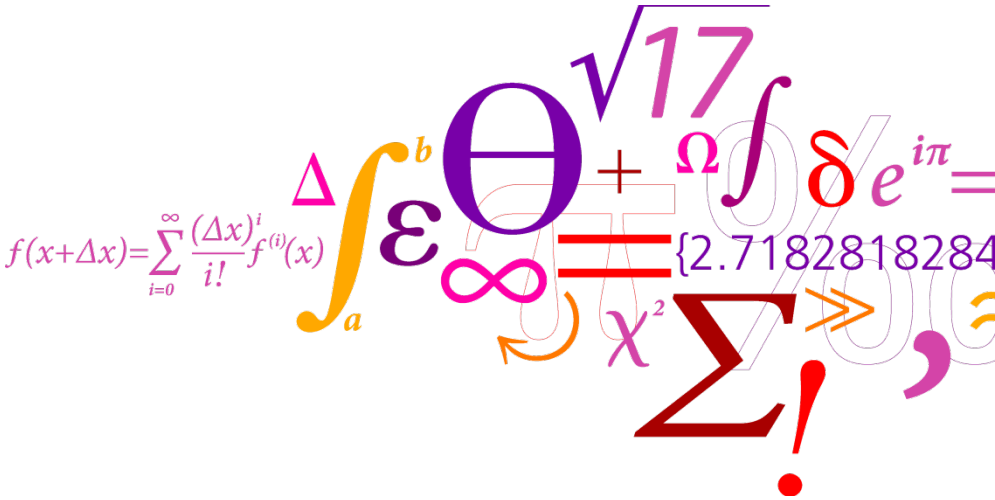
Results from EUPT-CF12
Incurred pesticides in hay flour

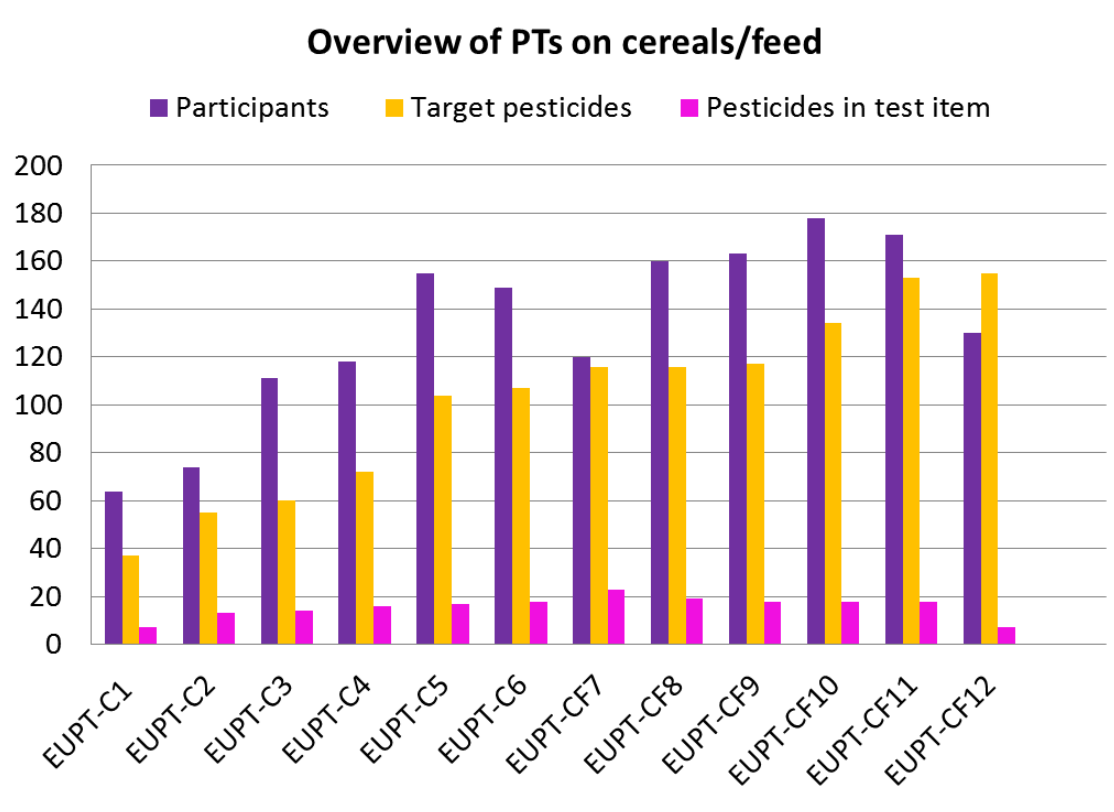
Mette Erecius Poulsen and Susan Strange Herrmann

Webinar, 9 July 2018



DTU Food
National Food Institute

PTs on cereals / feed



	2018 **EUPT-CF12**
Test material	Hay flour
Participants	**130 (120)**
Compulsory target pesticides	**155**
Voluntary target pesticides	**23**
Incurred pesticides	**7**
Spiked pesticides	**0**
Total no. of pesticides	**7**

Advisory Group

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EURL EUROPEAN UNION REFERENCE LABORATORY PESTICIDE RESIDUES IN CEREALS & FEEDING STUFF

DTU

29 October 2017

EUPT-CF12 - Announcement

The European Union Reference Laboratory for Pesticides Residues in Cereals and Feedingstuff (EURL-CF) announces its twelfth European Proficiency Test on for pesticides in cereals/feeds requiring multi residue methods (MRMs), EUPT-CF12.

The aim of the test is to provide laboratories with an assessment of their analytical performance and the reliability of their data - in comparison to other laboratories. This will hopefully result in positive changes and quality improvements at each of the laboratories.

Test Items

The Test Material will be **hay flour with incurred pesticides**. The participants will receive approximately 20 g of a treated and approximately 50 g of a blank test material.

Activity	Dates
Announcement Calendar Target Pesticide List	November 2017
EUPT-Registration Website open	5 December 2017
Deadline for registration	12 January 2018
Specific Protocol published	15 January 2018
Website for selecting pesticide scope open	15 January 2018
Website for selecting pesticide scope closed	26 January 2018
Distribution of Test items	29 January 2018
Deadline for receipt and acceptance of Test Materials	within 24 hr on receipt
Deadline for Result Submission	1 March 2018 at 24.00 CET
Deadline for submission of additional method information for false negative results	5 March 2018 at 24.00 CET
Preliminary Report (only compilation of results) published	30 April 2018
Final Report published	December 2018

Target list – 2 new pesticides

- Formetanate
- Prosulfocarb

Voluntary pesticides from working document

- Ametoctradin
- Benalaxyl including other mixtures ….(sum of isomers)
- Benzovindiflupyr
- Chlorfluazurone
- Clomazone
- Cyazofamid
- Cyflufenamid
- Etoxazole
- Fenpyrazamine
- Isopyrazam
- Novaluron
- Penflufen
- Penthiopyrad
- Proquinazid
- Pyridalil
- Pyriofenone
- Spinetoram
- Spirotetramat
- Spirotetramat metabolite BYI08330-enol
- Spirotetramat metabolite BYI08330-ketohydroxy
- Spirotetramat metabolite BYI08330-monohydroxy
- Spirotetramat metabolite BYI08330 enol-glucoside
- Sulfoxaflor

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EUPT-CF12 Pesticide Target List

(last updated: 30.11.2017)

Pestide no.	Pesticides	MRRL
	Compulsory Compounds (will be considered in Category A/B class	
1	2-phenylphenol	0.05
2	Acephate	0.05
3	Acetamiprid	0.05
4	Acrinathrin	0.05
5	Aldrin	0.05
6	Azinphos-methyl	0.05
7	Azoxystrobin	0.05
8	Bifenthrin	0.05
9	Biphenyl	0.05
10	Bixafen	0.05
11	Boscalid	0.05
12	Bromuconazole	0.05

Participation

Country	# labs	Country	# labs	Country	# labs
Albania	1	Greece	3	Singapore	1
Argentina	2	Hungary	4	Slovakia	1
Australia	1	Iceland	1	Spain	20
Austria	1	India	1	Sri Lanka	1
Belgium	3	Ireland	1	Sweden	2
Bulgaria	1	Italy	20	Switzerland	1
China	1	Latvia	1	Thailand	1
Croatia	5	Lithuania	1	United Kingdom	2
Cyprus	1	Luxembourg	1		
Czech Republic	2	Netherlands	5		
Denmark	1	Norway	1		
Estonia	1	Poland	12		
Finland	1	Portugal	1		
France	6	Romania	5	**No. of countries**	**38**
Germany	15	Serbia	2	**No. of laboratories**	**130**

