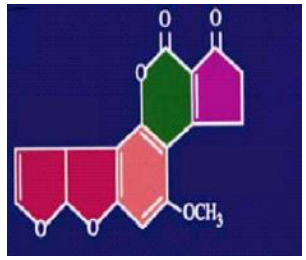
Aflatoxin Control: Experience from Poultry Industry



Prof. G. Devegowda
devegowdag@gmail.com

Poultry Industry in India is a vibrant Agribusiness with an annual turnover of over Rs.1,00,000 crore

Poultry feed production in million tons/year

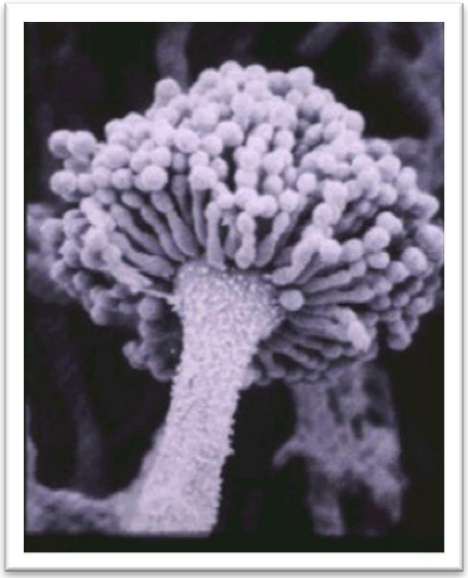
Broilers	12.0
Layers	10.0
Breeders	3 .0
	25. 0

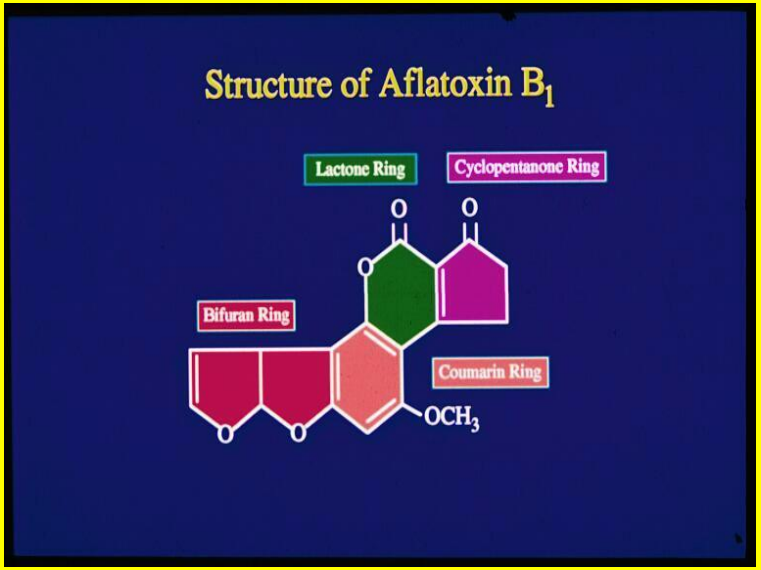
Aflatoxins B1, B2, G1,G2 are produced by

Aspergillus Flavus: Storage Fungi

Aspergillus Parasiticus:

Aflatoxin B1 is the most prevalent and toxic Aflatoxin





Aspergillus Flavus on Rice

Murthy and Devegowda, 2004

Aflatoxin reference standards

Murthy and Devegowda, 2004

Molds and Aflatoxins Production…

High moisture

High humidity

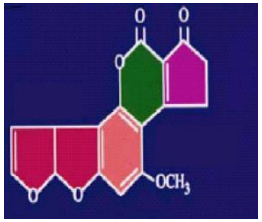
Temperature

Improper drying

Improper storage

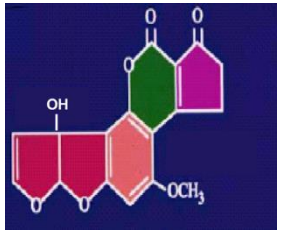
Aflatoxins

Regulations for Aflatoxins for Dairy and Poultry feeds



Aflatoxin B_1

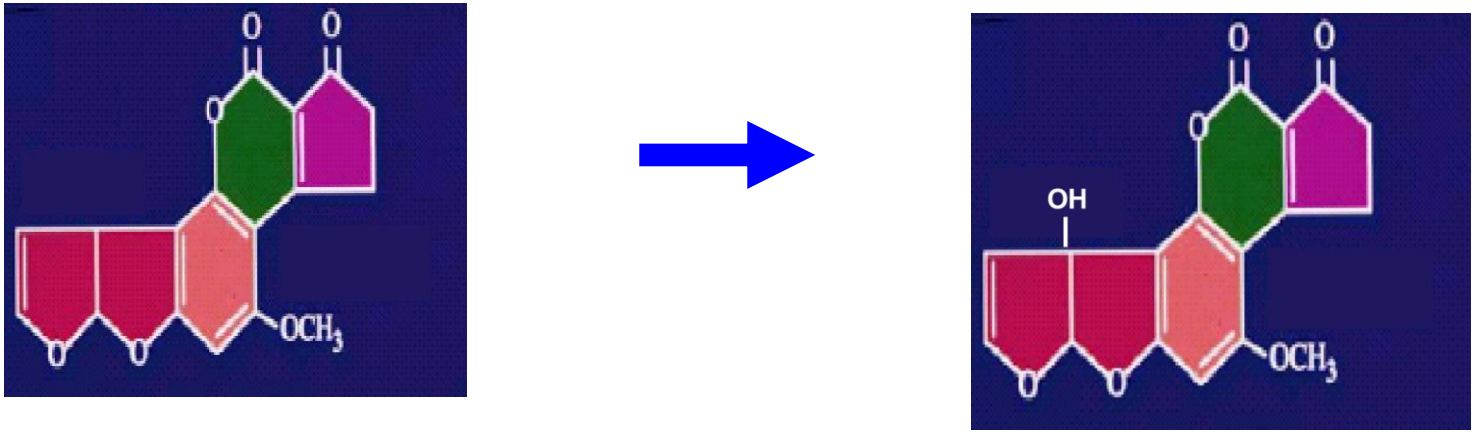
Food Safety….



Aflatoxin M_1

Aflatoxin M_1 in Milk and Milk Products and in Eggs and Chicken Meat

Aflatoxin B1 to Aflatoxin M1



- 1 to 6% of Aflatoxin B1 present in the Dairy feed is transferred to milk as Aflatoxin M_1

- 0.1 to 0.2% of Aflatoxin B1 present in Poultry feed is transferred to eggs and meat as Aflatoxin M1.

BIS Regulations for Aflatoxin B1 both in Dairy and Poultry feeds

20 mcg / kg (20ppb)

Source : BIS 2052 (2009) Compounded feeds for Cattle,

BIS 1374 (2007) Poultry feeds

U.S. FDA Regulations for Aflatoxin B1 both in Dairy and Poultry feeds

20 mcg / kg (20ppb)

Source : FDA (U.S), Ministry of Agriculture (Canada)

European regulations for Aflatoxin B1 in Feeds

	µg / kg (ppb)
Dairy Cows	5
Calves	10
Poultry	20

If the world adopts EU standard for Aflatoxins

what will be the economic impact on Dairy Industry and raw material exporters?

- US and BIS 20 mcg/kg (20ppb) in Dairy feeds
- EU 5 mcg/kg (5 ppb) in Dairy feeds

Good Manufacturing Practices and Storage of Feeds

Stop Fungal growth in Feeds by good Storage Practices

Source : Sriya Poultry Feed plant, Bengaluru

Moisture Optimization Program(MOP) to control mold growth in feeds

Contains Mold Inhibitor and Surfactant

- Optimize the moisture in the final feed to 11-11.5%
- Convert free form of moisture into bound form
- Spraying of mold inhibitor and water varies depending on raw materials moisture content

Silos for Maize Storage with Aeration: Pre-cleaned and Dried Maize is stored for 3 to 4 months

Maize pre-cleaner

- Removes dust (contains Aflatoxins)
- Removes all waste materials

Storage Conditions of grains…..

