

Joint Research Centre (JRC)

Developments in the EU on mycotoxins including risk assessment, regulatory and analytical issues



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<http://www.jrc.ec.europa.eu/>



Conclusions

- **Regulating contaminants in food :**
 - **Challenges for risk assessment, risk management, risk communication.**
 - **Challenges for enforcement**
(representative sampling / availability of reliable routine methods of analysis / sampling uncertainty/ measurement uncertainty)
 - **Research/science as driving force for regulation versus science as limiting factor for regulation**
 - **All possible options to meet the objectives are considered**

Historical perspective

Current Situation

Elements and Developments

Conclusion

Experience is like a lantern behind us, it only illuminates the past rather than the future...Confucius So I'd like to TRY to show you where the shadow might lead.

Aflatoxins₇ Regulated European wide since 1998:

COMMISSION REGULATION (EC) No 1525/98

of 16 July 1998

amending Regulation (EC) No 194/97 of 31 January 1997 setting maximum levels for certain contaminants in foodstuffs

COMMISSION DIRECTIVE 98/53/EC

of 16 July 1998

laying down the sampling methods and the methods of analysis for the official control of the levels for certain contaminants in foodstuffs

And what came with it (even before).....?

Development and Validation of analytical method for mycotoxins:

- **Aflatoxins in Paprika, Peanuts, Figs and Pistachios**
- **AfB1 in baby food**
- **AfM1 in milk**
- **Ochratoxin A in barley and coffee**
- **Ochratoxin A in baby food**
- **Patulin in apple juice and puree.**

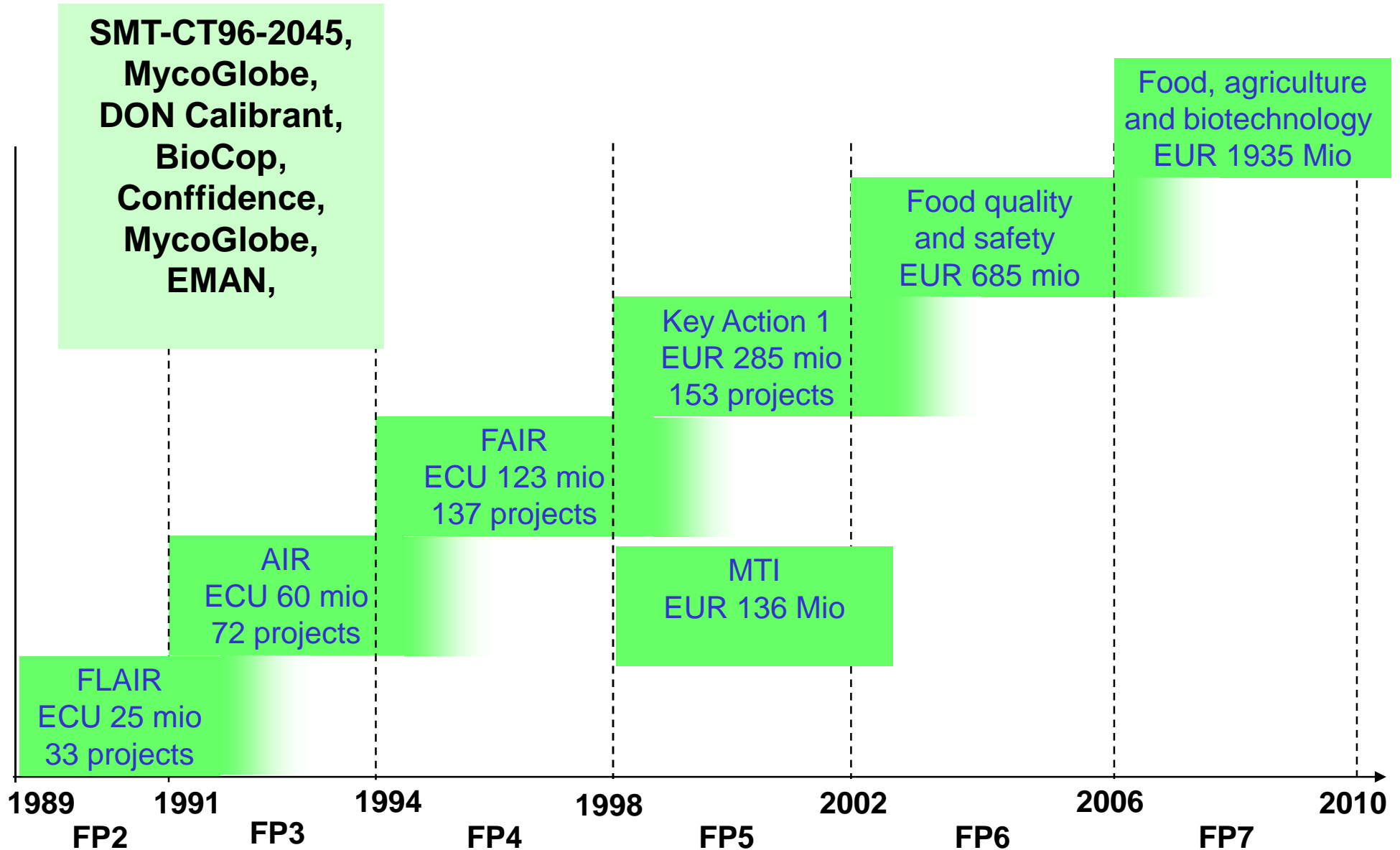
Transformation of validated methods into European Standards:

e.g. EN 14123 / EN 14177 / EN 14132

→ SMT-CT96-2045

e.g. EN 14352

→ SMT4-CT97-2193



Regulation 1881/2006 + amendments:

Aflatoxins:

12 prod. groups w/ var. levels
(0.1-15 µg/kg AfΣ & 25 - 50 ng/kg AfM1)

Fusarium toxins (**DON**, **T-2/HT-2**, **ZON**, **Fumonisin**):

9+?+10+6 prod groups @ var. levels

Ochratoxin A:

13 prod. groups @ var. levels (0.5-80 µg/kg)

Patulin:

5 prod. groups @ var. levels (10-50 µg/kg)

→ **55 + ?** specific limits

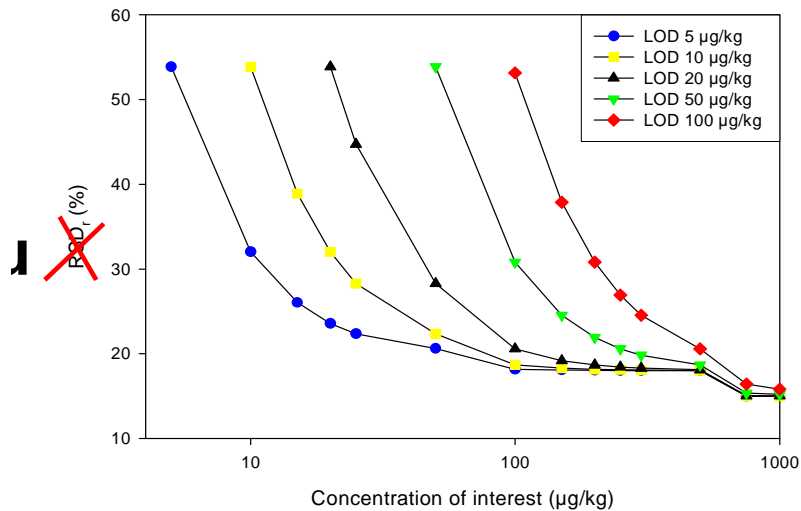
COMMISSION REGULATION (EC) No 401/2006

of 23 February 2006

laying down the methods of sampling and analysis for the official control of the levels of mycotoxins in foodstuffs

