

1 IN THE CIRCUIT COURT
2 TWENTIETH JUDICIAL CIRCUIT OF ILLINOIS
ST. CLAIR COUNTY

3 FRANCES E. KEMNER, et al.,)
4 Plaintiffs,)
5 vs.) NO. 80-L-970
6 MONSANTO COMPANY,)
7 Defendant.)

8 Before the HON. RICHARD P. GOLDENHERSH, Judge
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12 TESTIMONY OF DR. FRANK DOST

13 November 5, 1985
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17 APPEARANCES:

18 MR. REX CARR, Attorney at Law, and
19 MR. JEROME SEIGFREID, Attorney at Law
Appeared on Behalf of the Plaintiffs

20 MR. KENNETH HEINEMAN, Attorney at Law, and
21 MR. JOSEPH NASSIF, Attorney at Law
Appeared on Behalf of the Defendant

22
23 PATRICIA GANDY, CSR, RPR
Official Court Reporter
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DR. FRANK DOST

Direct Examination by Mr. Heineman 2

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1 BE IT REMEMBERED AND CERTIFIED that heretofore, on
2 to-wit: Tuesday, November 5, 1985, being one of the regular
3 judicial days of this Court, the matter as hereinbefore set
4 forth came on for hearing before the HON. RICHARD P. GOLDEN-
5 HERSH, Circuit Judge in and for the Twentieth Judicial Circuit,
6 State of Illinois, St. Clair County Building, Belleville, St.
7 Clair County, Illinois, and the following was had of record,
8 to-wit:

9 * * * * *

10 THE COURT: Good morning.

11 CONTINUED DIRECT EXAMINATION

12 BY MR. HEINEMAN:

13 Q Dr. Dost, I'd like to take you briefly back for a
14 minute to the discussions we had yesterday on porphyria and
15 the no observable effect level, the NOEL. Have you caused
16 a chart to be prepared which would assist you in explaining
17 this situation?

18 A Yes.

19 (At this time Defendant's Exhibit 1281 was marked
20 for identification.)

21 Q All right, sir, I'll show you what's been marked
22 as Defendant's Exhibit 1281 and ask you to examine that and
23 identify it for me, please.

24 A This is a chart prepared from tabular data that was

1 published by Smith, et al., in the journal Biochemical Pharma-
2 cology, Line 30 in 1981, and this is a description of the
3 effect of an acute or of acute doses, single doses of TCDD
4 as distinguished from the experiments that you have mentioned,
5 I believe, yesterday in which lower doses were given on a
6 weekly basis over an extended period of time.

7 Q Now, would you be so kind as to step down here and
8 explain that to the jury please?

9 A This is another dose response curve. It looks a
10 little different from the ones that we talked about the other
11 day. You recall that in the ones that we've seen earlier
12 there is kind of an S shaped arrangement. This is really
13 the same, the only difference that the, the change is very,
14 very rapid. The slope here is extremely steep, and what it shows
15 is that at a dose, and this differentiates between males
16 and females because there is some difference in the response
17 of males and females to most of the effects of TCDD. It isn't
18 much, but it's measurable. And again this is a logarithmic
19 scale and this would be a dose, I believe, of about 6 micro-
20 grams per kilogram. And you can see that the diamond and the
21 circle are superimposed here, they both have got the same
22 doses, this I think would be 12 micrograms per kilogram and
23 this would be 16. And what it shows here is that these
24 rather substantial single doses, now this is just a single

1 dose, not a chronic exposure but a single dose, that it requires
2 a very substantial single dose before this kind of response
3 emerges. The measurement that is being made is the appearance
4 of porphyrin actually depositing in the liver. You recall
5 yesterday I mentioned that they can appear in the urine and/or
6 in the liver, and here we are talking about hepatic porphyrin,
7 that is these intermediate product in the synthesis of heme
8 accumulating in liver cells and in the response the animals
9 manage a degree of impairment in heme synthesis very, very
10 well until they get up to some substantial dosage, and then
11 the accumulations in the tissues becomes very, very rapid be-
12 cause the -- presumably because the impact at the enzyme
13 level becomes so substantial that the material is not being
14 disposed of very readily.

15 Q Now, Dr. Dost, yesterday you spoke about the chronic
16 level or the no effect level for chronic dosage being at what
17 level?

18 A Well, it is at a level down on the order of a tenth
19 of a microgram per kilogram per week or less. Statistically
20 if you run, if you run the dose response curve all the way
21 down, the no effect level approaches a hundredth of a micro-
22 gram per kilogram per week. Statistically the no effect level
23 is on the order of a tenth of a microgram, and by that I mean
24 that it comes up a little but it is not enough that a statis-

1 tician would say that it is different from the control.

2 Q And that is what period of time does that chronic
3 test cover?

4 A The two that describe this most effectively, one
5 by Goldstein, it ran for sixteen weeks. And another by
6 Cantoni for forty-five weeks, one injection a week at a
7 variety of doses. They ran through a dose response or dose
8 levels of a hundred to one micrograms per kilogram. In other
9 words there would be four doses, .01, .1, it went to 10
10 micrograms per kilogram, actually, per week.

11 Q So it would be .01, .1, 1, and 10?

12 A Correct.

13 MR. HEINEMAN: Thank you, your Honor, at this point
14 we would move the admission of Defendant's Exhibit 1281.

15 (Defendant's Exhibit 1281 was offered into evidence.)

16 MR. CARR: No objection.

17 THE COURT: Admitted without objection. Thank you.

18 Q Dr. Dost, at the close of testimony yesterday we
19 were talking about the cardiovascular system and the effects
20 of dioxin on serum lipids, do you recall that, sir?

21 A Yes.

22 Q And what is the -- what do the studies demonstrate
23 with respect to the effect of TCDD on the production of serum
24 lipids?

1 A In general, at substantial doses, the fats and
2 cholesterol in the serum will rise as the dose is decreased,
3 and this is substantial doses experimentally. Single doses
4 at a dose of around 10 micrograms per kilogram in the rat.
5 The serum lipids, the fats will rise, cholesterol esters
6 tend to decrease, not drastically, but they tend to drop
7 below normal levels. And then if you follow this over a
8 period of time, all of these measurements tend to return
9 toward normal in about, in twenty-one days they are approaching
10 normal. The slopes of the data would suggest that by thirty
11 days their measurements would be in the normal ranges.

12 Q Now, are elevated lipids a clear indicator of
13 disease? How does one interpret elevated lipids?

14 A Well, to determine whether they are an indicator
15 of disease depends on far more than elevation of lipids,
16 because we all have elevated serum lipids after a meal,
17 depending on the nature of the meal. The increased lipids
18 may be just typical fats, it could very well be high
19 cholesterol, and that is going to be a relatively transient
20 process because this is the normal thing that we do in
21 processing our dietary intake. Any time that we eat some-
22 thing there is going to be reflected in the circulation
23 elevated levels of sugar, amino acids, fats, depending on
24 what is in the diet. That is fairly straight forward. When

1 we start dealing with a disease state, again, increased blood
2 lipids all by themselves could not provide a diagnosis. A
3 diabetic will often have elevated blood lipids, but that
4 wouldn't diagnosis diabetes. It depends on other kinds of
5 effects. It is associated with diabetes. Smokers may have
6 elevated blood lipids, but they may not, too. But it seems
7 to be -- there seems to be some association. People with
8 hypertension often have elevated blood lipids, but here we're
9 dealing with a chicken and egg situation in which the condition
10 that led to elevated blood lipids may have caused over a long,
11 long period of time a change in the arteries that we would
12 call arteriosclerosis that resulted in the hypertension, so
13 what I am saying here is that a long term, a long sustained
14 elevation in blood lipids and blood cholesterol could very
15 well lead to and in fact the statement is sometimes made that
16 prolonged elevation of blood lipids causes cardiovascular
17 disease. Other people are more conservative and say that it
18 is associated with it. Whether that's a weasel word, I don't
19 know. In any case, there is at least an association with
20 cardiovascular disease, and that's why we're talking about
21 it perhaps in the context of cardiovascular disease. But
22 there are a variety of other conditions that will cause either
23 a prolonged or a relatively foreshortened period in which blood
24 lipids are elevated.

1 Q And are the results in the animal data dose dependent?

2 A Yes, they are dose dependent. The difficulty is that
3 in the case of lipid abnormalities with respect to TCDD at least,
4 the doses have not been carried downward all the way to clear
5 no effect levels. The dose response is very clear, but we
6 don't identify a specific no effect level below which no
7 abnormalities in lipid metabolism appear. Many of the studies
8 use only a single dose because they are trying to understand
9 the mechanism by which this event occurs. But it would appear
10 that the no effect level is substantial upon the order of a
11 microgram per kilogram.

12 Q Now, would you say it would appear that it is sub-
13 stantial?

14 A By that I am just making a judgment on what the dose
15 response information is. You recall that the other day I told
16 you that if you don't have enough points on a dose response
17 curve to carry all the way to a no effect level, it is possible
18 to project within limits from what data there is where the dose
19 response, where the no effect level might be expected. However,
20 one cannot simply say here is where the dose, here is where the
21 no effect level is. I would judge on the basis of the existing
22 data that it is on the order of a microgram per kilogram.

23 Q Now, based upon a judgment of a microgram per kilogram
24 and the dose that you calculated with respect to the Sturgeon

1 exposure of the 10 grams of soil, would that Sturgeon dose be
2 sufficient to generate elevated blood lipids?

3 A Well, we calculated that a dose of .001 micrograms
4 per kilogram per day would represent a 222nd, if you will --
5 I beg your pardon, that is 222 times higher than that exposure
6 that we postulated, and since we are talking about 1 microgram,
7 then that number would be expanded by a thousand.

8 Q So the Sturgeon dose which you calculated would be
9 222,000 times 2 low?

10 A It would be that much less than the 1 microgram per
11 kilogram that I am postulating.

12 Q Now, I'd like to talk to you this morning about
13 neurological effects of TCDD. Now, briefly describe the
14 neurological system that we are talking about.

15 A Well, the neurological system is all of the --
16 it is the central nervous system, the brain and the spinal
17 cord, and all of the peripheral nerves. So the neurological
18 system is our system for transmitting rapid messages from
19 one part of the body and for integrating them and acting
20 upon them in the central nervous system. It is all of the
21 nerves of the body.

22 Q Now, had there been neurological effects observed
23 in humans exposed to dioxin?

24 A To my recollection, no. I am not entirely familiar

1 with all of the epidemiology, but I do not recall any in
2 humans.

3 Q All right, are you familiar with the examination
4 by Dr. Suskind of workers at the Nitro Plant who were exposed
5 to in the cleanup of an accident there in 1949?

6 A I remember some of that information, yes. I remember
7 one individual that from which a nerve biopsy was taken that
8 was found to have suffered some demyelination.

9 Q Now, do you recall whether or not there were reported
10 neurological effects in those individuals who were examined by
11 Dr. Suskind?

12 A I'm sorry, I don't remember.

13 Q All right, let's deal with the animal data. What did
14 the studies show with respect to animal data and the effect of
15 TCDD in causing neurological effects?

16 A It shows that TCDD is essentially inactive as far as
17 either the central or the peripheral nervous system. There
18 are reports of animals at high doses showing listlessness and
19 effects like that, but that can hardly be described as an
20 effect on the nervous system. These are very, very sick
21 animals and any sick animal is going to be listless, and so
22 I don't think that data like that or statements like that can
23 be interpreted as reflecting neurological change in the
24 experimental animals. What we have to rely on are the --

1 primarily the pathological examination of animals in which the
2 central and the peripheral nervous system are examined micro-
3 scopically in detail to determine whether there has been any
4 change in the architecture of the nerve cells.

5 Q Now, when you say that someone reports that the
6 animals look listless, at what doses have those reports been
7 made?

8 A Well, those have been at high doses that are pro-
9 ducing, they are on the verge of a lethal effect. These are
10 animals that are clearly intoxicated by the TCDD, they have
11 been badly injured.

12 Q All right. And so the list -- to what do you
13 attribute the listlessness?

14 A A very sick animal, and I would not try to assign
15 any specific -- any sick animal is going to be listless.
16 These animals don't convulse, they don't show signs that would
17 suggest that their central nervous system has been stimulated
18 as far as depression is concerned. There is no way of
19 assigning that kind of effect. However, those animals have
20 suffered a great deal of other kinds of damage, and a sick
21 animal is listless.

22 Q All right. Now, what do the studies demonstrate
23 with respect to neurological effects on animals due to TCDD?

24 A Really, nothing. The only effect that, for example,

1 that Kociba reported in his lifetime studies on rats was, and
2 this is at a dose range that was actually increasing the
3 mortality of the animals, there was a good deal of pathology
4 generally in those animals. There was, and I believe I men-
5 tioned something about this yesterday, some capillary break-
6 down in the central nervous system so that there was bleeding
7 around nerve cells, around nerves, but that's not a function
8 of effect on the nervous tissue. That's a function of an
9 effect on the circulatory system.

10 Q All right, now, Kociba, he carried on two different
11 sets of studies?

12 A This is the two year studies. In the thirteen week
13 studies in which they ran still higher doses of microgram per
14 kilogram per day, which again caused a good deal of other
15 kinds of pathology, there was no mention, they specifically
16 describe the parts of the brain that they examined pathologically
17 and they reported no information about any effects on the brain.
18 The custom is in a pathological study to identify every tissue
19 that is examined, and then to report only on the tissues in
20 which some kind of abnormality has been found, and for this
21 reason there is often a statement in the methods of the paper
22 describing the methods of examination and the tissues exami-
23 nation and no further discussion of certain organs, and that
24 means that there has been nothing identified as being abnormal.

1 Q Have there been any studies done? You mentioned
2 yesterday a Dr. Calder having done some studies?

3 A Yes, well, he was concerned about effects on the
4 peripheral innervation, that is out in the legs and arms and
5 so forth, of course he is studying rats, so they have four
6 legs, and I looked specifically for evidence, pathological
7 evidence of injury to peripheral nerves after substantial
8 doses of TCDD and found none.

9 Q Now, with respect to myelinization, we talked briefly
10 yesterday about myelin, what had the studies demonstrated with
11 respect to the effect on the myelin surrounding the nerves
12 after exposure to TCDD?

13 A Demyelination is one of the easier morphological
14 -- by that I mean anatomical changes to detect in nerves,
15 in nervous tissue, and there have been no reports whatsoever
16 of changes in the myelin sheath of nerves in animals intoxi-
17 cated with TCDD.

18 Q This is an effect that can be observed upon exami-
19 nation?

20 A As it happened I am not a pathologist, you must
21 understand, but I've done research on hexachlorophene, which is
22 a demyelin agent, and while I had a pathologist direct and
23 analyze pathological effects on those animals, I was able to
24 identify without any instructions at all just with the pathology

1 that I learned as a veterinary student thirty years ago or more,
2 I was able to identify the areas of demyelination. It is a
3 very easy thing to see.

4 Q Now, Dr. Silbergeld has testified to this jury that
5 there's been work to demonstrate that 2,3,7,8 TCDD causes
6 peripheral neuropathy and she cites the work of animal studies
7 done by, among -- she talks about the Strik Group in the
8 Netherlands, Elovaara's Group in Finland, and workers at
9 NEIHS, are you familiar with Dr. Elovaara's work?

10 A Elovaara was, I believe, a postdoctoral -- in any
11 case, I was a worker in the laboratory of Harry Vanio in Finland
12 who has done a good deal of work on TCDD and phenoxy herbicides.
13 It had nothing to do with myelin. They were working with a
14 strain of rat that has a genetic defect that -- let me see
15 if I can explain this. When hemoglobin or heme proteins are
16 used up, when they break down, the heme has to be, has to be
17 also destroyed. And one of the products of the heme breakdown
18 is a pigment called bilirubin, and it is excreted in the bile,
19 by and large. That has to be done actually in the cells just
20 as heme is made in each cell that uses it, that breakdown has
21 to take place in the cell when finally that enzyme becomes
22 ineffective and has to be disposed of. But you don't repair
23 proteins, you don't repair enzymes, discard them and start
24 over again. The strain of rat that Elovaara was using, and

1 he did this deliberately, was a strain that does not dispose
2 of bilirubin very well. I think I mentioned earlier that in
3 mice, why, there are strains and strains and strains of rats.
4 There are probably a hundred different strains of mouse used
5 in research, and they are almost designed for certain kinds of
6 investigation, and the Gun rat, which is what Elovaara was
7 using, is a rat with a genetic defect that doesn't permit it
8 to dispose of bilirubin properly, and what they found was in
9 those rats there was in effect, in rats treated with TCDD
10 with that genetic defect, they were able to find a somewhat
11 amplified toxicity, some biochemical changes in the nervous
12 system of the rats with that genetic defect. The doses again
13 were fairly substantial, we are not talking about fractions
14 of microgram as we have been.