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Australia	1	Iceland	1	Spain	20
Austria	1	India	1	Sri Lanka	1
Belgium	3	Ireland	1	Sweden	2
Bulgaria	1	Italy	20	Switzerland	1
China	1	Latvia	1	Thailand	1
Croatia	5	Lithuania	1	United Kingdom	2
Cyprus	1	Luxembourg	1		
Czech Republic	2	Netherlands	5		
Denmark	1	Norway	1		
Estonia	1	Poland	12		
Finland	1	Portugal	1		
France	6	Romania	5	**No. of countries**	**38**
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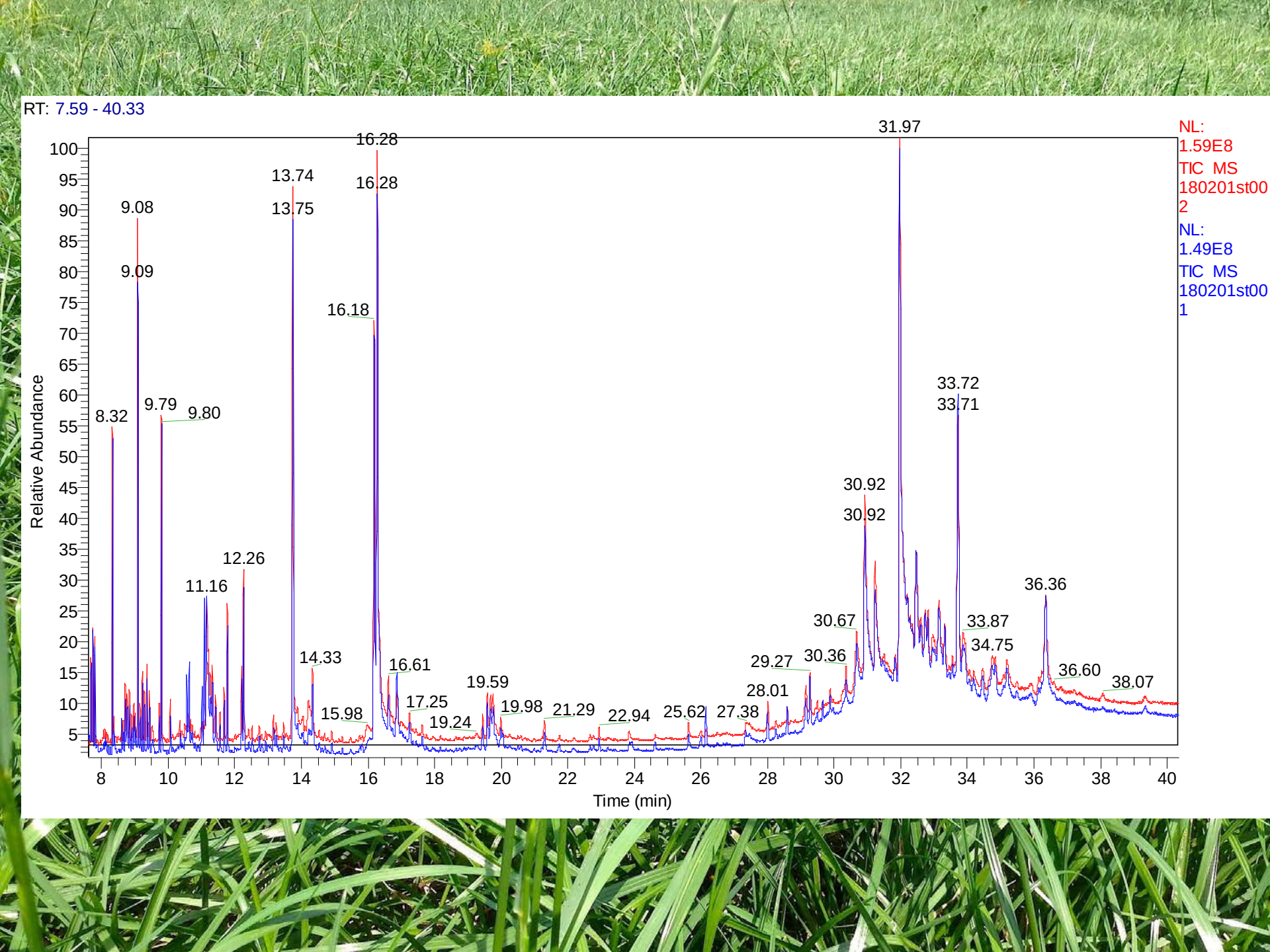
Hay from ryegrass

- Ryegrass was grown in Denmark in 2017
- Field treated by Aarhus University, Research Department Flakkebjerg
- Untreated ryegrass was by mistake combined with dirty plots
  - Purchased ryegrass hay from a organic farmer - but also this contained residues – maybe due to contamination from the mill
  - Purchased organic produced hay from a pet store.


RT: 7.59 - 40.33
NL:
1.59E8
TIC MS
180201st002
NL:
1.49E8
TIC MS
180201st001
Relative Abundance
Time (min)
8.32
9.08
9.09
9.79
9.80
11.16
12.26
13.74
13.75
14.33
15.98
16.18
16.28
16.28
16.61
17.25
19.24
19.59
19.98
21.29
22.94
25.62
27.38
28.01
29.27
30.36
30.67
30.92
30.92
31.97
33.72
33.71
33.87
34.75
36.36
36.60
38.07

Pesticides in Test Material

Pesticide	Included in the Target List	Formulation or standard	g.ai/ha
Azoxystrobin	x	Amisiar	250
Boscalid	x	Viverda	350
Epoxiconazole	x	Viverda	125
Fluxapyroxad	x	Imtrex	125
Metrafenon	x	Flexity	150
Pirimicarb	x	Pirimir G	100
Proquinazid	x	Talius	50
Pyraclostrobin	x	Viverda	150
Tau-Fluvalinate	x	Mavrik	48
Chlormequat		Cycocel extra	1518
Chlorotalonil		Bravo	1000
Fluroxypyr		Starane 333 HL	133
Glyphosat		Roundup Bio	1440
MCPA		Metaxon	750
Trinexapac-ethyl		Modus M	200

Spike procedure

- 20 gram Test Item with pesticide residues
- 50 gram Blank Test Item

- Instruction to use only 1 gram sample and 10 ml water.

Sample shipment

- Samples were distributed on Monday 29 January 2018
  (17 January to non-European countries)
- Most samples were delivered to EU laboratories within 1 day

Homogeniety test

	Mean, mg/kg	S_s^2	c	$S_s^2 < c$
Azoxystrobin	0.991	0.00108	0.0127	Pass
Boscalid	11.9	0.00000	2.387	Pass
Epoxiconazole	2.90	0.02887	0.1177	Pass
Metrafenon	1.030	0.00314	0.0140	Pass
Fluxapyroxad	1.48	0.00055	0.0280	Pass
Proquinazad	0.871	0.00077	0.0093	Pass
Pyraclostrobin	6.25	0.00912	0.5877	Pass
Tau-fluvalinate	0.812	0.00219	0.0086	Pass

Stability test

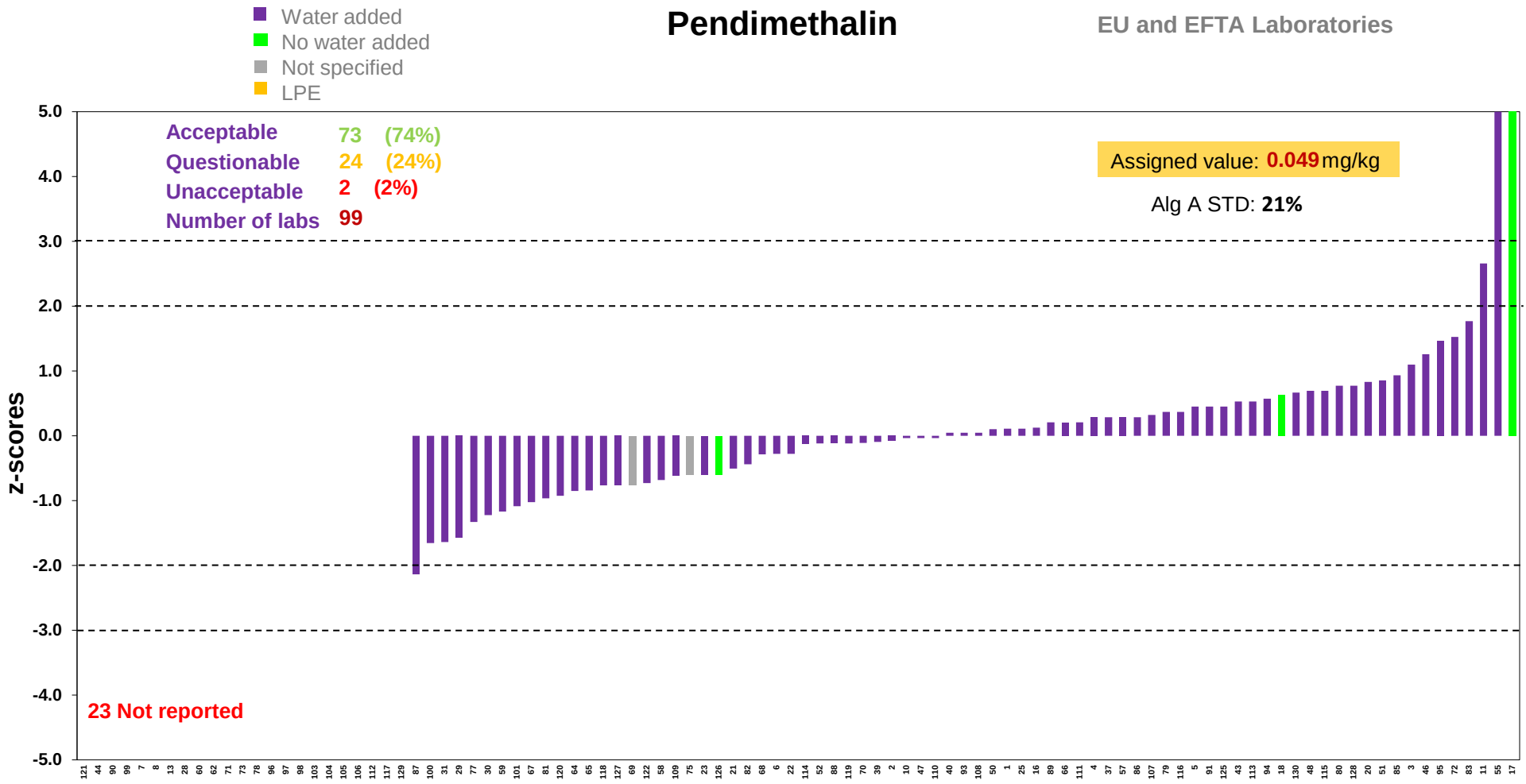
		Storage at -18 degrees			Storage at room temperature		
	Mean, mg/kg	| x1 - yi |	0.3×σ	| x1 - yi | ≤ 0.3×σ	| x1 - yi |	0.3×σ	| x1 - yi | ≤ 0.3×σ
Azoxystrobin	0.777	0.047	0.060	Pass	0.023	0.060	Pass
Boscalid	11.9	0.079	0.870	Pass	0.220	0.870	Pass
Epoxiconazole	2.49	0.125	0.185	Pass	0.065	0.185	Pass
Fluxapyroxad	1.01	0.020	0.101	Pass	0.007	0.101	Pass
Metrafenone	0.843	0.049	0.067	Pass	0.003	0.067	Pass
Proquinazid	0.754	0.044	0.060	Pass	0.023	0.060	Pass
Pyraclostrobin	4.04	0.195	0.444	Pass	0.098	0.444	Pass
Tau-Fluvalinate	0.586	0.006	0.046	Pass	0.045	0.046	Pass