Fitness for purpose MU:

$$Uf = \sqrt{(LOD/2)^2 + (a \times C)^2}$$



List with performance criteria of methods:

Performance criteria for zearalenone

Level µg/kg	Zearalenone		
	RSD _r %	RSD _R %	Recovery %
≤ 50	≤ 40	≤ 50	60 to 120
> 50	≤ 25	≤ 40	70 to 120

123

COMMISSION DECISION

of 12 August 2002

implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results

(notified under document number C(2002) 3044)

(Text with EEA relevance)

(2002/657/EC)

2.1.2. Performance of tests

2.1.2.1. Recovery

2.1.2.2. Specificity

2.2. SCREENING METHODS

2.3.1. Common performance criteria and requirements

2.3.2.1. Trueness of quantitative methods

In the case of repeated analyses of a certified reference material, the guideline ranges for the deviation of the experimentally determined recovery corrected mean mass fraction from the certified value are as follows:

2.3.2.2. Precision of quantitative methods

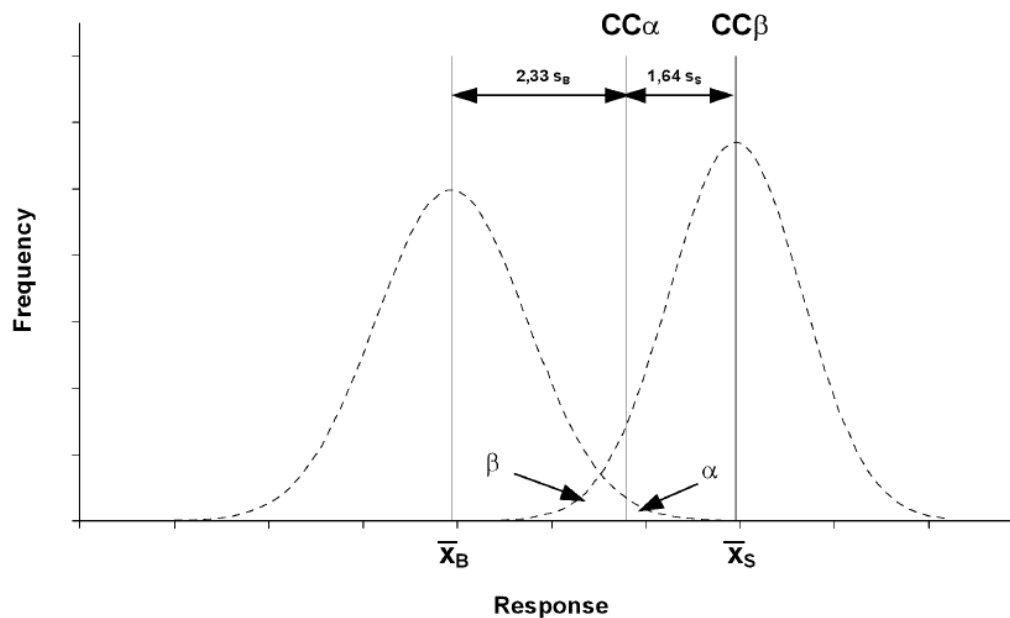
The inter-laboratory coefficient of variation (CV) for the repeated analysis of a reference or fortified material, under reproducibility conditions, shall not exceed the level calculated by the Horwitz Equation. The equation is:

$$CV = 2^{(1 - 0.5 \log C)}$$

The relationship between a range of classes of mass fragment and identification points earned

MS technique	Identification points earned per ion
Low resolution mass spectrometry (LR)	1,0
LR-MS ⁿ precursor ion	1,0
LR-MS ⁿ transition products	1,5
HRMS	2,0
IIR- MS ⁿ precursor ion	2,0
HR-MS ⁿ transition products	2,5

Substances for which no permitted limit has been established



New limits:

Ergot alkaloids!!!

Enniatinins???

Other mycotoxin/matrix combinations???

New group-parameters???

New “legislative guidance” if needed:

However interpretation of results and method performance requirements are regulated: whats next???

- In many parts of daily life: Tendency to consult higher instances to solve problems (e.g. Legislation, high-court).
- Legislation must kept “general” to also consider future cases, while being specific enough to regulate only what shall be regulated.
- This requires either a general understanding (interpretation texts) by all involved facing effects of legislation or the legislative designation of authorities bodies responsible for the scientific interpretation.
- However, as performance parameters are laboratory specific, the interpretation of results will be a case to case decision and requires sufficient understanding.

My conclusion: **Harmonised Training² (with prove of effectiveness)!!!**

BioCop: <http://www.biocop.org/>

New technologies to screen multiple chemical contaminants in food (IP)

MoniQA: <http://www.moniqa.org/>

Monitoring and Quality Assurance in the Food Supply Chain

Confidence: <http://www.confidence.eu/>

Contaminants in food and feed: inexpensive detection for control and exposure

MycoRed: <http://www.mycored.eu/>

Novel integrated strategies for worldwide mycotoxin reduction in food and feed chains

Several Approaches:

Research Projects:

- http://bookshop.europa.eu/is-bin/INTERSHOP.enfinity/WFS/EU-Bookshop-Site/en_GB/-EUR/ViewPublication-Start?PublicationKey=KINA23184

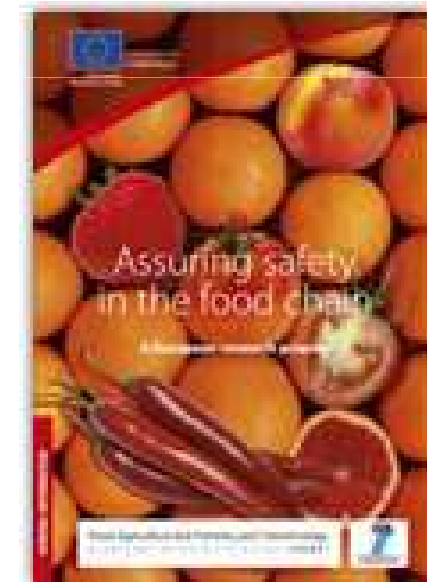
National Activities:

e.g. BfR,
FSA, others....



The screenshot shows the Food Standards Agency website search results for the term "mycotoxin". The page features the FSA logo, a navigation menu on the left, and search results on the right. The search results show 10 items out of 362, with a search time of 0.04 seconds. The results include links to various articles and reports related to mycotoxin risk management and surveys.