15 Q Did those studies demonstrate peripheral neuropathy?

16 A No, they had nothing to do with the peripheral nervous
17 system. They were working with the brain.

18 Q Now, I'd like to deal next with the immune system.
19 I wonder if you would describe the immune system in animals
20 and humans.

21 A Well, the immune system is in animals and humans is
22 in essence the same. There are differences among species but
23 the function and the general concept are the same. The immune
24 system first is the protection system that the body has for

1 dealing with infection, that is external organisms that can
2 cause damage to the organism. It is also intended to deal
3 with foreign substances, foreign proteins. One of the reasons
4 that oftentimes an insect bite can cause such a ferocious response
5 is because of foreign proteins that the insect injects into the
6 skin and the body immediately sends cells in to try to deal
7 with that foreign material. The mechanism that rejects organ
8 grafts, and I think everyone is aware of the present medical
9 advances in grafting organs, kidneys, heart, etc. And one
10 of the problems that they have is preventing rejection. That's
11 an immune response to the foreign protein of the new part. The
12 immune system apparently also responds to cells which are be-
13 coming cancer cells because they have become new and strange.
14 So what we have here is a protective system that is primarily
15 circulated in the blood that responds to external assault,
16 primarily infectious attack. It has two general divisions,
17 it is an exceedingly complex system, and I would never claim
18 to be immunologist, but it is a very complex system that can
19 be divided into two parts. One part is known as the cell
20 mediated immune response, which means that for the most part
21 it consists of cells which attack foreign organisms, which
22 attack foreign cells. If we have cancer cells starting to
23 develop, it may attack those cells and it in general responds
24 to, if you will, things that are particulate in nature by

1 oftentimes foreign inert material, some components of the cell
2 mediated immune response may actually engulf them, may actually
3 surround them and remove them. The other part of the immune
4 system is called the humoral immune system. Humoral arises
5 from the same root as hormone, and what it means is that this
6 is related to materials that are actually in solution in the
7 blood, primarily antibody, primarily antibodies, and antibodies
8 are proteins that have -- that arise from experience. In the
9 immune system there are some proteins that are very nonspecific,
10 the gamma globulin is a nonspecific immune response that will
11 tend to interact with foreign organisms, and it's there all the
12 time. If you have been vaccinated against some disease or if
13 you have had exposure to some disease, you will develop anti-
14 bodies and you also develop a memory in the immune system, so
15 that if you are exposed at another time, the body immediately
16 makes more antibodies to respond to the challenge. In other
17 words, if you have had smallpox or chickenpox, almost everyone
18 my age at least had chickenpox at sometime or another. You
19 don't get it a second time, and the reason you don't get it
20 a second time, it happens to be a viral disease, but the reason
21 you don't get chickenpox a second time is because the body has
22 learned about chickenpox and it is sitting there poised to
23 respond to the next infection. So the humoral immune system
24 has a memory that responds to the second assault, let's say,

1 or if you have been vaccinated, what we've done is instruct the
2 body to learn to make those antibodies, and you respond to the
3 first assault. So that in a very general sense is the immune
4 system. Now, the components of it are exceedingly complex.

5 Q Now, you say in a vaccination, what do you inject in
6 a vaccination? How does that work?

7 A Well, what you would probably do is in most cases
8 it is a killed vaccine, so-called, which is essentially a
9 suspension of the organisms that has been killed by some
10 chemical means, usually, and that is all of the proteins of
11 the organism that the immune system learns to respond to, but
12 it doesn't have any of the activity, it can't be infectious.
13 So if you inject it, then the immune system makes antibodies
14 and it also prepares itself for the future production of
15 antibodies. So whenever, after the vaccination has taken,
16 so to speak, you come in contact with those organisms, the
17 body immediately creates a response and fights off the
18 organisms.

19 Q What is the role of the thymus gland in all of this?

20 A The thymus gland is critical to the cell mediated
21 immune system, because it is a -- it processes certain kind
22 of cells called lymphocytes, and in some fashion changes their
23 character so that they will then go out into the lymph nodes
24 and they will reproduce by typical cell division, as cells do,

1 there is probably a genetic change in these cells as a result
2 of some interaction with the epithelium of the thymus and that
3 is not clearly understood, the mechanism. However, these cells
4 are given the capability of responding to the kind of challenge
5 that the cell mediated system has to meet. That is, they will
6 go and attack any organisms that are foreign, that don't belong.
7 It will attack cells that don't belong, and these lymphocytes
8 leave the thymus with this change in their genetic instructions.
9 They go to the thymus and they form clones. Now clones are
10 just collections of cells. A cell has divided, made another
11 cell just like it, they divide and make cells just like them,
12 and so we have a colony of cells in each lymph node and they
13 are continually shed into the circulation. There are also
14 cells in reserve, and when some kind of infection that requires
15 their services arises, then they move into the -- sometimes
16 right out of the blood vessels into the tissues to respond to
17 the challenge. These are the kinds of -- I would imagine
18 that when you have an infection on a skin wound and it gets
19 pusey, that primarily is lymphocytes of this sort, as well as
20 certain kinds of cells called macrophages that actually engulf
21 foreign material and they move into the area to clean up the
22 mess.

23 Q Now, what is the relationship between the thymus
24 gland and the age of a person, whether he be in infancy or

1 adulthood?

2 A The thymus has its greatest importance in early life,
3 very early. And in the human it begins to function, and as a
4 matter of fact has accomplished a major part of its activity
5 probably before birth. It begins to function several months
6 before birth and is going through this process, because the
7 fetus has its own circulation, it is processing lymphocytes
8 for a long time prior to actual birth. So that the human infant
9 is born with a fairly effective immune system, it isn't perfect,
10 but it is very nearly in place. And the thymus probably has,
11 will have disappeared in a child of perhaps ten years. In the
12 rat, and most other species in which we do experimental work,
13 the animals are born very very early, I think I mentioned
14 yesterday that the rat pup is born at about the same stage of
15 development as a two and a half month human fetus, so the rat
16 has got a long way to go in terms of its development before it
17 actually is competent. And this function of the thymus in the
18 rat is not really fully developed, and the thymus as a con-
19 sequence is really very sensitive in a very young rat.

20 Q Now, are T cells those cells that are coded or pro-
21 grammed in some way in the thymus gland?

22 A Yes.

23 Q Humans are born with circulation T cells?

24 A Apparently, yes.

1 Q How about rats?

2 A No, they don't seem to be, although at the time of
3 birth it is a little difficult to examine them. But they appear
4 not to have started the process of T cells until after birth.

5 Q Now, what does that mean then in terms of whether
6 a chemical would have an immune effect on a rat as opposed to
7 a human? Does this difference in development between the rat
8 and the human have any effect on your ability to extrapolate
9 data from one to the other?

10 A In the rat during the earlier stages of development
11 up until weaning and even for a period afterwards, the thymus
12 is extremely important, and if you interfere with thymic
13 functions, then almost certain, well, you're going to probably
14 interfere with development of T cells. The human infant is
15 probably past the stage of maximum dependence, keep in mind
16 that the thymus still functions in a young child. But the
17 initial function of delivering coded, to use your term, in-
18 structed lymphocytes has been done, and those lymphocytes
19 once out in the periphery maintain their own population.
20 There are still some being processed by the thymus, but a
21 lot of the effect, a lot of the T cell activity arises from
22 the reproduction of T cells out in the lymph nodes. So a
23 good part of the work has been done, much of the work has been
24 done as far as the thymus is concerned in an infant.

1 Q Well, does dioxin have an effect on the immune system
2 in experimental animals?

3 A Oh, yes.

4 Q All right, and what sort of effect does it have?

5 A Well, it has a general depressing effect on the immune
6 system. It has a more -- it has a greater effect on cell
7 mediated immunity than it does on humoral immunity. However,
8 there are some effects on humoral immunity. A lot of it
9 depends on what kind of tests are done, and there are a very
10 large number of different kinds of tests available to examine
11 the function of the immune system, and many of them do show
12 effects of TCDD and there are some of course that do not. But
13 I think that it is, as a general statement, yes, TCDD is able
14 at sufficient doses to have an effect on the immune system.

15 Q Because of the differences between the rat and the
16 human being in this particular parameter, the immune system,
17 how reliable is that data on rats in terms of predicting what's
18 going to occur in a human being?

19 A Well, there are two kinds of information. One is,
20 of course, what I just mentioned, that the cell mediated system
21 is fairly functional in a newborn human. The other is that the
22 impact of maternal treatment, that is treatment of the mother
23 prior to birth in experimental animals does not seem to be as
24 substantial as dosing afterward when the animals are nursing

1 because it is well-known that TCDD is able to find its way into
2 milk, that's not at all a secret. And there seems to be a
3 greater effect on the rats after birth during the period when
4 the thymus is actually starting to do its work. So I would
5 suggest that the human infant is not as vulnerable to infection
6 to failure of immune function, it is protected by the maternal
7 immune system of course prior to birth, just as the rat fetus
8 is, after birth it is in considerably -- it is much more
9 competent in terms of its immune function. You have to keep
10 in mind that the effect on the immune system in the -- on
11 the cell mediated immunity apparently is not an effect on
12 T lymphocytes per se. They don't seem to be, they don't seem
13 to be injured, even though the thymus is very severely affected,
14 it is one of the most sensitive organs to TCDD.

15 Q So the effect is on the -- is on the thymus gland
16 and not on the lymphocytes themselves?

17 A It would appear so.

18 Q What happens to the -- what do the studies indicate
19 happens to the thymus in animals?

20 A Well, if you follow almost any experimental animal,
21 and young animals are usually used because the thymus is
22 functional in young animals, and therefore more vulnerable,
23 you find that after TCDD treatment, assuming that the dose is
24 sufficient, the thymus shrinks, it becomes smaller and smaller.

1 One of the reasons it shrinks is because all of the lymphocytes
2 that are contained in it leave. Now, there is a continual,
3 probably it isn't that the lymphocytes leave, it is more than
4 likely that no new lymphocytes are coming in for processing and
5 the ones that were going to leave do. It appears that the
6 effect is on the, well, the thymus is a gland that is comprised
7 primarily of epithelial cells. In other words they are cells
8 interestingly enough that are related back in early development
9 to the cells that form the skin. Those are the cells apparently
10 that are affected by TCDD. There is recent work that is
11 described in Dr. Greenlee's Group has been working on this for
12 quite some while. So the effect on the thymus is really to
13 stop or interfere with the function of the cells that line
14 the thymus, and the lymphocytes that it processes are simply
15 not brought out anymore and the thymus shrinks way down.

16 Q What are the clinical manifestations of an impairment
17 of the immune system in animals?

18 A Well, there is a, you know, we are exposed constantly
19 to every kind of infection that you can imagine, just about.
20 And we occasionally catch cold, we occasionally get gastro-
21 enteritis, or we may even get an infection in some tissue.
22 The manifestations of a failed immune system would be infectious
23 disease. Now, that shouldn't be interpreted to mean that every
24 infectious illness is a function of a failed immune system

1 because we do get infections. I have been fighting off a cold
2 for three weeks and I haven't fully succeeded yet, but I'm
3 fighting it off. And that cold got a foothold, now maybe if
4 my immune system had been perfect I wouldn't have caught cold.
5 But if it requires perfection, then we are all defective be-
6 cause I don't know of anyone who never gets colds. But once I
7 got the cold, then all of my immune functions are coming to
8 bear in trying to get rid of it. So if we are looking at the
9 experimental context, we would do experiments in which we
10 administer an infectious organism to an animal and observe
11 its ability to respond, and lots of experiments like that are
12 done. We continually monitor animal colonies to see whether
13 there is an increased incidence of this. If we are seeing an
14 increased incidence, far more animals with the sniffles or
15 other kinds of disease, we know that we've either got a massive
16 infection in the colony or that we imaginably might have some
17 kind of an immune dysfunction.

18 Q Well, when the animals are treated with TCDD, does
19 the immune system just fail completely?

20 A No, it appears that it doesn't fail, because at doses
21 -- there are effects, and again the dose response relationship
22 applies in immune function just as it applies everywhere else.
23 It appears that there is an upper limit. It is known very clearly
24 that animals that die of TCDD intoxication do not die of infection.

1 Animals that are maintained, it seems to me that I made some
2 mention of this earlier, animals that are maintained without
3 any microorganisms in their environment at all are just as
4 sensitive to TCDD as animals maintained in a normal environ-
5 ment. Animals that are maintained over a lifetime, we con-
6 tinually seem to mention Dr. Kociba's work, but lots of other
7 long term studies as well do not show any increase in infectious
8 disease, and even though they don't speak specifically of
9 infectious disease, infectious disease causes pathology just
10 like chemicals do, and effects in the lungs and so forth would
11 be identified in the pathology reports on these kinds of studies.
12 So there is a clear upper limit to the degree of impairment
13 that TCDD might cause. And we can see this in experiments in
14 which animals are infected and their mortality is measured
15 following TCDD intoxication.

16 Q Well, is there a no observed effect level for immune
17 response?

18 A In every experiment where there has been an adequate
19 range of doses, no observed effect level becomes quite apparent.
20 Whether there are examinations of specific components of the
21 immune system or whether they are looking at the ability of
22 animals to withstand infection.

23 Q In connection with what you are discussing there, Dr.
24 Dost, have you caused to be prepared a chart dealing with that

1 subject?

2 A Yes, I have.

3 Q Would the use of that chart assist you in explaining
4 it to the jury?

5 A Yes, it would.

6 (Defendant's Exhibit 1282 was marked for identification.)

7 Q All right, sir, let me show you what's been marked as
8 Defendant's Exhibit 1282 and ask you to examine that and identify
9 it for me, please.

10 A This is a graph that is prepared from data, tabular
11 data that was reported by a researcher named Thigpen, that's
12 an unfortunate name, in the journal Infection and Immunity,
13 Volume 12, 1975 in which he treated mice with TCDD for four
14 weeks and then infected them with an organism, that is, with
15 a salmonella, the species is burned and this is an organism
16 that when administered to mice at sufficient doses is in fact
17 lethal, and this is one of several experiments that he did,
18 but it does represent the general findings in the studies.

19 Q Now, S-burn?

20 A Salmonella, salmonella burn, salmonella is a very
21 common genus of microorganisms. This particular one is one
22 that is used in the laboratory and it does appear out in the
23 general environment also. This is again a dose response curve,
24 and again it looks a little funny because the dose response is

1 so steep. As usual, the dosage is expressed logarithmically,
2 that is .1, 1, and 10 micrograms of TCDD given weekly. And
3 these are in young mice. And what we see here is that there
4 is an upper limit to the -- this is the mortality, this is
5 the percentage of animals that died given that treatment.
6 This is the control level, the number of animals, the percentage
7 of animals that were given no TCDD which succumbs to the infec-
8 tion. What we have here is at this highest dose which is
9 clearly an upper limit because you can see at the next highest
10 dose we are up here on somewhat less than 70 percent, and then
11 we are up here to 75 percent, something like that, and that's
12 as high as the mortality would go. And when we're up into
13 doses like this at, let's say 50 or 60 micrograms, some of the
14 animals will simply die of the TCDD at the lowest dose that they
15 use here, which is at about, what's that going to be, something
16 on the order of probably .4 micrograms per kilogram per week
17 in four weekly doses, there was no effect. In other words,
18 that dose of TCDD did not increase the susceptibility of these
19 animals to infection. And that is a common finding wherever a
20 dose response has been worked through.