False positive results

Lab code	Pesticide	Concentration mg/kg	Determination technique	RL, mg/kg
17	Cadusafos	0.05	LC-MS/MS QQQ	0.01
28	Lambda-cyhalothrin	13.3	GC-MS/MS (QQQ)	
43	Thiophanate-methyl	0.17	LC-MS/MS QQQ	0.01
60	Biphenyl	0.357	GC-MS/MS (QQQ)	0.02
75	Metolachlor	0.54		0.01
95	Fenpropathrin	0.07	GC-MS/MS (QQQ)	0.05
97	2-phenylphenol	0.185	GC-MS/MS (QQQ)	0.05

Pendimethalin

EU and EFTA Laboratories



Low findings of pesticides

Pestcide	No.	Min.	Max
Pirimicarb	7	0.0058	0.011
Pirimicarb-desmethyl	11	0.0012	0.014
Prosulfocarb	21	0.011	0.028
Prothioconazole-desthio	14	0.0081	0.0215
Tebuconazole	5	0.0054	0.012
Fluopyram	1	0.0014	
Lindane	1	0.0012	
Pirimiphos-methyl	1	0.0013	
Zoxamide	1	0.0248	

False negatives

Labcode	Fluxapyroxad	Metrafenone	Pyraclostrobin	Tau-Fluvalinate	Proquinazid	
5	FN					
44	FN		FN	FN		No scope selection
87					FN	
90	FN	FN				No scope selection
102				FN		

Calculation of assigned values and uncertainty of assigned values

- Algoritm A mean of results from EU and EFTA laboratories
  - Only result from participants that
    - Reported to add water to the samples before extration or
    - used mixture of solvent and water or
    - used LPE
- Outliers
  - Only obvious uncorrect results
    - ***(2 results for azoxystrobin and boscalid - 1000x)***
- Uncertainty
  - $u = 1.25 * (s^* / \sqrt{n})$
  - s^* is robust standard deviation (Alg A standard deviation)
  - N is the number of participants

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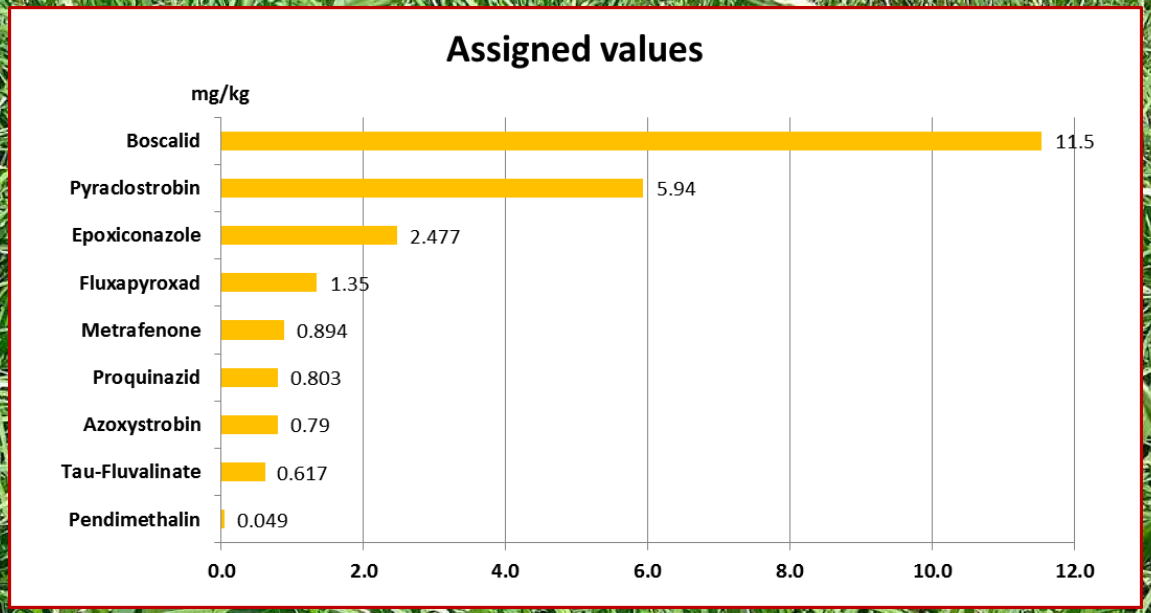
Assigned values

PESTICIDES	only water addition, mg/kg
Azoxystrobin	0.794
Boscalid	11.5
Epoxiconazole	2.48
Fluxapyroxad	1.35
Metrafenone	0.894
Pendimethalin	0.049
Proquinazid	0.803
Pyraclostrobin	5.94
Tau-Fluvalinate	0.617



Reported results and false negatives

Pesticide	No. of reported results	No. of NA	False negatives	% results
Azoxystrobin	104	10	0	91
Boscalid	102	12	0	89
Epoxiconazole	99	15	0	87
Fluxapyroxad	68	46	3	60
Metrafenone	69	45	0	61
Pendimethalin	101	13	0	89
Proquinazid	46	68	1	40
Pyraclostrobin	91	23	1	80
Tau-Fluvalinate	89	25	2	78

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Calculations of z-scores

- For each laboratory/pesticide combination :