Category	Search Results
Home	Results 1 - 10 of about 362 for mycotoxin . Search took 0.04 seconds.
News Centre	
Nutrition	Food Standards Agency - Managing mycotoxin risk
Safety and Hygiene	... Managing mycotoxin risk. The Agency has developed two codes of practice to reduce fusarium and ochratoxin A mycotoxins in cereals. ... www.food.gov.uk/foodindustry/farmingfood/fusarium/
Labelling and Packaging	
GM and Novel Foods	
Consultations	Food Standards Agency - Mycotoxin codes of practice published
Food industries	... Mycotoxin codes of practice published. Tuesday 27 February 2007. The Food Standards Agency has developed codes of practice to help ... www.food.gov.uk/news/newsarchive/2007/feb/mycocop
Enforcement	
Science and Research	
About us	Food Standards Agency - Mycotoxin Survey
Scotland	
Northern Ireland	... Mycotoxin Survey. Four-Year Surveillance Programme for Mycotoxins in Food First year – survey of mycotoxins in a range of cereal-based products. Area of work. ... www.food.gov.uk/science/surveillance/surveymulti200405/chemcontammulti/myco
Wales	
Cymraeg	
RSS	What is RSS? Food Standards Agency - Mycotoxins: answers for food business ...
	... 5. What do I have to do if I want to import foodstuffs susceptible to mycotoxin contamination? ... 15. Can Fusarium mycotoxin contamination of crops be reduced? ... www.food.gov.uk/safereating/chemsafe/mycotoxins/questions/operators/




EFSA – European Food Safety Authority:

Established 2002 (located in Parma, IT)

- to assess and communicate on all risks associated with the food chain.
- advice to risk management and policy-making processes.
- works in panels with experts (CONTAM panel for mycotoxins)

Overview of CONTAM Panel's risk assessments related to mycotoxins in food and feed

	RA for EU-Consumers from contaminants in FOOD	RA for animals from contaminants in FEED and impact on human health
No of Outputs	4	5
Topics/deadline	<p>Ochratoxin A in food (opinion: April 2006)</p> <p>Aflatoxins in three tree nuts (possible changes of maximum levels for pistachios, hazelnuts and almonds based on CODEX decision) (opinion: Jan. 2007)</p> <p>Aflatoxins in tree nuts other than pistachios, hazelnuts, and almonds (statement: June 2009)</p> <p>Ochratoxin A in food (recent scientific information on the toxicity of OTA) (statement: May 2010)</p>	<p>Aflatoxin B1 as undesirable substance in animal feed (opinion: Feb. 2004)</p> <p>Deoxynivalenol as undesirable substance in animal feed (opinion: June 2004)</p> <p>Zearalenone as undesirable substance in animal feed (opinion: July 2004)</p> <p>Ochratoxin A as undesirable substance in animal feed (opinion: Sept. 2004)</p> <p>Fumonisin as undesirable substance in animal feed (opinion: June 2005)</p>

Risk management follow up based on CONTAM Panels outputs



EXAMPLE – **Mycotoxins** as undesirable substances in animal feed

Changes in / recommendations to EU legislation for undesirable substances in animal feed - (Com. Directive 2002/32/EC):

Aflatoxin B1:	EU legislation. No need to change current maximum level (ML) in Com. Directive 2002/32/EC
Deoxynivalenol	No EU legislation. Recommended guidance values e.g. for cereals, maize products and compound feed (Com. Recommendation 2006/576/EC)
Zearalenone	No EU legislation. Recommended guidance values e.g. for cereals, maize products and compound feed (Com. Recommendation 2006/576/EC)
Ochratoxin A	No EU legislation. Recommended guidance values e.g. for cereals, cereal products and compound feed (Com. Recommendation 2006/576/EC)
Fumonisin	No EU legislation. Recommended guidance values e.g. for maize, maize products, and compound feed (Com. Recommendation 2006/576/EC)



Mycotoxin requests to be expected from the European Commission in 2010



FOOD

- T-2/HT-2 toxin,
- Zearalenone in breakfast cereals,
- *Alternaria* toxins,
- Stigmatocystin,
- Moniliformin,
- Diacetoxyscirpenol,
- Phomopsis A.

Mycotoxin requests to be expected from the European Commission in 2010



FEED

- T-2 and HT-2 toxin,
- Nivalenol,
- *Alternaria* toxins,
- Stigmatocystin,
- Moniliformin,
- Diacetoxyscirpenol,
- Phomopsis A.

The analytical tools used must be

“fit-for-purpose”

This means that the “tool” must...

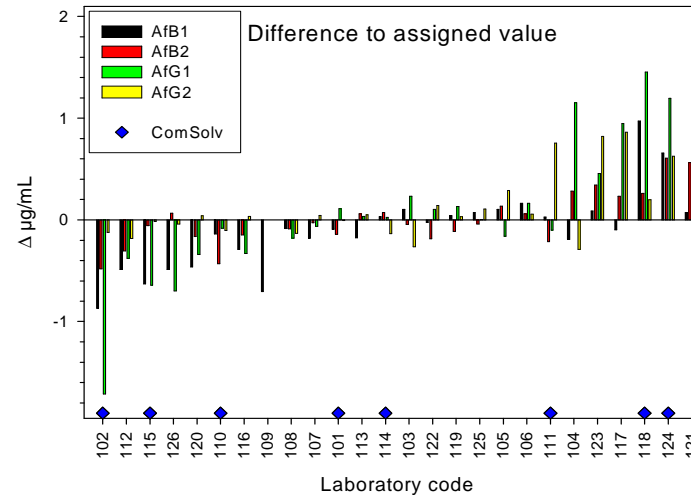
...as such give sufficiently reliable results

...be under control by the operator

...must be verified by blind experiments

To *evaluate* and help where necessary to *improve* the *proficiency* of laboratories to determine correctly the target analyte

To organise *training courses* for National Reference Laboratories (NRL)