- Moisture content less than 13 %
- Avoid damaged and broken grains(susceptible for fungal growth)
- Avoid Insect damaged grains

(susceptible for fungal growth)

SNEHA

KARIMNAGAR FEED PLANT WITH 800 TPD

New maize having high moisture more susceptible for fungal growth

https:en.Wikipedia.org/wiki/maize

Use it immediately and use mold inhibitors to prevent fungal growth in feeds

- Dilution of new maize with old maize
- Dilution of contaminated maize with good quality maize

Good Quality Raw Materials: Energy Sources

Good Quality Raw Materials: Protein sources

- Avoid using Distillers dried grains (DDGS)

  (It contains Aflatoxins 3 times more than the grain)
- Avoid using Brewery waste (high level of Aflatoxins because of poor quality grains used)
- Avoid Self mixing of feeds

The levels of Aflatoxins(ppb) in feedstuffs		
	Aflatoxins	
	0 -50 ppb	50-100 ppb
Cereals, samples	88	4
Cereal byproducts, samples	48	8
Oilseed meals, samples	152	8

Source: G. Devegowda, K.L Aravind and C.K Girish

Prevention and Control of Aflatoxins

Prevention and Control

An integrated approach is needed

- Use of new technology
  - Good Agricultural Practices (GAP)
  - Good Manufacturing Practices (GMP)

- Drying in Sunlight

- Inorganic adsorbents :
  - Clay materials

- Biotechnological :
  - Organic adsorbents -Yeast cell wall, Algae

Inorganic adsorbents: Clay materials

✓ **Zeolites (1:1)** Sodium Aluminosilicates

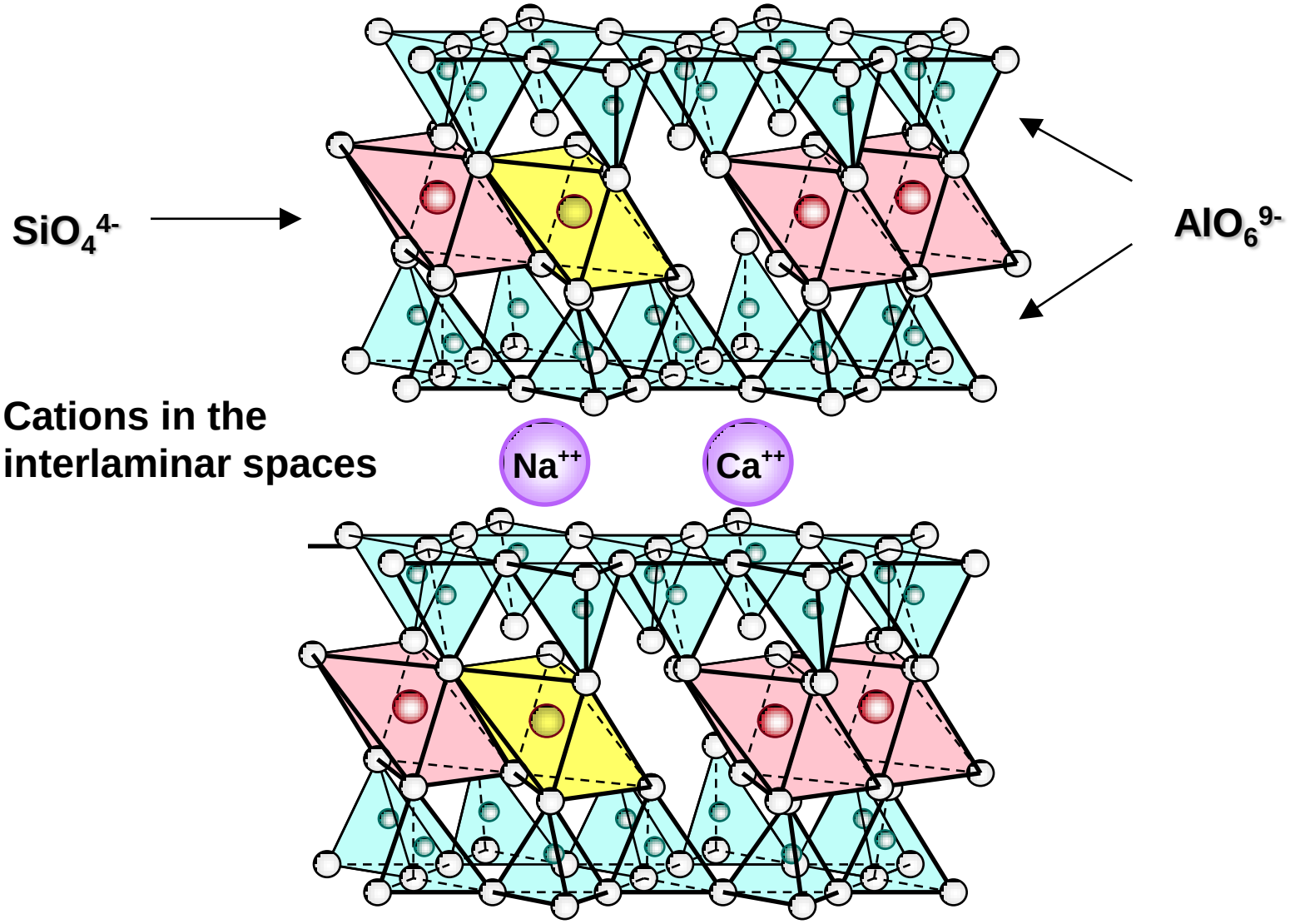
✓ **Bentonites (2:1)**

Sodium and Calcium Bentonite (2:1)

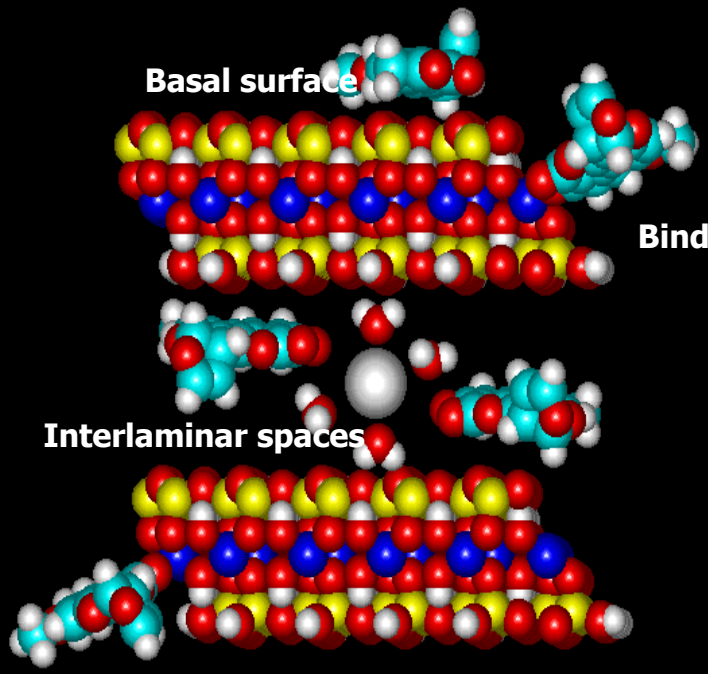
Montmorillonites (Na, Ca) x (Al, Mg) $2Si_4O_{10}$ (OH) $2.nH_2O$
Hydrated Sodium Calcium Aluminosilicates **(HSCAS)**

Sodium and Calcium Bentonite (2:1)

Hydrated Sodium Calcium Aluminosilicates **(HSCAS)**



MODE OF ADSORPTION AFLATOXINS ON HSCAS (BENTONITE) SURFACE



- 90% of aflatoxins are adsorbed between the interlaminar surfaces, the remainder, in the basal ends

