

Preparation of an incurred rice sample and evaluation of pesticide residues in the commodities obtained after processing.

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Overview



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- Introduction.
- Objectives.

2

- Preparation of incurred rice.
- Evaluation of the pesticide residues by LC-MS/MS and GC/MS.

3

- Comparison of the efficiency of the different methods.
- Conclusions and Future work.



Introduction



✓ Cereal crops comprise more than 60% of agricultural production worldwide.



✓ Among cereals, rice is the most consumed and its consumption has increased in the recent decades, with a consequent rise in the use of pesticides to improve the production yields.





Rice ecosystem in Uruguay



✓ The cultivation of rice in Uruguay is an unique production system based on the rotation of prairies and alternative crops, integrated with livestock production, in order to ensure a global sustainability and health safety.



✓ This system reduces the environmental impact of rice crop.



Rice pesticides in Uruguay

Insecticides



Diflubenzuron

Lambda cyalothrin

Cypermethrin

Thiamethoxam



Fungicides



Azoxystrobin

Tebuconazole

Epoxiconazole

Kresoxim methyl

Difenoconazole

Trifloxystrobin

Tricyclazole

Pyraclostrobin



Herbicides

Byspiribac
sodium

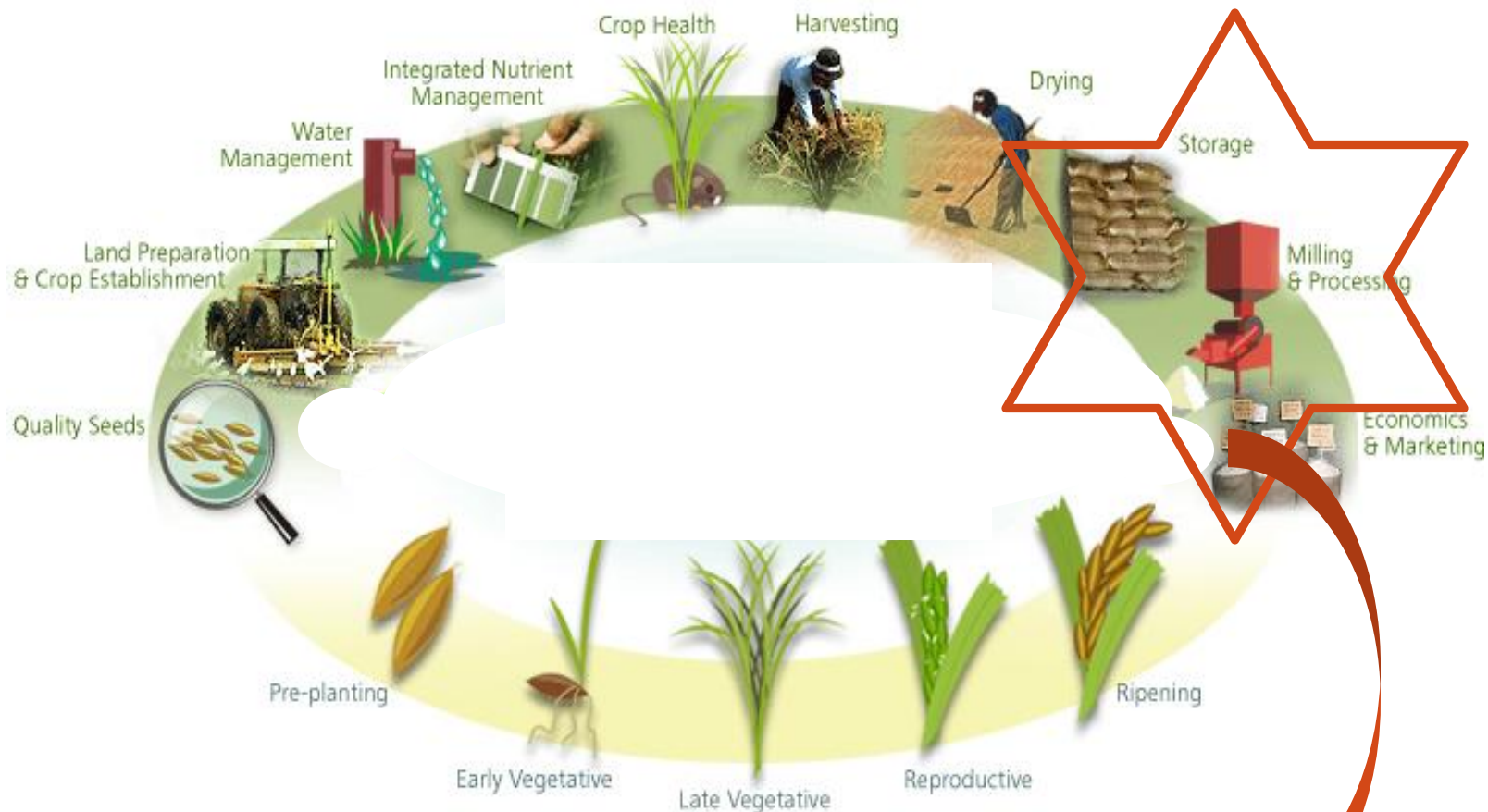
Quinclorac

Propanil

Clomazone

Glifosate

Pyrazosulfuron
ethyl



Is it safe for the consumer ?

CHECK COMPLIANCE WITH THE MRL.





Rice is not consumed
as raw material...



Which matrices do we have ?



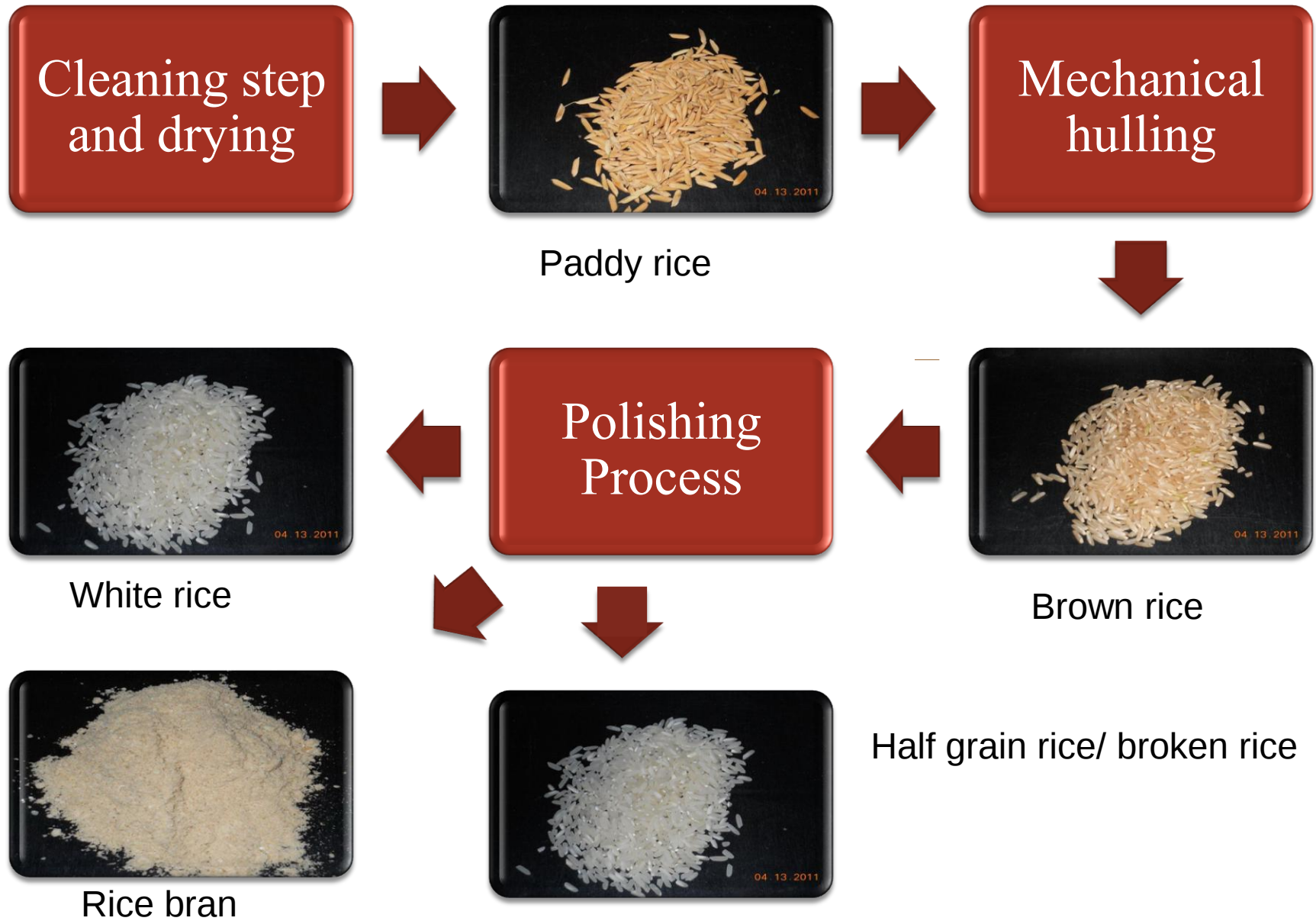
→ Bran Rice

→ White Rice

→ Brown Rice

→ Paddy Rice

Rice processing.