21 Q What is a direct challenge test?

22 A Well, a direct challenge test, if you are familiar
23 with a tuberculin test, what's done in tuberculin tests is to
24 inject into the skin, I think they do it on the forearm if I

1 remember correctly, into the skin, not under the skin, the skin
2 is thick, you realize, and so they inject right into the dermal
3 layer of the skin, either -- well, usually it is a killed
4 preparation of tuberculin organisms, and then what we're looking
5 for is to see how the body responds to it. Now if you have
6 never had tuberculosis the response will be minimal because
7 the body has never learned about tuberculosis. If you have had
8 an infection relatively recently and it's been active, why there
9 may be a very vigorous response. And if you had an infection
10 twenty years ago it may be somewhat lesser because there has
11 been a lot of forgetting going on during that period of time.
12 So what we're looking at here are invasion of white cells to
13 attack that presumed invader.

14 Q Now, what does the line on this chart mean, the
15 horizontal line in the middle of the chart?

16 A That just identifies the control level of mortality.
17 In other words, animals that were not given TCDD but were given
18 the information, and this is a challenge, too, for that matter,
19 it is an infectious challenge. We are given the microorganisms
20 in 25 percent of the animals that were not treated with TCDD died
21 of the infection. The no effect level is .5, I don't remember
22 what I estimated it at, but it was calculated.

23 Q As .5 micrograms per kilogram?

24 A Yes, it intersects the .5.

1 Q With respect to the dose that you calculated from
2 Sturgeon soil, would that dose be sufficient to effect the
3 immune system in the mice that were --

4 A As reflected here, no, this no effect level is a
5 half a microgram, and this is a thousandth of a microgram,
6 that's a difference of, what, 500 fold.

7 Q So it would be 500 times less than the dose necessary
8 to cause an immune --

9 A I beg your pardon, I'm looking at this other no
10 effect level. I misstated myself. We have, or have I? No,
11 we have with respect to the dose at, that we calculated at
12 Sturgeon, for the sake of simplicity call this .5 or change
13 it up to 5, and that means that we have a 10,000 fold difference.

14 Q 10,000 fold difference?

15 A Well, the difference between point -- we have the
16 difference between .5 and .00005, essentially.

17 Q So the Sturgeon dose calculated by you would be
18 10,000 times less than that required to begin to show an effect
19 in the mice?

20 A In this direct challenge experiment, yes.

21 Q Now, when the animal's immune system is affected by
22 a sufficient dose of TCDD, is this a permanent effect?

23 A No, it will recover, but it takes -- it takes a
24 substantial amount of time in an experimental animal, it will

1 take months.

2 Q To recover?

3 A To recover.

4 Q But does recovery occur?

5 A Eventually these return to normal.

6 THE COURT: Before you get into your next subject,
7 is this a good point for a short break?

8 MR. HEINEMAN: Yes, sir, fine.

9 THE COURT: Okay, we will take a short break at this
10 time. I would remind you, and this would go for any other breaks
11 we take during the day, that you are not to discuss this matter
12 among yourselves, with anyone outside the jury panel, or as of
13 yet form any opinions or conclusions about the matters on trial.
14 The Court is in recess.

15 (At this time Court was in recess.)

16 MR. HEINEMAN: Your Honor, at this time we would move
17 the admission of Defendant's Exhibit 1282.

18 (Defendant's Exhibit 1282 was offered into evidence.)

19 THE COURT: Any objection?

20 MR. CARR: None, your Honor.

21 THE COURT: Admitted without objection. Thank you.

22 (Defendant's Exhibit 1282 was admitted into evidence.)

23 Q (By Mr. Heineman) Dr. Dost, in your review of the
24 testimony of Dr. Zahalsky on behalf of the plaintiffs, do you

1 recall his testifying about a study by a man named Boorman?

2 A Yes.

3 Q Let me show you what's been marked as Plaintiffs'
4 Exhibit 260, yes, it is 260. Have you seen that before, sir?

5 A I've seen a photograph of it.

6 Q All right, is this the photograph that you have seen,
7 sir?

8 A Yes, it is.

9 (Defendant's Exhibit 1283 was marked for identification.)

10 Q What has been marked as Defendant's Exhibit 1283, sir?
11 Is that a photograph of Plaintiffs' Exhibit 260?

12 A Yes, it is.

13 Q Now, are you familiar with the Boorman article itself,
14 sir?

15 A Yes, I am.

16 Q And did you cause a chart to be made of some of the data
17 from some of the data in there --

18 A There is a table in the paper, and it was copied directly
19 from the, that is, it isn't a photocopy of the page, but the
20 data has been copied in essence in a table.

21 Q And based upon your review of the testimony of Dr.
22 Zahalsky, is that the table from which Dr. Zahalsky purported
23 to create this exhibit?

24 A Yes, it is.

(Defendant's Exhibit 1284 was marked for identification.)

Q Now, let me show you what's been marked as Defendant's Exhibit 1284, sir. Can you examine that and identify it for me, please?

A Yes, it is a copy of a table that was in an article by Boorman that was published in Environmental Health Perspectives, and we do not have a specific reference on here, but it is the same article.

Q It is adapted, it's taken directly from this Boorman article?

A It is taken from the article and it is the data from which Dr. Zahalsky's chart or table was derived.

Q All right. Now, do you agree, sir, with the testimony of Dr. Zahalsky with respect to that chart?

A No, I do not.

Q All right, I wonder if you would step up here and explain to the jury why you don't agree with this chart.

A I'll put this up here where it can be seen. The reason that I don't agree is because some very important data has been left out of the chart and there has been an incorrect representation of the dose, and also in reading the testimony, I saw no description of the experiment and the nature of the experiment is of considerable importance because the animals -- and it was necessary to go to the original article, Boorman's article

1 is a review, and they reproduced from an earlier paper done by
2 the same group, these data, and in the earlier paper they des-
3 cribe more completely the manner of treatment of the animal.
4 What they did was to administer to pregnant mice TCDD seven days
5 prior, I believe, to the birth of the pups, and then at seven and
6 fourteen and twenty-one days following during the period when
7 they are nursing, so that the animal, the pups were presumably
8 exposed while in the uterus, and then exposed through the milk,
9 and this a common way of exposing very young rodents in the
10 experimental context, that is, to treat the mother and deliver
11 the material in milk. There is a very serious error made in
12 representing the dosage. When we speak of parts per trillion
13 or million or billion, we are talking about concentration. We
14 do not use this representation in discussing dosage, because the
15 only thing that dosage tells us is how much went into the animal
16 for unit of body weight. It doesn't tell us anything about the
17 concentration in the liver or in bone or in the kidney or the
18 brain or any other tissue. The concentrations within organs is
19 going to vary with the organ. So this tells nothing about what
20 is, it doesn't represent what's happening in the animal, neither
21 does the dose in milligrams per kilogram, but it is the only
22 information we have when we administer a chemical to an animal.
23 We don't know where it went, we know enough about a chemical we
24 can say, yes, so much of it is in the liver and the kidney and

1 so forth. The other error which is extremely critical, even if
2 we accept this idea after parts per trillion as representing the
3 dose, if it were parts per trillion, it would mean that the dose
4 was .005 micrograms per kilogram and .015 micrograms per kilogram,
5 accepting for the moment that this improper reference is the way
6 to use the language. The fact of the matter is that the doses
7 that were administered were a thousandfold higher. This is
8 milligrams per kilogram, a thousandth of a milligram per kilogram
9 is one microgram. This is an erasable -- the dose is one micro-
10 gram per kilogram per week. So the dosage here has been repre-
11 sented as a thousand times lower than it really was. I don't
12 have a great deal of objection to changing the terminology
13 because Dr. Zahalsky was trying to explain the nature of these
14 cells. He did use a different convention than the authors did
15 for describing the results. He talks about percentage of in-
16 hibition, that is, that means how much of the activity of the
17 those cells was decreased. Whereas, the authors speak to how
18 much activity was left. That's not serious, that's just a
19 convention. But I want to point out that he is talking about
20 in this case the loss of competency and the loss of competency
21 obviously would be greater as the dosage increases. The impor-
22 tant thing is however that he left off the lowest dose, he left
23 off the lowest dose. Now, this information, this is the number
24 of cells in the bone marrow. It is not just cells making red

1 blood cells, but all cells in the bone marrow. These are cells
2 that will eventually form red blood cells as he states, these
3 are cells that eventually will form macrophages, similar cells
4 that actually, I mentioned cells that go out and surround,
5 engulf foreign material. The important information in this
6 paper is the dose response relationship. Now, these other
7 chemicals, don't worry about -- we simply wanted to reproduce
8 all of the material so that you can see what the original table
9 looks like. This is the data that we're concerned with, and
10 surely enough there is a dose response, one microgram per
11 kilogram per week, 5, 15. And as you might expect, here is an
12 effect, 15 micrograms is a good size dose for any animal, any
13 rodent, and here only 61 percent of function has been left,
14 and they describe the statistical validity of that. In
15 other words, that is a result in which they have great confidence.
16 This is also a result that they have confidence in, but somewhat
17 less because it isn't that far removed from the control. These
18 numbers are percentage of control, and that's the reason we
19 don't see a separate line for the control information. In this
20 one, they didn't determine the effect for whatever reason. At
21 the lowest dose, but again there is 68 percent of the function
22 left here, 37 percent at this higher dose, which is not surprising.
23 In this case there was a more severe effect even than the others
24 at 15 micrograms per kilogram. Very little effect at 5, and you

1 can see here that they consider, or the statistics tell them
2 that 90 percent is so near to a hundred percent in this experi-
3 ment that you can't tell the difference. And here at one micro-
4 gram per kilogram is actually these animals have more than the
5 normal amount of cells, don't take that as meaning that a low
6 dose of TCDD makes the bone marrow work better. This is the
7 kind of variation that one is going to expect in an experimental
8 context. What I want to show you here, though, is we have a
9 no effect here at one microgram per kilogram. A plot of a dose
10 response curve which suggests that you are going to have a no
11 effect level at this point, but the data isn't there and all we
12 can do is say that is suggested. Here we clearly have a no
13 effect dose with given the statistics, the animals are even
14 better than controls. Again, when I see information like that
15 it doesn't mean they are better, it means that there is enough
16 statistical variability, but given the location of this point
17 point and this point, it is very clear that there is a no effect
18 dose here. So at the dose level that he did not include in
19 his testimony there is clearly a dose, a no effect dose.

20 Q Now, what is the no effect level there, Dr. Dost,
21 is one microgram per kilogram?

22 A In this experiment, yes.

23 Q Now, was this an acute exposure?

24 A Well, it took place over a period of, I think, four

1 weeks in which the material was administered prior to birth of
2 the pups, and administered periodically after the birth of the
3 pups, so that there were four doses at this level and I think
4 the total period was over four weeks.

5 Q So would that be called a chronic exposure?

6 A No, it would be -- of course when you define those
7 terms, why everybody has his own definition of what is acute,
8 I guess I would call it, it is a short subchronic experiment,
9 but it could even be called a long acute experiment. It is a
10 little difficult to say when you have that kind of a dosing
11 schedule. Their objective was to maximize, at a given level
12 of input they wanted to maximize the exposure to those pups.

13 Q Now, what's the difference between the no effect
14 level of TCDD demonstrated on that dosing of those rat pups
15 and the concentration or the exposure level or the dose,
16 pardon me, the dose computed from the Sturgeon incident?

17 A Well, we're back at microgram per kilogram again,
18 and if I remember that would be, the difference would be
19 222,000.

20 Q 222,000 what?

21 A 222,000 fold difference between, that is ~~the dose~~ that
22 we've just been speaking of here at one microgram per kilogram
23 per week, or whatever the period would be, is but one of those
24 doses, one of those four doses is 222,000 times higher than

1 this exposure that we hypothesized.

2 Q From Sturgeon?

3 A From Sturgeon.

4 Q Now, and that was a no effect level when it was
5 administered -- when that dose, 220,000 times more was
6 administered once a week for four weeks?

7 A It is my recollection that that was the schedule,
8 it was one time prior to the birth of the pups, and then three
9 times afterward.

10 Q Now, Dr. Zahalsky testified about proteases. Do
11 you recall that testimony?

12 A I remember something about it, yes.

13 Q He suggested that proteases --

14 MR. CARR: What page, Counsel?

15 MR. HEINEMAN: 135.

16 MR. CARR: What day?

17 MR. HEINEMAN: May 3rd.

18 Q He suggested that proteases act on proteins and attack
19 tissue. They attack protein in tissue, do you recall that?

20 A Yes.

21 Q Do you agree with what Dr. Zahalsky said?

22 A Only in part.

23 Q All right. Would you tell us about your disagreement
24 there?

1 A Well, proteases are in fact enzymes that break down
2 protein. They do not act, when you say tissues, I am assuming
3 that is living tissues.

4 Q Yes, sir.

5 A They do not act in living tissues except under extra-
6 ordinary circumstances. They only act, let me explain. Proteases
7 are present in all cells. Now, I'm using the term very generally,
8 it is my recollection he mentioned a couple of proteins in here.
9 But proteases exist in cells for the purpose of degrading
10 proteins, enzymes, other proteins in the cell when they are no
11 longer working and have to be disposed of. In fact, proteases
12 are responsible for regulating the lifetime of certain proteins
13 in the cell, and this is a very effective system. If a protein
14 should not stay there too long, it will be in the course of time
15 depending on its size be degraded by the proteases that are
16 always in the cell. There are also proteases in certain cells
17 in the circulation and their responsibility, and they are part
18 of the immune response, too, in a sense because their function
19 is to break down. If a cell engulfs another cell, it's got to
20 do something with it, it can't just continue to hold onto that
21 cell. It's got to break it down, it's got to digest it so the
22 cell eventually, that is picked up eventually is digested. There
23 are a certain amount of free protease in the circulation as well
24 because they will tend to leak out of cells that have failed,

1 they will leak out of the macrophages that are in the tissues
2 and in the blood, just in the normal course of attrition of
3 the cells. So there is always a level of a variety of proteases
4 that break down various kind of protein in the circulation. And
5 if a cell is in, a cell is severely damaged, for example, a
6 lung cell, if a heavy smoker, the cell is alive and it is
7 functional but it is not well. And in that circumstance a
8 cell in that marginal zone between function and nonfunction or
9 life and death could very well be attacked by proteases that
10 circulate. But they don't attack healthy tissue.

11 Q Now, what is the purpose of them? As I understand
12 it, a protein is a chain of amino acids, didn't you tell us
13 that the other day?

14 A That's correct.

15 Q Okay, now when the body -- and does the body
16 normally dispose of protein as an everyday occurrence?

17 A Oh, certainly, because they either wear out or they
18 are no longer needed, and since we do not repair a protein
19 that isn't working properly, the body degrades it, makes use
20 of those amino acids, starts over again, uses them for what-
21 ever it needs amino acids for.

22 Q Is it something like using building blocks?

23 A Well, the structure of a protein, yes, is like building
24 blocks. There are a relatively limited number of amino acids,

1 probably twenty or so, that are incorporated into proteins, and
2 the proteins are very, very long, the molecular weights I think
3 I mentioned the P450 enzymes as having the cytochromes as having
4 molecular weights on the order of 50,000, well, the molecular
5 weight of a typical amino acid may be anywhere from 50 to 150
6 perhaps, or somewhat greater. They are small components, but
7 the pattern, the sequence in a given protein is very, very
8 specific. And in fact it is even specific across species.
9 Insulin, for example, which is a complex protein from the pig
10 works perfectly well in the human because it is essentially
11 the same structure. There are minor differences, but it is
12 possible to go through and identify every amino acid and
13 establish its sequence in a protein. So we have in all our
14 cells all of these proteins that keep us functioning, and
15 that are really the structural framework of the cell. If
16 there is failure of a molecule, and every molecule is going
17 to fail sooner or later, something has to be done with it.
18 We can't leave the debris lying around like the trash in my
19 back yard. It is necessary to dispose of it, get it out
20 of the way and make use of the parts.

21 Q And is that what the proteases do?

22 A And that's what the proteases do, and they are within
23 cells, normally. I suppose in a perfect organism we wouldn't
24 find any in the circulation, but no organism is perfect, there

1 is always leakage and they appear in the circulation.

2 Q Now, Dr. Zahalsky on about the same page or the next
3 page, I guess 136, talks about some materials called Alpha 1
4 anti-trypsin and Alpha 2 macroglobulin, do you remember?

5 A Yes.

6 Q Those terms in that testimony?

7 A Yes.

8 Q Now, do you agree with what he said about those
9 molecules in that testimony?

10 A It was a little difficult to understand, but basically
11 no, I don't agree.

12 Q All right. Point out to the jury if you would what
13 Dr. Zahalsky said about these materials and the area of your
14 disagreement.

