we have tRNA (video highlights blue grape cluster mid top) which is donating the amino acid into the growing protein.

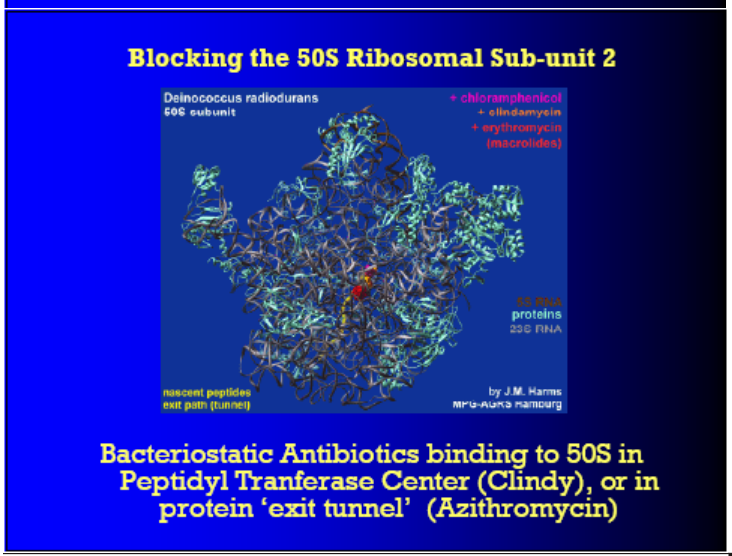
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SLIDES: **Blocking the 30S Ribosomal Sub-unit**
1 58:24 / 1:23/02

And this is an Xray structure, which was produced from the Max Planck group in Germany, and this x-ray structure shows a number of antibiotic molecules found in, you can look at them in your syllabus', but the one's we're interested in primarily are the tetracycline. And in fact, the tetracycline binds right at the top (video highlights top of 30S Sub-unit, see Orange color), and inhibits RNA translation.

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SLIDES: **Blocking the 50S Ribosomal Sub-unit 2**
58:56 / 1:23/02

On the 50S side, you can see how the protein is assembled and exits the exit tube a little more easily. And in particular, the important thing is here, you will notice that Clindamycin (video highlights center formation), just the light orange one, sits right at the PTC. And the Azythromycin sits a little bit below it, so they are actually symbiotic antibiotics.

All three antibiotics that we use are symbiotic. That means that each of them progressively reduces the function of the [bacterial] ribosome. That means that when you give them together, they are

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not competing with each other, they're complementary. It is symbiotic. Very important. Something that the genome, the bacterial genome, has told us quite clearly.

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Plasmids Make for a Toxic 'Pea-Soup'

367 Microbial Genomes have now been sequenced, and another 619 have been partially completed.

It has become obvious that most Bacterial species are not homogenous, consisting not only of a single chromosome, but also possessing a variable number of self-replicating plasmids carrying DNA, and Genes.

Borrelia burgdorferi has a large number of plasmids, indeed, nearly half its genome exists on its 21 self-replicating plasmid sub-units

Yet even common species, like *Staphylococcus epidermidis* ATCC 12228, give rise to plasmids (*S. Epidermidis* has 10% of its DNA spread over six self-replicating plasmids)

These plasmids are not targeted by antibiotics, and, unless destroyed by the immune system, will persist in chronic intra-cellular infections – think of it as a 'DNA Pea-Soup'

SLIDES: **Plasmids Make for a Toxic "Pea-Soup'**
59:53 / 1:23/02

So now we talk about the species.

Which species of bugs is it?

Well, it is not a single species. We're dealing with a polymicrobial disease. If you look at the various diseases, and the bacteria that have been reported with them all, the common ones like Staph-aureus and Propionibacterium-acnes, Propionibacterium-granulosum, seem to be the ones that crop up most of all, with the nasty bugs, micobactera cropping up far less frequently. And certainly less than 100%.

So we're almost certainly dealing with a polymicrobial disease. And depending on the exact mix of pathogens, determines which of the Th1 diseases, which of the Th1 symptom syndromes the patient will progress to. Whether it turns into Rheumatoid Arthritis, Multiple Sclerosis, or Anorexia Nervosa.