✓ It is important to study the distribution of the pesticide residues in the different commodities after rice processing.



To evaluate the pesticide distribution properly:
incurred residues study



Objective



Preparation of rice samples with known incurred residues and their evaluation in the commodities obtained after processing.



How to reach this objective?



1. Preparing the incurred sample.

2. Studying the distribution of the pesticide residues in the different matrices after processing.



Selecting a suitable analytical methodology for the different matrices.

An aerial photograph of a lush green rice field. A dark, winding irrigation canal or ditch cuts through the field, separating a section of tilled, brown earth on the left from the rest of the vibrant green rice plants. The field is divided into rectangular plots by narrow paths or ditches. In the background, a dirt road runs horizontally across the frame.

In the field

Preparation of the rice sample.



Which pesticides were applied???



Herbicides

- Bispyribac sodium
- Quinclorac
- Propanil
- Clomazone

Fungicides

- Tebuconazole
- Epoxiconazole
- Trifloxystrobin
- Difenoconazole
- Azoxystrobin
- Kresoxim methyl
- Isoprothiolane
- Tricyclazole
- Carbendazim

Insecticides

- Lambda-Cyhalothrin
- Thiamethoxam

Field treatment: Herbicides

Active substance	Commercial product	Application time	Application rate
Propanil	Propanil 480	1 application January	15 L/ha
Bispyribac sodium	Byspiriné	1 application January	0.5 L/ha
Clomazone	Cibelcol	1 application January	4 L/ha
Quinclorac	Exocet	1 application January	6 L/ha

Field treatment: Fungicides

Active substance	Commercial product	Application time	Application rate	Elapsing time (days)
Epoxiconazole	Allegro	2 applications March	2.4 L/ha	35
Difenoconazole	Convect	2 applications March	0.5 L/ha	28
Azoxystrobin	Amistar	2 applications March	0.5 L/ha	21
Tebuconazole	Bucanner	2 applications March	1.2 L/ha	35
Carbendazim	Agrizim	2 applications March	2.0 L/ha	14-35
Isoprothiolane	Ipetec	2 applications March	2.5 L/ha	14
Kresoxim methyl	Allegro	2 applications March	2.4 L/ha	35
Trifloxystrobin	Nativo	1 application March	1.6 L/ha	21
Tricyclazole	Tricyclazole 75%	2 applications March	0.36 L/ha	-

Field treatment: Insectides

Active substance	Commercial product	Application time	Application rate
Lambda cyhalothrin	Engeo 247	1 application March	0.36 L/ha
Thiamethoxam	Engeo 247	1 application March	0.36 L/ha

All the application rates were higher than the recommended.



Pesticide	Status under Directive 91/414/EEC MRL (mg/kg) Reg. (EC) No 396/2005	Status under Directive 91/414/EEC
Quinclorac	5	out
Propanil	0.2	out
Bispyribac sodium	---	pending
Clomazone	0.01	included
Lambda cyhalothrin	1	included
Thiamethoxam	0.05	included
Epoxiconazole	0.1	included
Difenoconazole	0.05	included
Azoxystrobin	5	In Uruguay is banned for rice!
Tebuconazole	2	
Carbendazim	0.01	included
Isoprothiolane	---	out
Kresoxim methyl	0.05	included
Trifloxystrobin	0.02	included
Tricyclazole	1	out

Obtained Material



**Yield: 150 kg of
paddy rice
(homogeneized)**

120 kg Paddy rice

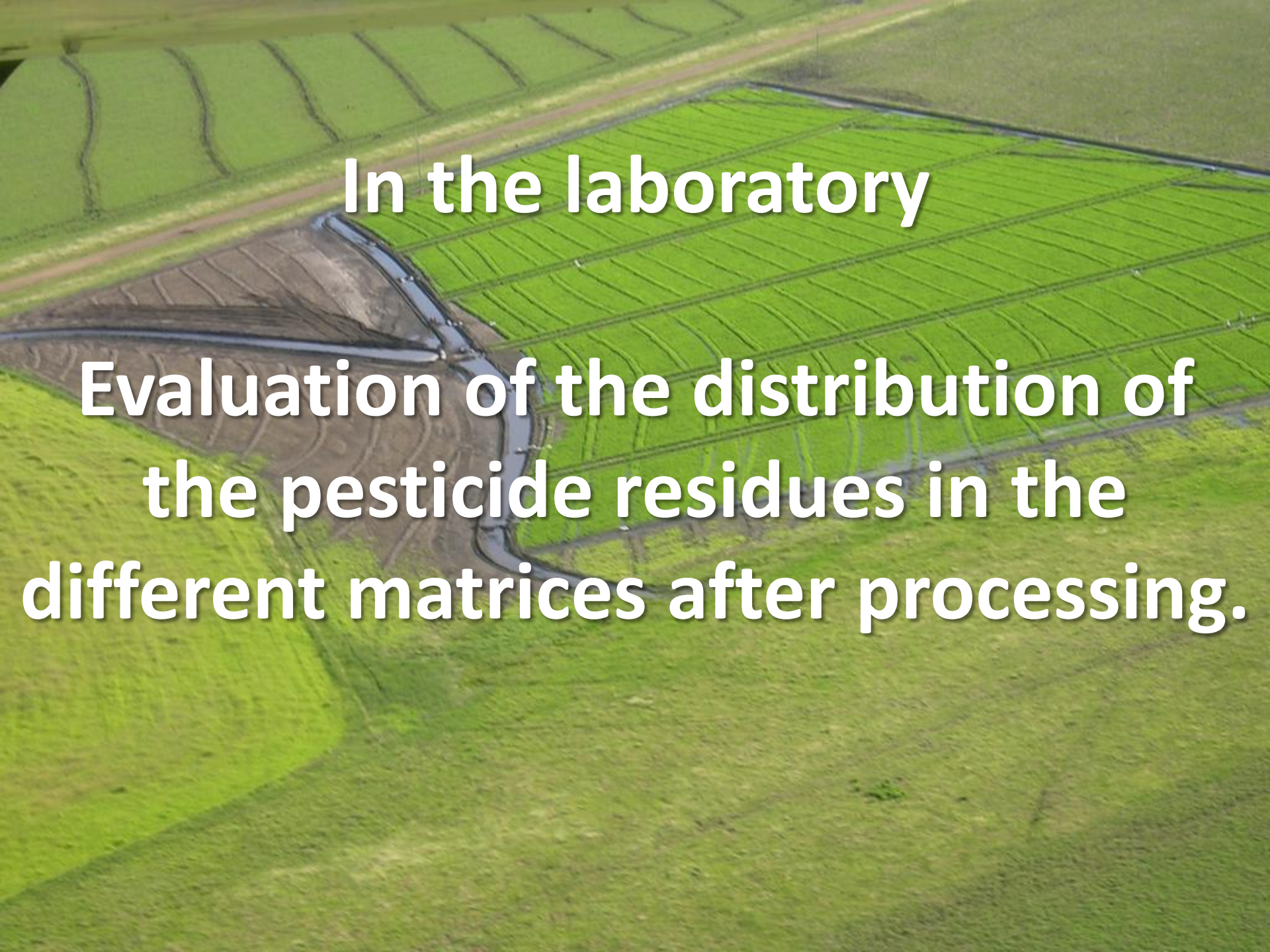
30 kg Paddy rice

Brown Rice

White Rice

Bran Rice

Broken Rice

An aerial photograph of a vast agricultural landscape. A prominent, winding canal or irrigation system cuts through the fields, which are divided into a grid-like pattern of smaller plots. The vegetation is a vibrant green, suggesting a healthy crop. The canal itself appears to be a mix of dark water and lighter, possibly silted or recently tilled, earth.

