On the Farm Diagnostic Procedures for Specific Toxicants

Gavin Meerdink, *D.V.M.*
Department of Veterinary Science
University of Arizona
Tucson, AZ 85721

I think I vastly underestimated the number that are here. I have a handout and if there are any of you who would like it you can leave your name and address on a piece of paper or something and give it to me and I'll make sure Gloria sends it out. I don't think its anything you can't find anywhere else, but at any rate, the topic that was given to me was on-the-farm-type diagnostic procedures, but the more I thought about this, the first thing that would come to mind is the difilamine test for nitrates and we have some sulfuric acid difilamine type reagents and we put it into some forages and what not and we get a blue color. I remember giving a talk on this eight years or so ago and within the next two weeks in the diagnostic lab we must have had fifteen calls relating to what did the color mean, did that mean there was too much? These cattle weren't necessarily showing anything. I think that these tests are perhaps more confusing than they are helpful. I don't think there are a lot of real shortcuts to toxicology diagnosis. I think we still have to come back to clinical signs. That is still our most accurate indicator of what is going on. Another way of looking at this I think is that we have a public that's becoming more sophisticated. They want numbers. Our clientele are more specialized. We're more specialized. I don't think I have to tell anybody our liability insurance has doubled this year. I guess I was reminded of a court case I was in a few months ago in another state and the case was precipitated by the fact that the veterinarian thought the feed quality was not very good. I think he was absolutely right, but that's all he had. I don't think that feed quality is very good. I think it is poor quality feed. By the time the opposing attorney got done with that he had an extra oriphice I think! He needed some numbers.

This subject I think was brought up at the AAVLD last month in Fort Worth. And I asked them, Is there anything on the scene? No, not really. It's a marketing type of thing and we don't have a lot of call for many of these tests. And I said, Well, if you had a chance to talk to a practitioner and explain the problems in diagnostic toxicology, what would be the subject? And it was pretty unanimous. Everyone said sampling. So at any rate I would like to twist my title just a little bit and talk about more sampling problems or some ideas and bring up a few points. I hate to be so melodramatic about this. It's a slide I had for a talk before...a study that we made on sampling problems. I thought at least to give you a few examples of some very common type sampling problems.

This was some corn that was being fed to some animals we were quite sure had a mycotoxin problem. The owner or the veterinarian sent samples of the stuff in three different times and they never could find it. So we went out with an ear corn probe, did a sort of schematic drawing of this thing, started it off just a little bit, took 15 samples and did find xylinone in pretty good significant levels in 3 out of the 15 samples. Of course we have a situation here where the feed that was causing the problem had already gone to the animal. So that particular sample was gone. Mycotoxin sampling is something that gets us into trouble all the time because, in fact we have three insurance cases on that right now that have boiled up within just the last few months. It has to do with sampling. It's not always the fault of sampling. Mycotoxins are very focal in occurrence. They are hard to find or at least hard to explain.

This is something that happens to us about every day. This is some formalized tissue and what I did was take the liver out. I made a thin section out of a piece of it, through it in some other form, and then I took that same liver, put it in another jar of formalin, and kept it for three days. Now this has been fixed at least three days in a chunk. You can see we've got penetration of formalin about three millimeters or so. And the inside of that tissue is essentially rotten. There is just nothing to be gained from doing histopath on that. Unfortunately, the normal post mortem decomposition of liver tissue very rapidly covers up some of the very things that you want to see. So this isn't perhaps good, but I guess ideal is 1-10. But at least these tissues will fix a little bit better and if you look closely for the most part we have some kind of thin sorts of sections here.

Here is another example of sampling problems in a diagnostic laboratory type setting. Convalescent serology. We get the first sample, not the second, the two-week sample. What does it mean? What does the first one mean? Usually we don't know what the first one means. It has to be in comparison with the second one and this is something I know you all are aware. So, just to look at samples for a subject I guess we have three things to think about. What, and how much, and how do we preserve it? Most chemists would say give us all you have and that might be enough. That's kind of a facetious remark but often true, particularly when you are on a bit of a scouting expedition.

The sheet that I handed out has more or less a summary of things and does have some levels listed on it. But very, very

Specimens to Submit for DIAGNOSTIC TOXICOLOGY

(For specific information contact your referral diagnostic laboratory.)

(All submissions should include thin tissue slices in formalin for toxicology; bloods and fluids should be chilled and fresh tissues frozen.)