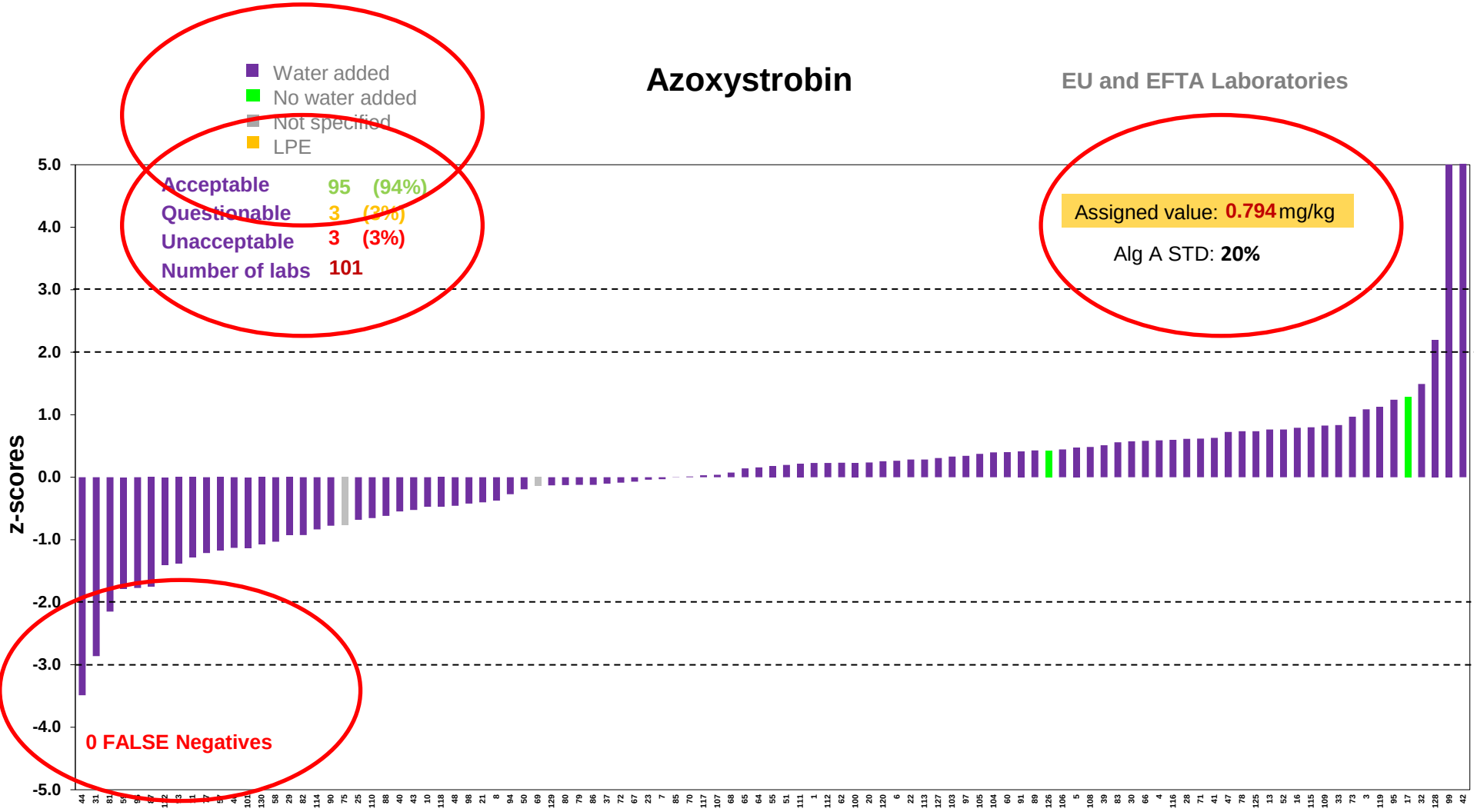
$$z = (x-X) / \sigma$$

  - x is the result reported by the participant or the specific reporting limit of the lab for those labs not having detected the pesticide present in the test material
  - X is the assigned value, Alg A mean without outliers
  - σ is the target standard deviation obtained by multiplying the median by the FFP RSD of 25%

- $|z| \leq 2$ Acceptable
- $2 < |z| < 3$ Questionable
- $|z| \geq 3$ Unacceptable