15 A Those two substances are inhibitors of proteases and
16 they exist in the circulation. They are very important com-
17 ponents of the plasma protein profile. In fact, they're even
18 greater quantities than, let's say, the immunoglobulins, which
19 we depend on for immune response. Their function is to inhibit
20 the activity of these proteases that we have been talking about
21 that inevitably escape from cells. In other words, they protect
22 us from proteases. And there are a number of genetic diseases
23 in which these protease inhibitors, particularly the Alpha 1
24 anti-trypsin, the reason it is called anti-trypsin is because

1 it is very efficient of the enzyme trypsin which is found in
2 cells and that is also an enzyme that is secreted into the
3 digestive tract to digest meat. But there are genetic defects
4 in which there is a deficiency of the anti-trypsin and people
5 with that deficiency seem to have a greater incident of
6 pulmonary emphysema and probably the reason for that is in the
7 normal course of assaults that our lungs take, whether we're
8 smokers or not, there are injured cells and those cells are
9 vulnerable or if trypsin and other enzymes have access to those
10 cells, there's going to be some other kind of break down and
11 that results in emphysema. The main thing about his statement
12 is that he describes anti-trypsin more or less effectively, but
13 then he talks about the Alpha 2 macroglobulin which is just
14 another kind of inhibitor as being some kind of a regulator
15 for anti-trypsin, and it is not. He also makes a peculiar
16 statement that if you increase the amount of Alpha 2 macro-
17 globulin, and I don't know how that is going to come to pass,
18 but if you increase the amount of Alpha 2 macroglobulin you
19 increase the activity of the proteases, and it is just the
20 other way around, it is just the other way around.

21 Q What do you mean "just the other way around"?

22 A Well, if you increase the amount of Alpha 2 macro-
23 globulin, its function is to inhibit those proteases, not to
24 increase them. It protects the body against the proteases.

1 Q Well now, if proteases don't attack healthy tissue,
2 why does the body have inhibitors, protease inhibitors?

3 A Well, I already mentioned that even in the best of
4 circumstances cells lose, they break up, they're going to lose
5 their -- the proteases, the enzymes that break down protein
6 that are inside the cell, that are there for normally to dispose
7 of unwanted or nonfunctional proteins in cells, and I think I
8 mentioned the other day about when the liver is injured, it
9 spills some of its enzymes into the circulation. The cells are
10 not broken, but they've become leaky. The same thing can happen,
11 a leaky cell presumably can lose its proteases, its trypsin, etc.,
12 or a cell that completely breaks up. So there's always going to
13 be some level, even though in an ideal state there wouldn't be
14 any, there's going to be some, and we have evolved that kind of
15 protection against that activity because we always have cells
16 in our body that are perhaps not perfectly healthy, and a cell
17 that is marginally healthy and has been injured in some way
18 would quite imaginably be affected by an enzyme that will break
19 down protein. And so we need to protect against that.

20 Q Now, Dr. Dost, is there any evidence from animal
21 studies or from anywhere else that you are aware of that TCDD
22 affects proteases or the protease inhibitors Alpha 1 anti-trypsin
23 and Alpha 2 macroglobulin?

24 A I know of no evidence, I don't think the question has

1 even been examined.

2 MR. HEINEMAN: Your Honor, it's noon, I notice, if
3 it's all right with the Court.

4 THE COURT: Okay, fine. We will break for lunch at
5 this time. We will resume again at 1:15, and the admonishments
6 that I have given you earlier will apply during this break also.
7 The Court is in recess for lunch.

8 (At this time Court recessed for the noon hour.)

9 Q (By Mr. Heineman) Dr. Dost, when we broke for lunch
10 we had just finished talking about the Boorman study, and I
11 wanted to ask you a little more about that. Do you recall
12 that Dr. Zahalsky testified that the effect of dioxin on the
13 immune system could be initiated by one molecule of dioxin?

14 A Yes, I recall that.

15 Q All right. Now, do you agree with that statement?

16 A No.

17 Q Now, what would be the possibility of an exposure
18 to or a dose of one molecule causing an adverse reaction in
19 an organism?

20 A No possibility at all.

21 Q Why is that, sir?

22 A There are a number of reasons, even if the substance
23 was highly reactive it is necessary for a reaction to take place
24 that there be a population of molecules. There would have to be

1 a lot of molecules present, a reaction that involves a single
2 molecule out of a population, a large population might imagin-
3 ably take place, but there are time factors. Molecules are
4 very, very lively things. They don't just sit in one place
5 all by themselves. And for a reaction to take place, time is
6 necessary. Chemical reactions are not just instantaneous. It
7 may seem like it when we look at them in a toy chemistry set
8 and pour two clear liquids together and they suddenly turn
9 purple, but there is a great deal of time involved. A molecule
10 has to get into a position where it can react, and then it has
11 to be held there for some finite period in order for whatever
12 it is going to interact with to react, for the two of them to
13 interact together. Otherwise, it may come into contact and be
14 gone again in an instant and more than likely never to return
15 again to that site.

16 Q Well, a molecule upon entering the body, what happens
17 to it? Where would it go?

18 A Anywhere. One molecule, we are not accustomed to
19 thinking in terms of one molecule. We are accustomed to thinking
20 in terms of large populations of molecules, and even a dose of
21 a microgram or a thousandth of a microgram is a very, very
22 large number of molecules.

23 Q Well, how does the fact that the Boorman study establishes
24 a no effect level affect Dr. Zahalsky's theory that one molecule

1 can initiate this immune function difficulty?

2 A Well, in terms of this particular study, we see a no
3 effect level as a microgram per kilogram. There are other studies
4 looking at other parameters that show somewhat lower no effect
5 levels. We are still talking about enormous numbers of molecules.

6 Q Now, Dr. Zahalsky has said, sir, that when this one
7 molecule would initiate this immune function problem, then other
8 dioxin molecules would be required to deal with the additional
9 messenger RNA and receptors, do you recall that?

10 A I recall something of that sort, yes.

11 Q Now, would a small number of molecules, a handful of
12 molecules have any possibility of causing the immune function
13 effect that is reflected in the Boorman study?

14 A Not really. Maybe I should illustrate what I talk
15 about when I say there would have to be an enormous number of
16 molecules, because --

17 Q All right.

18 A It happens that we know how many molecules are in a
19 given amount of a chemical. It may seem like an awfully esoteric
20 kind of thing to know, but I guess what I have to do is give a
21 brief lesson in chemistry. But we are talking about TCDD, and
22 I will draw the molecule and show you what I mean. Every atom
23 has a relative mass or relative weight. It isn't very much, but
24 it exists. And we've got a molecule here with chlorine and

1 oxygen and carbon and hydrogen. Now, you remember yesterday
2 I pointed out that at each of these corners there is a carbon
3 atom and there is also a hydrogen atom. And we rarely show
4 these in a structure unless they have some kind of meaning.
5 It happens that the atomic weight of hydrogen is 1 and the
6 atomic weight of carbon is 12 and of chlorine is 35 and a
7 fraction and oxygen is 16. And if we were to add those all
8 up, we would have the molecular weight, so to speak, well,
9 it is the molecular weight of 2,3,7,8 TCDD. We have four
10 hydrogens, and the atomic weight is 1, so that means we have
11 a total of 4, and we have two oxygens, and the atomic weight
12 is 16. So that means 32. And we have four chlorines, and
13 we will call that 35. It is a little more than 35, there is
14 a decimal in there, but let's keep the decimals out of it.
15 So we've got four chlorines, and then we've got twelve carbons
16 and they are each 12. So this comes out 4 times 35 is 140 and
17 12 times 12 is 144, and that adds up to 320. Now, that's the
18 molecular weight of TCDD. And a man named Avigodril back in
19 the last century somehow determined that for every chemical
20 compound, if we take its molecular weight in grams, in other
21 words, the gram molecular weight, if we take 320 grams of TCDD,
22 it has in it a very, very large number of molecules, as you
23 might imagine, the number is 6.023×10^{23} . What that
24 means is then since 10^{12} is a trillion, this means that there

1 is 600 billion trillion molecules in this much TCDD. Now, if
2 we were talking about, if we were talking about phenol, phenol
3 has a molecular weight of 94 and we would arrive at this in the
4 same way, 94 grams of phenol has the same number of molecules
5 in it, 180 grams of glucose has this many molecules. This is
6 a constant for all molecules, and it is a very useful number
7 as you can imagine. I guess to make use of that --

8 Q Dr. Dost, let me ask you a question. Originally when
9 Avigodril came up with that number, there couldn't have been
10 any way for him to count that. How in the world did he arrive
11 at it?

12 A I'm not exactly sure. He certainly didn't have
13 instruments to count that. He deduced it from the Table of
14 Elements and other information known at that time to chemists.
15 It was a rather astonishing intellectual feat, really.

16 Q Has it been established now as being accurate?

17 A It's still valid, and apparently now will remain that
18 way. In other words, his calculation was correct. So if we
19 want, we have 360 grams equal 6.023×10^{23} .

20 Q Excuse, was that 360 or 320?

21 A I'm sorry, 320. Then 320 milligrams which is a
22 thousand less would have times 10^{20} , 300 micrograms is 10^{17} .
23 Well, if we wanted to find out how many molecules are in one
24 microgram, well obviously we would take 1/320th, and I don't

1 do arithmetic like that in my head. What we really do is divide
2 this by 320. I've got the wrong exponent here, I'm sorry, I
3 would be giving you a million more than you really want. Okay,
4 we come up with a number that is 1.8822, we don't care too much
5 about these late decimals, times 10^{15} molecules in one microgram.
6 Now, we're talking about a microgram per kilogram, so we are
7 talking here about the number of molecules in a kilogram of
8 mouse, there are about forty mice to the kilogram. So if we
9 wanted to know how many molecules were in each of these mice,
10 then we would divide that number by approximately 40, a mouse
11 weighs about 25 grams. So since we are talking in this experi-
12 ment about a dosage per kilogram, we can leave it at that.
13 Here is the number of molecules per kilogram in this experiment
14 which did not produce an effect. Now, if we wanted, let's say,
15 to consider how many molecules, if our no effect level had been
16 a thousand times lower, then the number would be 1.9, I'm just
17 rounding this off, times 10^{12} , which is 1.9 trillion molecules
18 in a thousandth of a microgram, which is more than I can count
19 in an afternoon.

20 Q Well, how does that relate to the Boorman study?
21 I'm not sure I understand.

22 A I'm sorry, the Boorman study found a no effect level
23 at one microgram per kilogram. So the number that we are con-
24 cerned about is 1.9 times 10^{15} molecules.

1 Q So at no effect at all in the animal there was nearly
2 2 trillion molecules of dioxin.

3 A Per kilogram?

4 Q Per kilogram.

5 A Of body weight.

6 Q All right. Well, what does that number, how does that
7 effect what Dr. Zahalsky testified to as to the causation of this
8 immune effect?

9 A Well, it says that one molecule or even a few or even
10 a thousand are not likely to have any effect or will not have an
11 effect.

12 Q All right, now I believe you've testified the other
13 day that the Food and Drug Administration has arrived at a no
14 effect level in a human being, in a 70 kilogram man, is that
15 right?

16 A They have arrived at a conclusion. This is not measured
17 in human toxicity studies, this is information derived from animal
18 experiments and they have determined or decided that the no effect
19 level in man -- that they can establish a no effect level in man.

20 Q All right, what is that no effect level?

21 A It is one nanogram per kilogram. They use a figure of
22 70 grams just on a general assumption of a 70 kilogram person.

23 Q So that in a 70 kilogram man, which is what you say,
24 150 some pounds?

1 A 154 pounds.

2 Q There is a 70 nanogram no effect level?

3 A That's correct.

4 Q All right, how many molecules would that be?

5 A Well, we will multiply this figure by 70, and that's
6 for a 70 kilogram individual, it would be 1.31 -- well, let's
7 call it 1.32 times 10^{17} molecules, which would be to put it
8 in manageable form, a trillion is 10^{12} , so we've got 1.32 times
9 10^5 or 1.32 thousand trillion.

10 Q Molecules?

11 A Molecules.

12 Q Of TCDD?

13 A Of TCDD.

14 Q At a no effect level?

15 A If we consider this to be a no effect level, yes.

16 Q Now, in terms of the immune system, sir, when you say
17 that an adverse effect on the immune system will cause additional
18 infections, is that correct?

19 A Yes.

20 Q What kind of infections? Are you talking about a cold
21 or a runny nose or what kind of infection is evidenced in an
22 immune suppressed organism?

23 A Well, an organism that has a significant -- well,
24 to start with there is a lot of excess capacity. We're always

1 fighting off infections of one kind or another. An individual
2 that has a noticeably compromised immune system is suffering
3 probably continuous assaults, wounds don't heal, probably con-
4 tinuous respiratory ailments. The other problem is that these
5 tend to progress, not only are they allowed to take root or
6 take foothold, but it would seem to me that they would progress.
7 And I think that the, once the barrier, so to speak, had been
8 overcome of our excess capacity for immune protection, it would
9 seem that the individual animal, human or any animal presumably,
10 or for that matter any organism that relies on an immune system
11 would, I think, suffer fairly substantial infectious disease.

12 Q Let me start, first of all, with your statement about
13 excess capacity. What are you talking about?

14 A Well, the immune system has far more capability than
15 is necessary to help us get over a simple cold or to get over
16 a case of pneumonia. Now, to be sure, if the infection is
17 massive enough, if it just absolutely overwhelming or if the
18 individual has a protein deficiency and is unable to synthesize
19 protein, and this is a significant problem with heavy protein
20 malnutrition, people cannot manufacture antibodies. It seems
21 to me that if the failure of immune function is really signi-
22 ficant it is going to mean pretty nearly a collapse of the
23 immune system. There is a great deal of excess capability
24 available. If we interfere with part of the immune system we

1 do not necessarily cause animals at least to succumb to infection.
2 We can show a lot of different parameters of immune function, we
3 can look at the ability of the rat, you can inject a challenge
4 into the rat paw and to see whether the paw swells as it ought
5 to because cells are being brought into the paw, or we can look
6 at titers of antibodies.

7 Q Come again now?

8 A We can look at the amount of antibodies available in
9 the blood.

10 Q What is a titer?

11 A A titer is a measurement of the amount that is avail-
12 able for reaction with an infectious system. So there is lots
13 of ways of quantifying depressed immune function, and we know
14 that TCDD at substantial doses depresses immune function, there
15 isn't any question about that. Not all immune function, there
16 doesn't seem to be any impairment of the response to viruses,
17 for example, to the extent that laboratory experiments have
18 demonstrated, but animals do not die of infection that are
19 heavily attacked by TCDD, and we know that their immune function
20 must be at least partially impaired, you see, so --

21 Q Now, when you say that there is excess capacity, why
22 then if the immune function has more capacity than is affected
23 by dioxin exposure, why is it that takes so long to fight off
24 a cold? Why is it that an infection can last a while in an

1 organism?

2 A If I knew why it took so long to fight off a cold,
3 I would have probably become famous long since. That's a problem
4 that has yet to be solved. The fact remains, though, is that
5 colds, the cold that I have didn't just sweep in and wipe me
6 out. There were a couple of days where I was worried about that,
7 perhaps, and then there was a day or two perhaps that I was worried
8 that I wouldn't die. But nonetheless, that interaction took
9 place, and it has protected me, and I am having a little trouble
10 getting the last of it resolved, but if I didn't have an immune
11 system, I would have dropped very quickly. Why it takes so long
12 is a good question. Certainly these, the antibodies are continuing
13 to be produced. The cells that are fighting off specific organisms,
14 lymphocytes and so forth, are continuing to be produced. Why
15 they can't just step in and mop up instantaneously is something
16 that I don't understand.

17 Q If the type of disease for a quenched immune system
18 would be something more than just a cold --

19 A Not necessarily. I guess what I am saying is that if
20 you were to reduce the function of some component of the immune
21 system by half, let's say, by intoxication, that doesn't mean
22 that you have eliminated capacity sufficient to cause that
23 animal to succumb to some kind of an infection. There is
24 reserve capacity. We have impaired the animal, but we haven't

1 impaired it to the point where his ability to fight off this
2 infection is really affected, and that's really very clear
3 in all of the experimental work, because when animals are
4 challenged with a lethal dose of, well, I showed a dose response
5 relationship earlier. At the highest dose of TCDD, there is
6 still survival. A quarter of the animals survived this impact
7 that in untreated animals killed a quarter of the animals, and
8 we see in animal colonies where TCDD is being administered to
9 animals, there is no change in their response to the host of
10 infectious diseases which exist in most animal colonies.