It's become obvious that most bacteria species are not homogenous. They've got not only the chromosome, but also the self-replicating plasmids which carry DNA and genes. Borrelia burgdorferi has got a large number of plasmids, in fact, nearly half of its genome exists on 21 self-replicating plasmids sub-units.

Now I'm not saying that makes it any more strong of a pathogen, but it certainly adds quite a lot to the "Pea Soup" which I like to think of as mix of DNA which accumulates in the chronic diseases.

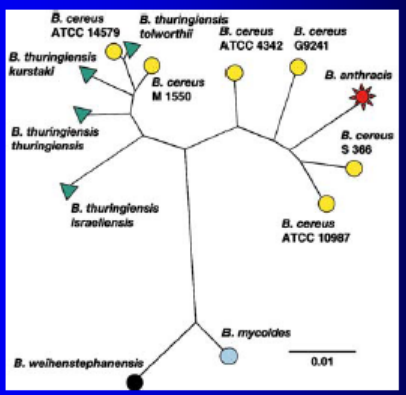
Yet even Staph. epidermidis gives rise to plasmids. It's got about 10% of its DNA spread over six self-replicating plasmids.

These can be shared between species. But the plasmids are not targeted by antibiotics. Antibiotics don't target plasmids. Only the innate immune system can go after the plasmids. So unless the plasmids are destroyed by the immune system, they will persist in chronic intra-cellular infections.

And I think of it as a "DNA Pea-Soup."

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Plasmids transmit DNA 'horizontally'



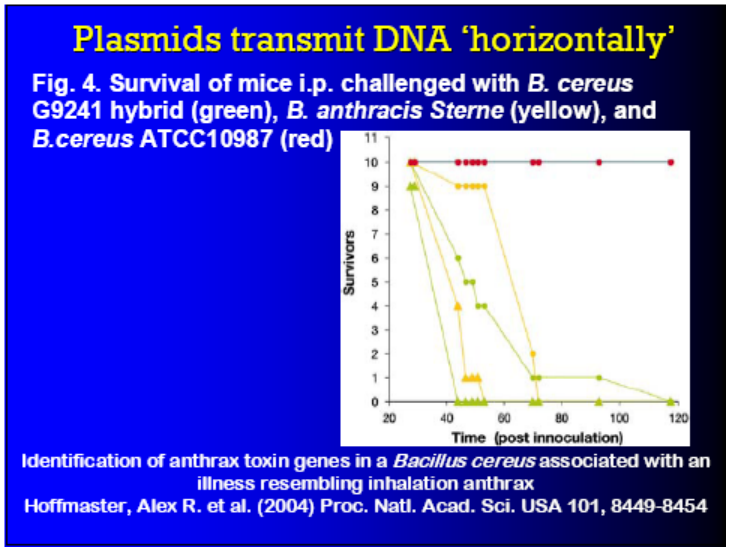
Identification of anthrax toxin genes in a *Bacillus cereus* associated with an illness resembling inhalation anthrax
Hoffmaster, Alex R. et al. (2004) Proc. Natl. Acad. Sci. USA 101, 8449-8454

SLIDES: **Plasmids transmit DNA 'horizontally' (1)**
1:02:04 / 1:23/02

Plasmids do transmit DNA horizontally. Notice the citation here, it was published in the Proceedings of the National Academy of Sciences in 2004. And what you had was, you had a patient that got very, very ill with a species of Bacillus cereus. Now, Bacillus cereus, we all know is a [relatively] harmless bacteria. It's a Bacillus that doesn't cause the body [much] trouble, it clears it (video highlights B.cereus yellow circle on figure, upper right of center).

Whereas just a little bit further around on the phylogeny here, we have Bacillus-anthracus (video highlights red spike circle far upper right). And what distinguishes Bacillus-anthracus is 2 plasmids. One plasmids contains the ability to evade the immune system, the proteins that give it the ability to evade the immune system, the other plasmids carries the ability to produce a cytokine storm and kill the host.