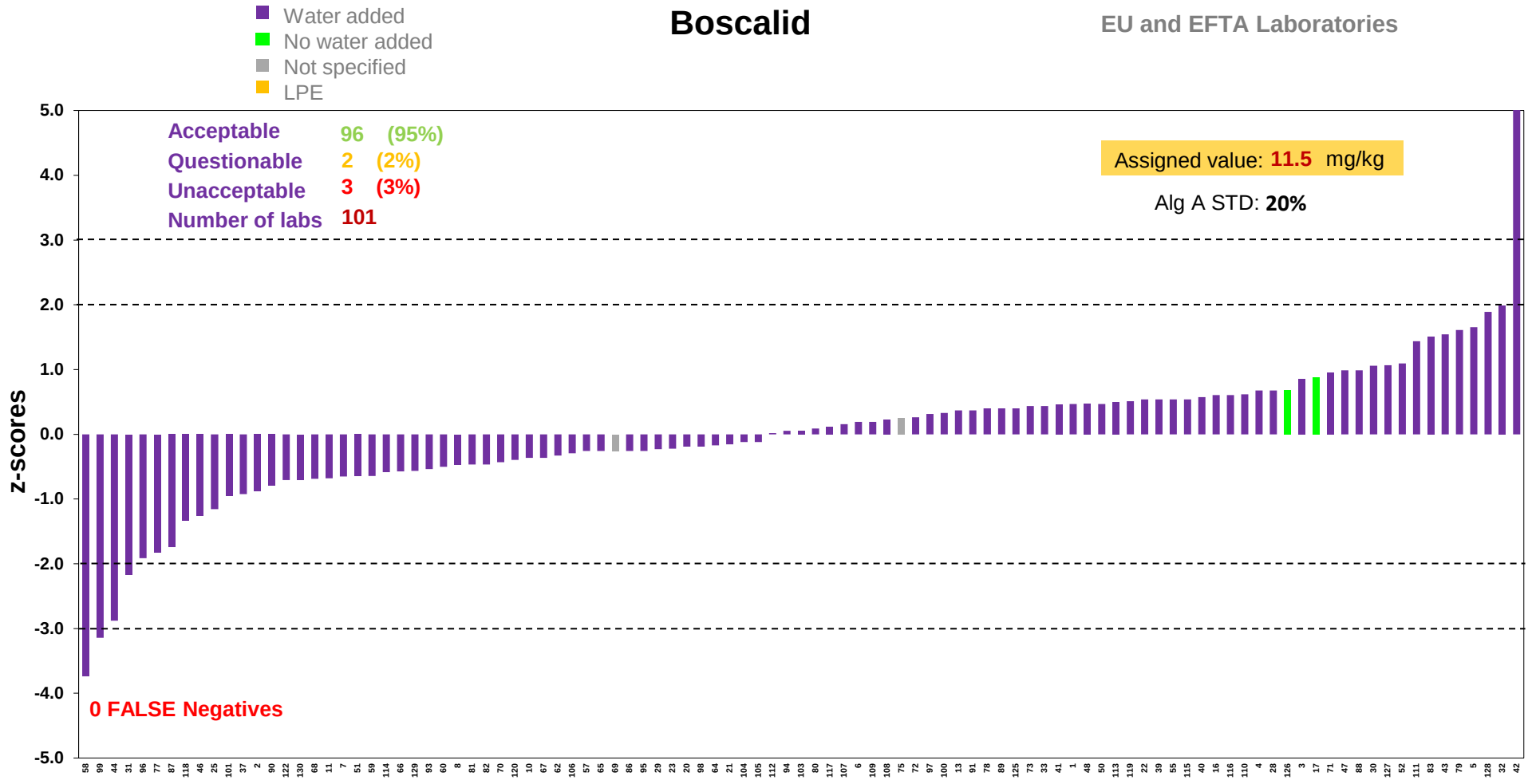
Azoxystrobin

EU and EFTA Laboratories



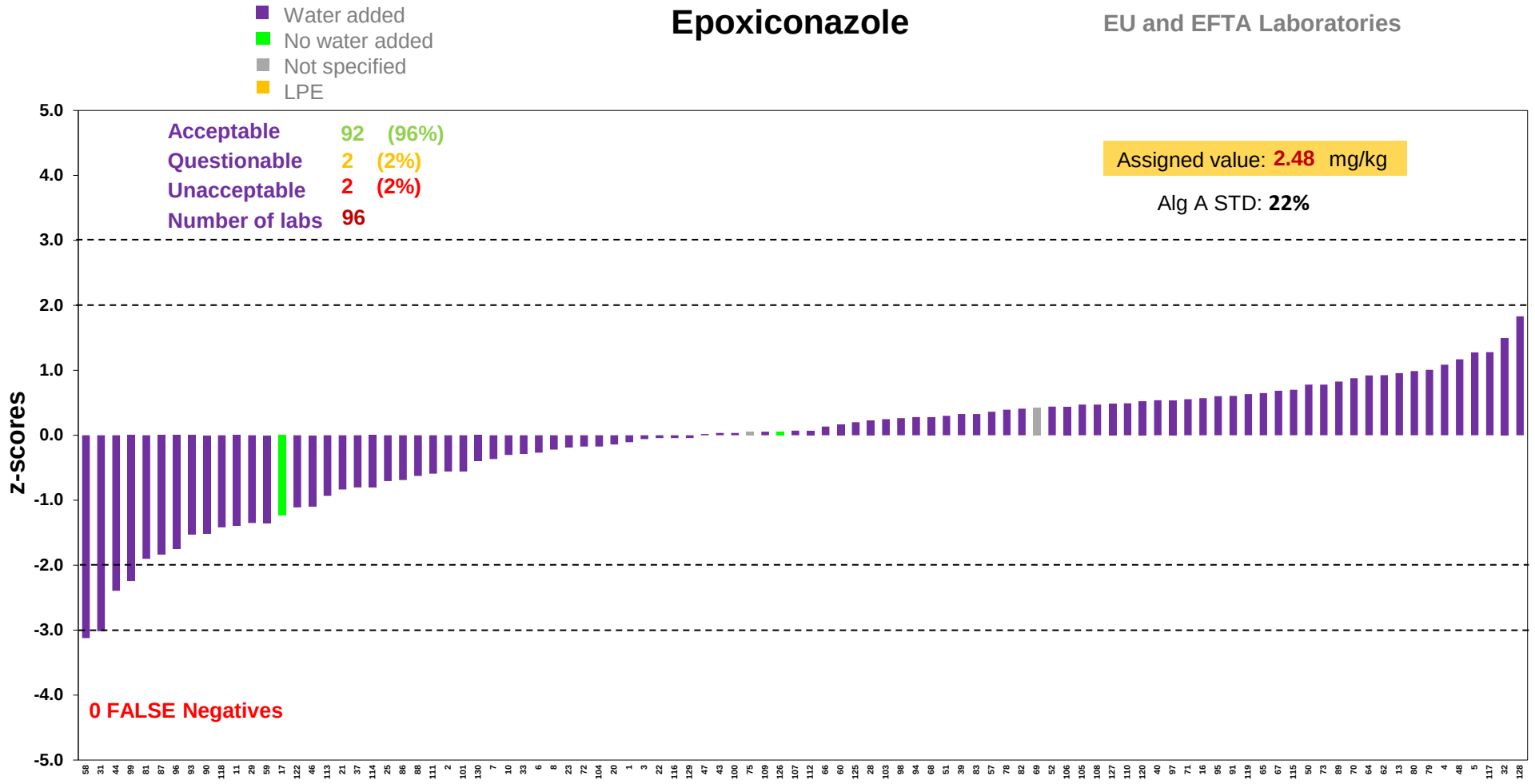
Boscalid

EU and EFTA Laboratories



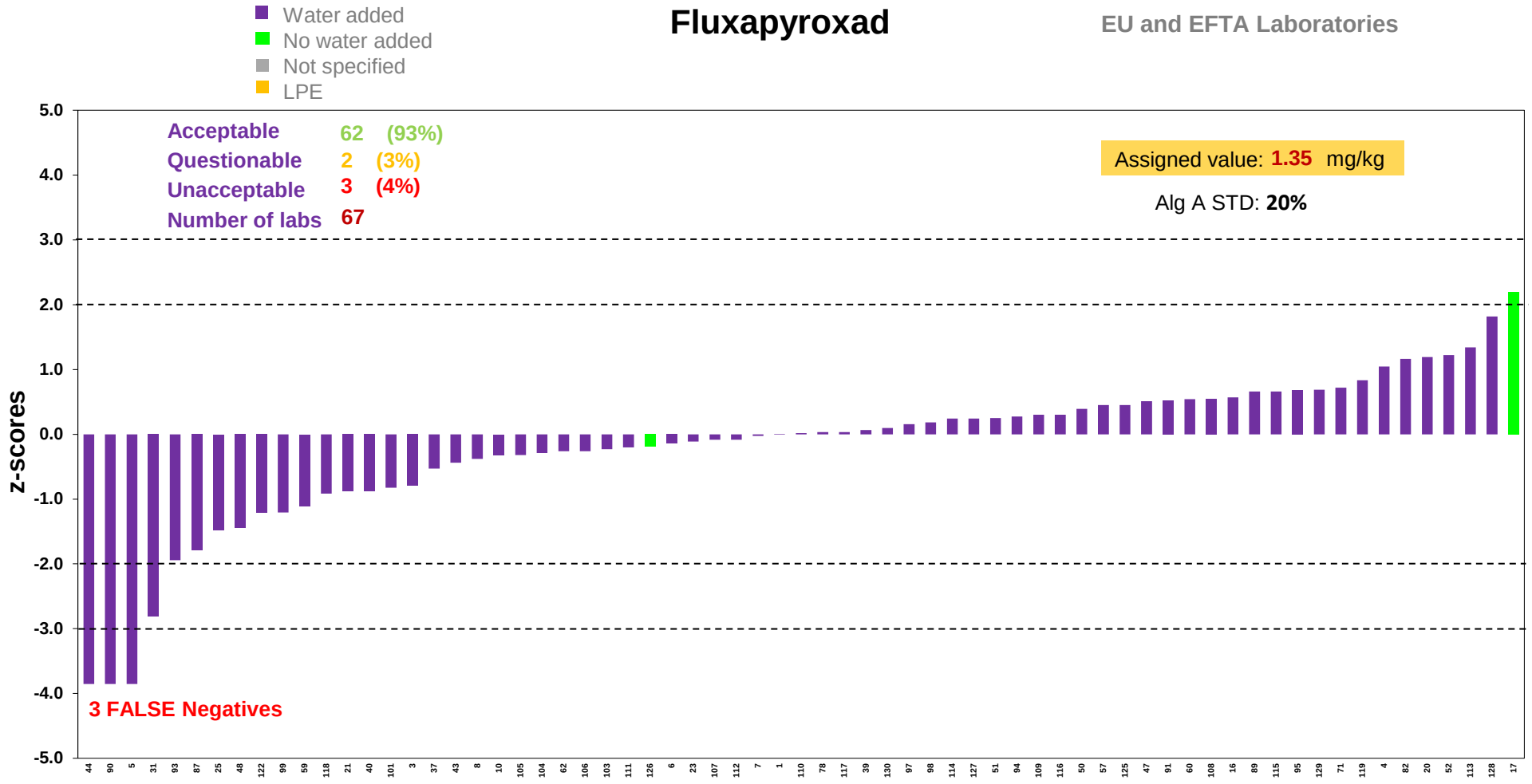
Epoxiconazole

EU and EFTA Laboratories



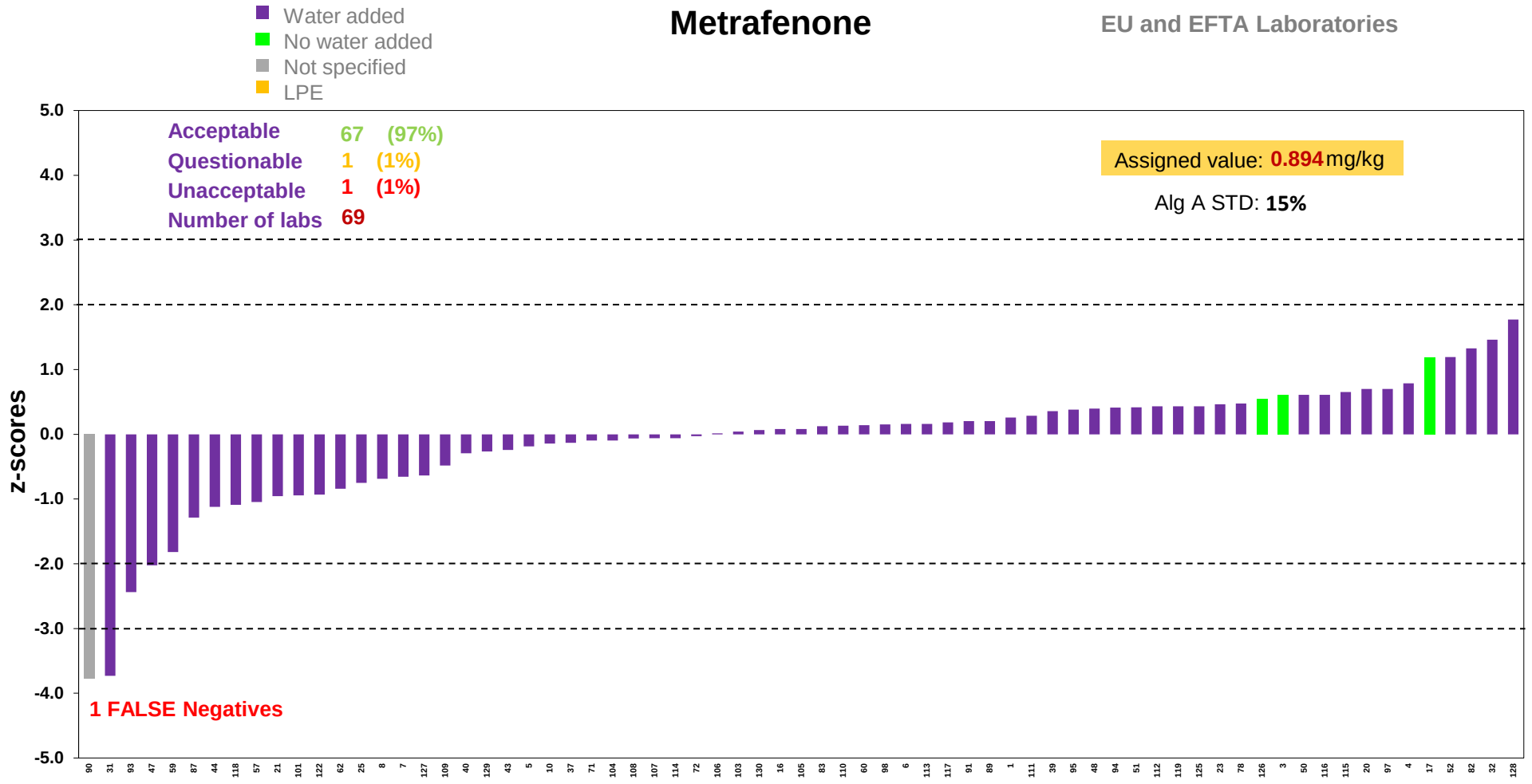
Fluxapyroxad

EU and EFTA Laboratories



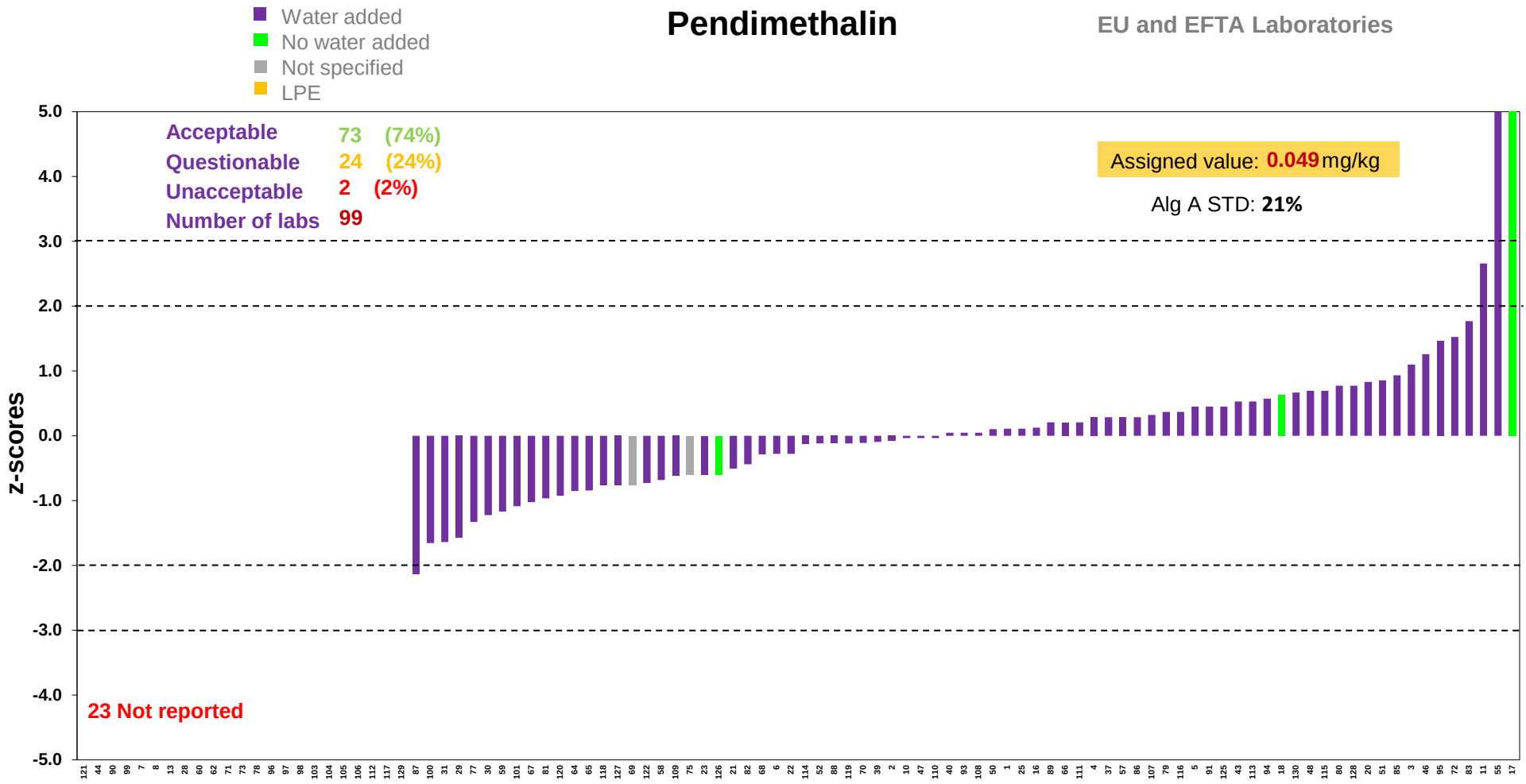
Metrafenone

EU and EFTA Laboratories



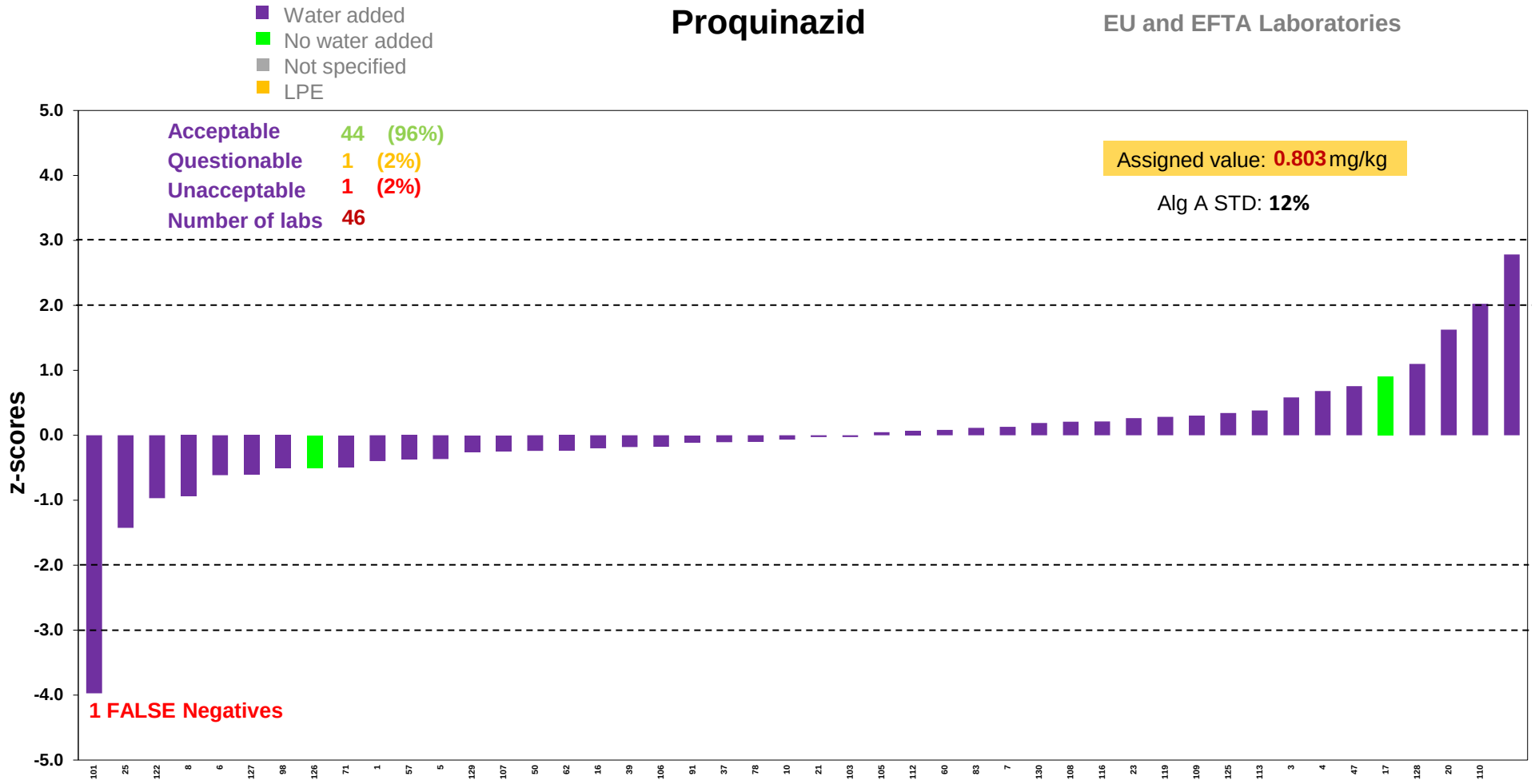
Pendimethalin

EU and EFTA Laboratories



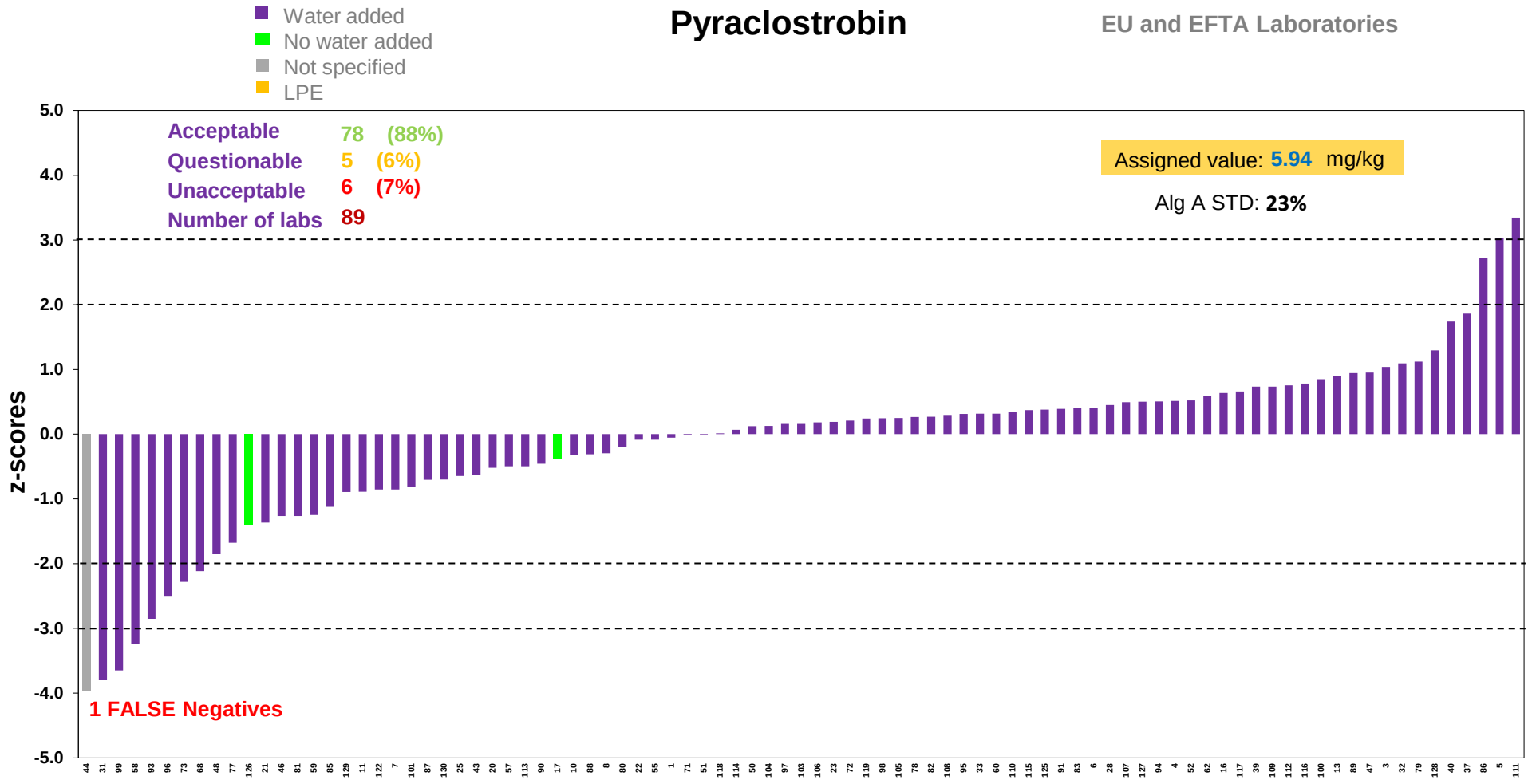