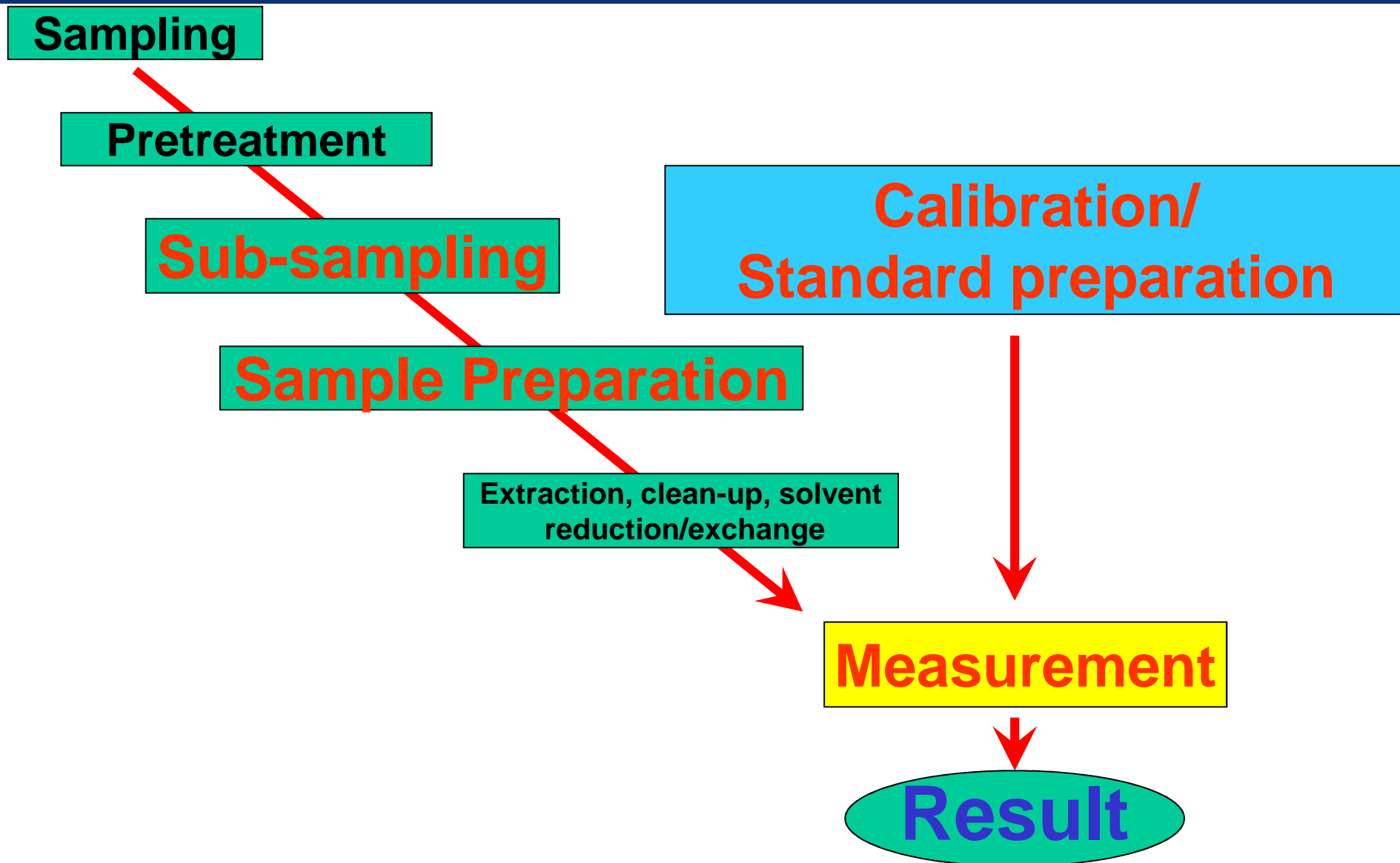


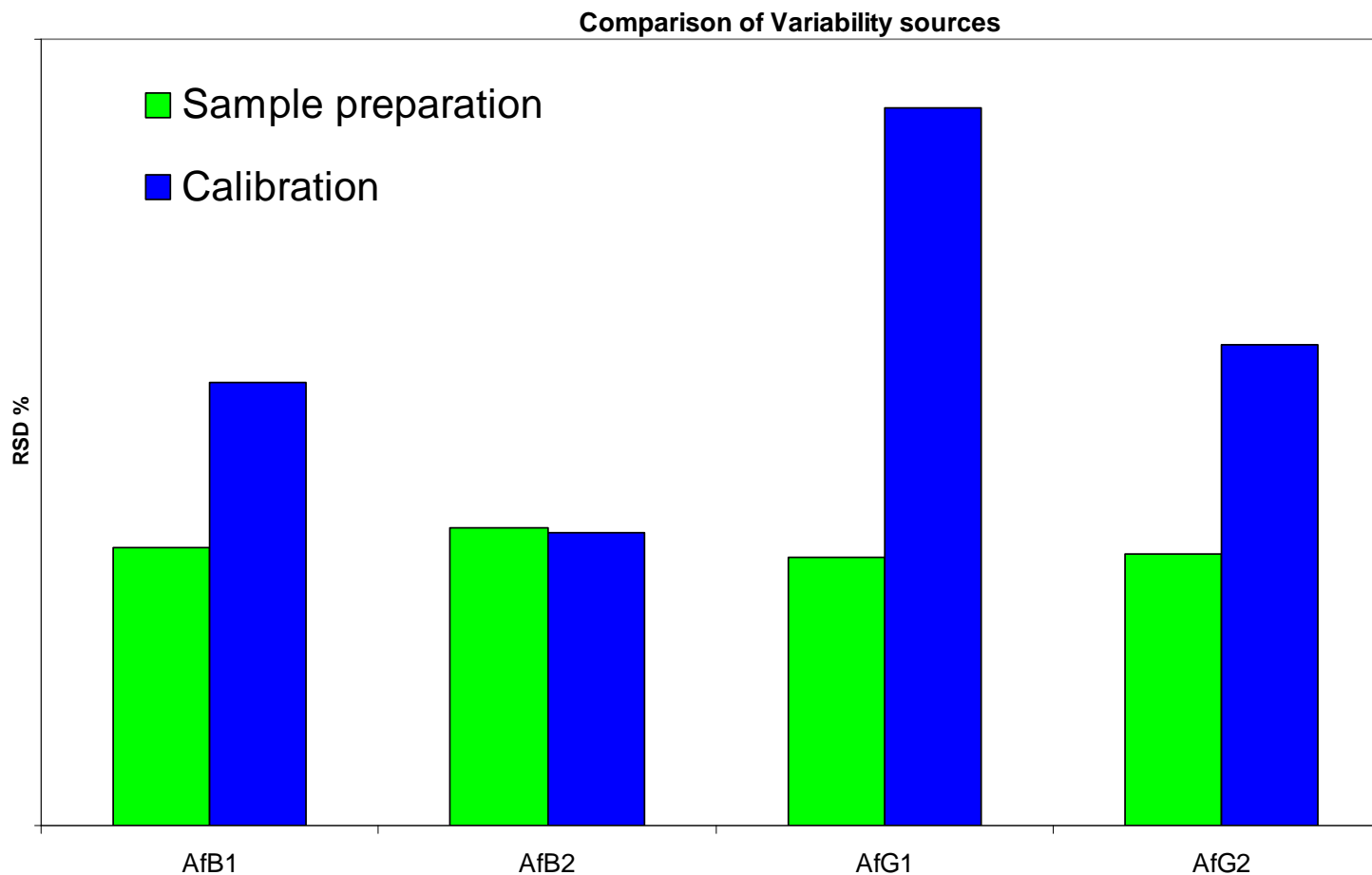
Source identification of errors in the determination of mycotoxins:

- On an individual case (classical PT approach)
- On overall performance with root-cause identification

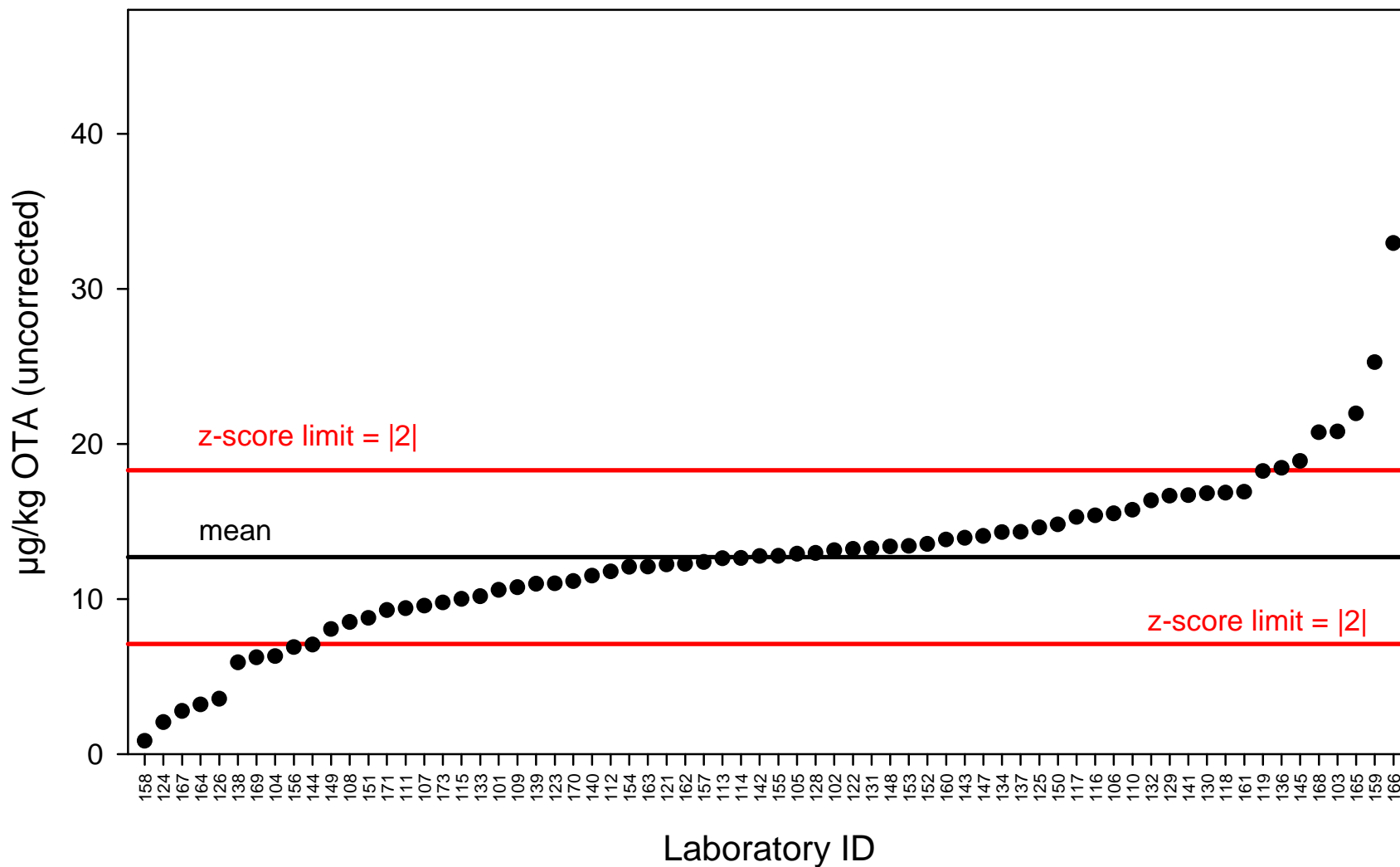
Interpretation of analytical results:

