Botulism of Cattle in Brasil
Diagnosis and Vaccination

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Brazil:
The 5th biggest country in the world
8,547,404 km²
~166 million Inhabitants
~160 million cattle
Main breeds:
- Nelore, crossbreds
- Gir and crossbreds
- Holstein Friesian
- Other Beefbreeds: Angus, Indubrasil, Simmental, Charolais
Ecology and Climate

Brazilian Highlands

Rainy season (summer): October-March

Dry season (winter): April-September
Typical Brazilian Pastureland
Brachiaria pasture in the rainy season

Brachiaria pasture in the dry season
Hygienical status

National eradication program for FMD

No national programs for the eradication of tuberculosis, brucellosis and IBR
Occurrence of Botulism in Brazil

Losses: app. 1.5 Billion US$ during the last 10 years

What is Botulism?

Botulism: in most cases a fatal intoxication with a neurotoxin produced by *Clostridium botulinum*:
- 8 types recognized on the basis of the produced toxins (A, B, Ca, Cβ, D, E, F, G)
- anaerobic gram positive rods, which produces oval, subterminal endospores
- endospores are distributed in soils and aquatic environments worldwide
- the types C and D can often be found in the intestinal-tract of domestic animals (cattle and fowl)
- germination of spores, growth of vegetative cells and toxin production occurs in anaerobic locations (rotting carcasses, decaying vegetation, wounds)
What is Botulism?

Intoxication occurs when preformed BoNT is ingested

- Osteophagia: phosphorus deficient cattle take up bones or carcases where C. botulinum has multiplied and formed its toxins
- Drinking of contaminated water
- Feed on contaminated silage or other feeding stuffs

Toxico-infectious botulism occurs when spores germinate in wounds or in the intestinal tract

- Intestinal toxico-infectious botulism is recorded in foals (shaker foal syndrome), broiler chickens, and human infants (infant botulism)
- Wound botulism is recorded in humans associated with the use of contaminated drugs or syringes
Mode of action of the BoNT

Botulinum Neurotoxin

1. Receptor
2. Binding Domain
3. Translocation Domain
4. Catalytic Domain

AXON

SNARE-PROTEINS

Synaptobrevin (VAMP) at the vesicle membrane cleaved by: BoNT B, D, F

SNAP-25 cleaved by: BoNT A, E

Syntaxin cleaved by: BoNT C1

intersynaptic space
Osteophagia

During the rainy season the fast growing and relatively low demanding pasture species *Brachiaria* contains very low amounts of phosphorus (0.12%) especially in the „Cerrado“ areas where the soil is phosphorus deficient.

*Brachiaria spp.*
Ostophagia

Inadequate phosphorus supplementation

Improper mineral supplementation sites
Osteophagia

High demanding cattle breeds

The result is a phosphorus deficiency in cattle especially in the lactation period or during growth when the demand of phosphorus is high.

Foto by: I.S. Dutra, Universidade Estadual Paulista, Campus Araçatuba
Ostophagia

Inadequate removal of carcases lead to a vicious circle
Other sources of intoxication

Cattle enter the holes, lose their droppings and contaminate the water with botulinal spores.

water holes: "cacimbas" which serve as a cistern

Fotos by: Aires Manoel de Souza, Escola de Veterinária, Universidade Federal de Goiás
Other sources of intoxication

Decaying carcass in a water hole

Foto by: I.S. Dutra, Universidade Estadual Paulista, Campus Araçatuba
Other sources of intoxication

Chicken litter:
Used for supplementation in the winter
Other sources of intoxication

Poorly fermented silage:

During a poor fermentation process *C. botulinum* is able to produce its toxin.

Foto by: I.S. Dutra, Universidade Estadual Paulista, Campus Araçatuba
Other sources of intoxication

Harvested maize fields with decaying material

Foto by: I.S. Dutra, Universidade Estadual Paulista, Campus Araçatuba
Diagnosis

Clinical signs:

Weak gait

Paralysis of the limbs
Diagnosis

Clinical signs:

Unability to get up
Diagnosis

Clinical signs:

- atony of the tail

Unshadowed sensorium
Diagnosis

Clinical signs:

Complete paralysis of muscles
Diagnosis

Clinical signs:

- Tongue flaccidity
- Dysphagia
- Decreased salivation

Foto by: I.S. Dutra, Universidade Estadual Paulista, Campus Araçatuba
Institut für Tropentierhygiene

Therapy

If available a polyvalent antitoxin is effective in neutralizing unbound toxin in early stages of the disease.

But: antitoxin is very costly and the action of already bound toxin is nonreversible.

Mildly affected animals sometimes recover over a period of weeks without therapy.

Good nursing is essential for recovery.
Research and results

80 samples (soil, carcass material, fodder, feces) were collected

64 on farms with known history of botulism (23 positive, 35%)

16 samples at sites of no known history of botulism (2 positive 12.5%)

25 were positive for the presence of *C. botulinum*, 24 of the CD complex group and 1 of *C. botulinum* type A.

<table>
<thead>
<tr>
<th>Type</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type C</td>
<td>4</td>
</tr>
<tr>
<td>Type D</td>
<td>6</td>
</tr>
<tr>
<td>CD complex</td>
<td>14</td>
</tr>
<tr>
<td>Type A</td>
<td>1</td>
</tr>
<tr>
<td>total</td>
<td>25</td>
</tr>
</tbody>
</table>

In this work *C. botulinum* types C and D were found in 96% of all positive samples.

Laboratory diagnosis in Brazil was done by mouse bioassay with neutralization tests.
Vaccine

There are several types of vaccine available on the market which are more or less effective.

The efficacy of three in Brazil available vaccines was tested.
A group of 60 young bulls mainly Nelore and Nelore crossbreds were selected.
15 animals each where vaccinated with one of the three vaccine types, 15 served as the negative control.
Blood was taken over a period of one year, once a month at the beginning and then every second month.
The immunologic titre was determined by ELISA using the toxin of reference strains of *C. botulinum* types C and D.
Vaccine

Two of the tested vaccines (T1, T3) produced a good antibody titre.

T2 had no measurable response compared to the control group.
Conclusions

The types C and D are playing a major role in causing botulism in Brazil.

Not every vaccine against botulism is producing a measurable antibody titre.
Prophylaxis

Vaccination: protection against botulism types C and D with an effective vaccine

Pasture management: supplementation of phosphorus removal of carcasses use of different grass species (*panicum spp*) proper preparation of silage exclude chicken litter as cattle fodder avoid overstocking