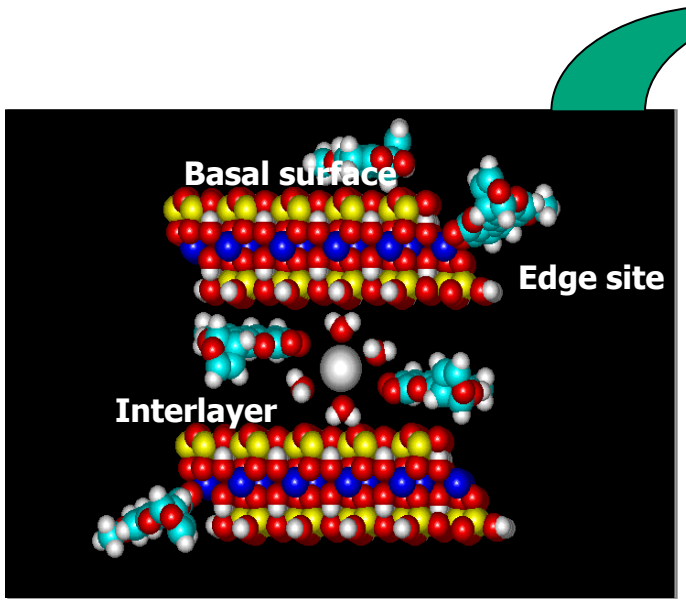
Source: Phillips, 2010

Clay Materials

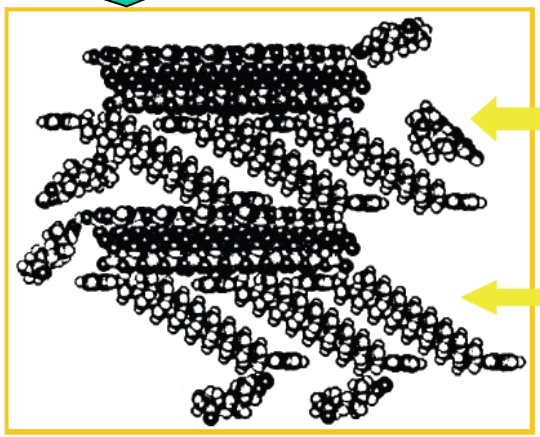
- High inclusion rates (10 to 12 kg/ton)
- Reduced mineral utilization: Manganese, Zinc, Copper, Phosphorous and Magnesium
- Reduced Vitamin utilization
- Contamination by Dioxins and heavy metals

Modified Organoclay

- Modified organoclays are synthesized from the bentonite / zeolite (natural clays) through substitution of inorganic cations for organic cations (Redding et al., 2002).



Natural Clay



Lemke et al., J Agric Food Chem 46:3789-3796, 1998

Modified clay

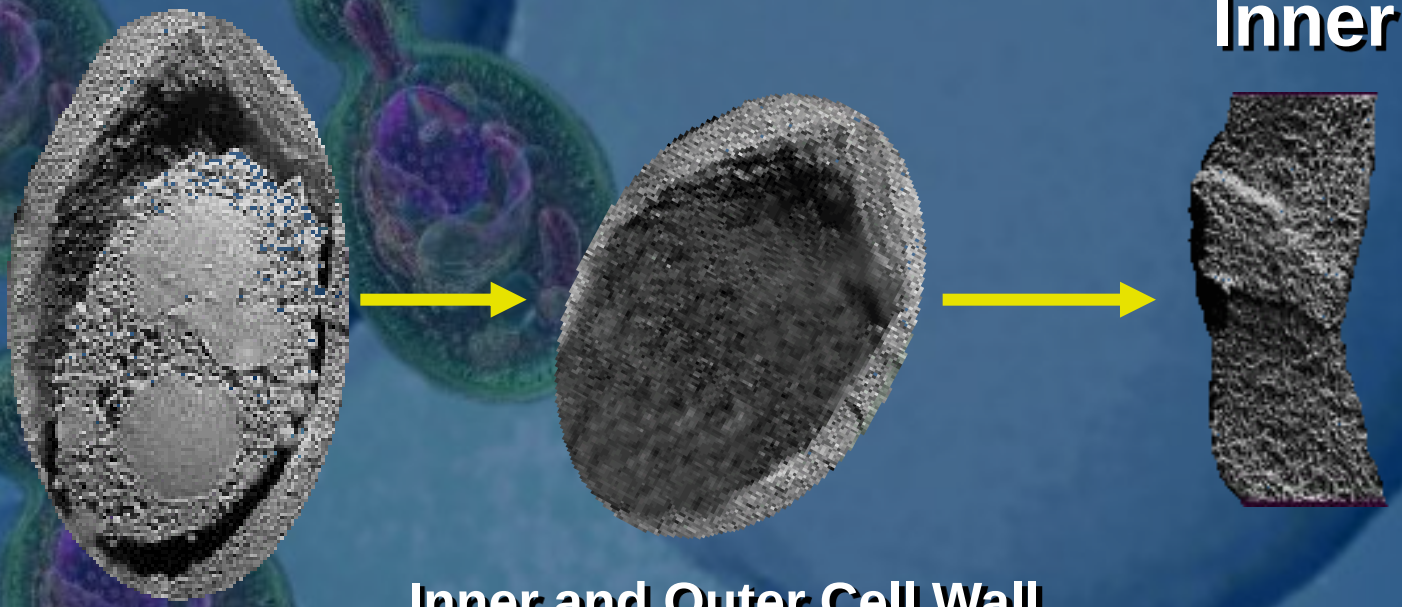
Quaternary amines
Risks to the Environment

- Due to health and environmental risks related to quaternary amines, modified clays are difficult to produce.

- Literature:The Role of Surface Interactions in the Synergizing Polymer /Clay Flame Retardant PropertiesSeongchan Pack, Takashi Kashiwagi, Changhong Cao, Chad S.Korach, Menachem Lewin, Miriam H. RafailovichMacromoleculesVol.: 2010, 43 (12), pp 5338-5351DOI: 10.1021/ma100669g

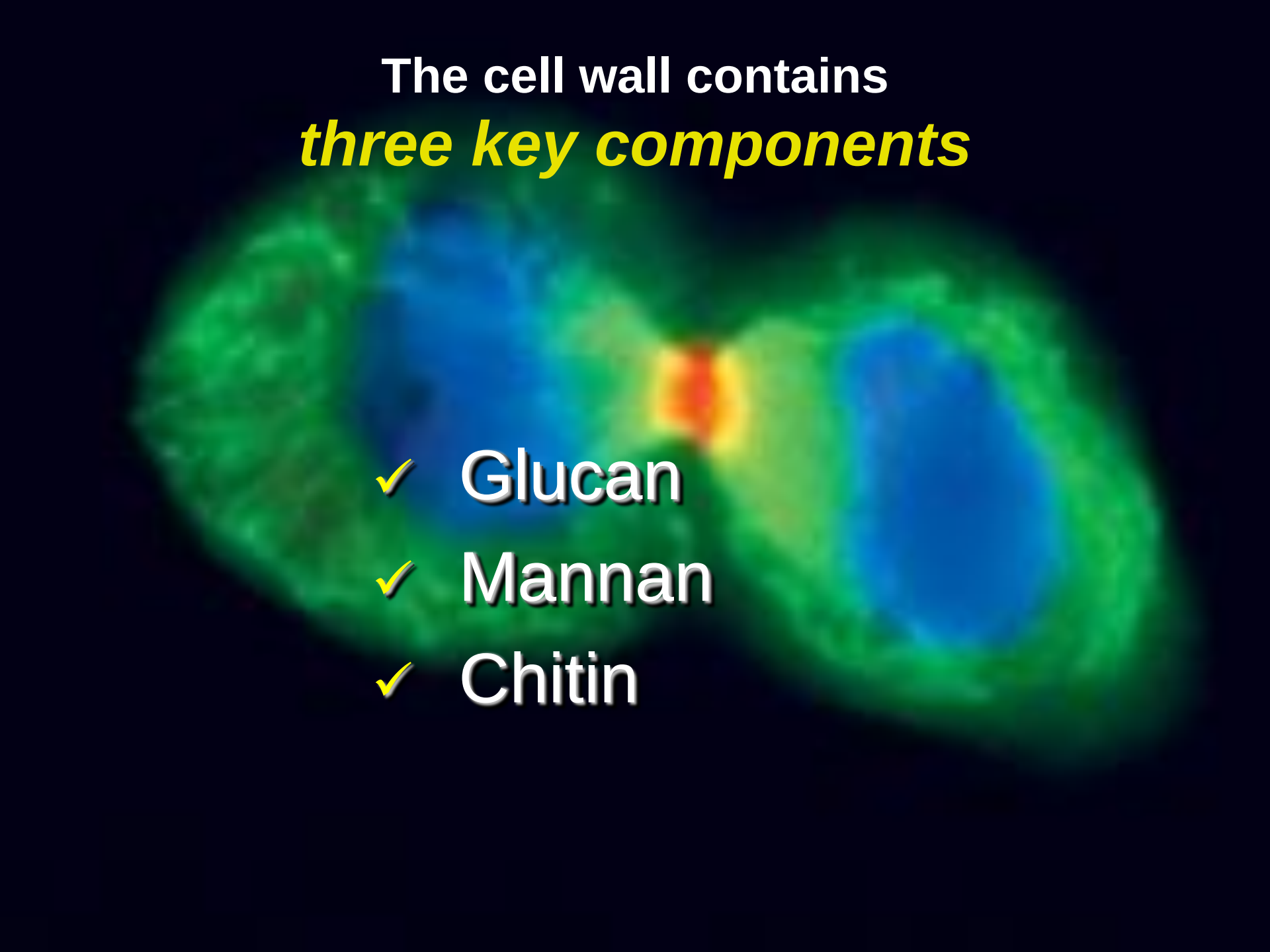
Biotechnological Solutions: It's based on yeast cell wall