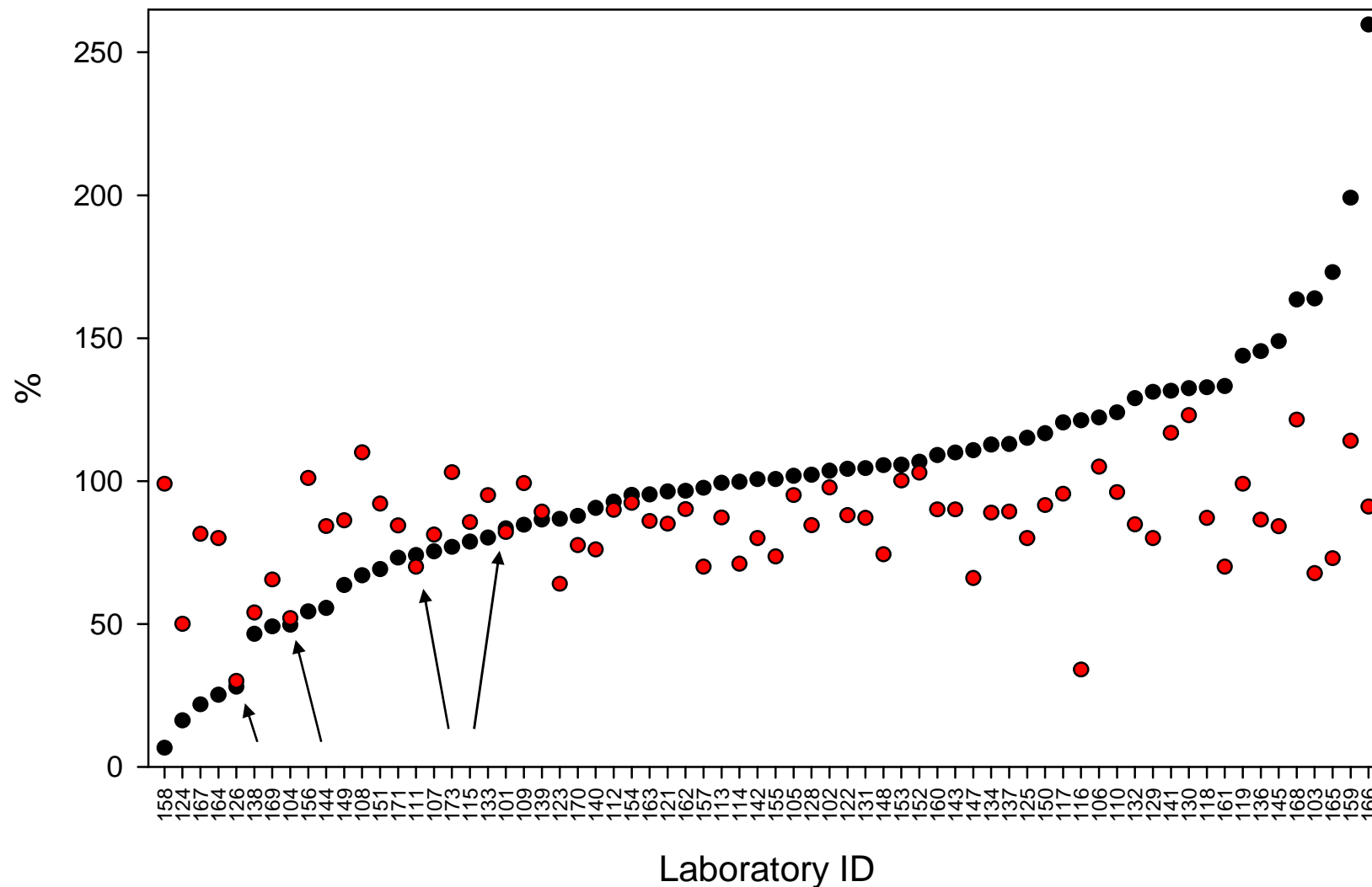
- Proper estimation of measurement uncertainty
- Proper estimation of method performance

