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Edward McCowan 41

To: USDA/FSIS
Fm: Edward McGowan, PhD, BSc Medicine
Re Comments on Following Dockets: 03 025IF [excluded parts]; 01 033IF [captive bolt]; 03 038IF [ARM]
Dt 1-10-04

I wish to pursue three topics: Prions in compartments not considered by Doc # 03 025IF; Stunning by methods other that air-injected captive bolt—Doc# 01 033IF; and ARM using high-pressure hydraulic stripping---Doc # 03 038IF. These are interrelated because they 1) affect human health directly through consumption of product, and 2) because waste-streams may carry prions which could wind up in products of commerce or in sludge put back on land, including top dressing of pasturage.

There are sufficient data from the published scientific and medical literature to show that prions are not confined to the brain and spinal cord of slaughtered cattle--they are in several other compartments and extra-CNS-Spinal nerve centers.

While the mode of slaughter with a captive bolt stunning device almost assures that if there are prions in the brain, this material will be circulated through the entire carcass*, other forms of stunning may also cause openings in the blood brain barrier. Thus, anything that uses head trauma can effect the same problem. A massive blow to the head sufficient to stun the animal will probably cause an intracranial bleed.

The high-efficiency meat salvage (ARM) using high-pressure hydraulic pressure is also capable of stripping out nerve tissues. The Coanda effect and Bernoulli effect need to be brought into the discussion.

* For the above see: Daly DJ, et al. Use of a marker organism to model the spread of central nervous system tissue in cattle and the abattoir environment during commercial stunning and carcass dressing. Appl Environ Microbiol. 2002 Feb;68(2):791-8.

I would also take exception to the following reported statements and ask, is the USDA really looking at a zero-risk product and non-spread of prions as its goal? If so, then the data gathered and the proposed regulations do not supply a proper underpinning for such a goal.

"Federal, state and local health officials were all cited as stressing that the USDA considers all the recalled meat from Washington safe, as it does not contain brain or spinal cord tissue thought to be able to harbor the infectious agent that causes mad cow disease, formally known as bovine spongiform encephalopathy."

"Steven Cohen, a spokesman for the USDA Food Safety and Inspection Service, was quoted as saying, "This is a zero-risk product."

Docket: 03 025IF [excluded parts]

A little anatomy seems warranted here. The drainage from the brain back through the lymph system seems not to be discussed. Further, is it USDA dogma that the issue is directional---i.e., the prions get to the brain via food intake—what if the movement of prions along nerves is bi-directional?

Originally, the USDA excluded the brain and spinal cord. These are not the sum and substance of this system. Now the proposed regs would add skull, trigeminal glanglia, vertebral column, including transverse processes, wings of sacrum, dorsal root glanglia (DRG) for animals over 30 months, tonsils, and distal ileum of small gut. These additional areas are based on limited studies.

The rationale for excluding these various parts is obscure and needs to be explained, notwithstanding the limited studies. The background information indicates that the specified excluded areas were noted to be contaminated on examination of a limited number of experimentally infected animals or animals that became infected. We do not know whether or not a statistically valid number of animals was included. For example, do the areas of contamination invariably show up in each animal—i.e., are these areas the usual sinks? If the issue is glanglia, then there are many more ganglia within the carcass.

When discussing the trigeminal nerve for comparative purposes, in Herpes zoster, for example, one finds that the ophthalmic branch of the trigeminal is often involved. If one had small sample theory projections for this disease, then based on that limited information, vesicular or bullous eruptions within dermatomes on the trunk might be considered as an entirely different disease. This is thus a comparative issue with a parallel study on a limited number of cattle.