Yeast



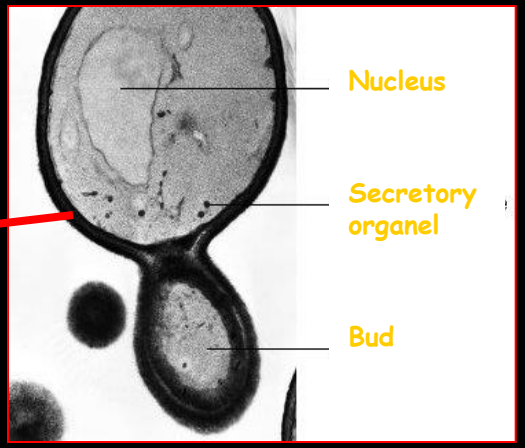
Inner

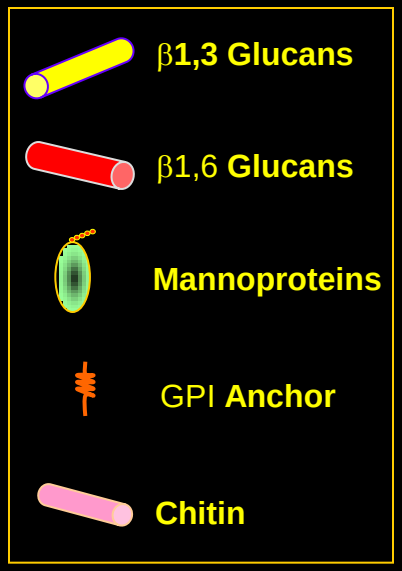
Inner and Outer Cell Wall

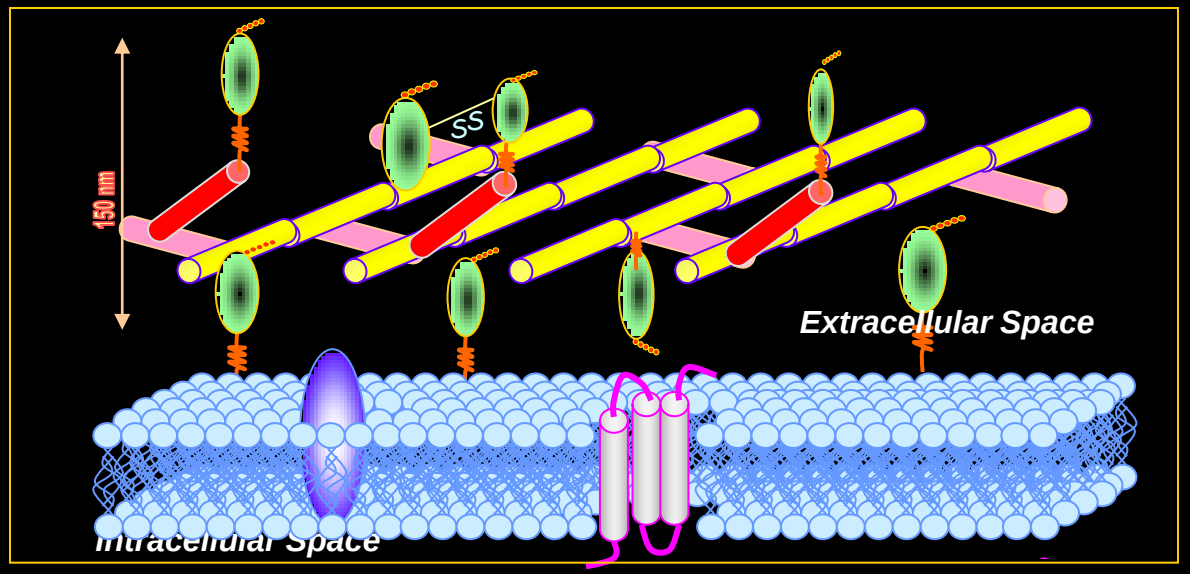

The cell wall contains
three key components
✓ Glucan
✓ Mannan
✓ Chitin

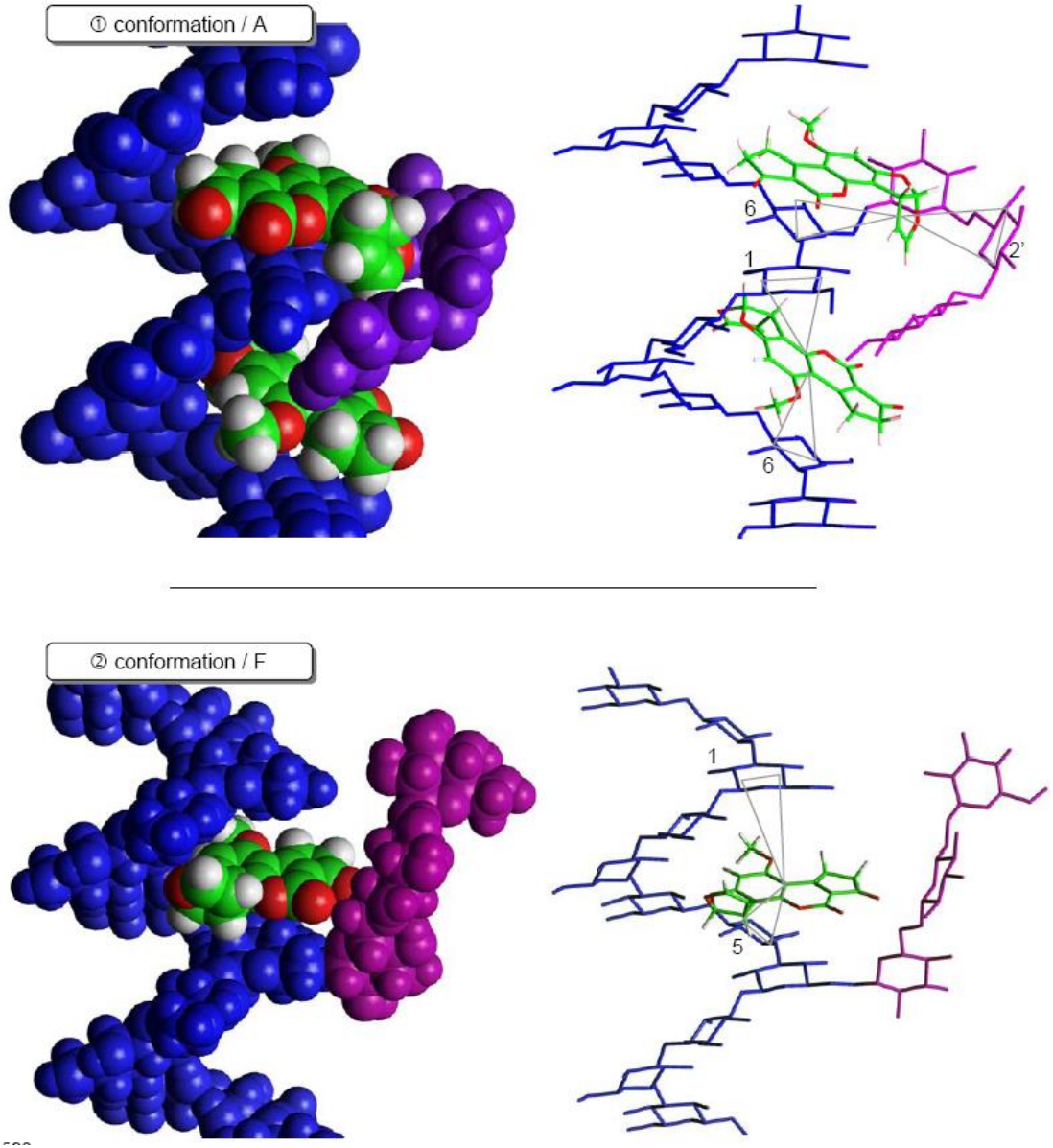
Yeast cell wall

⇨ Glucans







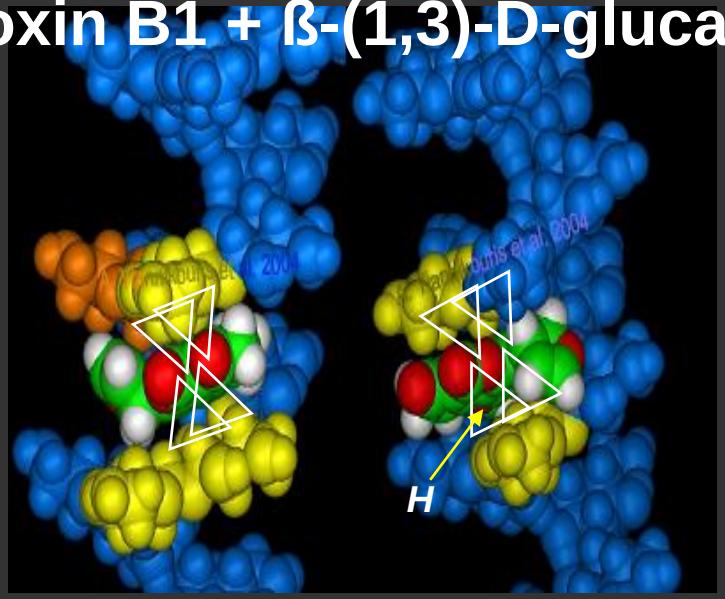


Adsorption of Aflatoxin B1 into the β-(1,3)-D-glucan chain

A. Yiannikouris et al. 2004. Biomacromolecules

ß-D-glucans binding Aflatoxin B1

[Aflatoxin B1 + ß-(1,3)-D-glucans]



Energy (kcal/mol)		
	Aflatoxin B1	
E (Van der Waals)	- 39,3	- 38,3
E (electrostatic)	- 0,6 (0 H)	- 2,7 (1 H)
E (docking)	- 39,9	- 41,0

High geometrical similarity between AFB1 and β-(1,3)-D-glucans

A. Yiannikouris et.al.,2012

Adsorbing aflatoxin : an efficient approach

MYCOSORB®

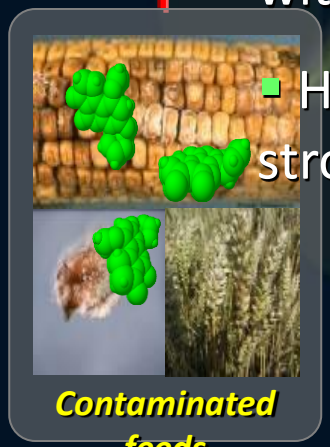
Gastro-intestinal tract

- Mycotoxins chemically bond to the adsorbent
- Mycotoxins pass through the digestive tract without absorption
- Harmful effects of mycotoxins strongly decreased

Ingestion

Excretion

Absorption



Contaminated feeds

fæces

Mycotoxin intrinsic toxic effects

milk
eggs
meat

urine

Activation

Detoxification

High toxic effects

Recycling

Low toxic effects

Transfer

Taylor-Pickard et al., 2009

⇨ Scientific collaborations

A. Yiannikouris, UR1213, Unité de Recherches sur les Herbivores, INRA, Centre de Clermont-Theix, France.

INRA/Alltech PhD Student

Dr. J François PhD, UMR-CNRS 5504, UR-INRA792, INSA, Toulouse, France.