	stomach or rumen contents-frozen	liver - frozen - 100 gm	kidney - frozen - 100 gm	kidney - formalin fixed	brain - ½ frozen	brain - ½ formalin fixed	heparinized blood (10 ml)	serum (no hemolysis) (2 ml)	urine - 20 ml	bone	feed - 1 quart	body fat - 100 gm	suspect material or bait	milk - 200 ml	ocular fluid (2 ml)	forage	cerebral spinal fluid	kidney - ½ in alcohol	water - 100 ml
Ammonia	*						*	*											
Anticoagulants (Warfarin, etc.)	*	*					*						*						
ANTU	*	*																	
Arsenic	*	*	*						*		*								
Blister beetle (Cantharidin)	*	*	*					*								*			
Calcium								*		*	*								
Carbon Monoxide							*												
Carbamates (see Organophosphates)																			
Chlorinated Hydrocarbon Insecticides	*	*	*		*						*	*	*	*					
Chromates	*	*	*				*						*						
Copper		*	*				*				*		*						
Cyanide		*					*									*			
Ethylene Glycol				*				*	*										
Fluoroacetate (1080)	*	*	*										*						
Fluorides									*	*						*			
Fuel Oils	*																		
Herbicides (Synthetic Organic)	*												*						
Lead	*	*	*				*			*			*						
Magnesium								*	*		*								
Mercury			*	*	*	*			*										
Metaldehyde	*	*											*						
Mycotoxins	*			*							*								
Nitrates/Nitrites	*						*								*	*			*
Organophosphates	*				*		*						*						
Oxalates				*												*			
Phenols	*	*											*						
Phenothiazines		*							*		*		*						
Phosphates								*		*			*						
Phorphorus (zinc phosphide)	*												*						
Pindone (Pival)		*											*						
Potassium								*											
Salt/Sodium (water deprivation)						*		*			*						*		
Strychnine	*	*							*				*						
Sulfa Drugs							*											*	
Sulfates											*								*
Thallium		*	*						*										
Urea (see Ammonia)											*		*						
Zinc		*	*								*		*						

briefly here, if you want some bacteriology or analytical work done, freeze it, freeze a big chunk of it. Usually world-pak bags are fine. Anything for histopath in thin slices. Formalin does not penetrate those tissues very well, and in fact, in human labs when they want some very, very finite histopathology of a brain, for example, they put the formalin in the refrigerator. The brain does not break down quite as fast and the formalin has a chance to penetrate before decomposure.

Now what I have done, this is more or less a copy of that sheet I handed out. I thought I would just point out about 2 or 3 things here and then pick up some new samples that we are beginning to use. One is lead, the blood test for lead. You see heparinized blood there. I think we can safely say now for almost any lab in the country, certainly in our lab, it doesn't make much difference what kind of anticoagulant you use. EDTA is fine. In fact, if all you can get is a clotted blood sample that's fine too, run it on the clot. The error that we have sometimes is people send serum. Lead likes to adhere to the red cell membranes. So we must have cells. But in anything that has red cells we can find a lead level. Magnesium? The problem is in the live animal it's fine. Serum. You need to separate the serum pretty rapidly because magnesium is an intracellular ion and with hemolysis you'll get some contamination. But what about the dead animal? Urine is fine if she hasn't been dead very long. Again, the cells in the bladder, as they break down, as they decompose, will leach magnesium into the urine. But there should be 60 mg or so in urine. If the serum level of magnesium is much below 1.7 which is a rough type of a threshold, that's below the renal threshold and there won't be any in the urine, or very little. So urine in a carcass for magnesium, when you suspect grass tetany, is not a bad sample, often times pretty good. Nitrates? The only thing I wanted to mention here is occular fluid. This is something that has been done by some labs now for almost a year and a half. I think done by most labs in the United States. It's an easy sample to get, with the needle injected just anterior to the limbus into the anterior chamber of the eye and pull all the fluid that you can get. I'm talking about a carcass here. It's hard on the eye of a live animal. And of course anything else. But that's a clean sample. It works quite well and fairly easy to hold. If you don't do that I think it's fine also to send in the entire eye. Chill it a little bit so it won't break down quite as fast. Organophosphates, carbamates or any other thing that inhibits cholinesterase, we've talked about brain for a long time, the caudate nucleus. That's fine, but we have some reactivation problems with it. If we had a long time we'd talk a little more about that. Blood is fine but I think a sample that will be on this list some day is retina, or eyes, something like that. But we continue to keep looking for a better indicator in the body for cholinesterase activity.

What are we going to see in the future? What type of samples can we use to find the things that we are looking for? Ideally we'd like to be able to pull a blood sample and do the whole thing. We are not there yet and not even close to it. We are finding that bile is a good indicator in a carcass of a lot of element levels. Certainly a lot of drug metabolites that are excreted in the bile. And again the eye, the body in the anterior vitreous chamber. Vitreous bodies are used in humans to determine how long the person has been dead, because the potassium leaches out of the tissue in the eyeball at a very constant rate. We're starting to look at that with race horses now to settle some insurance claims. But this post mortem chemistry is becoming more and more fascinating. It all depends on the integrity of the sample.

So, if I had one item to leave with you, it is, when in doubt pick up the phone and call the lab. I can't tell you when things are going to change or a new test is going to come about, but that happens quite often whenever someone gets a wild idea and starts looking at some things and all of a sudden we have a new method.

XIV World Congress on Diseases of Cattle

Dublin, Ireland
August 26-30, 1986

The Irish Cattle Veterinary Association is pleased to invite everyone to the XIV World Congress on Diseases of Cattle which it is sponsoring in collaboration with the World Association for Buiatrics in Dublin, Ireland on August 26-30, 1986.

Situated between Europe and North America, Ireland is easily accessible by international air transport from all continents. Dublin, "fair city where the girls are so pretty", while possessing all the attractions of a modern European capitol, still retains the charm and infimacy which make Irish hospitality a unique experience.

The provisional scientific programme will cover the following areas: herd health programs in intensive units, reproductive disorders, embryo transfer, ultrasonics, congenital defects and genetics, mastitis, respiratory diseases, nutritional metabolic disorders, enteric diseases, digital diseases, parasitism and miscellaneous topics.

Further details from Dr. H. J. Greene, Secretary, XIV World Congress on Diseases of Cattle, 44 Northumberland Rd., Dublin, 4, Ireland. (Telephone 688244.)