In Glatzel and Aguzzi below, one notes that within the same species, different lab animals behaved differently and the prion's pathology shows up in distinctly different areas. It was noted that transgenic mice exhibited deposition of the prion and infectivity in specific portions of the central and peripheral sensory pathways, but almost no splenic accumulation. If these were the limited number of cattle used for development of the USDA regulations, one could see that there might be an error in judgement here. For example in the wild-type mice, by contrast, the prions always demonstrated splenic accumulation, and had widespread deposition throughout the central nervous system. Based on the type of mouse, either a lympho-neural sequence of spread occurred or there was substantial predilection of intranerval over lymphoreticular spread.

In cattle we have information that at least one animal in Japan behaved differently, but again, there are not sufficient numbers to really draw conclusions. Additionally, the range of age puts the majority of frank infections above 30 months, yet there are examples well below this. In the brief research done by USDA, it appears that with the limited number of cattle used, there are insufficient data upon which to really draw conclusions that will adequately assure beef eaters that the product, is as attributed to Steven Cohen of the FSIS, a zero risk product.

Given the limited testing, are we assured that other neural tissue is "zero risk"? At least in lab animals it is shown that the prion will track down peripheral nerves before reaching the CNS and that some areas are repositories for prion

accumulation. There are many more ganglia in animals as well as lymphoid tissues. It also appears that tracking is distally. Thus an argument for bi-directional movement.

For example, there is the submandibular of CN-VII, The superior, inferior and otic of CN-IX, on the Vagus on its exit from the jugular foramen, middle cervical sympathetic, cervicothorasic (stellate), sympathetic trunk of intercostals, celiac, super mesenteric, inferior mesenteric, chain, etc, etc. **The splenic sympathetic nerves may act as extracerebral prion reservoirs. Thus, the peripheral nerves coursing through the carcass may be potential contaminated areas, but these are not discussed. Again, there is a need to understand the rationale behind these regs and why some areas are excluded and others are not. What is the mechanism that would allow a prion to contaminate CN-V and not CN-VII?**

If the eye and CN-V are only considered, what makes them uniquely different from the other cranial nerves?

The eye (optic nerve) is properly part of the CNS, but that is not all that sticks out from the skull. The cervical and facial nerves arise directly from and are essentially extensions of the brain stem. The cribiform plate has holes that allow the olfactory nerves to project into the nasal cavity; the olfactory bulbs are brain, and in cattle are large and well supplied. The tong interconnects and is often used as a meat source. The sympathetic and parasympathetic ganglia as mentioned above need consideration.

My question here is where is the shut-off in this---is there some kind USDA policy, i.e., a politically and expediently driven "biological gate" that shuts off the chance for prions to move along these nerves?—see below where they actually put in a "gate".

Sympathetic innervation of lymphoreticular organs is rate limiting for prion neuroinvasion.

Glatzel M, Heppner FL, Albers KM, Aguzzi A.

Institute of Neuropathology, University Hospital Zurich, Schmelzbergstrasse 12, CH-8091, Zurich, Switzerland.

Transmissible spongiform encephalopathies are commonly propagated by extracerebral inoculation of the infectious agent. Indirect evidence suggests that entry into the central nervous system occurs via the peripheral nervous system. Here we have investigated the role of the **sympathetic nervous system** in prion neuroinvasion. Following intraperitoneal prion inoculation, chemical or immunological sympathectomy delayed or prevented scrapie. Prion titers in spinal cords were drastically reduced at early time points after inoculation. Instead, keratin 14-NGF transgenic mice, whose lymphoid organs are hyperinnervated by sympathetic nerves, showed reduction in scrapie incubation time and, unexpectedly, much higher titers of prion infectivity in spleens. We conclude that sympathetic innervation of lymphoid organs is rate limiting for prion

neuroinvasion and that splenic sympathetic nerves may act as extracerebral prion reservoirs. Neuron. 2001 Jul 19;31(1):25-34. PMID: 11498048 [PubMed - indexed for MEDLINE]

McGowan's notes--- sympathectomy delayed or prevented scrapie---what they are saying here is that they inserted an artificial gate so the prions could not move along the nerves. So, here we have some indication that my premise above is correct---- since splenic sympathetic nerves may act as extracerebral prion reservoirs and there may be bidirectional movement.