In the laboratory

**Evaluation of the distribution of
the pesticide residues in the
different matrices after processing.**



Which methodology is the best for each commodity??



In a previous work we compared different based QuEChERS methods for the analysis of white rice.



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journal homepage: www.elsevier.com/locate/talanta



Evaluation of various QuEChERS based methods for the analysis of herbicides and other commonly used pesticides in polished rice by LC-MS/MS[☆]

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Recovery Results: 10 µg/kg

Table. Methods: 1. Original QuEChERS; 2. Citrate buffered QuEChERS 3. Citrate buffered QuEChERS without clean-up; 4. Acetate buffered QuEChERS without clean-up. N.A: not analyzed.

Pesticide name	Method 1	Method 2	Method 3	Method 4
Azoxystrobin	112.6(6)	108.7(14)	102.3(13)	94.5(4)
Bispyribac sodium	51.1(37)	81.6(22)	62.2(14)	84.1(27)
Carbendazim	97.5(7)	114.1(8)	81.9(8)	87.6(4)
Clomazone	104.3(5)	110.7(11)	104.7(11)	92.6(7)
Epoxiconazole	101.3(9)	102.1(18)	67.3(10)	74.3 (12)
Kresoxim methyl	NA	NA	NA	NA
Propanil	100.7(9)	88.5(9)	77.4(6)	86.3(5)
Tebuconazole	117.7(6)	102.1(8)	59.8(6)	91.0(14)
Thiamethoxam	101.6(11)	118.4(16)	101.7(24)	96.5(11)
Tricyclazole	101.3(3)	111.3(3)	112.1(8)	101.2(3)

Recovery Results: 300 µg/kg

Table. Methods: 1. Original QuEChERS; 2. Citrate buffered QuEChERS; 3. Citrate buffered QuEChERS without clean-up; 4. Acetate buffered QuEChERS without clean-up. N.A: not analyzed.

Pesticide name	Method 1	Method 2	Method 3	Method 4
Azoxystrobin	99.6(2)	90.6(4)	93.3(7)	89.5(1)
Bispyribac sodium	99.8(6)	80.5(20)	87.6(6)	68.1(3)
Carbendazim	99.1(3)	85.3(4)	85.6(10)	80.8(1)
Clomazone	102.7(7)	86.6(6)	77.8(14)	78.1(6)
Epoxiconazole	101.4(2)	85.9(5)	65.9(11)	71.1(8)
Kresoxim methyl	105.7(5)	82.5(8)	71.2(6)	75.2(4)
Propanil	101.1(1)	94.9(5)	82.2(1.3)	84.3(2)
Tebuconazole	101.3(5)	79.3(4)	66.7(18)	69.2(9)
Thiamethoxam	98.5(3)	87.8(4)	90.6(11)	90.1(3)
Tricyclazole	101.6(2)	88.2(3)	89.4(10)	82.9(2)



But....



What about quinclorac, isoprothiolane, trifloxystrobin, λ -cyhalothrin and difenoconazole which were not included in the previous work??



What about the performance of these methods in the other matrices???



We evaluated original QuEChERS and citrate QuEChERS for the different products.



Results - Validation.



White Rice



Paddy Rice



LC-MS/MS



Azoxystrobin
Byspiribac sodium
Carbendazim
Clomazone
Difenoconazole
Epoconazole
Isoprothiolane
Kresoxim methyl
Propanil
Quinclorac
Tebuconazole
Thiamethoxam
Tricyclazole
Trifloxystrobin

ESI +/-; 45/15 min

Column: Agilent Zorbax, Eclipse XDB – C8; 4,6 X 150 mm; 5 μ m particle size.

Mobile phase: MeCN/ 0.1% formic acid in Water.

Results - Validation.

Accuracy (% Rec)

Reproducibility (% RSD)

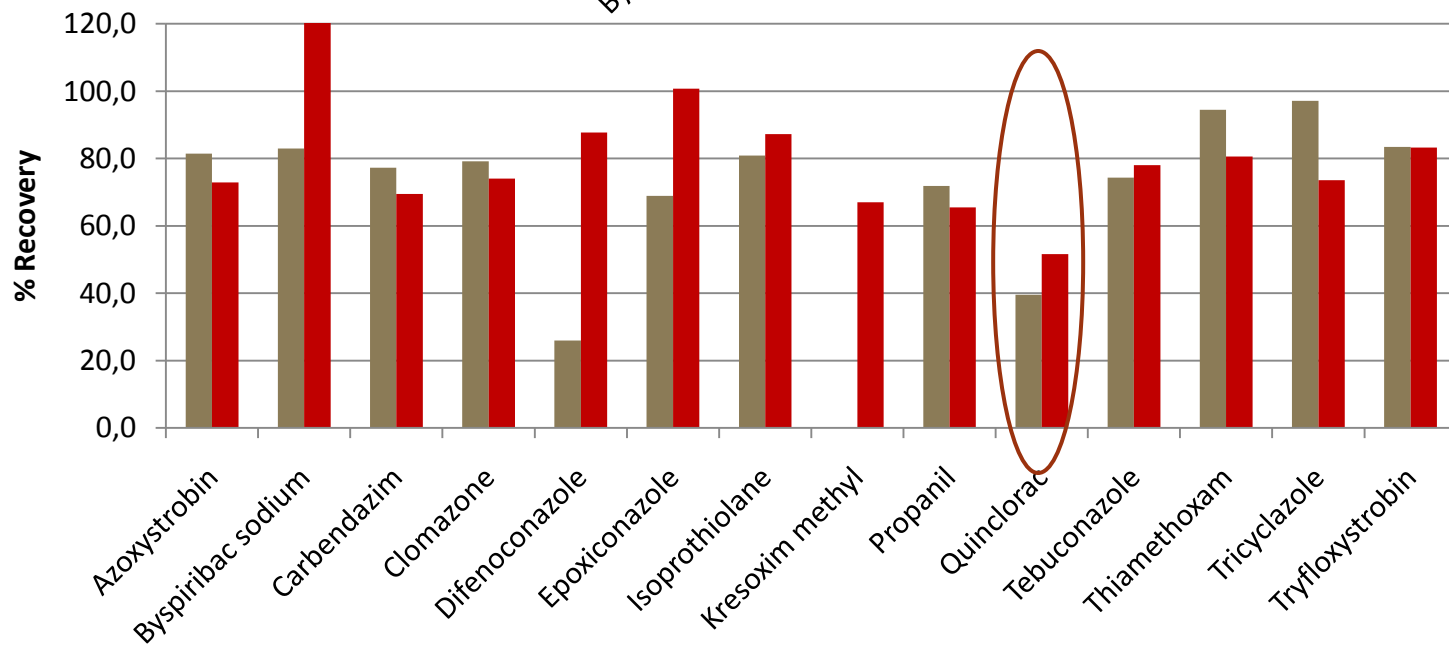
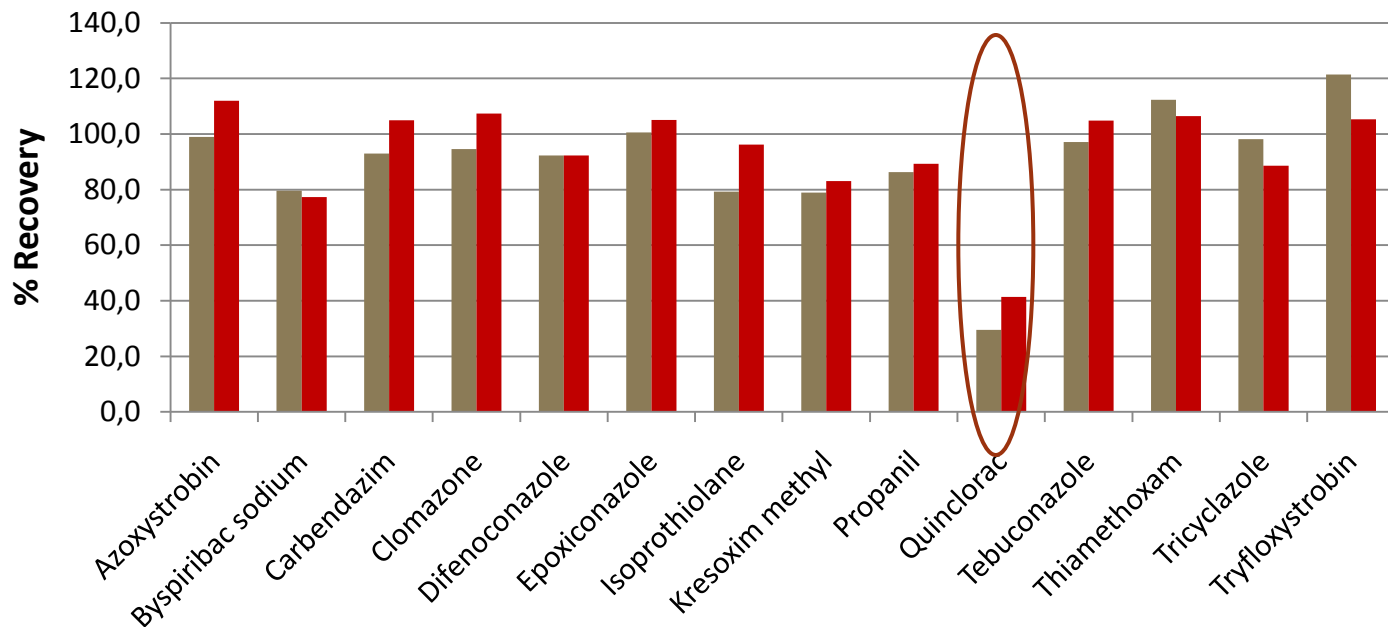
Detection and Quantification limits.
(LOD and LOQ)