11 Q Well, if the immune system were devastated, were
12 quenched, as I think Dr. Zahalsky used the term, what would
13 happen?

14 A We would have no, we would have no protective response
15 to infectious disease, and this is the kind of problem that
16 happened. People who have leukemia, who has a bone marrow
17 transplant, have to go through a process by which all of the
18 bone marrow is destroyed, usually by radiation or by chemotherapy,
19 and then new bone marrow is transplanted or, yes, new bone marrow
20 is transplanted into those bones, which is a very painful process
21 but interestingly enough, and that new bone marrow will then
22 proliferate and will form these kind of cells that we are
23 talking about here and repopulate with normal white cells. A
24 person who is in that stage where they have had bone marrow

1 destroyed have had virtually all of their other immune system
2 destroyed. They are very vulnerable to infection and it is
3 necessary to protect them with great care. People who have
4 organ transplants are usually maintained on drugs that depress
5 the immune system very, very sharply. There are many such drugs,
6 and they depress the immune system to the point where that heart
7 is not being rejected or that kidney is not being rejected. If
8 a person is not fortunate enough to find an organ that is com-
9 patible, they are in a continuous state of depressed immune
10 function, continuously vulnerable to major infection and have
11 to live a very special kind of life in order to make that
12 possible, a great deal of protection, antibiotics a lot, and
13 so forth. And of course we occasionally read about the children
14 born completely without an immune system, and no one has solved
15 that problem yet, and they live in a bubble and not all that
16 long. They have to be completely protected from any contact
17 with other humans or any kinds of organisms, and their food is
18 sterilized before it is sent into them and so forth. They have
19 no protection.

20 Q So that if an organism, an animal does not have a
21 catastrophic illness or if a scratch doesn't become blood
22 poisoning or if a minor infection doesn't advance to a very
23 serious infection, does that indicate that the immune system
24 is working?

1 A Yes.

2 Q Let me go next, Dr. Dost, to the kidney as an organ,
3 if I may. Dr. Silbergeld testified on April the 12th, 1984,
4 Page 97, that the animal demonstrates that TCDD destroys kidney
5 cells. Do you recall that testimony?

6 A Yes.

7 Q Do you agree with that?

8 A No.

9 Q Why not?

10 A Well, the animal data show really just the opposite.
11 TCDD at survivable levels does not have an appreciable effect
12 on the kidney even over long periods of time. If I may refer
13 again to Kociba's work and other chronic studies, long term
14 studies, the kidney is not significantly affected. One of the
15 interesting aspects of the work that Dr. Kociba did was that
16 he found that the normally occurring degenerative kidney
17 disease that occurs in all old animals was significantly
18 diminished at the high dose of TCDD. What that means, I don't
19 know, but it certainly does mean that there was no increased
20 incidence of kidney toxicity in those animals, nor was there
21 any at the lower doses.

22 Q Now, how exactly, I want to get back to these Kociba's
23 studies in just a minute, but how exactly does the kidney work?
24 You were talking before about organs in which -- that handle

1 large volume of blood and organs that have a lot of fat so that
2 TCDD can be stored in them. Where does the kidney fall in that
3 spectrum of types of organs in the body?

4 A Well, the kidney is our primary excretory organ for
5 soluble substances, and it would -- I think I'll draw a picture
6 if I might. This by no means is a precise rendition of a kidney
7 tubule, but the kidney is comprised of a large number of individual
8 units, kidney tubules, and the way they work is there is a filter,
9 any fat. The blood breaks up into small vessels and it comes in
10 and there is just a big clump of vessels here surrounding, that
11 is a kind of a cup, and it is really a filter, and as the blood
12 goes through there, probably as much as ten percent of all of
13 the fluid in the blood actually goes through that filter. The
14 cells don't come through, the large proteins like the plasma,
15 albumin, which is a normal part of the plasma, and the immuno
16 proteins that we were talking about, all those large proteins
17 don't go through. This is a rather convoluted tube, but I'm
18 just going to draw it straight. And it is surrounded by cir-
19 culation. And what happens is that all of the water, most all
20 of the water, probably 99.9 percent of all of the fluid that
21 went through the filter is brought back out. But in the process,
22 many of the materials that we don't want to bring back are left
23 in that fluid. And most of the materials that we don't want
24 to get rid of like glucose, you know, we have lots of glucose

1 in the blood. Normally about 100 million grams per 100 milliliters,
2 and that's going to come through here, too, because it is a
3 small molecule. So it goes down here and we reabsorb it back
4 out again if we are normal. Some people can't do that very well,
5 but we recover all of that glucose, unless we've just eaten a
6 candy bar or something like that, there may be too much to
7 recover, and some of it will escape. And then there are some
8 other cells that actually have the function of secreting certain
9 things into the urine, ammonia and so forth. But anyway, in
10 essence, it is a filter followed by a scavenging unit which
11 brings most of the fluid and valuable stuff back in, and those
12 thousands of units all converge in the center of the kidney.
13 And then from the kidney there is a tube that goes to the
14 bladder. So what we have here is a filter, and it deals --
15 if we are talking about a toxic substance or any kind of a
16 foreign substance, if it is at all soluble in water, it is
17 delivered to this point and hopefully left in the urine and
18 disposed of.

19 Q Now, the thing that you have drawn, the mass of
20 blood vessels at the top is called what?

21 A It is called the glomerulus. I had nothing to do
22 with naming these.

23 Q All right, and the tube is called the tubule?

24 A This is the kidney tubule, and it has a proximal

1 segment, actually it comes in kind of an S shape, and this is
2 the distal segment and there are different functions in dif-
3 ferent lengths of that tube, but --

4 Q All right. Now, how many of those operating units,
5 let's say, the glomerulus and the tubule with its reabsorption
6 collar, and that sort of thing, how many of those are in the
7 kidney?

8 A Quite a few million.

9 Q So the blood is coursing through this thing?

10 A These are little bitty vessels here, you see, the
11 kidney circulation has divided into an enormous number of
12 vessels. This thing is only one cell in thickness, you know,
13 the wall of this tubule is only one cell. So there is really
14 a collar of cells, you might say. In other words, if you were
15 to -- took a tube from the end, it would look something like
16 this, you see. So it is a very, very small structure.

17 Q And so the blood courses through the kidney, does
18 the kidney receive its own blood supply for energy and life
19 for the cells?

20 A Well, that's right. It happens, you see, all of
21 these tubular cells receive their circulation from the same
22 blood flow that is bringing, it is a very efficient system,
23 we don't have to have a whole separate plumbing system in
24 order to supply nutrient to the kidney itself. The same

1 circulation that brings fluids to the kidney and carries wastes
2 in here and comes back down here, the same circulation it
3 brings out the fluid that we don't want to dispose of, provides
4 the energy resources for those cells.

5 Q All right. Now, when Dr. Kociba studied the kidney,
6 how many occasions did he do it?

7 A I'm not sure what you mean, how many.

8 Q How many separate studies were there in which this was
9 observed?

10 A Well, they didn't find -- he had done two studies,
11 one thirteen week study that I think I mentioned yesterday,
12 and then the long term, lifetime study that they did.

13 Q Was one of them in 1976?

14 A The short one was reported in 1976. The longer one
15 was reported in 1978, and they had a substantial number of
16 animals at each experimental level, of course.

17 Q Now, what did the 1976 study show with respect to
18 kidney effects in animals?

19 A It didn't show any kidney pathology that I recall.

20 Q Were there any blood chemistry results?

21 A Oh, yes. They did a variety of blood chemistry,
22 and they didn't find, except -- I believe at the higher
23 dose they found some elevations in levels that would suggest
24 that might be interpreted upon the kidney, but there could

1 also have been effects upon the liver, too.

2 Q Now, you say the higher doses?

3 A They used a maximum dose in that study, in the 1976
4 study, of one microgram per kilogram per day.

5 Q All right, was that high enough to cause death in
6 some of the animals?

7 A It certainly did.

8 Q And at that high dose, there were some blood chemistry
9 results, is that right?

10 A Yes.

11 Q Did they do any kidney pathology on that occasion?

12 A Yes.

13 Q Did they find any pathological?

14 A To my recollection they found no pathology at all.
15 They found no change in the architecture or the shape of the
16 kidney cells.

17 Q All right, so that in 1976 Kociba did not show that
18 TCDD destroys kidney cells?

19 A That's right, and the same outcome emerged in the
20 study reported in 1978 where they had maintained it for two
21 years.

22 Q All right, now the 1978 was a chronic study for
23 the lifetime of the animal?

24 A Yes.

1 Q And how did the kidney effect in the 1978 study
2 relate between the dosed and the undosed animal?

3 A Well, I think I mentioned a short time ago that the
4 normal incidence of degenerative kidney ailments, all, parti-
5 cularly rats, as they age, have a substantial incidence of
6 kidney damage. The nephron here, this unit that we have been
7 talking about will just simply fail, and there will be areas
8 in the kidney where is scar tissue and a lot of the number
9 of functional units decreases with age.

10 Q That naturally occurs?

11 A That occurs in any, it occurs in all of us, of
12 course, but it is quite visible and easily quantifiable in
13 experimental animals because we are examining their pathology
14 all of the time. And the interesting thing in that particular
15 study was that the incidence of that kind of degenerative
16 image decreased at the highest dose, and decreased to an
17 extent that was statistically significant.

18 Q Decreased in the animal that had received the highest
19 doses of TCDD?

20 A Yeah, I'm afraid so.

21 Q Well, is there any evidence in the animal data that
22 the kidney is adversely affected by the 2,3,7,8 TCDD exposure?

23 A Well, really only at doses that have caused the
24 animal to collapse, and sometimes not then. That's where it is

1 seen, is at very, very high doses where the animal is probably
2 mortally affected.

3 Q At below that high dose?

4 A It appears that there isn't an appreciable impact on
5 the kidney.

6 Q All right, sir. Now, Dr. Dost, I'd like to talk next
7 about cancer. I wonder if you would define cancer for us.

8 A Cancer is a condition in which cells have lost their
9 regulation, usually have regressed in their differentiation.
10 By that, I mean that in all of our cells begin the same, and
11 as we develop, cells become, we use the term differentiation,
12 and that I think is self-explanatory. Eventually we have to
13 have a liver and we have to have bones and we have to have
14 kidneys, and as the animal develops, cells begin to take
15 direction toward the formation of the various organs, and so
16 as the embryo develops, the cells become more and more and
17 more different until finally we have a complete organism
18 with arms and fingernails and eyes and all of the structures,
19 but they all started with similar cells. In the case of tumor
20 cells, there is a tendency to go back part way down that
21 ladder and lose the differentiation. Now, they don't go all
22 the way back to the point where they are just like initial
23 embryonic cells, but cancer is a collection then of cells
24 that is reproducing by itself and forming usually a tumor

1 that has some form, and it has been removed pretty much from
2 the formal regulation that the body exerts on itself. These
3 cells have become autonomous. They are self-governing to a
4 large extent, so that they are no longer controllable and they
5 grow and grow and grow at a slow rate. But they grow all by
6 themselves, without regulation by the rest of the body.

7 Q Now, when you say regulation, how is it that the body
8 regulates a cell's growth?

9 A Well, we are really a very well-organized system
10 and not only does the body regulate each cell's growth, but
11 the companions, all of the cells in the liver have a common
12 mission, all of the cells of a given type, and they exert
13 regulation on one another. When they lose that regulation,
14 well, cells that have become a cancer cell may start to
15 multiply, and it's going to form a larger and larger colony
16 of cells that eventually it is independent of the cells that
17 are around it. But we regulate our cells, we have hormones
18 that turn cells on and off. If we want to cause sugar to be
19 absorbed by cells, we release insulin and that tells cells
20 to absorb the sugar. And that is a form of regulation that
21 arises from a central point. That particular phenomena doesn't
22 have anything to do with cancer except when you have a cancer
23 of the organ that produces insulin, and there might very well
24 be for a while a great deal of insulin produced so that we have

1 caused a problem elsewhere in the body. Generally, the problem
2 with cancer is it reaches a point where it invades and inter-
3 feres with the function or some of those cells may break off
4 and go to other parts of the body and continue to reproduce
5 by themselves without regulation.

6 Q What's the name of the latter condition you just
7 described?

8 A That's a phenomena known as metastasis, m-e-t-a-s-t-a-s-i-s,
9 and it simply means the movement of the cells from their origin
10 to some other location.

11 Q All right. Now, when you are talking about cells
12 developing, each cell nucleus contains, you mentioned a library
13 of material?

14 A Yes.

15 Q Called what?

16 A DNA.

17 Q Now, but you say it starts, the human embryo starts
18 from the joining of two cells, right?

19 A That's right, it starts from the fertilization of an
20 egg, and we have one cell, it divides into two identical cells,
21 and they divide. Early on, you have a structure that looks
22 rather like a raspberry that's comprised of a substantial
23 number of cells that are all identical.

24 Q Now, is the DNA in the nucleus in each of these cells

1 the same?

2 A Yes.

3 Q Now, what about when the differentiation begins to
4 occur, when cells start to become part of the bone or part of
5 the foot or skin or hair or whatever it is?

6 A All of the information is there, but by some process
7 that isn't really well understood, cells only utilize part of
8 the information, the part that's important to that function.
9 So if we have a liver cell, that is all the same information
10 that kidney cells have, but they don't use the part that's
11 related strictly to the kidney cells, you see.

12 Q And the kidney cell doesn't use the part of the DNA
13 cell that relates to what the liver parts use?

14 A Exactly.

15 Q So the DNA is the same in every cell?

16 A Yes, it is a question of which book we take off the
17 shelf.

18 Q Now, what does DNA have to do with cancer?

19 A Cancer is generally believed to start with some kind
20 of an injury or a lesion on DNA, so that DNA is changed and
21 not repaired, and that that change remains and somehow it
22 remains part of the genetic equipment in the cell, and ultimately
23 is triggered to develop into a tumor cell. It is a very long,
24 complex process. It doesn't just happen.

1 Q Have you develop a chart which would assist you in
2 explaining this?

3 A I have a chart I use in lectures for that very
4 purpose.

5 (Defendant's Exhibit 1285 was marked for identifi-
6 cation.)

7 Q Dr. Dost, let me show you what's been marked as
8 Defendant's Exhibit 1285 and ask you to examine that and
9 identify it for me, please.

10 A It is a -- it is the chart that I brought with
11 me that I use in describing the process. It is a general
12 description of the process by which a cell eventually can
13 become a tumor. And as it points out, it is hypothetical
14 steps, this is generated not from a specific body of research
15 data, but it represents the present understanding of the
16 process. All right --

17 Q Would you step up and use it, please? I am interested
18 in the term "hypothetical."

19 A Well, no one has determined fully the process by
20 which cancer is caused. That probably is the most, probably
21 it is the most heavily investigated problem in biology at the
22 present time is to try to establish just how cancer is caused.
23 There is a great deal known about how it is caused, there is
24 a great deal known about intermediate steps. But unfortunately

1 the process seems to vary with certain kinds of cancer, and it
2 also, and also there are some very specific and important pieces
3 of information that simply are not yet known, so it would be
4 presumptuous to say that this is the way cancer happens. It
5 does, I think, represent quite properly the general scope and
6 sequence of the process. So these are all, these are generali-
7 zations. They do not relate to some specific tumor type, nor
8 do they relate to any specific pattern that someone has laid
9 out. But the first thing that happens, you spoke of DNA, and
10 the first thing that happens is apparently a mutation of some
11 other damage to DNA in the cell, and that affects some part
12 of the genetic apparatus. Now, there is a concept of the
13 oncogene that has crept into the popular press, and it is a
14 very important issue of discussion among cancer researchers,
15 and that is the idea that there are segments of DNA, genes,
16 that by some mischance with very minor changes can be converted
17 into a gene that actually leads to cancer. In other words, the
18 idea is that we have in all of our cells genes that require
19 very little modification to touch them off and cause them to
20 take control of the cell, so to speak, and cause cancer. In
21 any case, there still has to be some kind of a change in the
22 DNA to start this whole process. And that step probably is
23 irreversible. But other things have to happen for it to be
24 expressed, and when I say, by express, I mean other things have

1 to happen for it to take effect. Now, one of the things that
2 apparently have to happen is that the cell has to divide, and
3 in dividing it, copies of its DNA for the second generation
4 cell. So that there is two identical sets of DNA. And if
5 that happens, the speculation is that probably it takes two
6 cell divisions in order to do that for that to be copied. So
7 at the same time I want to step back for just a moment. We
8 have talked about a DNA lesion, that is irreversible. There
9 is damage occurring to DNA all of the time in the cell, naturally,
10 and as a result of everything that we do in our environment.
11 And this lesion has to survive this repair process, and I
12 probably will have an opportunity to discuss the idea of DNA
13 repair a little later.