Yeast cell wall isolation + extraction

Dr X Cameleyre Ph.D., Toulouse, France.

Mycotoxin analyses

Dr L Poughon & Pr. CG Dussap PhD Laboratoire Chimique et Biologique, Clermont-Ferrand, France.

Mathematical modeling

Dr G Bertin PhD, Regulatory Affairs Manager, Alltech-France, Goussainville, France.

Alltech Scientific Coordinator

Dr I Canet & Dr G Jeminet PhD, Laboratoire SEESIB, UMR CNRS 6504, Clermont Ferrand , France.

NMR analyses

Dr G André PhD, Unité de Biochimie Structurale, INRA-Institut Pasteur, Paris, France.

Molecular modeling

Dr A Buléon PhD & Dr B Pontoire, UR783, Laboratoire de Physico-Chimie des Macromolécules, Nantes, France.

X-ray diffraction analyses

Dr P Galtier PhD, UR66, Laboratoire de Pharmacologie et Toxicologie, Toulouse, France.

Scientific support

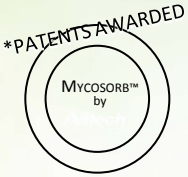
Evolution of Mycosorb

1988

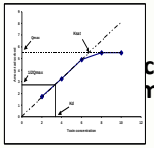
Mycosorb was created through the modifications to the glucomannan portion of the inner cell wall of yeast

1995

Mycosorb was awarded a patent by the US patent and trademark office.



1999



Dr. Karl Dawson discovered conditions for the maximum efficacy of Mycosorb

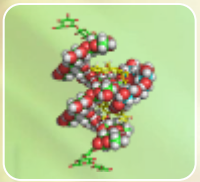
2000

The beginning of extensive research on Mycosorb took place in the lab of Dr. T.K. Smith, Guelph

2002

Dr. G. Devegowda showed the ability of Mycosorb to act within 30 minutes

2004



Dr. A. Yiannikouris described the physical and chemical mechanisms involved in Mycosorb mode of action.