Linearity

Matrix Effect

Recoveries White Rice

200 µg/kg
RSD: 3.3-21 %
n=5

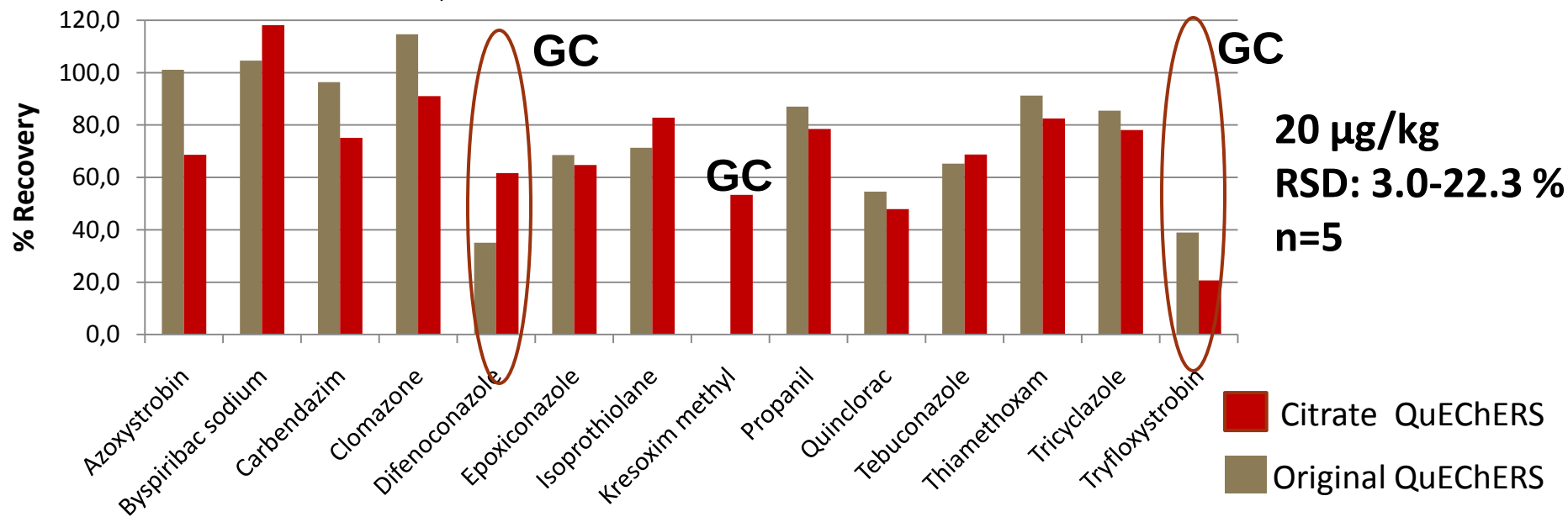
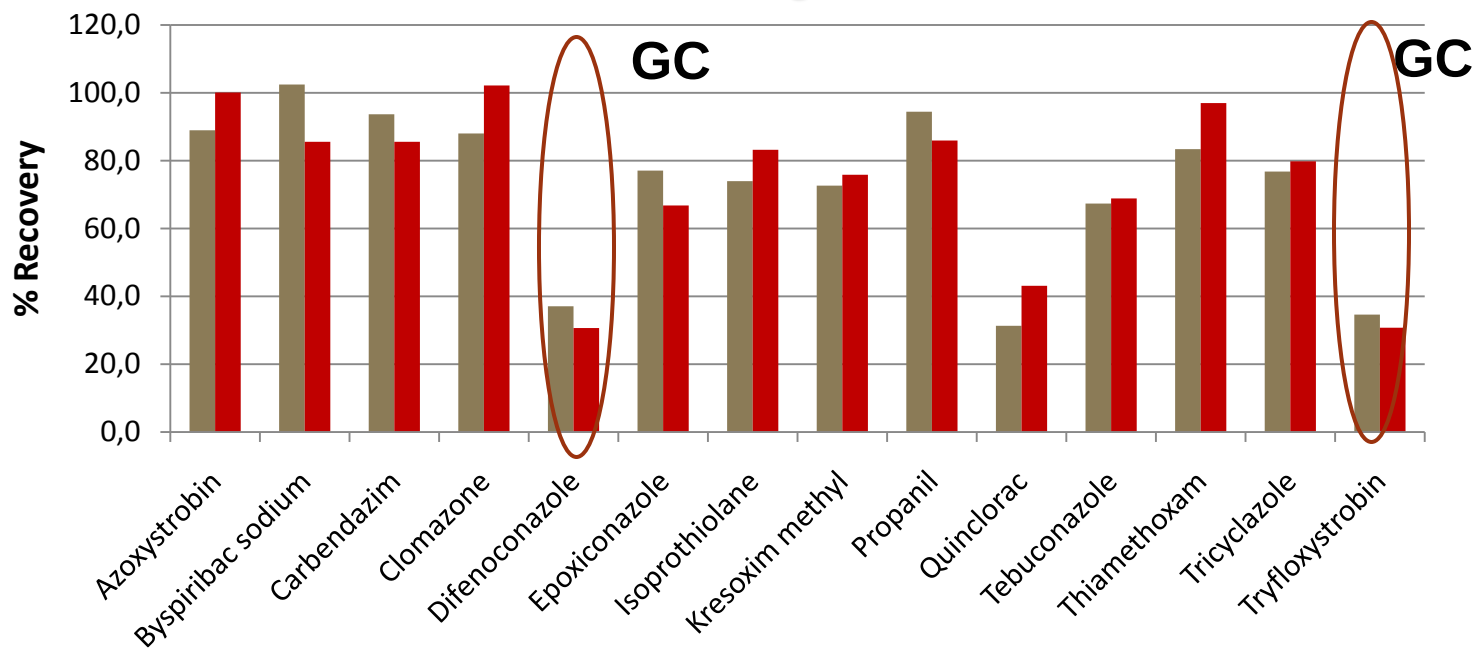