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Now what happened in this case was the two plasmids from anthracus were transferred into cereus and formed an extremely toxic combination.

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SLIDES: **Plasmids transmit DNA 'horizontally' (2)**
1:03:06 / 1:23/02

And you can see that the mice that were just challenged with cereus, they all survived (video highlights upper center double red circles on figure 4).

Those that were challenged with anthrax (video moves down to yellow circle dropping line) -which color is anthrax? yellow- most of them died.

BUT they died a lot quicker if they were the hybrid (video highlights green circle line) of the anthrax plasmids and the cereus genome. This is a big, big problem!

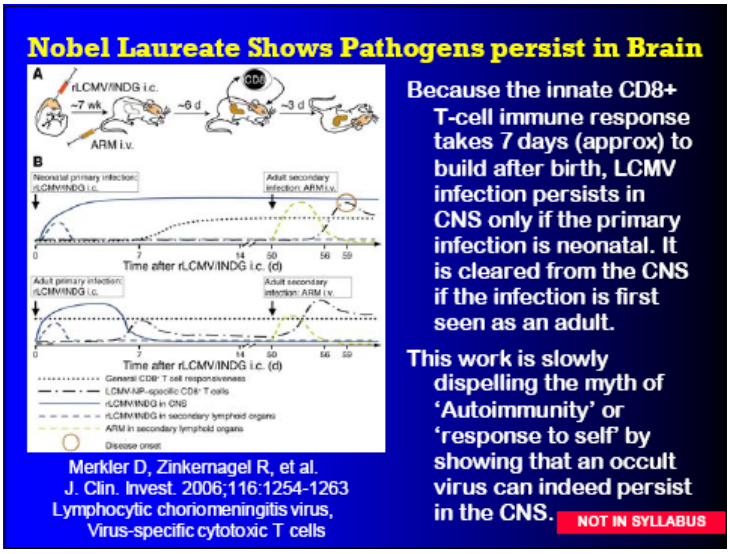
Why is it a problem?

Well, (video returns to first horizontally slide) if you look further around on this phylogeny (video highlights B thuringiensis israeliensis, mid far left green triangle), you'll find here Bacillus thuringiensis Israelensis. That one is sprayed onto agricultural crops as a herbicide. Sorry, it is an insecticide.

Anyway, you might have noticed, for example, Garth Nicholson found a higher incidence of autism in the agricultural areas. This fellow (video highlights B thuringiensis israeliensis) is sprayed in the agricultural areas.

If the chronic infection allows the plasmids to spread, these people will get very ill.

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SLIDES: **Nobel Laureate Shows Pathogens persist in Brain**
1:04:25 / 1:23/02

The final thing that I want to show is that the pathogens actually persist in the brain.

This [slide] is taken from a paper by Rolf Zinkernagle, whom I was lucky to meet at Karolinska, the home of Team Nobel. Rolf gave the keynote speech there back in May [2006]. And what they found they've done, is they injected the brains of mice at a very young age (video highlights A - neonate mice figure upper right figure), neonatal in fact, with a virus. And this is injected before the mice have an adaptive immune system.

As you all know, the adaptive immune system in humans and mice takes some time to kick in (video highlights top time line center, showing Time after rLCMV/INDG). I think these are helper cells coming up to indicate the adaptive immune system has kicked in after about 7 days.

But when the mice are born, the only thing they have to protect themselves is the innate immune system. You know, the thing we're knocking out with 'vitamin' D.