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PrP(C) expression in the peripheral nervous system is a determinant of prion neuroinvasion.

Glatzel M, Aguzzi A.

Institute of Neuropathology, University Hospital Zurich, Schmelzbergstrasse 12, CH-8091 Zurich, Switzerland.

Transmissible spongiform encephalopathies are often propagated by extracerebral inoculation. The mechanism of spread from peripheral portals of entry to the central nervous system (neuroinvasion) is complex: while lymphatic organs typically show early accumulation of prions, and B-cells and follicular dendritic cells are required for efficient neuroinvasion, actual entry into the central nervous system occurs probably via peripheral nerves and may utilize a PrP(C)-dependent mechanism. This study shows that transgenic mice overexpressing PrP(C) undergo rapid and efficient neuroinvasion upon intranerval and footpad inoculation of prions. These mice exhibited deposition of the pathological isoform of the prion protein (PrP(Sc)) and infectivity in specific portions of the central and peripheral sensory pathways, but almost no splenic PrP(Sc) accumulation. In contrast, wild-type mice always accumulated splenic PrP(Sc), and had widespread deposition of PrP(Sc) throughout the central nervous system even when prions were injected directly into the sciatic nerve. These results indicate that a lympho-neural sequence of spread occurs in wild-type mice even upon intranerval inoculation, while overexpression of PrP(C) leads to substantial predilection of intranerval over lymphoreticular spread. The rate of transport of infectivity in peripheral nerves was ca. 0.7 mm per day, and prion infectivity titres of sciatic nerves were much higher in tga20 than in wild-type mice, suggesting that overexpression of PrP(C) modulates the capacity for intranerval transport. J Gen Virol. 2000 Nov;81(Pt 11):2813-21. PMID: 11038396 [PubMed - indexed for MEDLINE]

USE OF EXCLUDED PARTS

While these excluded parts are prohibited from being included in the human food chain, what of other animals? If this goes to pet food and this material is later salvaged, can it be returned to the cattle feed-stream or to the rendering which could arrive at the human table or become products of commerce. There needs to be more discussion of these alternative routes. It has been demonstrated that both fecal material and urine can contain prions. What of the excrement from pets? Cat litter is composted as is manure. Movement of prions through the renal system into the surroundings (cat litter or manure) must be evaluated. What other animals can move prions through the renal system?

Further, what of the animal parts that wind up in sewer sludge or landfills where the drainage is recirculated through sewer works and back into sludge. There is

no way that sewer works would be able to disinfect a prion. More needs to be said of the land application of sewer sludge, especially the topping put on pastures. Here we have the circus route back to cattle.

As landfills age, their organic matter falls and thus the ability to capture charged particles is diminished. The USDA and EPA need to pay particular attention to landfill drainage and prions contaminating sludge. This is especially critical with the newly proposed EPA regs on compost and maximizing reuse. Recent EPA draft sludge regulations would appear to tie back into the Resource Conservation and Recovery Act (RCRA) and the Executive Order "Greening the Government Through Waste Prevention, Recycling, and Federal Acquisition". EPA is proposing to revise the current compost designation to include compost made from manure or sludge (biosolids). Thus, if these products of commerce are used in urban areas, there is a chilling prospect of passing around prions. Because the urine of cattle may include prions, this liquid is mixed with the fecal material and thus becomes the composite manure.