Proquinazid

EU and EFTA Laboratories



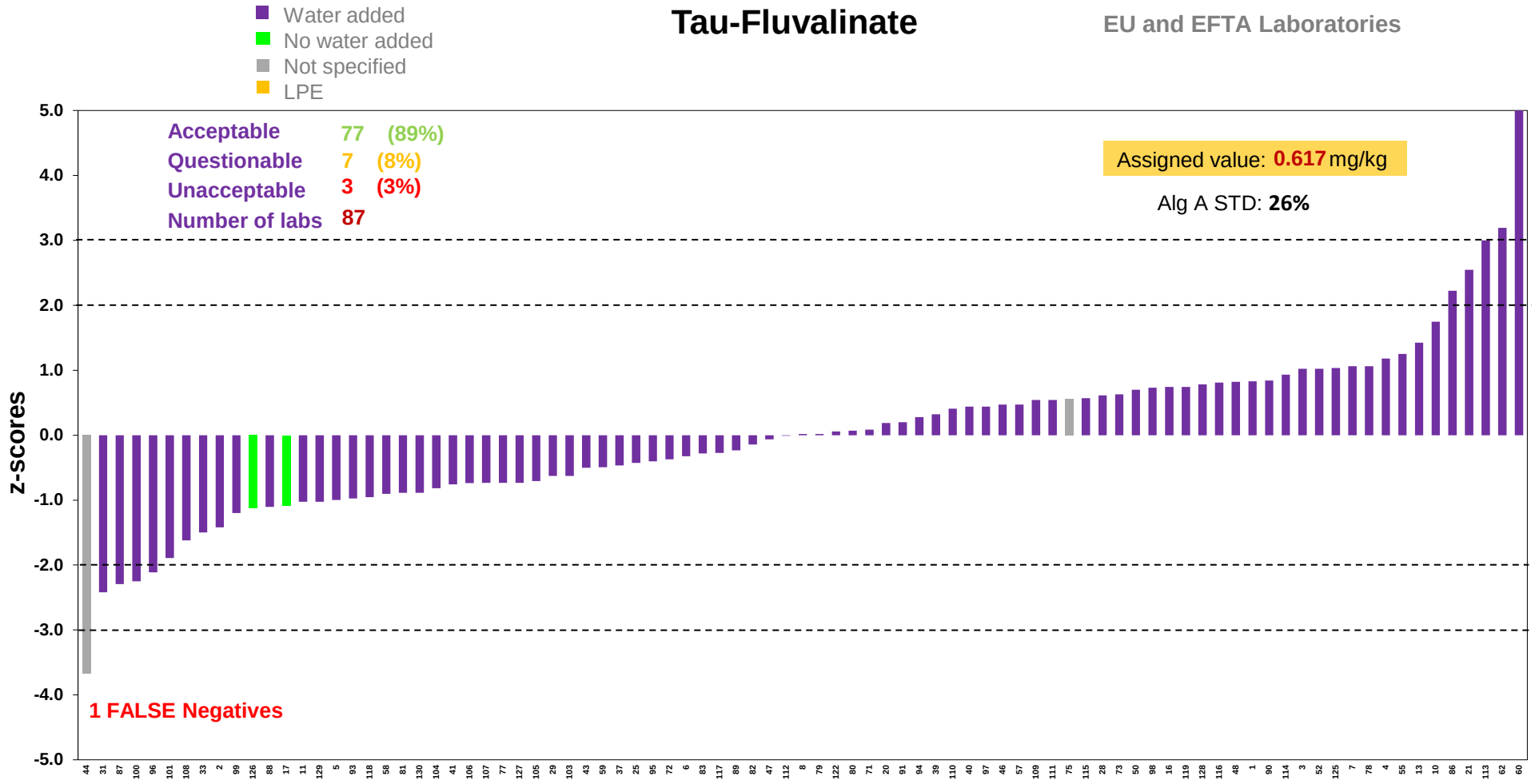
Pyraclostrobin

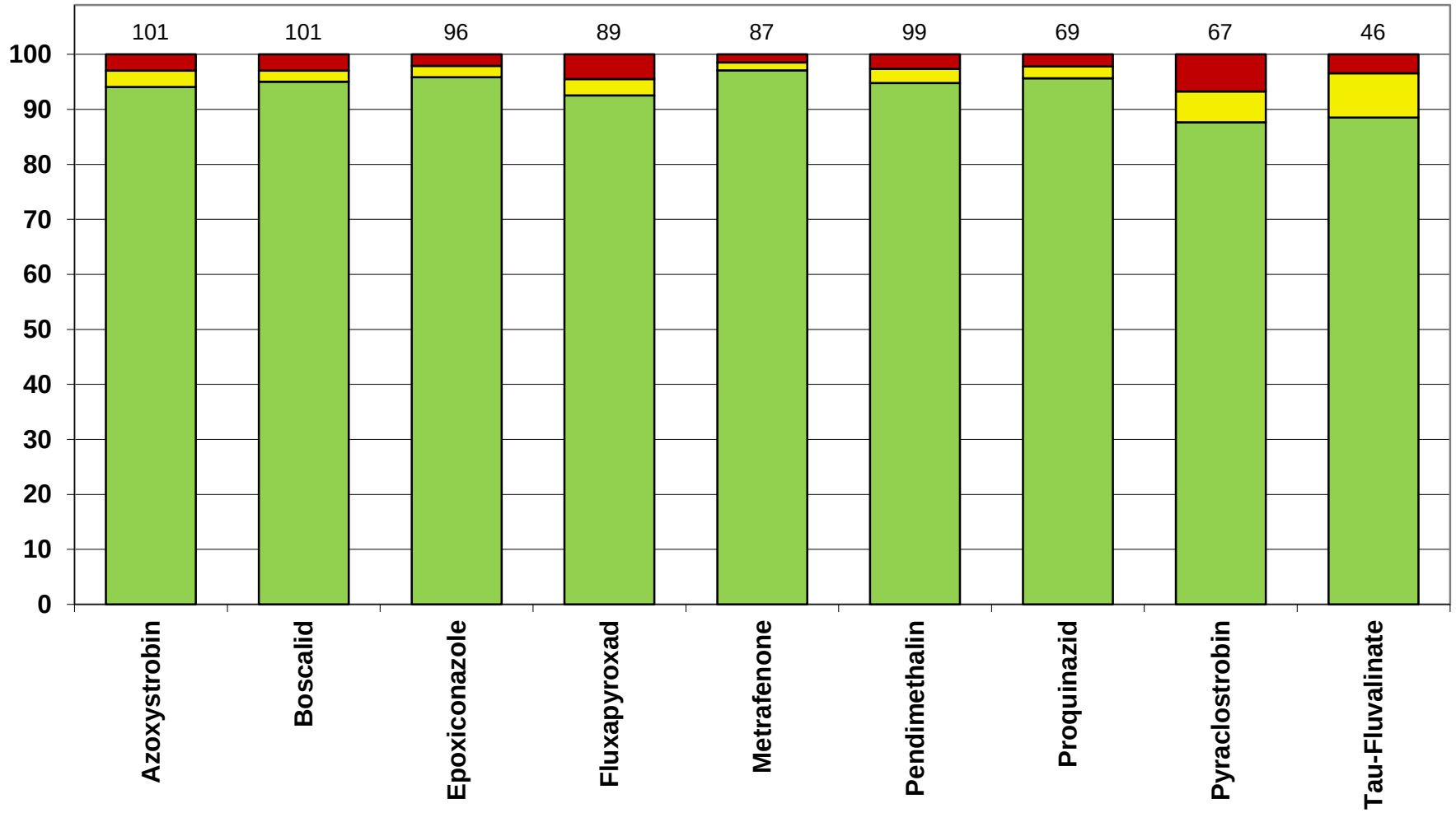
EU and EFTA Laboratories



Tau-Fluvalinate

EU and EFTA Laboratories




101
101
96
89
87
99
69
67
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100
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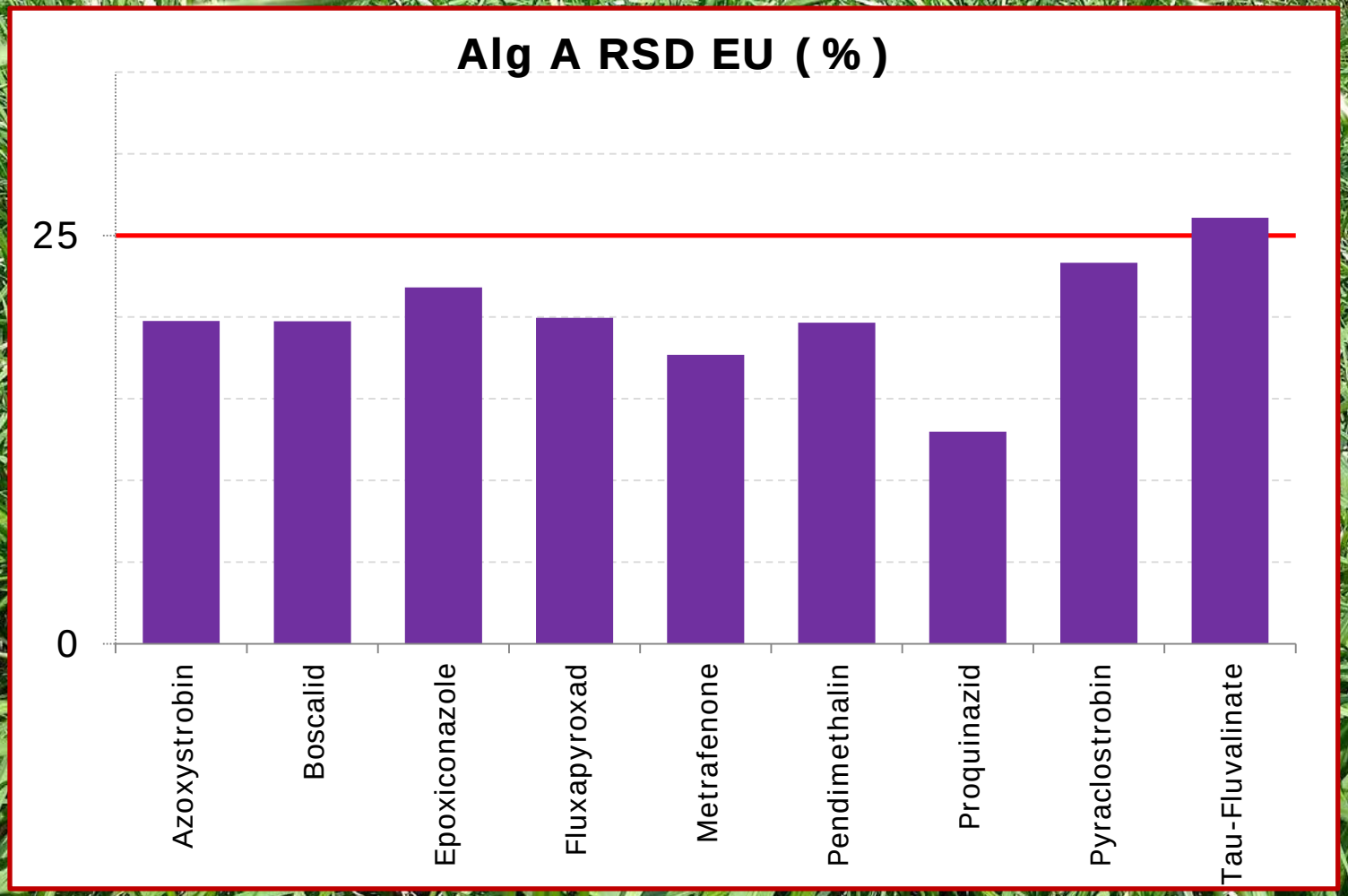
0
Azoxystrobin
Boscalid
Epoxiconazole
Fluxapyroxad
Metrafenone
Pendimethalin
Proquinazid
Pyraclostrobin
Tau-Fluvalinate

Alg. A RSD