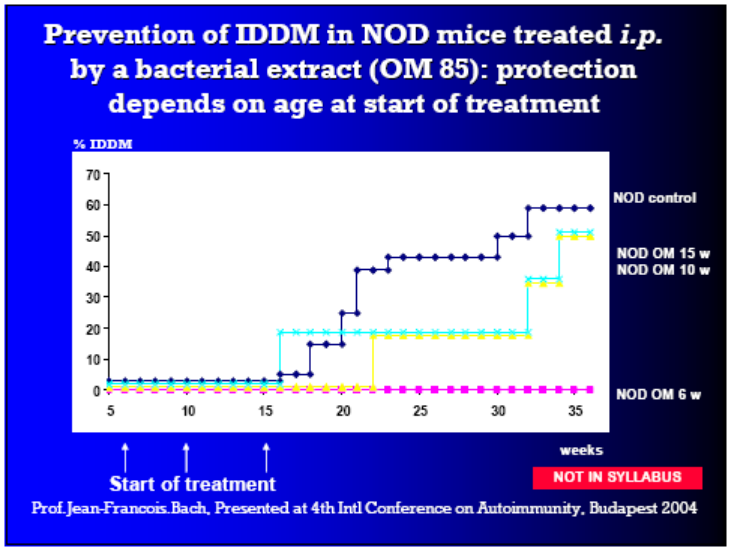
Anyway, what Rolf did was, he showed that if they injected this virus during this period, it remained dormant in the mouse. And later, when a similar virus was used to challenge the mouse, It killed them (video highlights upper right dead mouse figure).

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Whereas mice that were challenged in adulthood by the same virus (video highlights lower left time line, far left), injected by the same pathway were not killed when they were re-challenged, in fact, they survived primarily.

This is a fascinating paper, it is quite detailed with citations there at the bottom [of the slide]. And Rolf is working on dispelling a myth of autoimmunity — or response to self — by showing occult viruses can, in fact, can exist and they can indeed persist in the brain. Very important when we are trying to understand the symptoms of our patients.

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SLIDES: **Prevention of IDDM in NOD mice...**
1:06:30 / 1:23/02

And here is the slide from Professor Bach, Jean-Francois Bach, which was presented in Budapest in 2004. Jean-Francois Bach was the person who proposed the hygiene synthesis.

But what he found was that bacteria, the particular strain of mice called non-obese diabetic mice (NOD), where typically 60% of them died (video highlights upper right dark dot line) from diabetes — type 1 diabetes during their lifetime — that if he injected them early enough with a mix of common bacterial proteins, that they survived (video highlights cyan and yellow end point line).

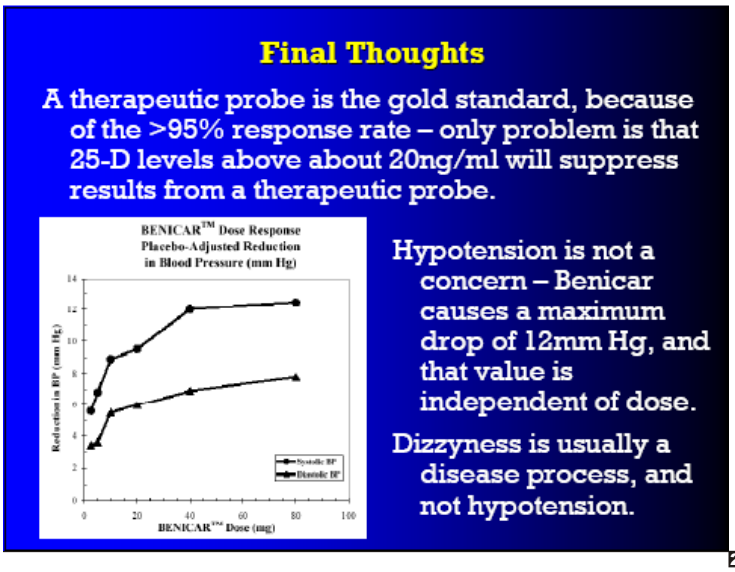
And the ones that survived best were the ones that got the bacterial proteins earlier in life than later in life (video highlights magenta square dotted line lower right).

Again, another fascinating insight into exactly how these diseases persist and why some people get ill, and other people carry the bacteria and don't get ill.

Because it is no secret that the NIH/NHLBI did a study in 2001 where they tested with PCR, they tested the blood of controls. No, they didn't use PCR, they cultured the L-form using Wayne State Lyda Mattman's technology. And they found 60% of their controls were carrying L-forms in the blood. Supposedly a sterile compartment.