With respect to land application of manure or sludge, transport mechanisms warrant considerable thought. Since in the dry portions of the nation, there will be movement of soil and sludge in dust, prion movement is also an issue. The setbacks under Part 503 will have little effect when considering down wind movement of dust. As a grad student at Davis, I worked with the aerial application of Propanil and we had prune tree damage at 50 miles. But 50 miles is nothing. There are excellent papers discussing pathogen laden dust arising in West Africa, lofting into the upper atmosphere, transiting the Atlantic and 2 to 3 weeks later falling out over the Caribbean, there causing respiratory problems. This dust is subjected to intense UV, yet the pathogens survive to cause disease. How is this different from the setbacks noted in Part 503? Is there some vastly different mechanism at work as recognized in Part 503, that one does not find in trans-Atlantic movement. If this is the case, has it been scientifically evaluated? Thus why are essentially the same pathogens able to move thousands of miles and cause disease, yet under EPA's Part 503, they are completely harmless after movement of a few hundred feet?

Particulate matter lodged in the upper respiratory tract will move up the ciliary escalator to be swallowed, hence the potential for ingestion of prions as well as a route through the lungs.

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Docket 01 033IF [captive bolt]

In the early 1970s, I did a series of audits for the Legislative Analyst's Office on slaughterhouses here in California. The captive bolt trashes the brain and part of the skull vault and these disrupted tissues are then picked up by the blood and circulated to the body--so whether or not the prion is in the meat to begin with,

brain tissue soon arrives in the muscle during the killing. Also, the larger non-penetrating captive bolt equipment produces sufficient concussion to cause intracranial bleeds and disrupt the blood-brain barrier. The background information related to Doc # 01 033IF discussing captive bolt also mentions macro emboli.

There is no discussion of micro emboli. Trauma sufficient to cause unconsciousness will cause damage ranging from contusions and ultra structural changes including the disruption of the blood-brain barrier to frank tissue destruction and accompanying hemorrhage. There may also be white matter shearing. Thus, there is an adequate opportunity for micro emboli to circulate. Since prions are small, there is no need for macro emboli. Additionally, the smaller emboli will reach deeper into the arborizing capillary beds. Penetrating and major head injuries also cause an almost immediate rise in intracranial pressures, thus augmenting movement of micro emboli into the damaged vascular net. With epidural hematomas, there is stripping of the tightly attached dura from the inner table of the skull---again an opportunity to open transport routes for prions.

Killing with electrocution can see micro petechiae to frank tissue destruction, again releasing material across the damaged blood-brain barrier.

Also since the killing and quartering equipment is not sterilized between killings (for prions, this is moot as surgical instruments used on vCJD patients are now disposable as it is virtually impossible to sterilize instruments contaminated with prions), it also contaminates the next few cows--that's why over 10,000 pounds of beef was pulled. If you back calculate this to number of head, the following crude analysis obtains. Assume the average steer weight at 1175 pounds (#), hot carcass weight 740#, yeild is about 54% or 405#. Thus, the 10,000+ pounds of pulled beef is about 25 head, somewhat less if a mature dairy cow is substituted.

But what of the cattle that have BSE/TSE and are not noted. Additionally there is a lot of fatty tissue to saw through with the chain saws, this stuff, tissue fluids, and the blood are aerosolized all over in a fine mist. It may be worthwhile to contact NIOSH on this as well as some of the larger worker's comp carriers. They are probably having their actuaries looking at this right now as they won't want to see increased claims payments down the line. Some one with spongyform can get very expensive to care for and this would raise hell with the funding for an insurance company.

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Docket 03 038IF [ARM]

Because of the numerous curved surfaces and foramen in the skull and spine, there is a need to consider both the Coanda effect and Bernoulli effect. These effects will cause, in the former case, fluids to follow contours into foramen, thus tending to wash out what ever is within that cavity. In the latter case, fluids moving across curved surfaces may create negative pressures, again tending to draw materials out. Since the material in question is nervous tissue and related material, high-pressure fluids may tend to contaminate the salvaged product. This may be behind the high rates of failure found in this product.

It is not specified, but is the nasopharynx, oropharanx, and laryngopharynx thus treated? I see that the tonsils are to be excluded. The reason for this inquiry is the cribriform plate and thus the above discussed fluid dynamic effects pulling CN-I into the product.

If there are questions. I may be reached by phone at (805) 968-0481