Comments from Dr. Glenn Songer
Department of Veterinary Science and Microbiology, University of Arizona

- Oral exposure to ETX (epsilon toxin) seems an unlikely scenario, due likely dose size and delivery problem, to say nothing of toxin inactivation at low pH in stomach.

- Aerosal exposure to ETX has apparently not been studied, so the precise BW (bioweapon) threat posed by ETX, given by this route, is unknown.

- Hemorrhage is uncommon in type D-induced disease in sheep and cattle (Griner et al 1956; Lulov and Angelov 1986), but Caprine disease frequently presents as chronic hemorrhagic entercolitis (Blackwell and Butler 1992; Blackwell et al 1991; Oxer 1956; von Rotz et al 1984). The basis for this species-to-species difference in clinical picture is unknown (Blackwell et al 1991), but its occurrence is noteworthy in the absence of documented human infections or intoxications.

- Furthermore, if two small ruminant species respond as differently as goats and sheep, there is good reason to expect that ETX effects in humans and other monogastrics may be different, as well. It is not absolutely inconceivable that humans are insusceptible to ETX.

- There is no such precedent for infection of humans by C. perfringens types B and D or intoxication by ETX. As noted, it may be that monogastrics are insusceptible to infection with types B and D, and/or that they are not subject to the effects of ETX.

- Laboratory work with C. perfringens type D dates to the early 20th century, but there are no reports (published or anecdotal) of ill effects due to laboratory exposure to C. perfringens types B or D or to ETX-containing fluids. The same can be said of the research/development and production divisions of veterinary biologics companies. It proves nothing, but it is unusual that no such cases have been reported if ETX is an important threat to humans.

- Thus, there is little or no justification in fact for including ETX on the list of select agents.

- Direct and indirect evidence do not support a need for strict regulation of ETX possession limits, particularly in the veterinary biologics industry. Furthermore, there is ample justification to reconsider the inclusion of ETX among the Category B Select Agents.

Comments from Dr. Keith Haffer
Advantage Bio Consultants

- Records I have reviewed from your firm indicate that you have been producing Clostridium perfringens type D for 30 years and during this time there has been no evidence of equipment failure or release of bacterial cultures into the work space or environment. Additionally, there have been no incident reports by production workers of any accidental exposures of clinical disease (incapacitation) associated with Clostridium perfringens type exposures.

- Although many studies using animal models reproduce the ETX (epsilon toxin) effects, correlation to human disease is based upon in vitro studies or extrapolation from goat, sheep, and calf models.

- The recent review article by Mantis (2005) provides an excellent overview of ETX. The review states, "There are no available reports examining the effects of purified epsilon on humans or non-human primates..... extrapolating from experiments performed in small animal models, it is estimated that the lethal dose of epsilon give intravenously is 100ng/kg. The actual lethal dose of epsilon as an aerosol or ingested will greatly depend on the interaction of toxin with the human respiratory and gastrointestinal mucosae, of which nothing is currently known."

- Paddle (2003) makes the following statement, :Ignoring such issues as the stability of the toxin in the environment, what these findings illustrate is that the toxicity data currently available from animal studies are generally insufficient to decide the extent of the inhalation threat to humans."

- The publication by OIE/Institute for International Cooperation in Animal Biologics/ The Center for Food Security & Public Health states, "There seems to be little or no information about the effects of epsilon toxin on humans."

- Although certain publication are used for extrapolation of ETX for humans, one that is not cited suggests the kidney contributes to host defense against lethal toxicity of epsilon toxin (Tamai et al, 2003). IN this mouse study, a highly purified, recombinant epsilon toxin (20 ng MLD50) was injected into mice and the progression of the toxin accumulation was followed. The authors conclude, "Epsilon toxin is accumulated predominantly in the kidneys, which attenuates its lethality... We propose that the kidneys contribute to the host defense against epsilon toxin by trapping the toxin and thereby protecting more susceptible organs, e.g. the brain, from lethal toxicity."

Comments from Dr. Keith Haffer (continued)
Advantage Bio Consultants

- No human or non-human primate studies have been documented, ranges are extrapolated from mice and aerosol exposure toxicity level are made equivalent to intravenous toxicity.

- A weaponized ETX is of no more of a threat than natural exposure.

- There has never been a documented case of Clostridium perfringens Type D infection in humans or ETX exposure.

- In conclusion, based upon published literature, past production history, production methods and personal protection afforded production workers, and risks of human exposure and weaponization, I strongly believe the risk working with Clostridium perfringens Type D is low. The following specific items are convincing for an exemption for this firm and this product: A thirty year history of safety and total lack of specific intoxication with ETX; the lack of any direct scientific evidence of actual human health risks associated with purified ETX. Much less the non-pure form which exists in the production methods of Clostridium perfringens Type D, as is conducted by Colorado Serum Company; The risk analysis of human exposure yields a risk rating of Low indicating very little concerns are associated with the production process; and, weaponization for animals or human with the actual material produced during a production run of Clostridium perfringens Type D at Colorado Serum Company is mitigated by available USDA- licensed biologicals (animals) and biological facts, uncertainity factors, physical and other safeguards (humans).