This has since been confirmed by the Relman Lab, Dave Relman up at Stanford published a paper in the last year, where he too has noted that 60-80% of the population are carrying around bacterial RNA in their blood. It should not be there. Many of them, most of them in fact, are not ill or certainly not diagnosably ill. Some however, regress to really serious illness. It all depends on the DNA mix and how the mutations progress in life.

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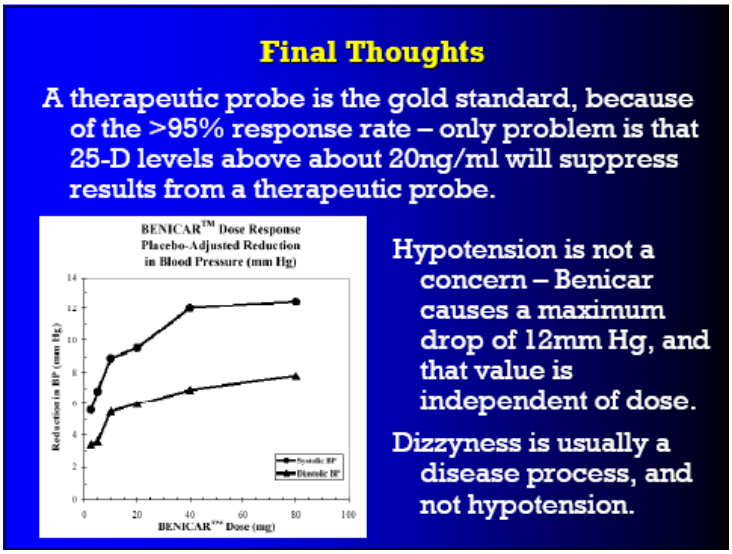


SLIDES: **Final Thoughts**
1:08:34 / 1:23/02

So just a few quick final thoughts that my colleagues at FDA felt I had to put in because I am talking to clinicians here, [are] just the simple stuff again.

Final thoughts, if you are thinking if your patient might in fact be suffering from Th1 diseases, then the therapeutic probe is really the gold standard. That's because it is greater than 95% response rate. Most of the patients will immediately feel immunopathology. They'll immediately feel more ill when they start taking the antibiotics. At that point you know that you are killing bugs. You know that these people are responding.

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The only thing I would say is 25-D levels above about 20ng/ml you will be getting immuno-suppression there and it will be suppressing the therapeutic probe. So you need to be somewhat careful about the 25-D level before initiating a therapeutic probe.

Hypotension is not a concern, even though we are using an angiotensin-receptor blocker. These are the graphs from the FDA describing insert, the package insert [see slide figure]. As you can see, the maximum drop is about 12 millimeters of mercury [12mm Hg], and that value is independent of dose. You can see the dose along the bottom here the response tops at 40mg. It is not a very good hypotensive. Benicar is not a very good hypotensive. Luckily, it does other things as well like protect the kidneys, and protect the eyes and all the other things that you are reading about in the clinical literature right now.

Hypotension is not a concern.

Dizziness. When the patients get dizziness, especially in the first month, it's usually a sign of the disease process. It is not hypotension. It's actually better to increase the Benicar to give them a better blockade than to back off the Benicar.

There is no way you can start with a low dose and work up. You've got to have those receptors being blocked, all the receptors being blocked, or it doesn't work properly. You will increase the immunopathology with only a low dose. That means the patient will get, "Herxheimer" if you wish, they will get ill form only a low dose of Benicar, but they won't get the palliative effects, they won't get the renal-protective effects, until the dose goes up.

So it is very important to give them the correct dose so they are actually going to make it through the therapy and kill those bugs off.

The only thing I didn't mention was how long it would take: three to five years for the seriously-ill patients to totally get their lives back. Most of them are happy with some form of recovery. They see the light at the end of the tunnel after about six months. Very few have not seen the light by twelve months but actually, they are symptom free or say they are symptom free at about two years. But all that means is they are getting along from day to day very nicely and much better than they were before. There is still a lot of disease present, they are still getting a lot of reaction to the antiobiotics.

It takes about another two years to where they can just guzzle the antiobiotics like candy. At that point, the bacterial load is low enough for them to pronounce recovery.

Thank you.

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Q&A

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