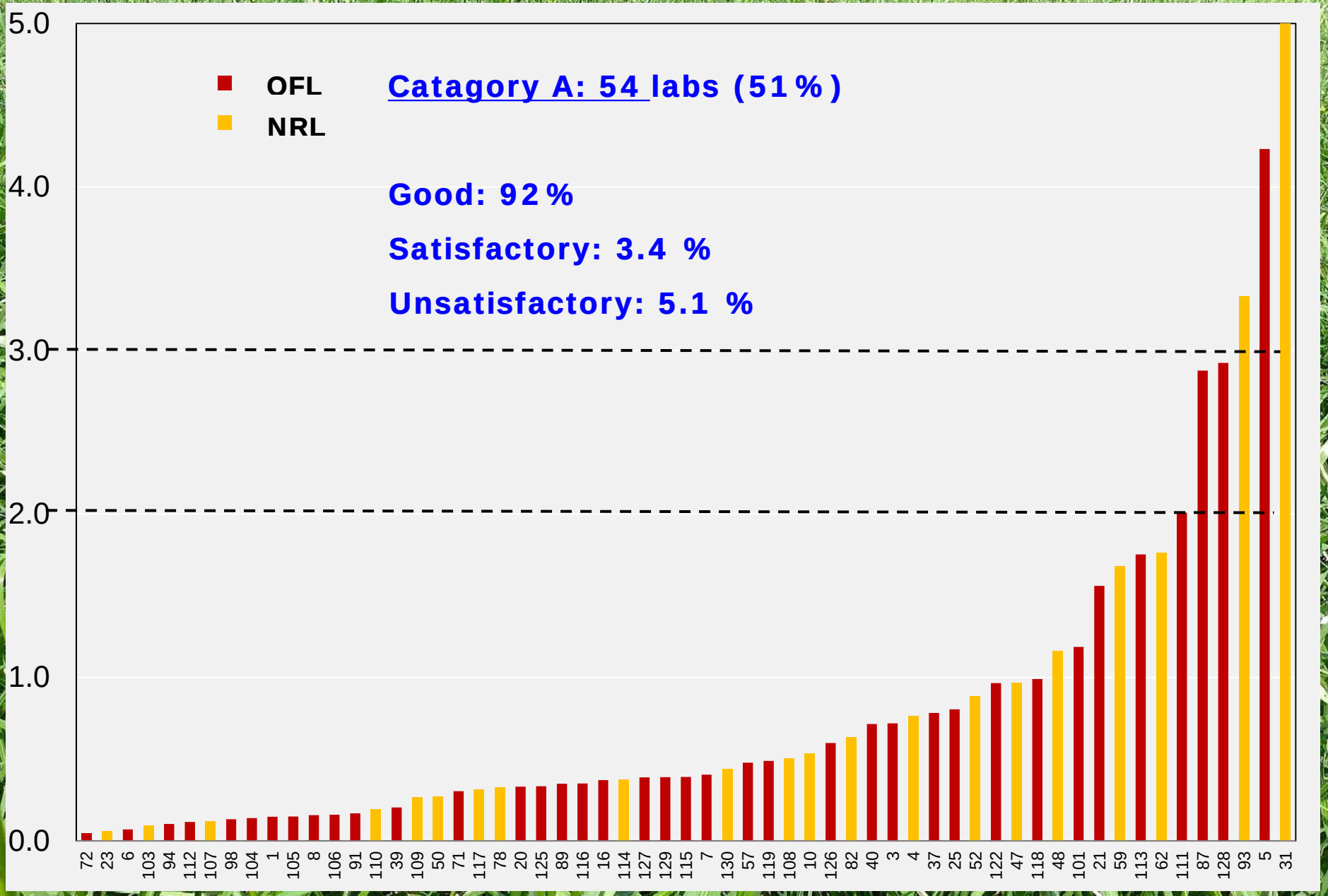

Alg A RSD EU (%)
25
0
Azoxystrobin
Boscalid
Epoxiconazole
Fluxapyroxad
Metrafenone
Pendimethalin
Proquinazid
Pyraclostrobin
Tau-Fluvalinate

Catagory A laboratories and combined z-scores

- To be classified as Category A laboratory the labs had to 1) be able to analyse for at least 90% of the compulsory pesticides on the target pesticides list, 2) have correctly detected and quantified at least 90% of the pesticide present in the test material (7≥ pesticide residues) and 3) report no false positive.
- Evaluation of the overall performance, the Average of the Squared z-Score (AZ2) are calculated:

$$AZ^2 = \frac{\sum_{i=1}^{n} Z_i^2}{n}$$

- z ≤ 2 Good
- 2 < z ≤ 3 Satisfactory
- z > 3 Unsatisfactory


OFL
NRL
Catagory A: 54 labs (51%)
Good: 92%
Satisfactory: 3.4 %
Unsatisfactory: 5.1 %
5.0
4.0
3.0
2.0
1.0
0.0
72 23 6 103 94 112 107 98 104 1 105 8 106 91 110 39 109 50 71 117 78 20 125 89 116 16 114 127 129 115 7 130 57 119 108 10 126 82 40 3 4 37 25 52 122 47 118 48 101 21 59 113 62 111 87 128 93 5 31

Thank you for your attention