20 µg/kg
RSD: 0.3-20.5 %
n=5

■ Citrate QuEChERS
■ Original QuEChERS

Recoveries Paddy Rice

200 µg/kg
RSD: 4.4-20.7%
n=5

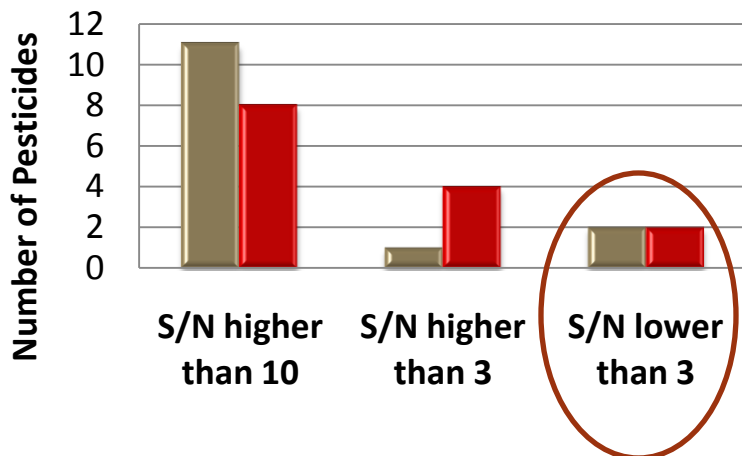




LOD & LOQ

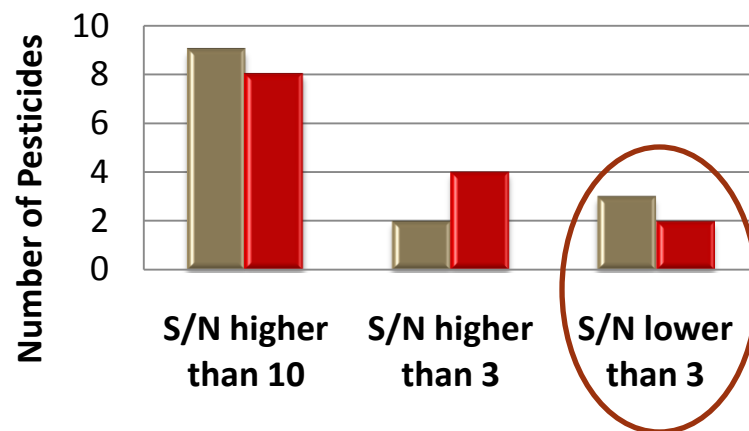
5 $\mu\text{g/kg}$

White Rice



■ Citrate QuEChERS
■ Original QuEChERS

Paddy Rice

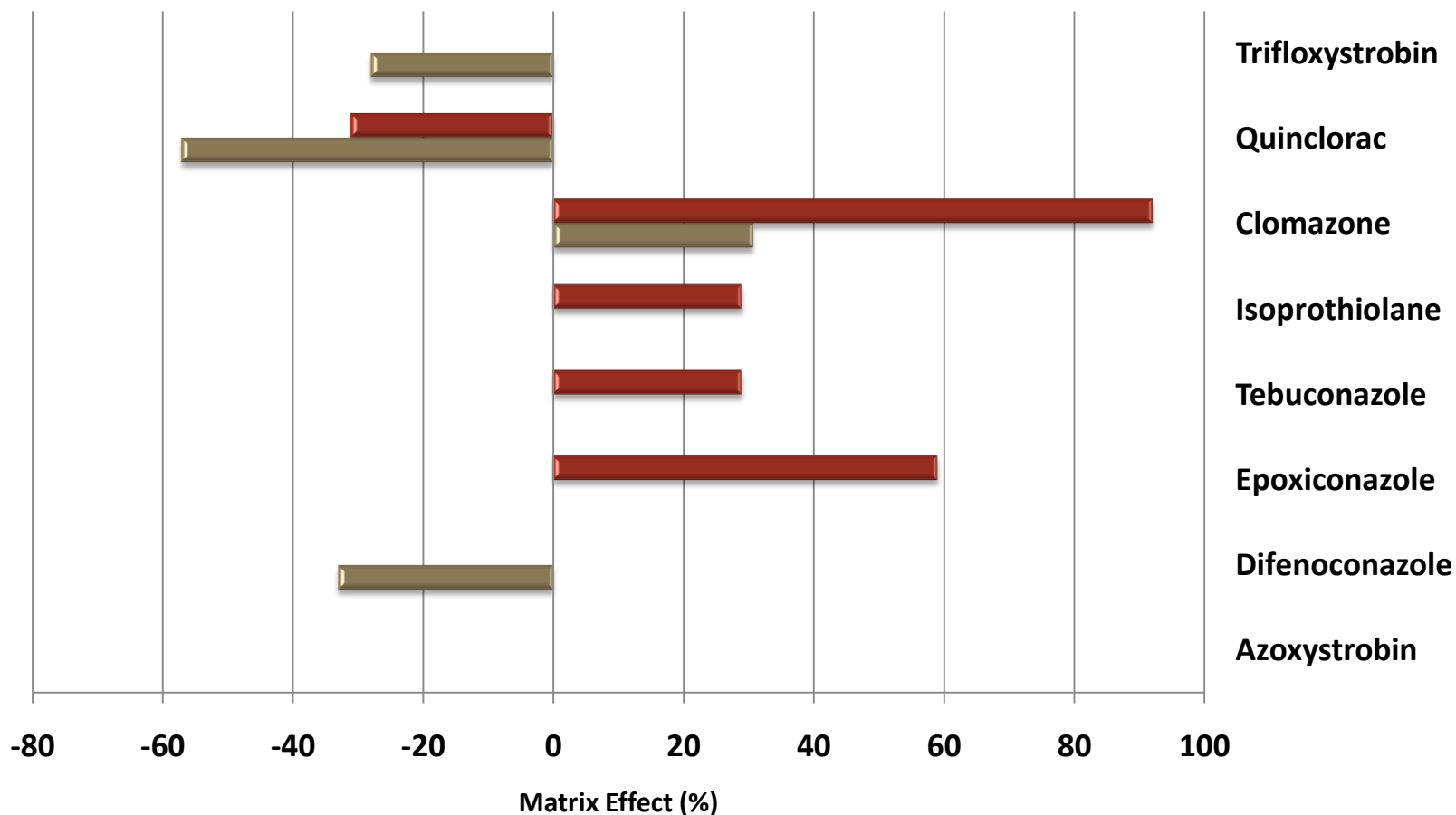


Linearity

Correlation coefficients were higher than 0.999 in all cases in the range 5-1500 $\mu\text{g/kg}$ except for quinclorac and kresoxim methyl.