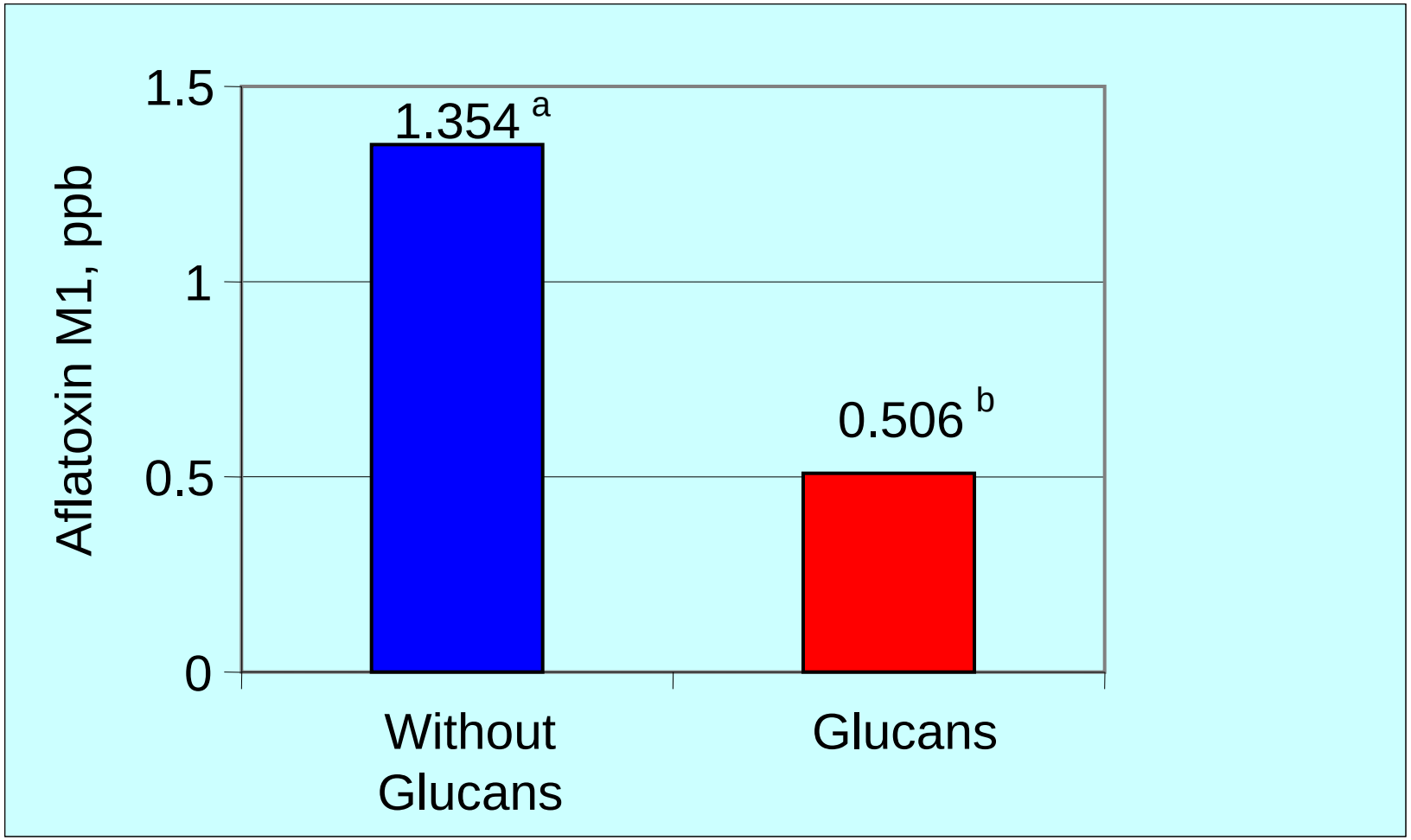
2007

Merrill and co-workers unveiled the efficacy of Mycosorb in new circumstances

2009

Mycosorb continue to be the leading technology worldwide with 13 PhDs and more than 50 peer reviewed papers

Reduction of aflatoxin M_1 in milk with Yeast Glucans

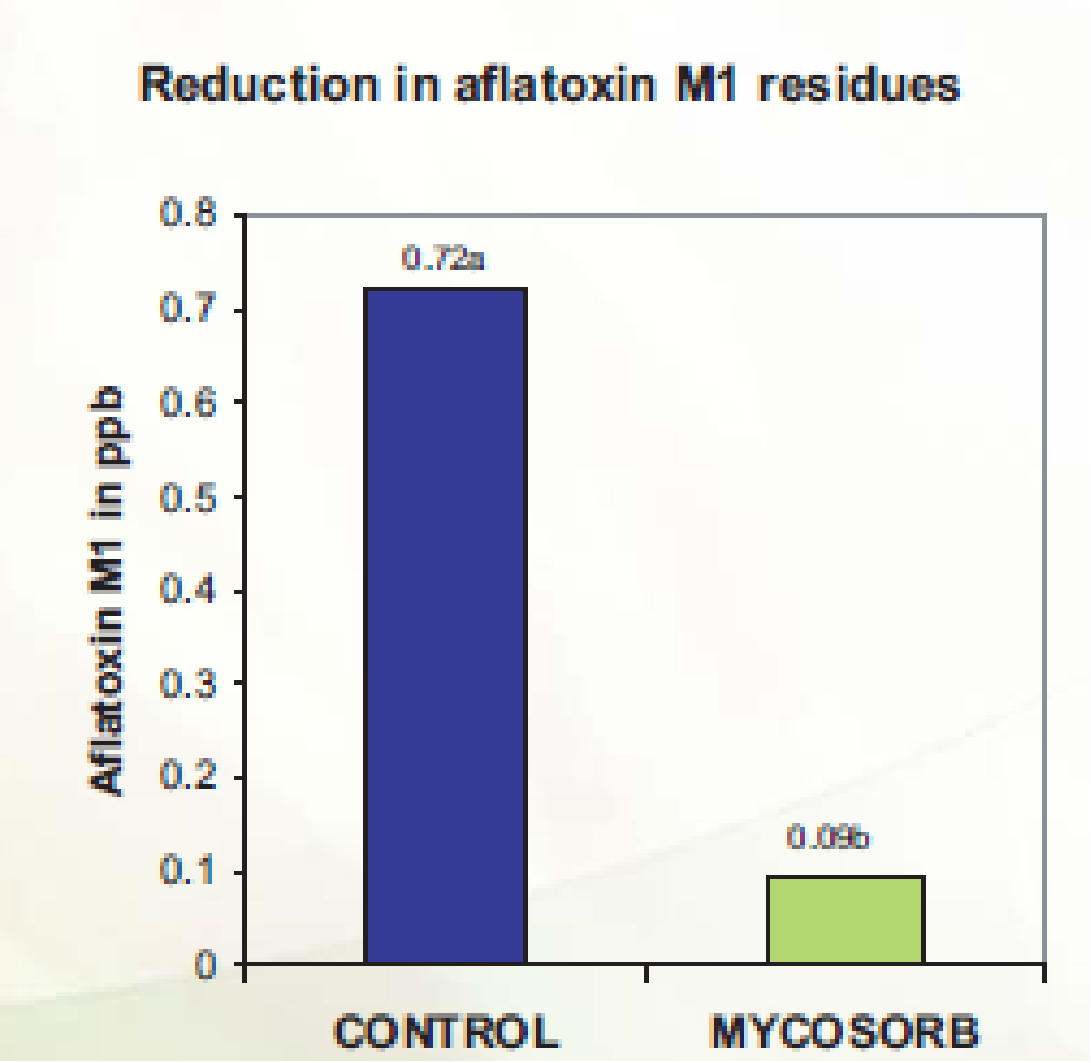


Aravind and Devegowda, 2004

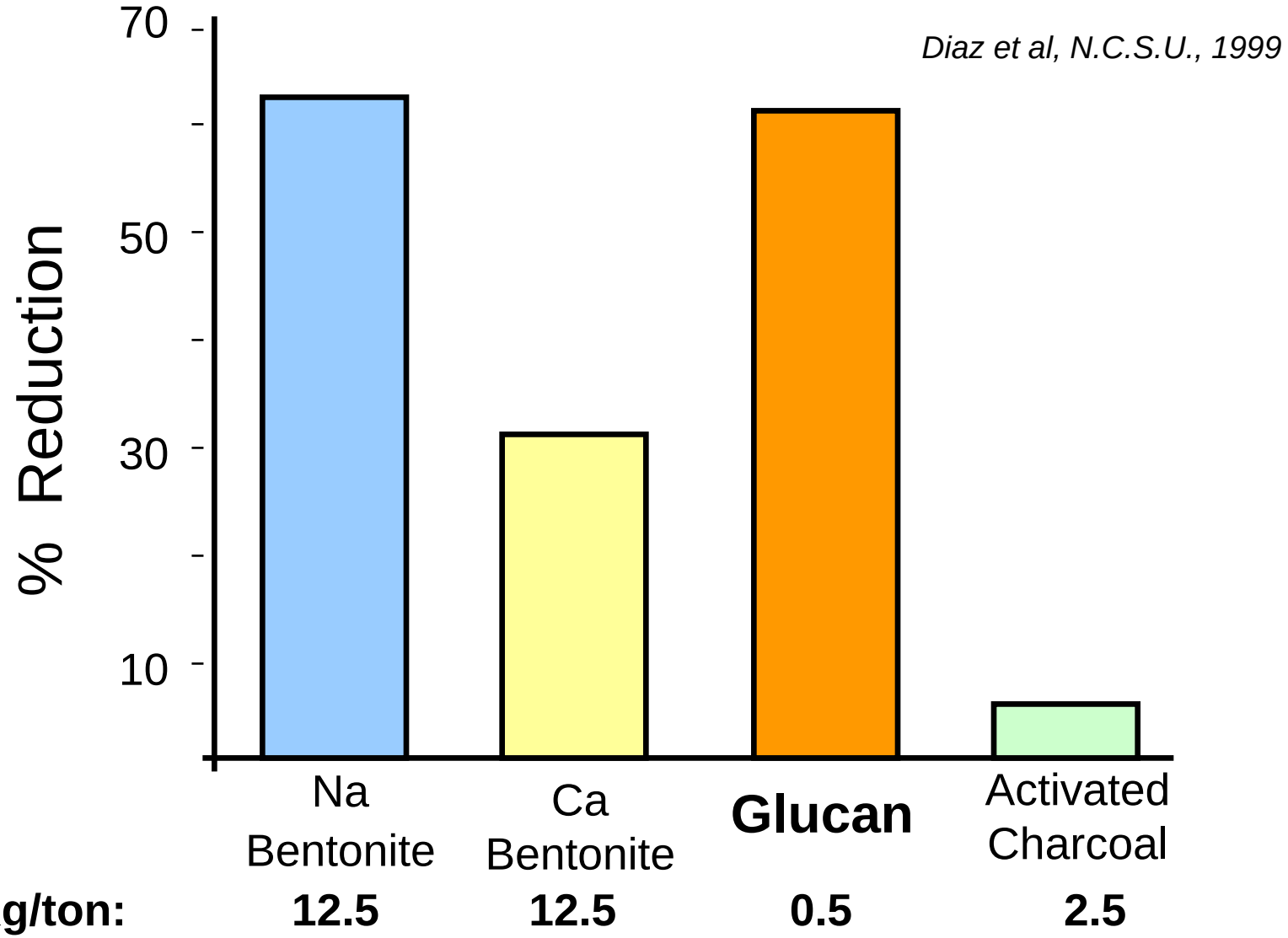
Mycosorb® modulated improvement in milk yield and reduction in milk residue of aflatoxin M_1 in dairy cows