14 Q Dr. Dost, excuse me, when you talk about our environ-
15 ment, what are you referring to specifically?

16 A Diet, everything we do, our ultraviolet radiation
17 from the sun, our environment is kind of hostile place, and
18 we are taking in all of the time a lot of genetically active
19 material in our diet. If you were to go through everything
20 that you eat and identify all of the substances that are
21 known to have some kind of genetic activity as determined
22 in microbial mutation tests and so forth, you would soon be
23 searching trying to find out what in the world you could eat
24 that will sustain you without creating a problem. Apparently,

1 we are not creating problems by eating our daily meals because
2 our life span is increasing, but the fact remains that there
3 are lots and lots and lots of materials in the natural environ-
4 ment, both physical and chemical, that has capacity to at least
5 theoretically cause injury. Not only that, within the cell,
6 the intercellular environment is also very violent and there
7 are lots of very reactive molecular species created in the
8 normal metabolism in the cell. Perhaps I can talk about that
9 a little later, too.

10 Q When you talk about genetically active, what do you
11 mean?

12 A I mean it has the ability to interact with DNA.

13 Q Now, we've heard talk here about DNA and RNA, and
14 messengers and all that sort of thing. What does all that
15 mean? Is DNA a ribbon or is it a piece of --

16 A DNA is, okay, DNA is the primary source of information.
17 The RNA that we have talked about, there are a number of kinds
18 of RNA, but the only number of RNA that we have particular
19 concern about is that is made as a copy of a segment of DNA
20 and goes out into the cell to the ribosome or to endoplasmic
21 reticulum to form a pattern for the synthesis of some protein.
22 DNA is a very, very, very long molecule. There are probably
23 -- it is comprised of pairs. I may have to stop and go back
24 now and then because this is a little complex, but DNA, you

1 may have heard of the double helix, which was a book that
2 Watson and Crick wrote about thirty years ago when they
3 finally discovered what the configuration of DNA was.

4 Q And won a Nobel prize?

5 A And they won a Nobel prize. The DNA, I guess the
6 best way to describe it is to think of a zipper, an ordinary
7 zipper. And you know that a zipper has teeth and they come
8 together like so. Well, DNA is set up as pairs of bases,
9 and I don't need to describe what these bases are, but there
10 are four kinds in DNA. And I guess we could say that those
11 four bases are the letters of the genetic alphabet, and you
12 can imagine that there is an unimaginable -- that's not very
13 good language -- there is an enormous number of possible
14 combinations because these molecules are very, very long.
15 They carry an enormous amount of information. And if we think
16 of this as being, as three, four letters in this alphabet, and
17 there are words in the DNA and they are all three letters long
18 and each of those words codes for an amino acid. Now, when we
19 go through this process of inducing, which we've talked about,
20 the DNA opens up and we form RNA as I've just mentioned. And
21 that RNA carries the same code, this series of three letter
22 words. It is very long and it codes for these thousand or
23 two thousand amino acids that come together to make this protein.

24 Q So it unzips it?

1 A Unzips, half of it comes apart and the RNA is copied
2 off the other half, or it comes apart and RNA is copied off of
3 one half. But if you think about this zipper, for every 10
4 teeth it has taken one twist, this thing is twisted, and I
5 mention the number of bases. There are in the cell about,
6 between 10^8 and 10^{10} base pairs. In other words, in each cell
7 there are, let's say, 10^8 words or letters, how would I put
8 that? No, we will divide it into three and make words. We
9 will say there are 3 times 10^7 words of information in each
10 cell. That's 30 million, probably more than that, but the
11 point is that in these rather interesting three letter words
12 are the codes for all of the amino acids. And so it is the
13 sequence on the DNA that governs the sequence of synthesis
14 out in the cell. All right, if there is an error, if there
15 is an error in that sequence, then incorrect information is
16 going to be transmitted. Now, that may not make any difference,
17 it may be a serious error or a big chunk of DNA is lost and
18 the cell dies. That, fortunately, is not an unhappy circum-
19 stance except in the nervous system, but it is happening all
20 the time. In this situation we have inverted some kind of an
21 incorrect message. All right, it may stay that way forever
22 and nothing ever happen, but if the cell divides before it is
23 repaired, that lesion may be thick in the gene so that it stays
24 there and the cell then is hearing that message. Well then,

1 there are a number of processes that go forward that come into
2 the category of promotion and enhancement. Some of these words
3 are used interchangeably, sometimes you find progression used
4 here and sometimes translation. This question of cell trans-
5 formation falls up here away, but let me read this to you.
6 "It is assumed that specific physiological or biochemical
7 phenomena are required to facilitate the expression of the
8 initiating event." Now, that may be cell injury, there are
9 certain chemicals that are known to promote this effect. I've
10 mentioned the phorbol esters the other day when we were showing
11 that chart on the difference between promotion and initiation.
12 TCDD, phenolbarbitol, smoke condensates having been suggested
13 as promoters. And the promotion process appears to be reversible
14 at least in its early stages, in other words, if it does take
15 place before some of the other events going on, why then that
16 cell may just revert back to normal. It also is clearly dose
17 dependent and clearly there is argument about whether it is
18 a threshold or not, but it is very clearly dose dependent.
19 Then, the next step is what is called cell transformation, and
20 the cell changes its whole genetic makeup in some fashion and
21 it does different things than it used to do. It becomes
22 juvenile, if you will. It reverts back to a less developed
23 form. And one of the problems that it may or may not be
24 recognizable as a strange cell, as a different cell, and if it

1 is recognizable, then the immune system will clean it up. And
2 this is probably going on in our bodies all the time. From
3 this point forward, those cells when they reproduce they carry
4 all of these bad characteristics with them, and they begin to
5 reproduce. If they do evade the immune surveillance and they
6 could very well repeat characteristics of the normal cell that
7 are sufficient that the immune, the cellular immune cells, the
8 killer cells don't recognize them as being tumor cells or nearly
9 tumor cells. Well, so there is a progression here where we go
10 from a genetically injured cell to a cell that has to be further
11 stimulated in some fashion. And then from that we go to cells
12 that are producing, changing their character and eventually
13 becoming a sufficient population of cells that either they
14 form a tumor that can be seen or they may form a tumor in the
15 bone marrow that is recognized as, say, leukemia or something
16 like that, which is a circulating tumor in effect. There is no
17 mass, no hard tumor. I don't know whether I have made that
18 sufficiently clear, it is a very muddy process, and there a
19 lot of different -- there is a lot of alternative descrip-
20 tions that can be inserted.

21 Q I'm interested in the initiating event that you've
22 got there at the top. I'm not sure I understand how this
23 occurs. Is this something that has to come in from the
24 outside, or is it something that naturally occurs in the cell

1 because you talked about DNA repair and errors in the cell before.

2 A Well, DNA is, to start with, we have an enormous
3 amount of material that has to be manufactured, and we considered
4 that the manufacture of DNA in the cell is a very accurate process.
5 But the error rate is about one in every million base pairs. Well,
6 I've already talked about a situation in which we've got what,
7 10^8 , that's 100 billion, I beg your pardon, a 100 million base
8 pairs in the cell. And if we are, actually the number is higher
9 than that, it is probably about 300 million. And if we have,
10 if we have an error rate of one in a million, that means that
11 for the growth of a single cell there will have been 300 errors
12 just in that cell alone. Just in manufacturing DNA. And that
13 has to be corrected. There is a repair process that inspects
14 DNA. It is fascinating. It literally inspects it, like going
15 over a string of beads and find defective spots, slices them
16 out and fixes them. Now, that is not a perfect process. Some
17 repair may have been missed or there might be a mistake made in
18 the repair process, but if you consider that there are 300
19 errors in a given cell and if, oh, in the liver there is some-
20 thing in excess of one billion cells per gram, so let's say
21 10^9 cells per gram for a gram of liver, which is maybe would
22 be a piece that size, perhaps, there would be 300 billion errors
23 committed in the process of making the DNA for those cells.
24 So this process is going on all the time of error in synthesis.

1 The other thing that is happening is that, I'll just write that
2 down, the other thing that's happening is, as I said, the cell
3 is a very violent place and there are some very violent reactions
4 going on in the cell in the process of generating energy. There
5 are some very reactive oxygen atoms that are generated in the
6 cell, and most of them are trapped and are, don't cause any
7 harm, but enough of them get away that there is a very signi-
8 ficant and measurable amount of DNA damage by some of these very
9 reactive oxygen species inside the cell. And peroxide, hydrogen
10 peroxide is made inside the cell, and there are enzymes that get
11 rid of that stuff. There are traps that collect it, but some of
12 it inevitably escapes and the rate of injury of cellular DNA
13 can be measured by the collection of some of the products, be-
14 cause as I said, they slice out pieces of DNA and bring them,
15 they are excreted into the blood and into the urine and they
16 are disposed of. People in Bruce Ames' lab at Berkley have
17 done some interesting work on this, and what they find is those
18 products emerge at a rate that indicates cell damage to the
19 extent of 320 DNA damaging events per cell per day. And when
20 you multiply that by the number of cells in the body, which has
21 been estimated, there are various estimates, but the one I
22 generally use is 6 times 10^{13} cells, which is 60 trillion cells
23 in the body. Then we have an enormous number of events taking
24 place in the cell every day in the body, every day that can

1 be potentially leading to genetic damage of some kind. Virtually
2 all are repaired. Now, if the system was absolutely perfect,
3 we might not have cancer. But unfortunately the system is not
4 absolutely perfect and there are inevitably some errors in DNA,
5 some damage to DNA that persists. Now, also, external functions.
6 I've talked about some of the mutagens that are in natural sub-
7 stances, black pepper, broccoli, and when I say broccoli, I don't
8 mean to isolate it, I think we can say that about most vegetables,
9 don't worry about it. Carcinogens, benzo[a]pyrene in automobile
10 exhaust, or gasoline, benzene, lots of chemical carcinogens
11 that can also result in this DNA damage that serves as the
12 initiating step. If the concentration is high enough it can
13 cause these kinds of events, too. So we have all kinds of
14 things happening, very high repair rate that presumably is
15 not absolutely perfect, so you can see, there is an incredible
16 amount of activity going on in the cell all of the time that
17 relates to the potential for DNA damage in this initiating
18 event.

19 THE COURT: Mr. Heineman, is this a good point for
20 a short break?

21 MR. HEINEMAN: Yes, sir, it will be fine.

22 THE COURT: We will take a short break at this time.
23 The admonishments that I gave you earlier will apply during
24 this break also.

(At this time Court was in recess.)

(Defendant's Exhibit 1286 through
1289 were marked for identification.)

BY MR. HEINEMAN:

Q Dr. Dost, for the record, I have had these sheets
marked. Defendant's Exhibit 1286 is the calculation of the gram
molecular weight of dioxin.

A That's correct.

Q Showing of Avigodril's number at the bottom?

A Yes.

THE COURT: What was that, 1286?

MR. HEINEMAN: 1286, your Honor.

Q 1287 is the calculation of the total number of cells.

A Total number of molecules.

Q I'm sorry, total number of molecules, and what?

A Well, I made an error when I calculated the number.

First, we are talking about the number of molecules in one
microgram, and then the number if we are talking about a
thousandth of a microgram, and then I carried that calculation to
the number of molecules that there would be in a 70 kilogram person
and I had made an error in the exponent and I had a thousand times
more than it should have been, so it should have been 132 trillion
rather than 132 thousand trillion. This is not like Senator
Dirksen's famous saying, "A billion here and a billion there and
you are talking real money." We need to be a little more precise.

1 So the number is 132 trillion, not 132 thousand trillion.

2 Q Okay, and that's exhibit 1287?

3 A If you want, I will put per 70 kilogram person.

4 Q All right. Then I have marked as Defendant's Exhibit
5 1288 the drawing you did of the--

6 A Kidney tubule.

7 Q Kidney tubule. You referred to it as a nephron?

8 A A nephron, yes, that's the word for the whole unit.

9 Q And 1289 is the calculation you did for what, sir?

10 A For the number of natural errors in the formation of
11 the DNA in cells as cells grow, as tissue grows, and then a
12 statement of Dr. Bruce Ames' finding of about 320 DNA or finding
13 the residue that has arised from the repair of about 320 damaging
14 events per cell per day, and the number below is the number of
15 cells in the estimated in the human body, 60 trillion. Perhaps I
16 should identify that number so in the future--

17 Q All right, thank you.

18 MR. HEINEMAN: Your Honor, at this time we would
19 offer into evidence Defendant's Exhibits 1283 through 1289.

20 (Defendant's Exhibits No. 1283 to 1289 were
21 offered into evidence.)

22 MR. CARR: The Plaintiffs have no objection to these
23 exhibits, your Honor.

24 THE COURT: Fine, they are all admitted without

1 objection. Thank you.

2 (Defendant's Exhibits 1283 to 1289
3 were admitted into evidence.)

4 Q Dr. Dost, if you would resume your place there by
5 your chart, discussing the mechanisms of cancer, you did some
6 detail on the initiation, and to make sure I understand it, the
7 initiation can be an internal event?

8 A It is an internal event in the cell.

9 Q All right. But it can be caused perfectly naturally
10 without any outside intervention at all?

11 A It can be caused, well, we assume that all of the
12 events that take place in the cell that damage DNA also have the
13 capacity to cause a DNA mutagen. Now, how does that relate to the
14 event of initiation that you are describing?

15 A Well, first I should point out that a cell has
16 initiated does not behave any differently in the organism than a
17 cell that has not been initiated. So its function has not
18 changed, its function hasn't changed, and it appears that even
19 when it divides, while that initiating lesion has stayed in the
20 cell and then carried to the next cell, the daughter cells also
21 will not function any differently than the normal cell. So
22 we have got a cell here whose nature hasn't really been changed in
23 terms of what it does in the organism. Other events are necessary
24 to make that happen.

1 You asked about mutation. Mutations are changes
2 in the genetic material of a cell such that the cell in either
3 its next generation or possibly some subsequent generation will
4 be altered. Generally when we are speaking at the cellular level,
5 we are talking about a change that appears in the next generation.
6 If we are speaking about the whole animal, then we would be
7 discussing a change that is induced in germinal tissue in a
8 reproductive organ, and then again there is some complex genetics
9 involved here. It could very well be that offspring of the parent
10 with damage might be similarly affected or if it is a recessive
11 change of some kind, it may appear several generations down the
12 family tree, in an adult organism. Now, we use, it is thought
13 and again pretty universally thought, that mutation, the event
14 that leads to mutation are rather similar to the events that
15 lead to the initiation of cancer. And it is possible for us
16 to study those kinds of events in the laboratory. Microorganisms
17 are very handy for this sort of thing, cultured cells. So
18 there are a variety of laboratory, of fruit flies, there are a
19 variety of systems available in the laboratory for learning
20 whether a chemical has the capacity to interact with DNA, it
21 happens that there is a good correlation between the ability of
22 a chemical to interact with bacteria and other bench-top kinds
23 of tests and the ability of chemical to cause cancer. Generally
24 chemicals that do not cause mutation does not cause the initiating

1 kinds of events.

2 Q Well then, as I understand mutation or mutogenicity
3 is not the same thing that you are talking about here?

4 A The initial event is probably very similar, just
5 the initial event. The only difference is that it's likely
6 that in mutogenesis there is sufficient change that the next
7 generation of the cell is changed, you see, and imaginably we
8 could have cells in, let's say, the liver that had a minor
9 mutation, and they functioned a little differently, and we
10 may accumulate a few cells of that sort in the liver, but it
11 wouldn't matter because it isn't transmitted to the next
12 generation, and those cells don't necessarily make a tumor,
13 they are just a little bit different from the cells, the normal
14 cells of the liver, and that presumably happens frequently.