Matrix Effect: White rice

Signal suppression ← Signal enhancement →

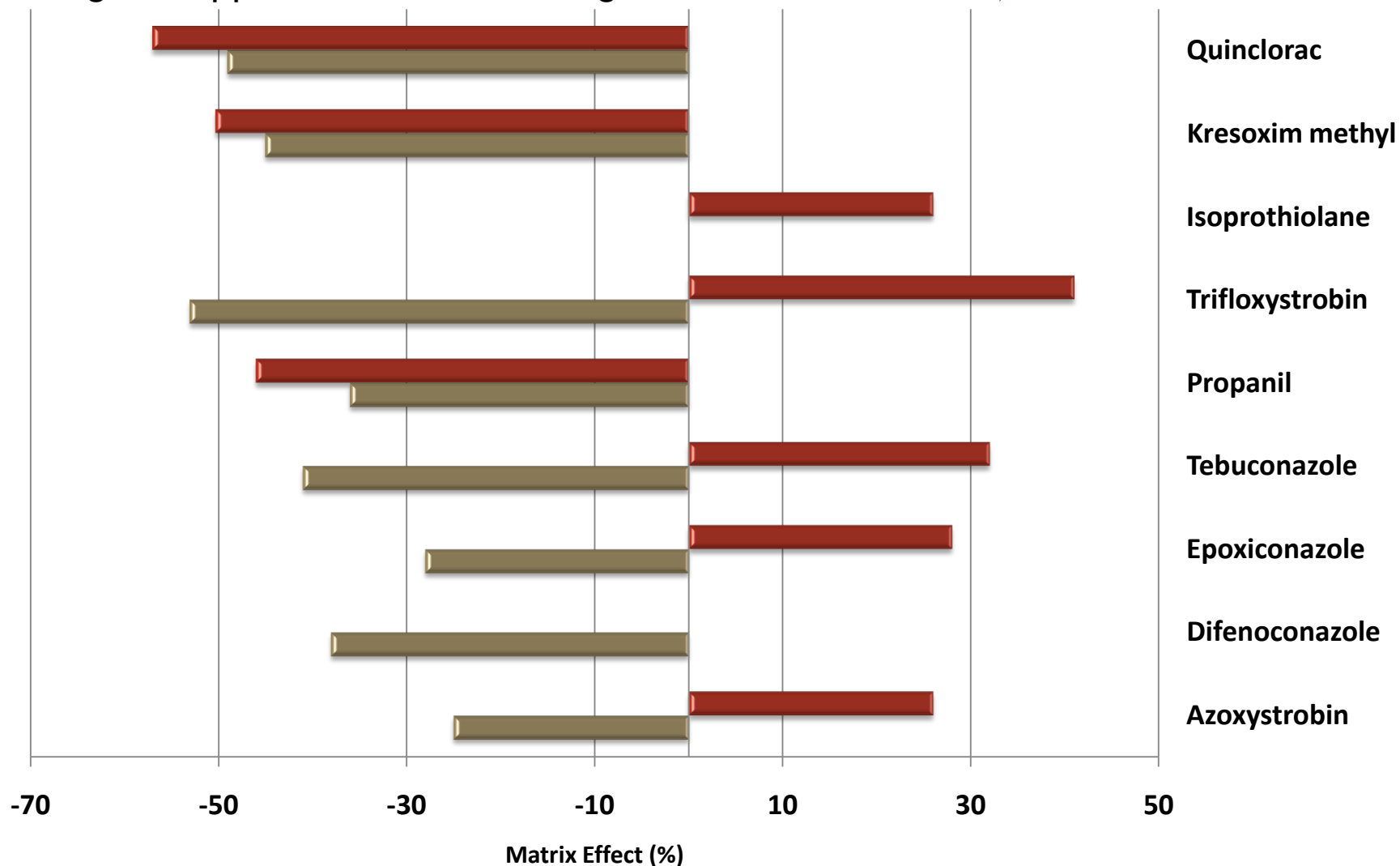


■ Citrate QuEChERS
■ Original QuEChERS

Matrix Effect: Paddy rice

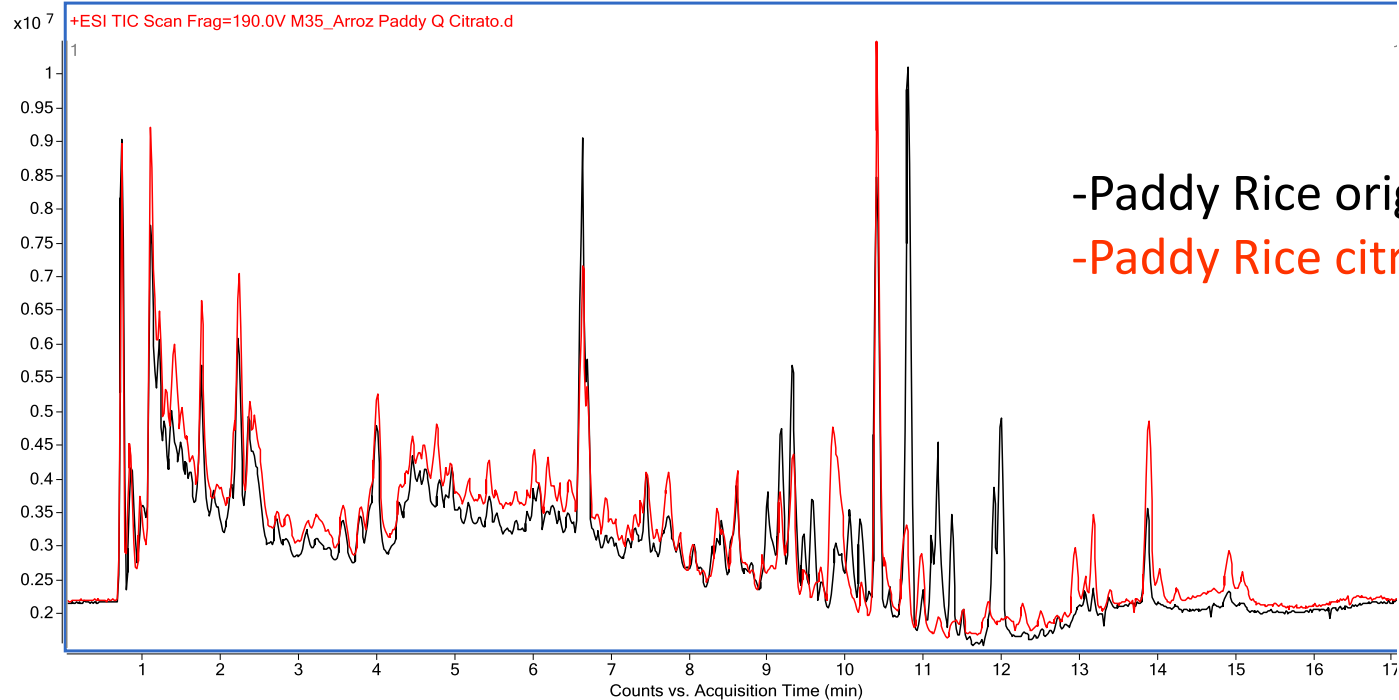
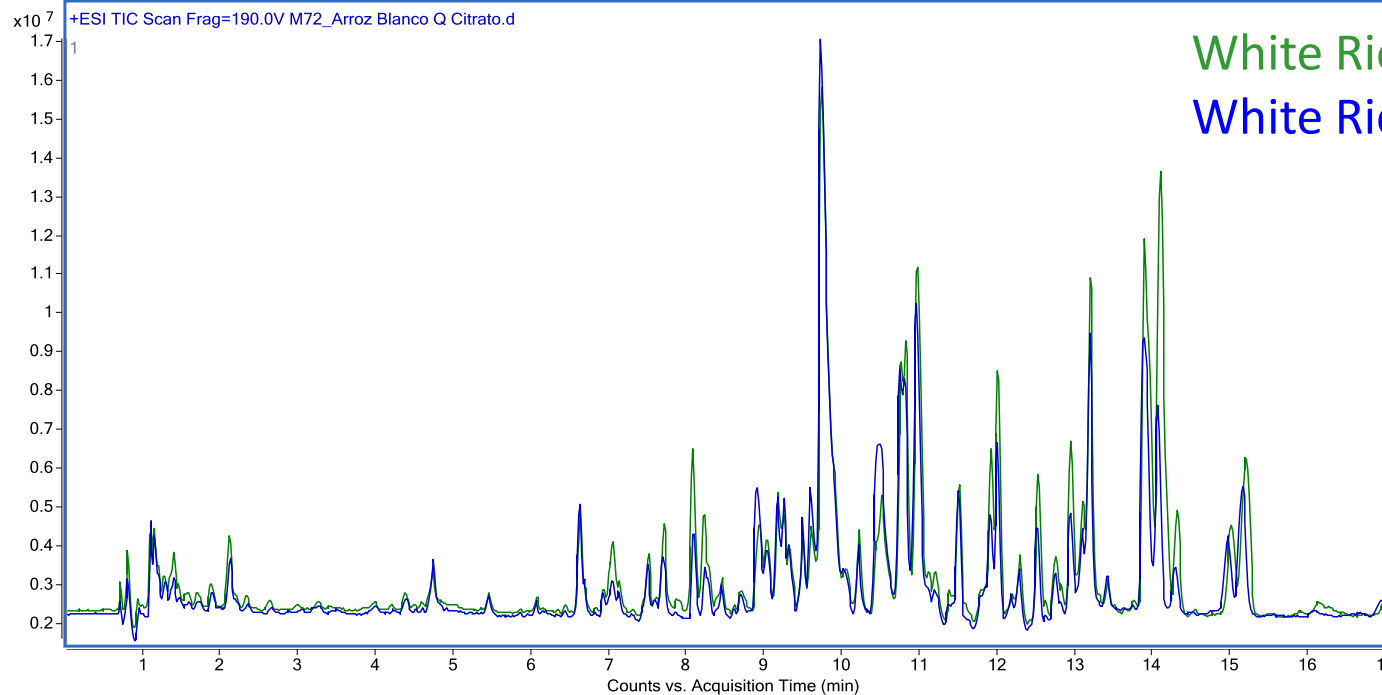
Signal suppression ←