G. Devegowda[1] and T. N. K. Murthy[2] and A. Ramesh[2]

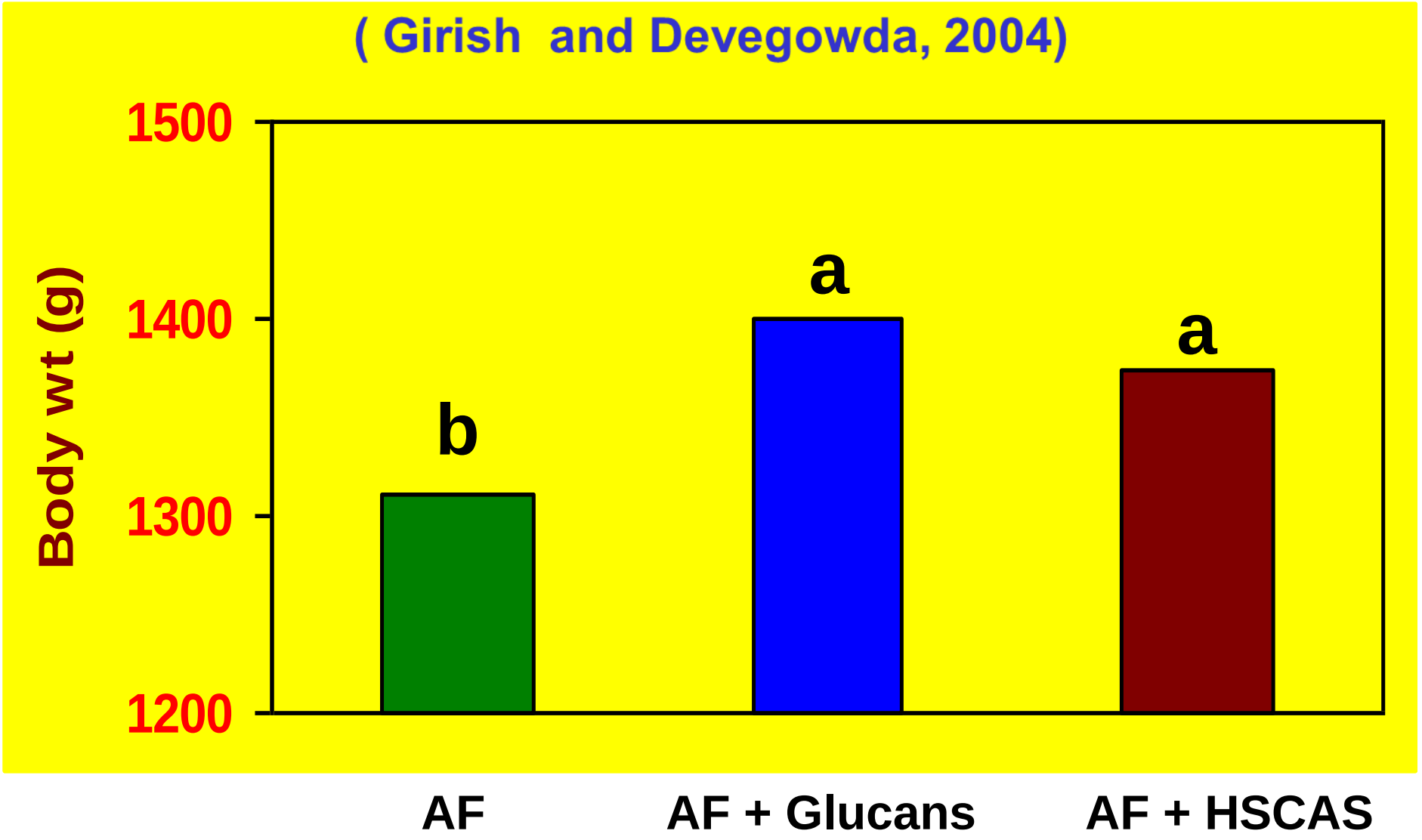
[1]University of Agricultural Sciences, Bangalore, India
[2]Veterinary and Fishery sciences University, Bangalore, India



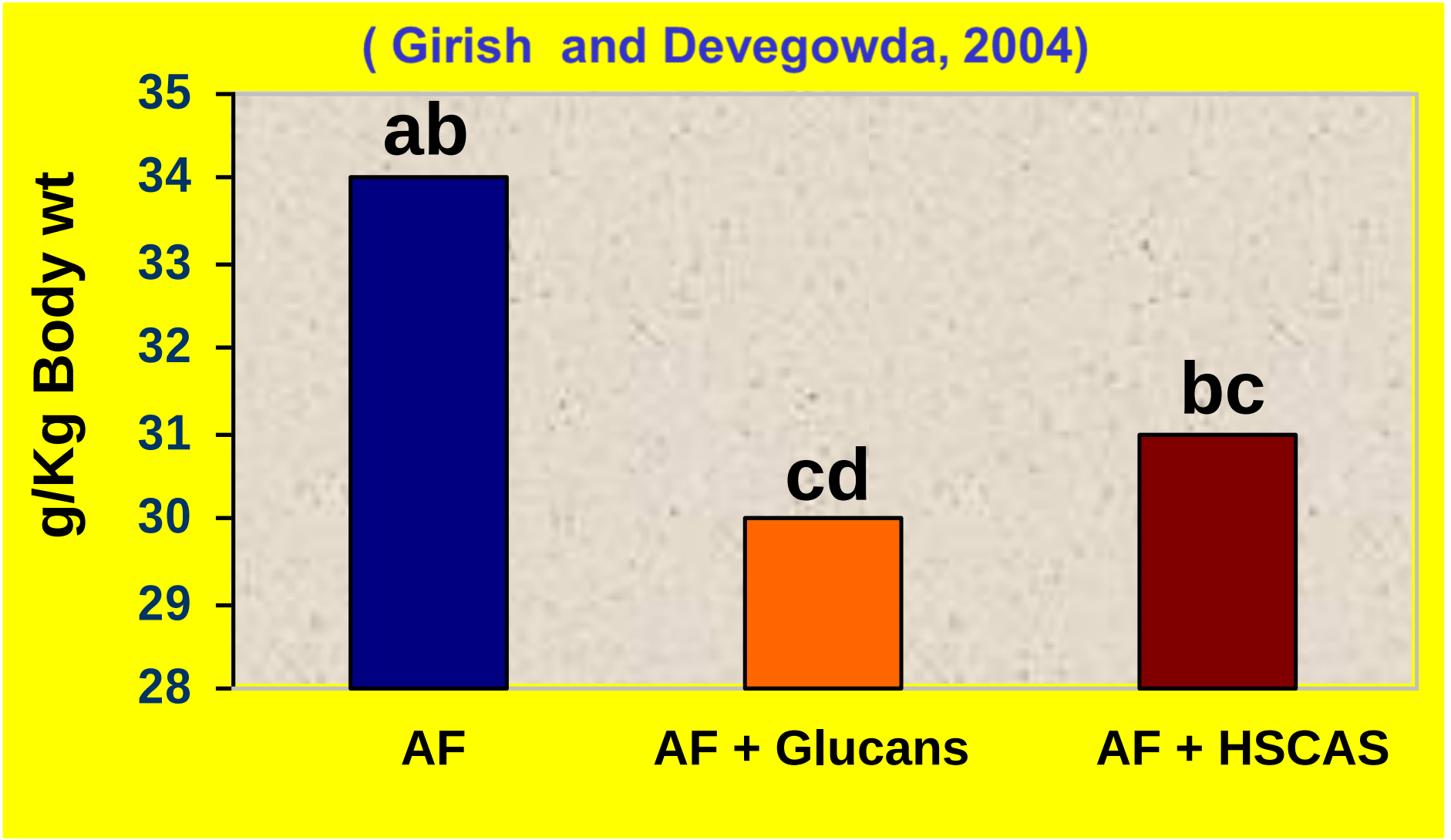
Effects of Binding Agents on Milk Aflatoxin Residues