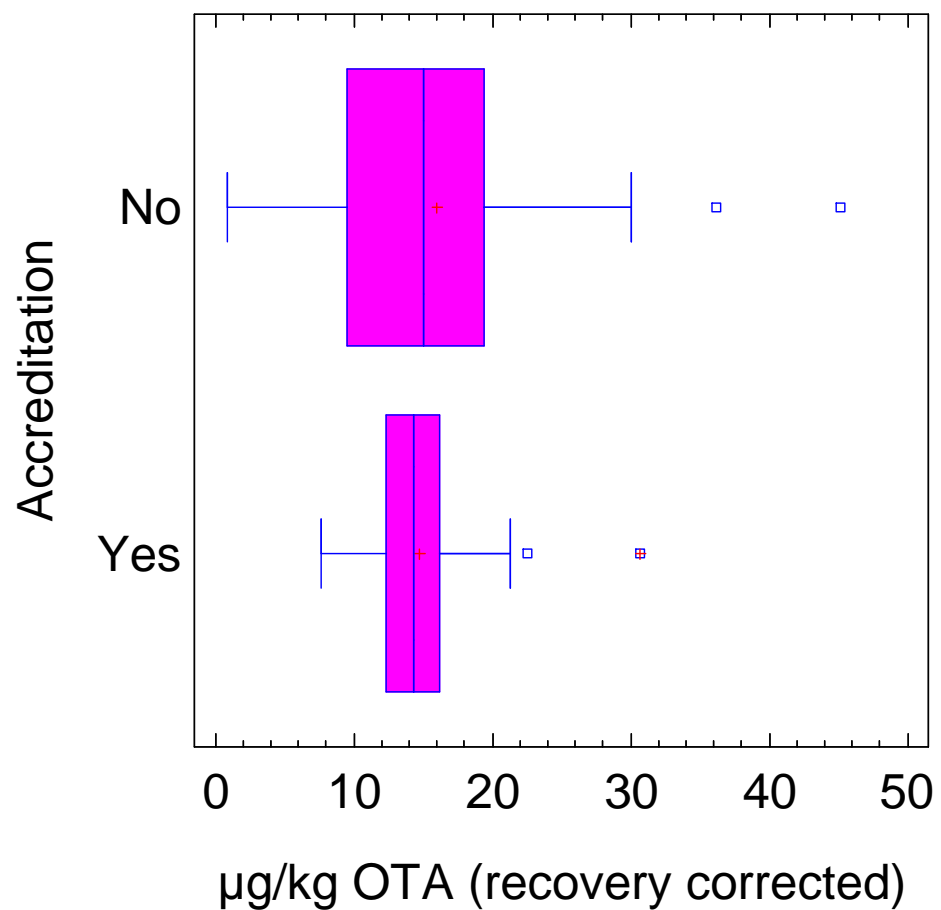


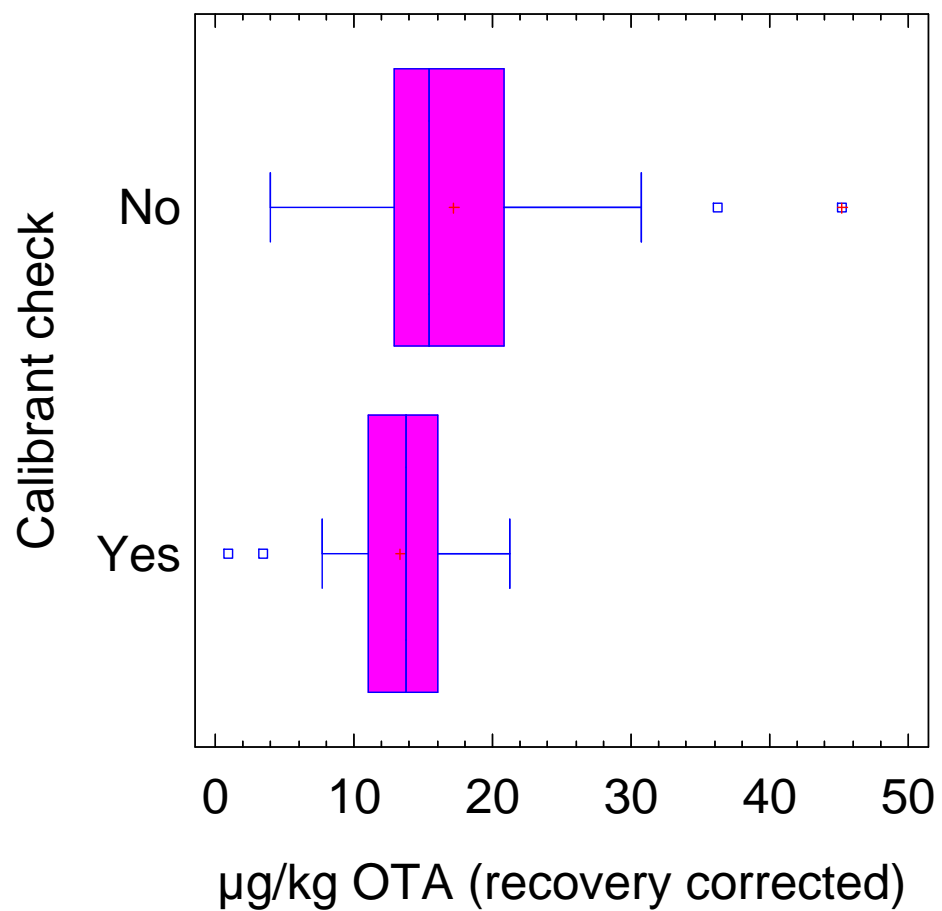


Source: TUBITAK Collaborative Trial Data by Dr. Hamide Senyuva for the extension of EN 14123



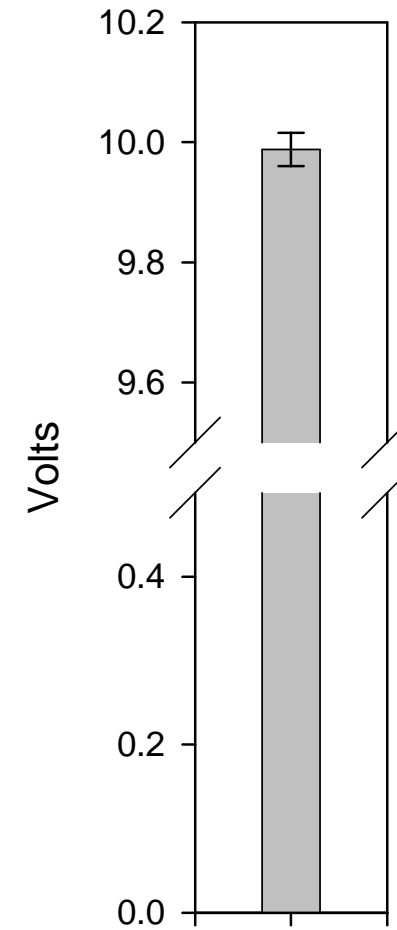






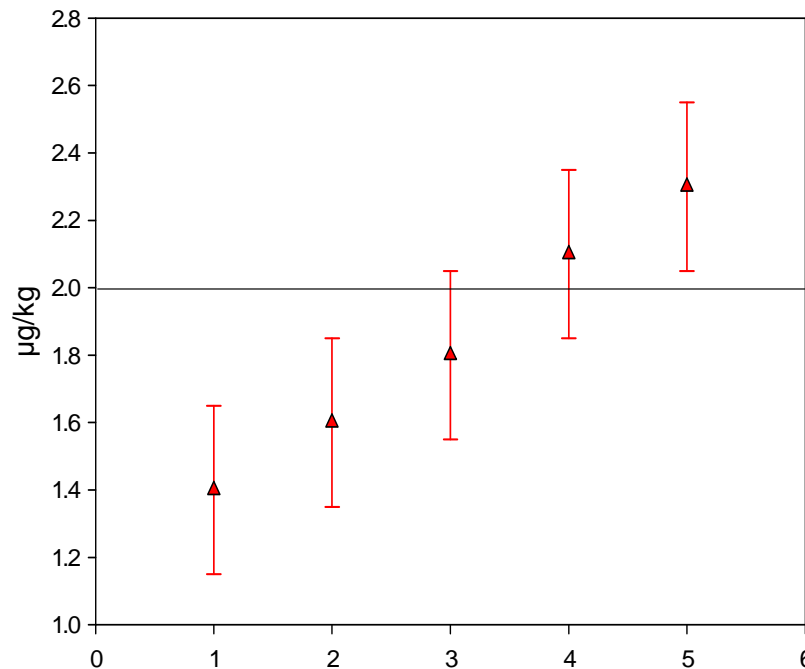
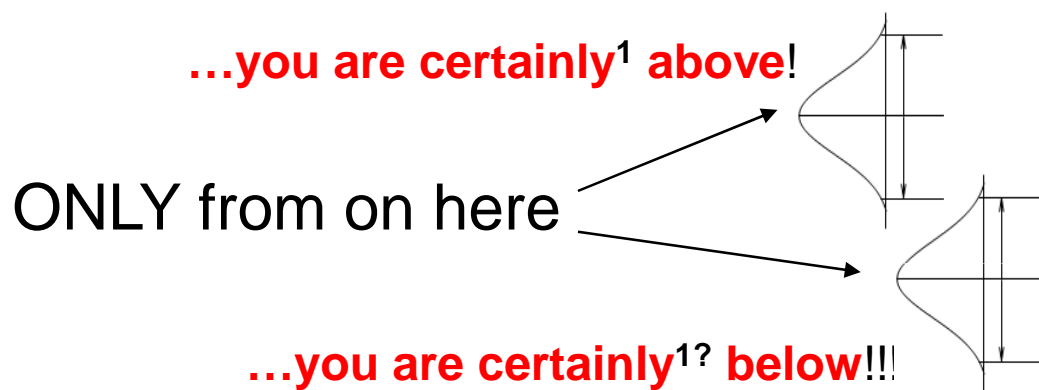