15 But the carcinogenic event may be sufficiently
16 specific that it only can be transmitted in this fashion to the
17 next generation of cells. The point about mutogenicity, though,
18 is that apparently the damage in the cell is about the same,
19 and there is an empirical correlation, by that I mean the
20 experimental evidence shows that chemicals that are mutogens
21 are frequently carcinogens. Okay, now, precisely what is
22 happening at the level of DNA in the two cases to link them
23 is not clearly known, but we know that DNA damage takes place
24 in both instances, and that there is a correlation. Chemicals

1 that cause one generally cause the other.

2 Q But is the opposite always true, or equally true?
3 For example, when you talk about whether or not TCDD is a mutogen,
4 as affecting the determination of whether it initiates cancer,
5 what do the two have to do with one another?

6 A Well, it is interesting, because the likelihood
7 that a chemical that is not mutogenic in modern test systems or
8 a chemical that is not mutogenic is not likely to be a carcinogen.
9 There is a better correlation from negative information than to
10 positive, because not all chemicals that are known to be
11 mutogens are in fact carcinogenic. Many are, and if we
12 find a chemical that is mutogenic, knowing nothing more about
13 it, we have to assume that that is a potential for carcinogenic
14 activity until it is proven otherwise, and this is one of the
15 bases by which chemicals are selected for a carcinogenicity
16 assay, because it is so expensive to do those kinds of tests
17 that we simply can't put every chemical that we have through
18 this very long, elaborate carcinogenicity test that costs several
19 million dollars. It is necessary to select chemicals which are
20 likely to be positive.

21 Q All right. Now, if you would, please, I am a little
22 confused about, a little confused, I am confused about this
23 fixation stage. What happens between the initiating event
24 and fixation?

1 A Fixation is just a term that is used to represent
2 that after, if damage occurs in the cell to the genetic
3 material in the cell, and is not repaired before the cell
4 reproduces. Our cells all are reproducing at some rate. We
5 have cell division going on in the body all of the time. Nerve
6 cells do not. But cells everywhere else are making new cells
7 at a slow rate. This is not going on at an enormous rate, but
8 it is going on all of the time. If the cell divides before
9 that lesion is repaired and some biologists believe that it
10 has to go through two sequential divisions, but we will just say
11 if the cell divides before that lesion is repaired. The lesion
12 then becomes locked into the genetic structure of the cell
13 and can't be repaired, and so it is there.

14 Q And the comment beside the term fixation uses the
15 term genome, fix the genome of the cell?

16 A What we have done is put information into the genome,
17 in other words the genome is just the total genetic pattern of
18 the animal and of the cell. And what has happened is that
19 information has been put into the genetic structure of the cell
20 that changes, potentially changes the way it behaves. It isn't
21 such severe damage that it alters the nature of the cell and
22 causes it to die, it is still functional, but it is sitting
23 there as a potential tumor cell. Now, it isn't a tumor cell,
24 probably won't be one, but there is always the probability some

1 slight probability that it can. So we have cells in the body
2 that are, that have been changed so that they are able to be
3 transformed or promoted and changed into active tumor cells.

4 Q And this again happens whether or not there has been
5 some influence from the outside. It can happen purely naturally?

6 A Yes.

7 Q And cells like this can be in the body and nothing
8 ever happen?

9 A Yes, that's correct. It appears that in mice there
10 is a very high population of induced or initiated cells in the
11 liver and mice make a very poor model for determining the
12 ability of chemicals to cause liver cancer because they have a
13 very, very high incidence of liver cancer anyway, just they are
14 very easy to throw into that biological state.

15 Q Because they have so many--

16 A They've got many initiated cells just by virtue of
17 being alive. Part of this is because the metabolic rate in
18 those very small rodents is very, very high and so there is a
19 high frequency of damage and there is a, they have a relatively
20 inefficient DNA repair system, much less efficient than human.
21 That's a parameter that can be measured.

22 Q Now, are all of these steps necessary before a
23 malignant tumor results?

24 A Yes.

1 Q All of them are necessary?

2 A All of them are necessary.

3 Q All right.

4 A And all of them, there is, all of them presumably
5 can be interrupted at some point until we get down here
6 where we have cells that have actually evaded all of the defense
7 systems.

8 Q And now, can there be a material that is a carcinogen,
9 for example, that initiates cancer that will lead to a tumor
10 without the need for a separate promoter?

11 A There are some chemicals that are considered to be
12 complete carcinogens. That is that they will not only cause
13 this kind of event, but if they are sustained in the organism
14 will also apparently carry through the promotion function and
15 will by themselves result in a certain tumor incidence.

16 Q Well, I want to get back to that in a minute. The
17 promotion aspect, what is it that happens in promotion?

18 A There is still a lot of questions to be answered
19 about promotion, but it appears that one of the characteristics
20 of promotion is that cells are stimulated to proliferate. Cells
21 become more active, they become--they in some cases cell division
22 takes place, the kinds of response that takes place in response
23 to injury. As a matter of fact, there are certain kinds of skin
24 tumors that can be promoted simply by cutting the skin in the

1 region of the application of the carcinogen, and by applying the
2 carcinogen and leaving it alone, nothing happens. If you wound
3 the skin, you get tumor formation.

4 Q Okay. So it is something to enhance?

5 A In some fashion, it enhances. Now, in the simplest
6 sense, maybe it is simply something that increases the rate of
7 multiplication of the cells and sort of magnifies this error that's
8 already shown up. The biochemical mechanism of promotion is
9 not clearly worked out yet.

10 Q Okay. Now then, what is transformation?

11 A Well, transformation is a process by which these
12 cells change their character. Up until this point, you probably
13 wouldn't be able to identify the cells as being all that much
14 different from the cells from which they arose. But in the
15 process of transformation they change their shape. They change
16 the way they carry out their business, they do things differently
17 and they become less, they become more primitive compared to
18 their very highly specialized form of the cells from which they
19 arose.

20 Q Now, is there a dividing going on all during this
21 period of time?

22 A Well, yes, and probably the process of cell division
23 becomes accelerated at this point to some extent.

24 Q And the new cells that begin at that point, do they

1 have to go through the process or do they begin that transformation?

2 A Well, the new cells, you mean cells that start up
3 here?

4 Q No.

5 A At here, at this point?

6 Q If they are dividing at the point of transformation.

7 A They are dividing, and the cells that arise from
8 that division are also cells that are more primitive. In other
9 words, if we have a population of cells here some place that
10 don't really resemble the original cells, and they are
11 reproducing on their own and they are making new cells just like
12 themselves.

13 Q All right. And then the tumor arises as a result
14 of that?

15 A Eventually there are enough cells that we would
16 call it a tumor.

17 Q Now, eventually?

18 A Eventually.

19 Q How long does that take?

20 A Often a long time. We talk about the latency period
21 for tumors. Part of that is just simply the time it takes for
22 a tumor to reach a size that it can be seen. But it also means
23 the time that these events take, which are not instantaneous.
24 But if we just consider the population of cells, tumors have a--

generally tumors have a doubling time, so called. That is the number of cells double in a period of--in most cases say from 30 to 90 days. Okay, in other words, if there are two cells at 30 days, there will be four at 60 and eight at 90, et cetera, et cetera. If it was a 90 day doubling time, we would have two cells, let's say the first division took place at the first of the year. There would be four cells in March and eight cells in June and 16 cells in September and by the end of the year there would be 32 cells. Now, that's not very fast. Thirty-two cells are not very many when you consider, you know, I mentioned that something may be a cell mass this big weighs probably less than that, weighs about a gram, and there are a billion cells in that. Sometimes tumor cells are larger, so there would be fewer cells. But you can see that it would take a long time to build up a mass at a rate, at a doubling rate of say 60 days. It would probably take five years before you could find that tumor if you were looking for it, you see, and to have a tumor suddenly be noticed with a doubling time like that, it has probably been there for quite a long while before somebody detected it.

Q Now, I'd like for you to talk about the difference between an initiator and a promoter, and is there a chart that would help you explain that that we brought with us here today?

A We showed one, did we not, the other day? There was

1 a chart, maybe not. We have a chart that described the difference
2 between initiation and promotion.

3 (Defendant's Exhibit 1290 was marked
4 for identification.)

5 Q All right, sir, let me show you what's been marked
6 as Defendant's Exhibit 1290 and ask you to examine that and
7 identify it for me, please.

8 A That's a chart that has been taken out of a paper
9 by Tom Slaga that was published in Environmental Health
10 Perspectives, Volume 12, in 1985. It is a really standard
11 representation of the difference between initiation and
12 promotion.

13 Q And Dr. Slaga is with what organization?

14 A He was at one time with the University of Washington,
15 and I believe at the present time he is at the National
16 Institute of Environmental Health Sciences, although I am not
17 absolutely certain. I haven't visited with him for a while.

18 Q Now, would this chart help you explain this
19 process?

20 A Yes, it would. The black square, and the black
21 squares of various sizes represent an initiator, a chemical
22 that can cause this, an early lesion in the cell, the early
23 lesion in the DNA. These arrows represent doses of a promoter.
24 Now, in the first situation we have a very high dose of the

1 initiator, and that very high dose of the initiator, even though
2 nothing has happened, nothing has been administered after that,
3 does cause some tumors. A much, much smaller dose, however,
4 causes no tumors if we have a small dose, and then follow it
5 with several smaller doses of the initiator, we may very well
6 get tumors. Most initiators have some capacity to carry this
7 process on if the initiator is continued in the system. Now,
8 if we take a still smaller dose of an initiator, we get no effect.
9 Now, we have a promoter, and we give that promoter that substance
10 that, and many, many chemicals will do this, that cause this
11 series of events in here between the time the lesion is locked
12 into the cell and the time that the cell goes out of control, we
13 give that by itself, nothing happens. They won't cause tumors
14 by themselves. If we administer a small dose of the initiator
15 and follow it with a promoter, and these are representing several
16 doses over a period of time, then we get a substantial number
17 of tumors, compared, for example, with this same dose of
18 initiator, and no tumors at all without a promoter. Now, the
19 effect wears off a little bit with time. If we do the same
20 experiment and wait for a period of time, perhaps a year or so,
21 to give the promoter, we still will get tumors. The reason for
22 that is because those initiator cells stay there. There has
23 been nothing to remove them, they are there, they are functioning
24 as part of the liver or whatever organ and they are still in that

1 state of, I suppose you would say genetic readiness to become
2 a tumor cell. If we should give the promoter first and then
3 the initiator, nothing happens, nothing happens. If we give
4 the initiator and we give less frequent doses of the prohibiter,
5 you may get a response. If you give still less frequent doses
6 of the promoter, no response. Now, what this shows is just
7 simply, it is a generalization that describes the relationship
8 between substances that initiate the tumor process and those that
9 promote it and they are separate functions.

10 Q Now, Dr. Dost, do you recall what Dr. Zahalsky
11 testified on the 3rd of May, 1984, that exposure to TCDD as a
12 promoter prior to initiation would cause cancer?

13 A Yes.

14 Q All right. Do you agree with that statement?

15 A No.

16 Q All right, why not, sir?

17 A Well, he has stated it upside down. If this is
18 the situation that he is talking about, the promoter by itself
19 or prior to an initiator doesn't do anything. The definition
20 of the terms makes it very clear which is the first step in the
21 process and which is a subsequent step. Promotion, initiation
22 is an action of some kind that starts a process. Promotion
23 is some kind of activity that causes a process to go forward.

24 Q And so if you put the promoter on first before the

1 initiator?

2 A Nothing happens.

3 Q And is that demonstrated by the data upon which that
4 table is based?

5 A That's demonstrated by a great deal of data. This table
6 does not represent specific data from a given experiment, it
7 is a representation that has been generated from a great deal
8 of experimentation in this general area. This is intended as
9 a generalization, but this is the situation that represents
10 the application or administration of a promoter prior to an
11 initiator, and there isn't going to be a carcinogenic result.

12 Q Now, what does that chart demonstrate with respect to
13 a dose response relationship for initiation?

14 A Well, there is a dose response relationship for
15 initiation. We think of it, though, not in terms of the
16 intensity of the response as we would in other kinds of events,
17 but rather the frequency with which the response will take place
18 because cancer is in effect an all or none situation in which
19 if the cell undertakes this process that we've described, a
20 tumor will at least potentially eventually form. So a tumor
21 can eventually form from a single cell, at least in theory,
22 and probably in fact, because it has to start someplace. There
23 is nothing, we don't deal with the issue of whether it is a
24 big tumor or a little tumor, it is either a tumor or it is not.

1 The cancer has either been caused or it has not. So we talk
2 about the incidence or the frequency with which cancer occurs
3 in a population so that we relate that to the number of animals
4 that have been exposed and the number of animals that have gotten,
5 that show tumors. And yes, there is a dose response. As the
6 dose increases, more animals in a population, if it is a carcinogen,
7 more animals in the population will show tumors. Now, it is
8 possible with certain kinds of tumors that occur in large
9 numbers in an animal to also deal with this in terms of number
10 of tumors per animal, et cetera, but that's really another form
11 of frequency. So we are talking about the frequency with which
12 this event occurs and that is dose related.

13 Q The first two items on the chart, you show the
14 big dose and the small dose?

15 A And I think we can say, yes, this in itself is a crude
16 dose response relationship in that the big dose caused tumors
17 and the little dose didn't. However, we wouldn't know the shape
18 of the dose response curve, all we would know is where the upper
19 end is.

20 Q Now, what do the studies show with respect to a
21 no observed effect level for promotion?

22 A For promotion, there appears to be a no effect level.

23 Q All right, and are there any particular studies
24 upon which that conclusion is based?

1 A There is a substantial amount of research on the
2 question of promotion. The data show very clearly the dose
3 response clearly is shown and there are no effect levels shown.
4 Now, the difficulty is that there has to be some assurance that
5 there were not some initiated cells there in the first place,
6 and so most people who discuss this issue believe that there is
7 a threshold, but they are not prepared to prove it, okay, and
8 I think that's the best way to describe it. It is very clear,
9 however, that the dose response is there and in experiments in
10 which the doses that show no effect are a long ways away from
11 doses which show an effect. It is fair to say that there is
12 in fact a threshold.

13 Q Well, what is the consensus with respect to TCDD?
14 Is it a promoter or initiator, or what?

15 A The consensus with respect to TCDD is that it is a
16 promoter.

17 Q And whose work demonstrates that?

18 A Allen Poland has worked in this area and has shown
19 with various strains of mice that TCDD is a promoter. Henry
20 Pitot at the University of Wisconsin, in fact, they are both
21 at the University of Wisconsin, has also worked very extensively
22 in this area. In fact, that's his primary area of research.
23 Slaga has worked in this area and has also shown that TCDD is
24 a promoter. The other evidence arises from the nature of the

1 carcinogenicity studies on TCDD in that the nature of the
2 tumor pattern, the behavior of the systems indicate quite
3 strongly that TCDD is in fact a promoter and is probably not
4 an initiator.

5 Q Now, what is the evidence that demonstrates that?
6 Is TCDD a mutogen?

7 A TCDD is not a mutogen. There were some studies
8 in the early 70s in which TCDD had some mutogenic effect. No
9 one has been able to reproduce that information, including in
10 one case one of the people who had done the work, J.P. Seiler
11 had made these kinds of findings in the early 70s and himself,
12 he himself has not been able to find that kind of evidence again,
13 and there is a good reason for this because our systems for
14 detecting, for managing the microorganisms, our system for
15 detecting mutogenic events is much more precise than it was
16 even ten years ago.

17 Q You referred to microorganisms, were these early
18 tests in bacteria or something?

19 A Yes, these are the so-called Ames tests in which
20 an organism, usually species of salmonella, these are often
21 found in the digestive tract, they are common organisms, but a
22 mutant has been derived that cannot live in the presence of or
23 without one of the essential amino acids. Okay, it is a mutant
24 and it is susceptible to being mutated back to an organism that

1 can live without the particular amino acid, in this case
2 histidine. Now, there are a number of strains of organisms,
3 well, the trick is very simple. If you have organisms that
4 have to have histidine to live and you cause a mutation, you
5 treat them and you transfer them to a medium that has no histidine.
6 Organisms that have been mutated back and can use histidine or
7 can make their own histidine will survive on the new media
8 and the ones on the old media, the ones that didn't mutate
9 can't survive. The concept is very, very simple and there are
10 a large number of microorganisms that have been modified in
11 this way so that they are very sensitive to chemical mutagen.
12 It is a very fortunate circumstance, because it means that we
13 can at least make preliminary screens of the ability of a
14 chemical to interact with genetic material without going to the
15 enormous expense of doing a whole cancer study. We can do a
16 bacterial mutagenesis study in a matter of days.