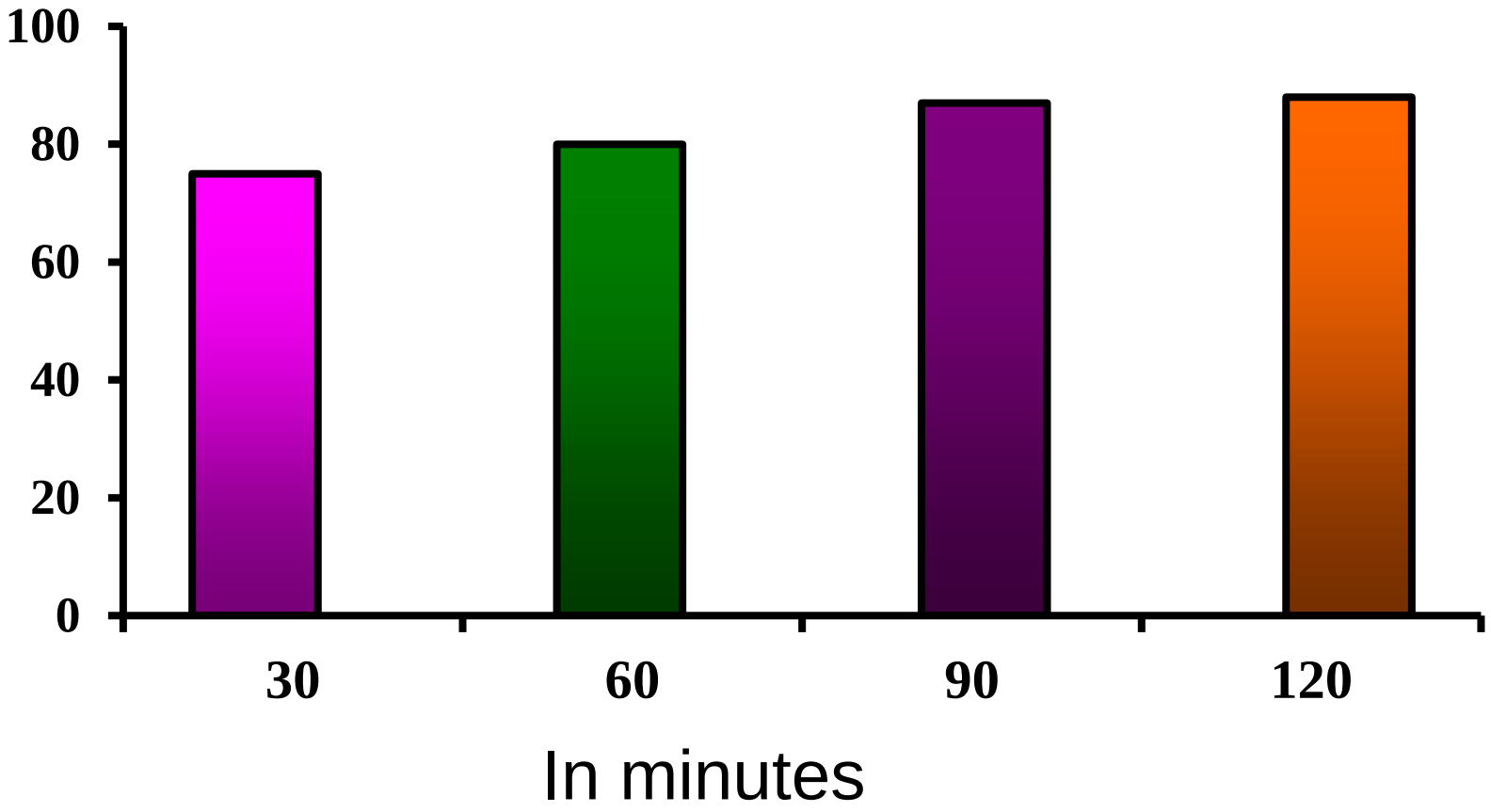
Efficacy of Glucans(1kg) and HSCAS(10kg) to alleviate the Aflatoxin toxicity in broilers on body wt



Efficacy of Glucans(1kg) and HSCAS(10kg) to alleviate the aflatoxin toxicity in broilers on liver wt



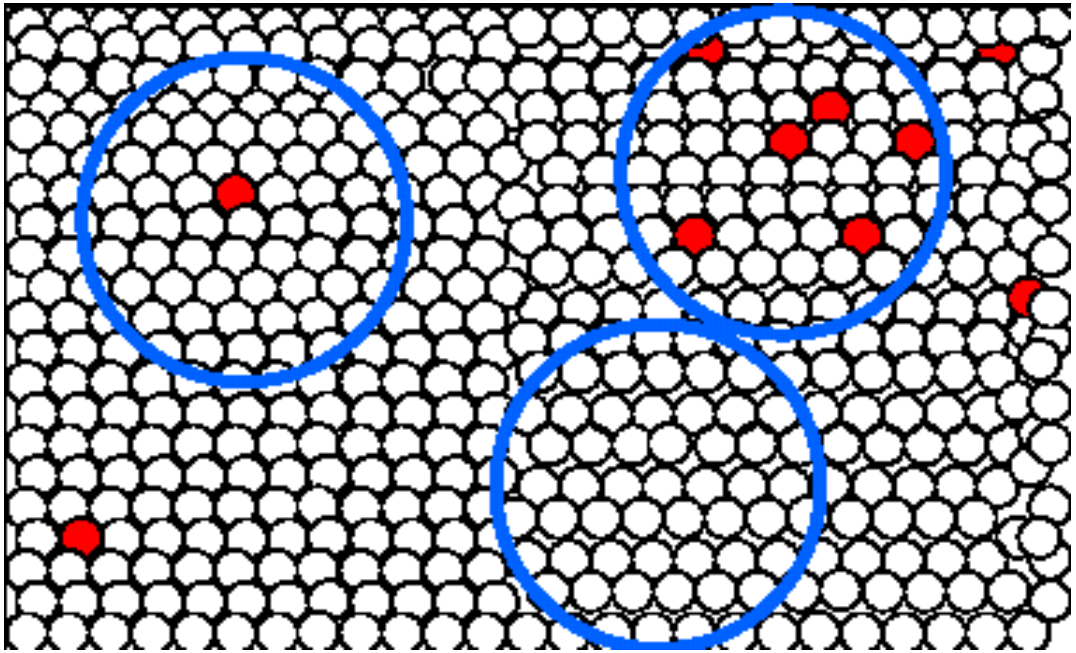
% Aflatoxin adsorbed by Glucans in broilers at different time intervals



Krishnamurthy & Devegowda, 2002

Problems with Sampling for Aflatoxins

- Mycotoxins are not evenly distributed
- Mycotoxins are present in very small amounts (ppb)



Conclusions

"Safe feed Safe Milk and eggs"