Picture: <http://www.sprut.de/index1.htm>

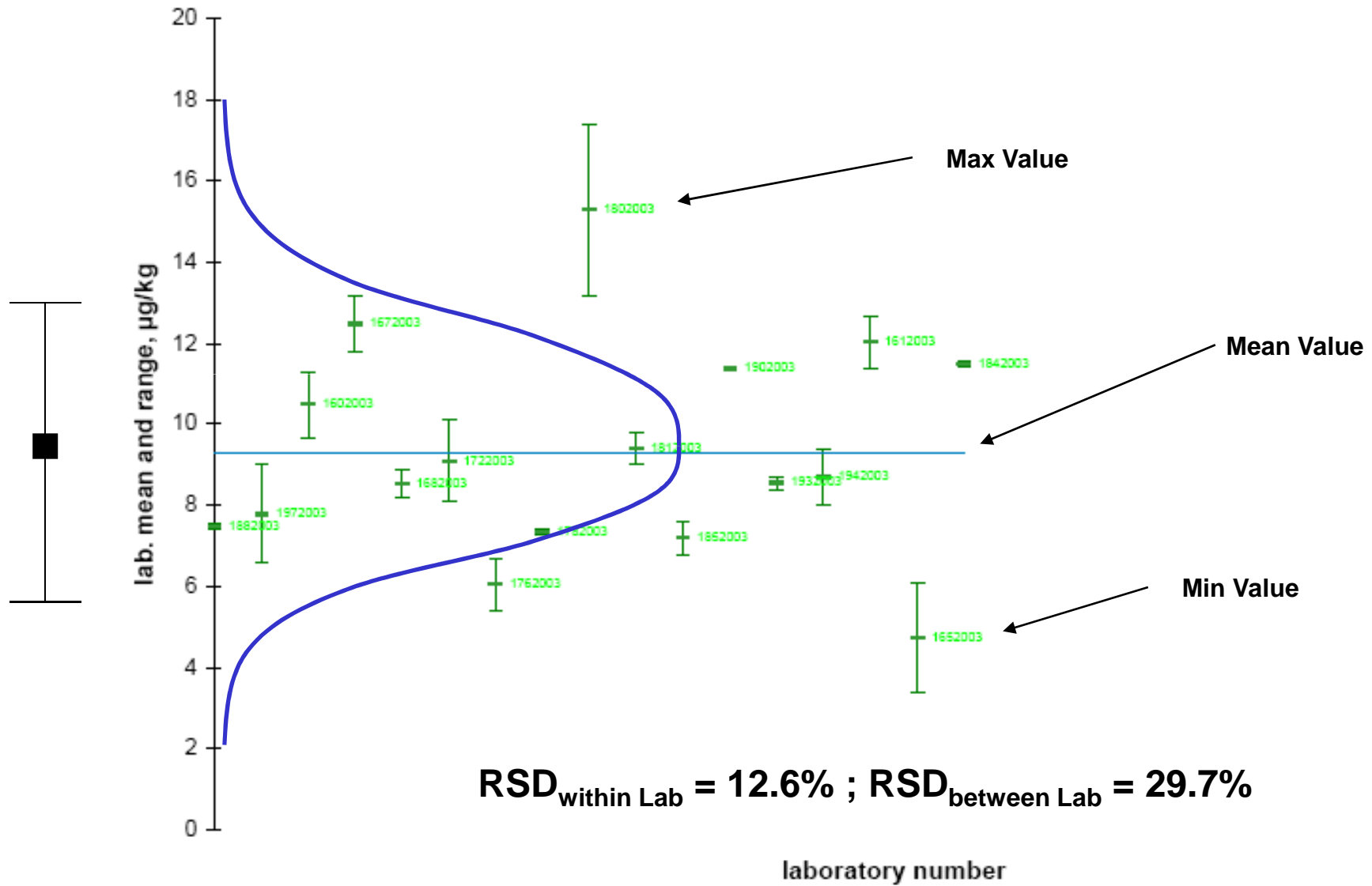


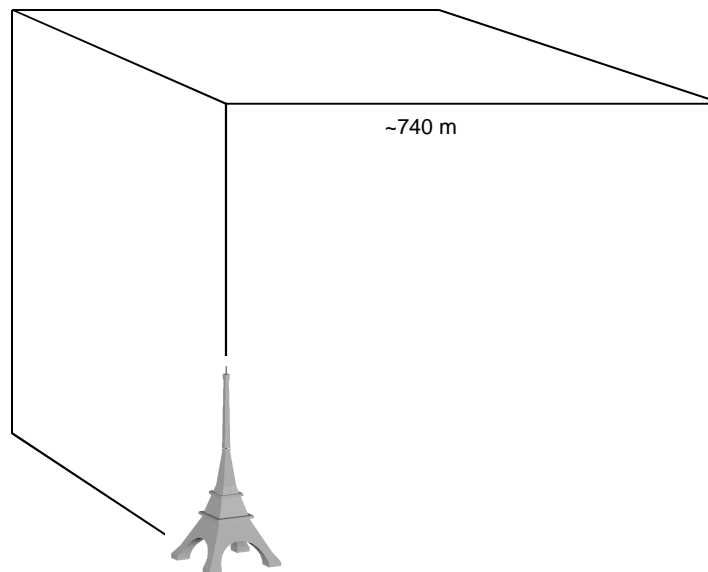
B.6. Acceptance of a lot or subplot

- acceptance if the laboratory sample conforms to the maximum limit, taking into account the correction for recovery and measurement uncertainty;
- rejection if the laboratory sample exceeds the maximum limit beyond reasonable doubt taking into account the correction for recovery and measurement uncertainty.