→ Signal enhancement



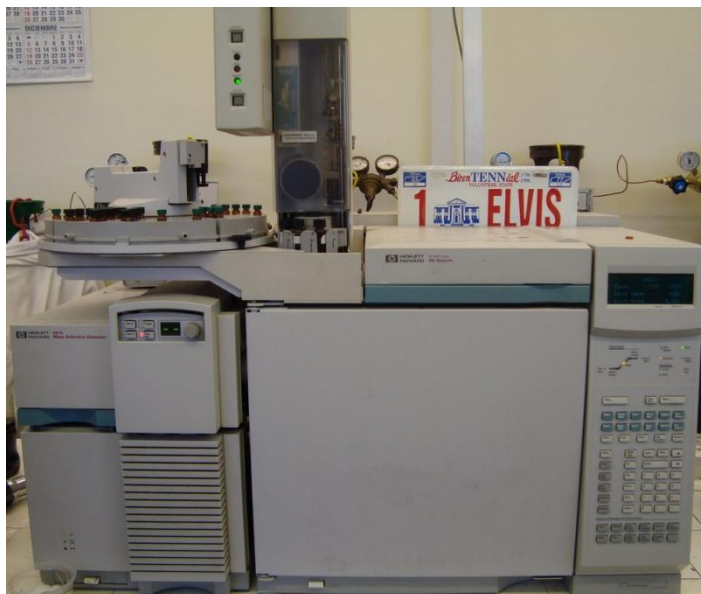
 Citrate QuEChERS

 Original QuEChERS





GC/MS



Difenoconazole
Kresoxim methyl
Trifloxystrobin
λ cyhalothrin

GC-MS HP 6890-5793

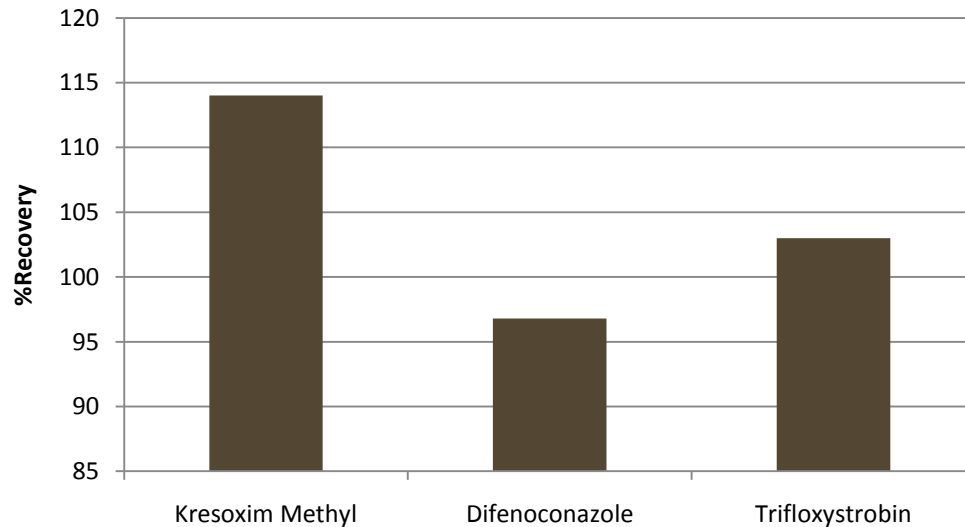
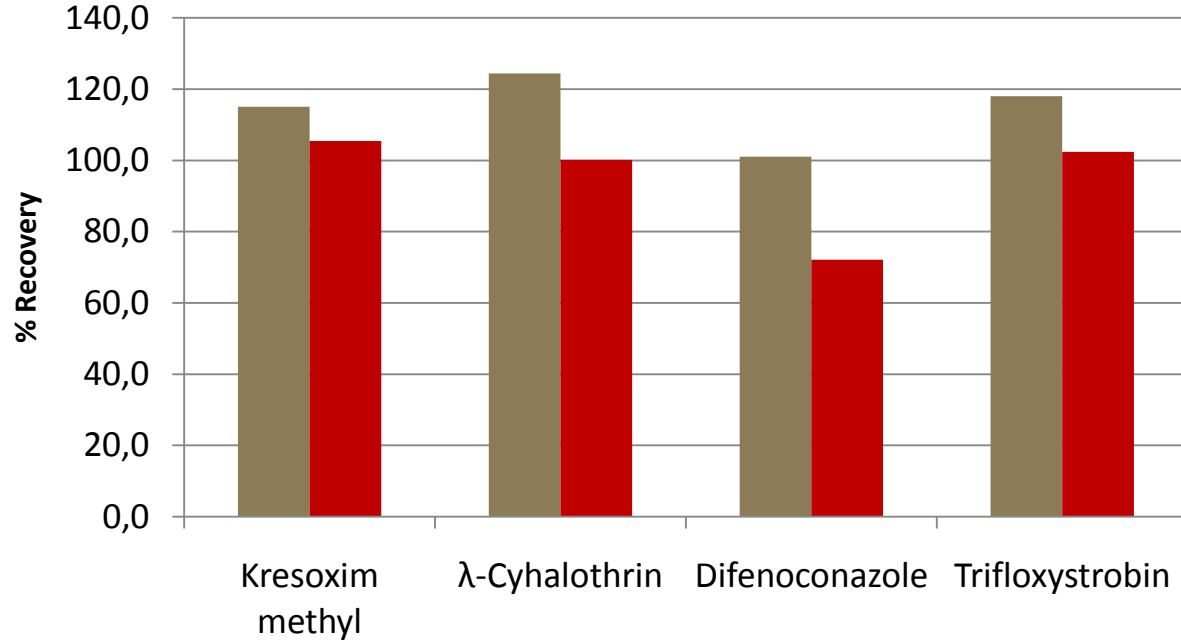
Column: HP-5: 5% Phenyl 95 % Polydimethylsiloxane.

Run Time:41 min.

El mode.