17 Q All right. So that what has been found in these
18 studies is that TCDD is not a mutagen?

19 A TCDD does not cause DNA damage. There are some
20 systems where some cell preparations where it can cause some
21 chromosome rearrangement. This is not a mutation.

22 Q Well, does TCDD interact with DNA?

23 A No, not at all.

24 Q And has that been demonstrated in studies as well?

1 A That has been demonstrated by again, Dr. Poland
2 and Dr. Gunther. Poland has used, has done experiments in
3 which he has used labeled TCDD and has found no evidence of
4 binding to DNA, much evidence of binding to protein and other
5 structures, but none to DNA.

6 Q Now, Dr. Silbergeld in her testimony here
7 stated that TCDD--

8 MR. CARR: What page, counsel, and day?

9 MR. HEINEMAN: April the 12th, Pages 120 and 149.

10 Q States that TCDD causes cancer itself. It is the
11 most powerful chemical carcinogen identified, and is a complete
12 carcinogen. Do you recall her testimony in that regard?

13 A Yes, I do.

14 Q Do you agree with that, sir?

15 A No, I don't agree that it is a complete carcinogen.

16 Q And on what basis do you disagree?

17 A I've already discussed really the evidence that
18 shows that TCDD is in fact a promoter and it does not
19 interact with DNA either by direct chemistry or by any ability
20 to cause a significant mutation incidence, and also by studies
21 that have been done that show that in organisms that are treated
22 with TCDD there is no increase in the DNA repair activity,
23 which is a measurable, a measurable factor in cells.

24 Q Now, at the same time in her testimony she suggested

1 that the Kociba studies show quite clearly that exposure to
2 TCDD alone causes cancer, and that's why it is a complete
3 carcinogen. Do you agree with that statement?

4 A No, I don't. What the Kociba studies show in
5 that light is that at high doses, at very high doses up at a
6 point where the animal is suffering really substantial injury,
7 there is a lot of effects on the liver, particularly in those
8 animals, that the incidence of a variety of tumors changes, a
9 variety of tumors, and when I say change, I mean just that.
10 There were increases in the number of hepatocellular carcinomas
11 and also nodules in the liver that probably are reversible by
12 the definitions of most pathologists. There were many kinds
13 of tumors. Now, you must understand that in any population of
14 animals there is going to be a high population of tumors, a
15 high incidence of tumors of any kind. We all know unfortunately
16 in our population that most cancers appear in older people.
17 The same is true in laboratory animals. It is interesting that
18 in those experiments the incidence of many kinds of tumors,
19 particularly those of the endocrine system of the glands,
20 actually it decreased statistically. In other words, it wasn't
21 so much an increase in total numbers of tumors, but an
22 alteration in the whole tumor pattern. In another study that
23 was done in the National Toxicology Program, there was an
24 increase in some tumors in the thyroid at the highest dose.

1 When the National Toxicology Program study was done, instead of
2 feeding TCDD in the diet, they introduced it by stomach tube
3 once a week. Long term experiment, they gave the rats their
4 treatment once a week on an exact basis with a stomach tube.
5 Those rats did not get the somewhat--the slight increase in
6 tumors of the palate and the nasal passages and the upper
7 respiratory tract that Kociba's rats did. The reason for that
8 is because the material was not in contact with those areas.
9 Rats when they eat tend to inhale quite a bit of food, particularly
10 when it is in that powder form as is used in these kinds of
11 experiments. The fact that these tumors occurred at the site of
12 contact is suggestive of promotion, particularly when we compare
13 with the studies that were done on the skin, TCDD has also been
14 tested as a skin carcinogen, again in the National Toxicology
15 Program, and the only tumors that they found were at high doses
16 again, and they were a tumor called a fibrosarcoma of the skin.
17 It is a connective tissue tumor, and there were no other tumors
18 anywhere in the animals. That, too, is suggestive of a promoting
19 response rather than initiation, and promoting at the site of
20 application.

21 MR. HEINEMAN: At this point, we would move the admission
22 of Defendant's Exhibit 1290.

23 (Defendant's Exhibit 1290 was offered into
24 evidence.)

1 MR. CARR: No objection, your Honor.

2 THE COURT: Okay, 1290 is admitted without objection.

3 Thank you.

4 (Defendant's Exhibit 1290 was admitted
5 into evidence.)

6 Q Now, Dr. Dost, I'd like to talk to you again about some
7 of Dr. Zahalsky's testimony, and in that connection, I'd like to
8 show you what's been marked as Plaintiff's Exhibit 253. Have
9 you seen that before, sir?

10 A I've seen pictures of it.

11 Q All right. Or a picture of it?

12 A Yes.

13 (Plaintiff's Exhibit 1291 was marked
14 for identification.)

15 Q I'll now show you what's been marked as Defendant's
16 Exhibit No. 1291 and ask if that's a blow-up of a photograph of
17 Plaintiff's Exhibit 253.

18 A Yes, it appears to represent it.

19 Q Now, Dr. Dost, you have read, have you not, sir, the
20 testimony of Dr. Zahalsky in which this chart was discussed?

21 A Yes, I have.

22 Q All right. Do you agree with that testimony and this
23 chart?

24 A No, I don't, really.

1 Q All right. Would you please come up and explain to the
2 Jury why you disagree with the chart and the testimony relating
3 to it.

4 A This as I recall appeared in Dr. Zahalsky's testimony at
5 a time when he was discussing effects on the immune system, and
6 if I remember correctly began talking about this representation,
7 and I guess that I would ignore that and take for granted his
8 representation that this scheme represents the way that dioxin
9 causes cancer, and it is clearly incorrect. There is, the
10 schematic that we're dealing with here--

11 MR. CARR: Mr. Heineman, what would be the date the
12 testimony was taken and the page number that he is referring to?

13 MR. HEINEMAN: It starts at 101, I think, and following
14 that, I think is where he starts talking about it.

15 MR. CARR: What date?

16 MR. HEINEMAN: May the 3rd.

17 A Well, the scheme that's represented here is clearly a
18 scheme that is apparently related through the induction of
19 microsomal enzymes, the P450 enzymes. And what we have here,
20 this looks very much like a diagram that Dr. Eisen published at
21 one time or a part of that diagram, but I'm not prepared to say
22 that it was adapted from it directly. When TCDD or any of a
23 number of other polycyclic aromatic hydrocarbons, and we've discussed
24 this earlier, I believe, when it moves into the cell in order to

1 cause the induction process, it has to have a way of starting
2 the process, and just by itself in the cell it can't do that.
3 It attaches to a receptor. I don't like the term recognizer,
4 because that implies that this is so specific that it identifies
5 each molecule that comes into the cell. The fact is that this
6 receptor will bind a very large variety of substances. It happens
7 that it binds TCDD more intensely than it does any other, and I
8 think that's just an accident of the very flat structure of TCDD.
9 It turns out to have provided us with an excellent tool, however,
10 for studying this process of induction, and it has led to an
11 understanding of the way that all of these complex chemicals cause
12 induction. The two join and they form a complex between TCDD and
13 the receptor, and that receptor moves into the nucleus of the cell.
14 Now, this receptor is not out on the surface of the cell, it is
15 free within the cytosol or the body of the cell. That's why they
16 call it a cytosolic receptor. It is in the cytosol of the cell.
17 And it is correct that it does move from the cell body into the
18 nucleus of the cell. But it starts from in here. The TCDD comes
19 into the cell and joins with the receptor to form this complex,
20 and the complex then moves into the nucleus. There is a whole
21 series of these, and I suppose that we should bring them all
22 up one by one.

23 Q Let me hand you, sir, what's previously been marked as
24 Plaintiff's Exhibit 254, 255, 256, and 257, and ask you if you have

1 seen those documents before.

2 A These represent the photographs that I have seen and
3 apparently they are in the sequence that they originally appeared.

4 (Defendant's Exhibits 1292 to 1295 were marked
5 for identification.)

6 Q Now, sir, would you compare these photographs by number
7 with the Plaintiff's exhibit to be sure I've got them right?
8 Defendant's Exhibit 1292 is a photograph of what, sir?

9 A Of Plaintiff's Exhibit 254.

10 Q All right.

11 A And this is the next step.

12 Q And Defendant's Exhibit 1293 is a copy of what, sir?

13 A Let's see, is a copy of Plaintiff's 255.

14 Q All right, sir. And Plaintiff's Exhibit 12--I'm sorry,
15 Defendant's Exhibit 1294 is a copy of--

16 A Plaintiff's 256.

17 Q And Defendant's Exhibit 1295?

18 A Plaintiff's 257.

19 Q Now, would you like to go through these one at a time?

20 A Maybe in order to maintain sequence I will put this over
21 on this easel. This complex does in fact go into the nucleus
22 and it interacts with the genetic material in the nucleus. I
23 don't know what really is meant by a critical target here, because
24 what is interacted with is a segment of DNA, and this complex, I

1 presume when he says nuclear complex does not act as a message.
2 The message, what it does it cause in some fashion, causes the
3 DNA in a particular part of the cell, in a particular part of the
4 genome to go through the process of opening up and making a
5 copy of messenger RNA. Now, it is a fact, yes, that it does
6 result in the making of messenger. We are talking here about a
7 form of nucleic acid called RNA, and it is very similar to the
8 DNA. Then the messengers, the messenger RNA goes to the structures
9 in the cell that manufacture protein, the endoplasmic reticulum,
10 and they provide instructions to make certain protein, and in a
11 sense, I suppose we could say this is correct, except that as he
12 has it depicted here and as he states it here, they are making
13 new receptors. If I understand that, he means recognizer as
14 representing the receptor or TCDD. That does not happen. What
15 is made are the group of enzymes that we have discussed at various
16 times, the P450 enzymes, the other non-P450 enzymes that we saw
17 illustrated on that chart of Dr. Eisen's, I think yesterday, but
18 we do not, the cell does not make additional receptors, they do
19 not go to the outside lining of the cell or the cell wall. I am
20 not sure what is meant, and they don't react with dioxin except
21 that those are the very enzymes that are responsible for changing
22 dioxin, metabolizing it so that it can be disposed of. They are
23 not a new set of receptors that are here in the cell body which
24 interact with TCDD. There is a clear limit to the number of those

1 receptors. There are a few compounds that are known to be able
2 to cause the formation of additional receptors to a limited
3 extent, TCDD is apparently not one of them. And these proteins
4 that we are talking about that are formed do not go to the
5 outside of the cell. They go--they are attached to this
6 endoplasmic reticulum, where they are synthesized. They stay
7 there and are held in place. These are what are known as
8 membrane bound enzymes as differentiated from some of the
9 enzymes that I mentioned on another occasion that are soluble
10 that can actually leak out of the cell. These P450 enzymes
11 can't leak out of the cell because they are stuck on larger
12 structures that can't leave. Now, apparently Dr. Zahalsky
13 believes that these TCDD, these new TCDD receptor complexes go
14 back through the same cycle again, according to this diagram,
15 but then they do something different. According to his
16 description, they react and form reactive intermediates. There
17 are some real problems with that because to begin with the metabolism
18 of chemicals that might or might not take place and might proceed
19 to "reactive intermediates" takes place out in the cell body on
20 that endoplasmic reticulum. There are no such reactions taking
21 place in the nucleus. The nucleus has other kinds of work to do.
22 So somehow or other we have two cycles described here that start
23 out exactly alike and then are presumed to end up differently with
24 a different function in the second cycle. So this part of the

1 description is really completely incorrect. We don't have
2 formation of new receptors. There is a great deal of binding
3 of TCDD to the proteins in the liver cell, particularly the
4 endoplasmic reticulum, but it is nonspecific. It is where most
5 of the TCDD that is stored in the liver ends up and it is
6 just stuck there, it is not really doing anything in particular,
7 and it can be taken back out in a biochemistry lab so we can
8 tell where it is. There is just a very small amount of receptor.
9 Measurements that have been made indicate that no more than one
10 percent, and probably much less than that, of the TCDD in the
11 liver cell actually has bound to a receptor. The rest of it is
12 all bound nonspecifically to other proteins. So we don't have
13 reactive intermediates formed in the nucleus, and this complex
14 between the receptor and dioxin has nothing to do with the
15 metabolism of dioxin except as that receptor causes the formation
16 of the enzymes that eventually can break dioxin or it can alter
17 dioxin so that it can be disposed of.

18 The last chart says that now the reactive intermediates
19 bind to critical targets. We know that TCDD and its metabolites
20 do not bind to DNA. The implication here is that it has bound to
21 DNA inside the nucleus of the cell. This has been demonstrated
22 not to occur. It has also been demonstrated that we do not have
23 a problem of reactive intermediates that are thrashing around in
24 the cell, which is 3-methylcholanthrene or dimethyl bez anthracene

1 the reactive intermediates of those substances do a great deal
2 of harm and they do bind anywhere they can find a point in their
3 path. The implication is this is what results in toxicity, and
4 we've discussed this I think sufficiently to show that there isn't
5 a relationship between reactive intermediates and their binding
6 to toxicity because we don't have reactive intermediates as
7 part of the system. The implication is also that this initiates
8 the process by which cancer cells, cancer is caused, starting
9 with the initiation in the nucleus of the cell, and it also
10 results in transformation. And I am not sure what the relationship
11 of these two events is. We would have to concede that TCDD is
12 capable of the promotion process which, depending on definition,
13 may or may not include transformation. But it has nothing to
14 do with the initiation process. So the representation that this
15 scheme is a description of the way that TCDD causes cancer is, I
16 think, erroneous, entirely erroneous. I don't know where it arose.

17 THE COURT: Gentlemen, could I see you at the bench for
18 a minute, please?

19 (At this time an off-the-record conference was
20 held at the bench.)

21 THE COURT: Ladies and gentlemen, we are going to break
22 for the day at this time. We will resume again tomorrow morning
23 at 9:30. I would remind you as I do for any overnight break that
24 you are not to read, listen to, or watch anything about this case

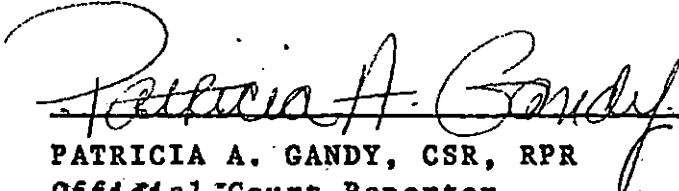
1 in particular or subject matter in general in any of the media,
2 print or electronic. Thank you for your attention and cooperation.
3 Court is adjourned. We will start at 9:30 tomorrow morning.

4 (At this time Court was adjourned for the day.)
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1 STATE OF ILLINOIS)
2) SS.
3 COUNTY OF ST. CLAIR)
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8 I, Patricia A. Gandy, CSR, RPR, Official Court Reporter
9 in and for the Twentieth Judicial Circuit, and the Official Court
10 Reporter who transcribed the above-styled cause had on November 5,
11 1985, do hereby certify that the foregoing transcript of proceedings
12 is a true, correct and complete transcript of the proceedings had
13 on said date.

14 DATED this 10th day of November, 1985.
15
16

17 
18 PATRICIA A. GANDY, CSR, RPR
19 Official Court Reporter
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1 STATE OF ILLINOIS)
2) SS.
3 COUNTY OF ST. CLAIR)
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8 I, RICHARD P. GOLDENHERSH, Circuit Judge in and for the
9 Twentieth Judicial Circuit, hereby certify that the above is a
10 true and correct transcript of the proceedings had in the case
11 captioned: FRANCES E. KEMNER, et al., v. MONSANTO COMPANY,
12 Cause No. 80-L-970, heard on November 5, 1985.

13 DATED this _____ day of November, 1985.

14
15 ENTER:
16
17

18 _____
19 RICHARD P. GOLDENHERSH, Circuit Judge
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24