¹ “beyond reasonable doubt”

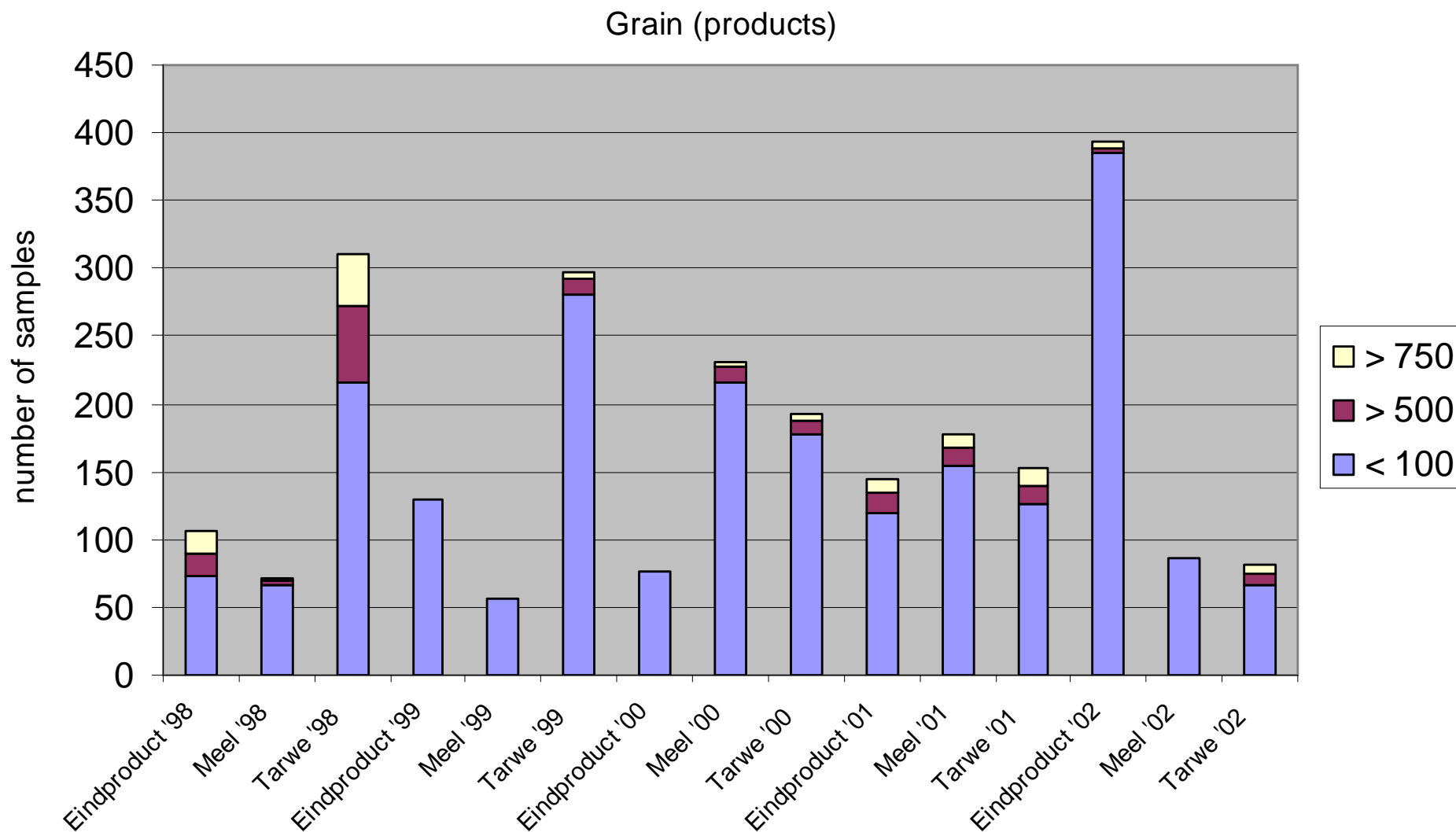




Coceral: ~286,000,000 MT

World Peanut Production: ~34,000,000 MT

Source: Martien Spanjer



Lateral flow devices

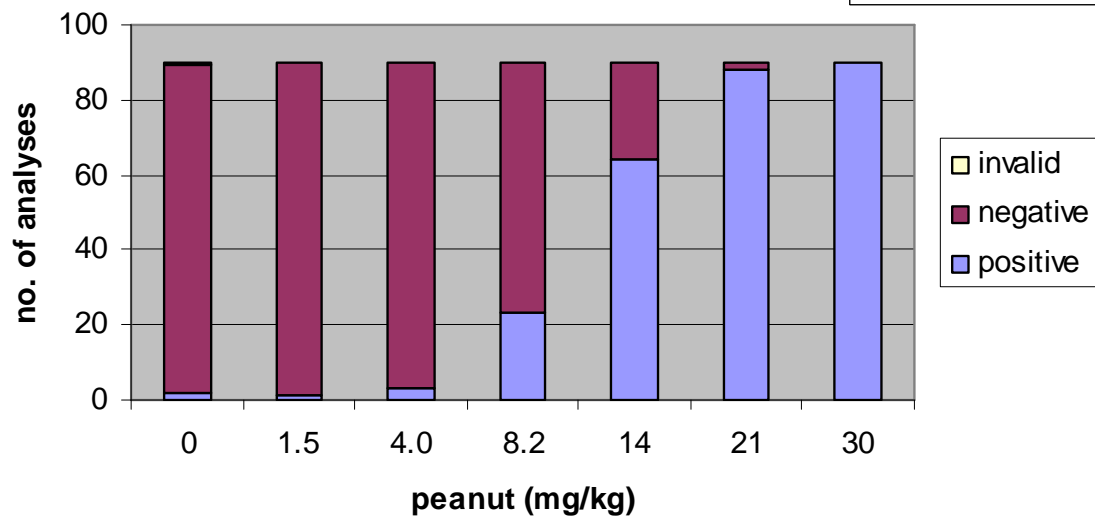
Dip-stick tests

Membrane ELISA

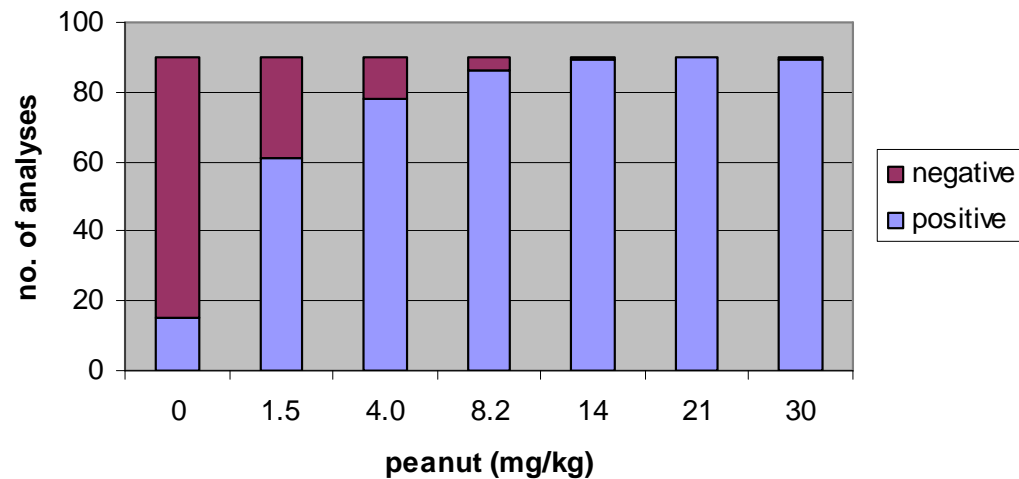


Result	Control band	Test band	Incubation time
negative	++	--	5 – 15 min
20 ppb	++	+ (++)	15 min
10 ppb	++	+ (++)	15 min
4 ppb	++	+	15 min

Test kit A



Test kit B



Decision 657 → general requirements for screening methods

In the case of mycotoxins:

**LC/MS,
ELISA (incl. Dip-stick),
Fluorescence Polarisation,
Other spectroscopic methods...**

→ Need for more “specific” guidelines for these methods

- **More sensitive?**
- **Faster?**
- **More reliable?**
- **Cheaper?**
- **More analytes?**
- **Earlier control?**
- **Mutually accepted procedures?**
- **Common agreement on Interpretation of Results?**

Thank you!!!