Recoveries White and Paddy Rice

200 µg/kg
RSD <20%

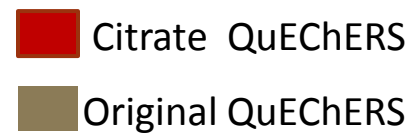


200 µg/kg
RSD <20%

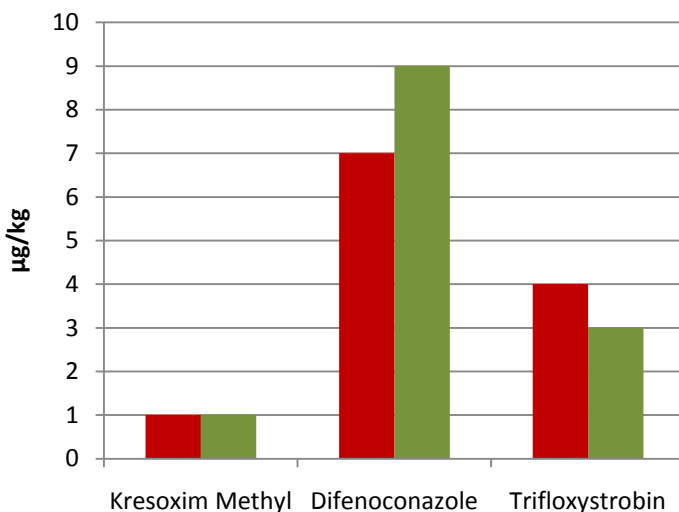
 Citrate QuEChERS
 Original QuEChERS



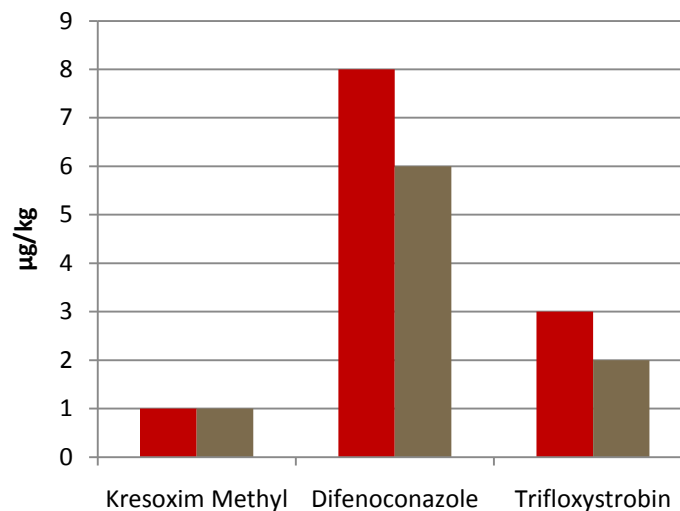
LOD & LOQ



White Rice



Paddy Rice

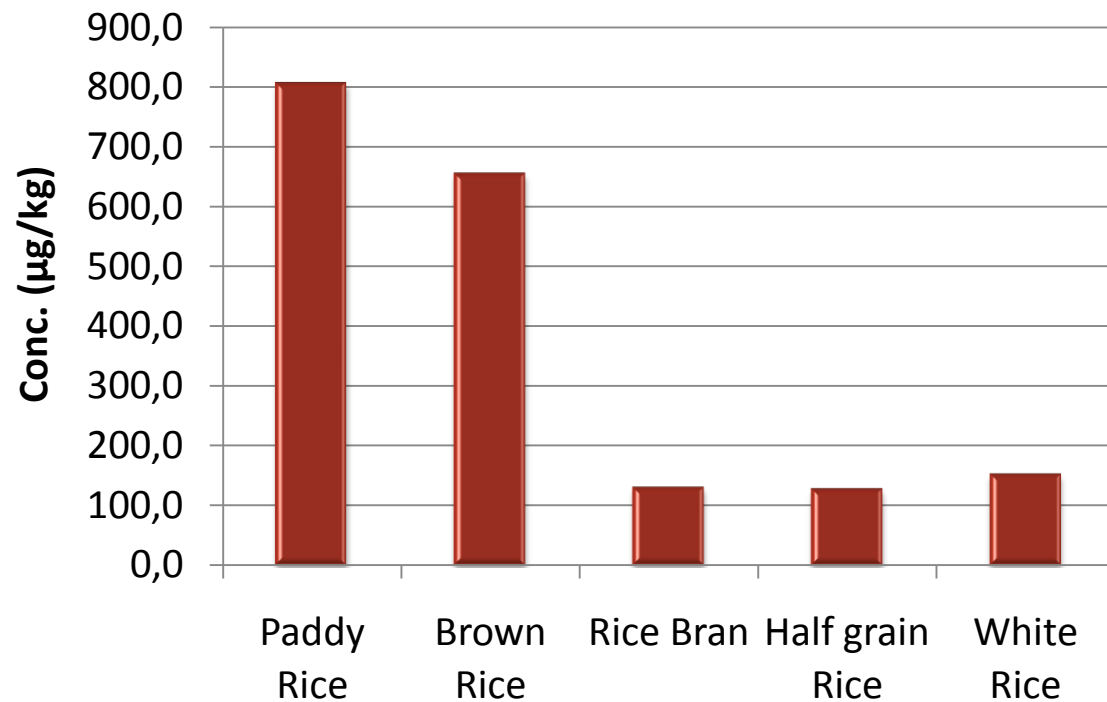


Linearity

Correlation coefficients were higher than 0.999 in the range 10-1500 µg/kg.

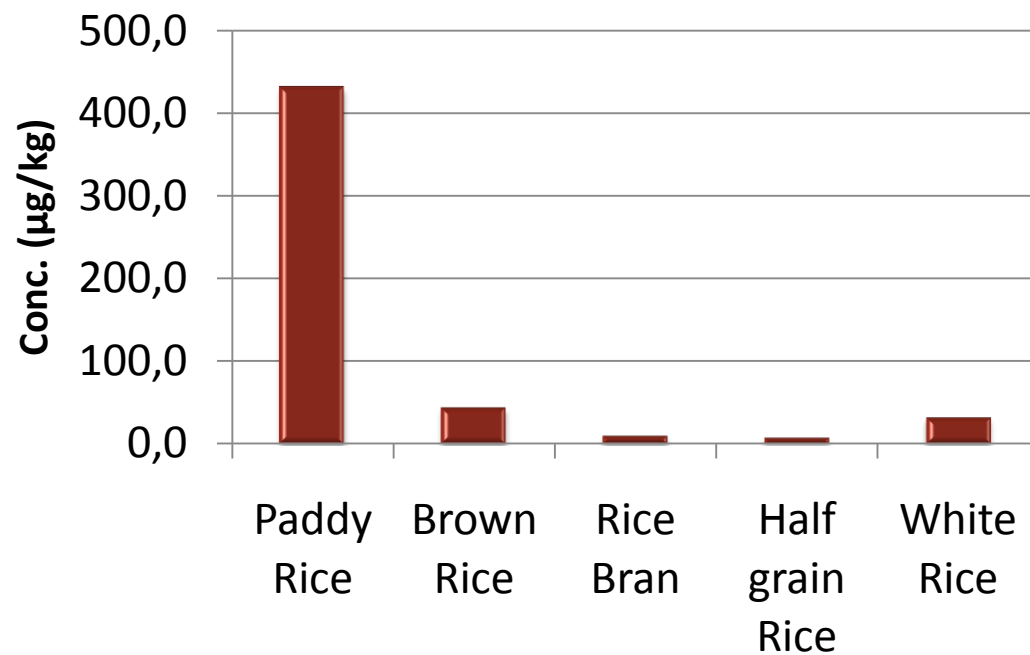
Pesticide Distribution

Pesticide	Method	Paddy Rice μg/kg	Brown Rice μg/kg	Rice Bran μg/kg	Half grain Rice μg/kg	White Rice μg/kg	pKow	MRL
Azoxystrobin	Original QuEChERS	210.3	8.9	16.8			2.5	✓
Carbendazim	Original QuEChERS	718.9	80.8	110.3	10.4	11.7	1.4	✗
Difenoconazole	Citrate QuEChERS	138.6	21.7	19.3			4.4	✓
Epoxiconazole	Original QuEChERS	431.4	44.2	10.1	24.0	32.2	3.3	✓
Isoprothiolane	Citrate QuEChERS	806.7	655.6	131.4	128.5	153.1	3.3	✓
Tebuconazole	Citrate QuEChERS	774.2	178.2	5.9	25.4	29.4	3.7	✓
Thiamethoxam	Citrate QuEChERS	31.9		20.3			-0.1	✓
Tricyclazole	Citrate QuEChERS	638.6	34.2	262.3	9.0	8.5	1.4	✓



Isoprothiolane
distribution
Citrate QuEChERS

Epoxiconazole
distribution
Citrate QuEChERS



Conclusions and Future work



✓ A distribution of the applied pesticides on the different rice products was demonstrated.



✓ Different factors could explain this distribution: type of application, mode of action, weather conditions, matrix composition, etc.



✓ None of the herbicides studied were found in the different commodities.



Conclusions and Future work



- ✓ Paddy rice presented the highest concentrations.
- ✓ In general the concentration decreased with rice processing.
- ✓ No direct relationship was found between the concentration of residues found and their lipophilicity (K_{ow}).

This work should be continued, trying to find the best extraction conditions in our laboratory to analyse λ -cyhalothrin.

Acknowledgements

✓ AECID



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✓ MGAP

✓ UNIVERSIDAD DE ALMERÍA

✓ UNIVERSIDAD DE LA REPÚBLICA



**Thank you for your
attention !!**

Hope you enjoy Montevideo!!!