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Validated Method for the Determination of 46 Pesticides in Surface, Subterranean and Wastewater with No Sample Preparation

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1. Introduction

- Pesticides are commonly used with both agricultural and non-agricultural purposes worldwide and may have potential toxic effects for both the environment and communities in the surrounding areas, making them important contaminants to bodies of water.[1, 2.]
- The highly toxic potential of the pesticides brings together a big number of international and local legislations demanding the monitoring of this class of compounds in a variety of types of aqueous matrices.[3, 4]
- Several regulations demand the monitoring of these components in different types of bodies of water including EPA and Brazilian Ministry of Health.[1, 2, 3, 4]
- This work brings a fast and robust method measuring 46 pesticides in less than 6 minutes without sample preparation for three different water matrices.

2. Methods

- Analyses were performed using a Nexera™ XR UHPLC coupled with LCMS-8050 (Shimadzu®, Japan), equipped with an electrospray ionization (ESI) source (**Fig 1**). All conditions can be seen in Table 1.
- The LabSolutions software was used for sample injections, MRM event optimization, and data processing. The Connect and Insight software platforms were employed for interface condition optimization and data analysis, respectively.
- The water samples used in these analyses were filtered through 0.45 µm and 0.22 µm PVDF filters, transferred to 1.5 mL vials, and subsequently injected into the LC-MS system with no sample preparation.

Fig 1. Nexera™ XR coupled with LCMS-8050.

Table 1. UHPLC (Nexera™ XR system) and LCMS 8050 conditions.

Analytical Column:	Shim-Pack GIST C18AQ HP (50 x 2.1 mm, 1.9 µm)
Mobile Phases	A: 5mM ammonium acetate + 0.05% acetic acid in water
	B: 5mM ammonium acetate + 0.05% acetic acid in acetonitrile
Time Program:	0%B (0 -1 min)→ 98%B (1-2.3 min)→ 98%B (2.3-4.0 min) → 0%B (4.0.-4.01 min) → Stop (5.5 min)
Flow Rate:	0.35 mL/min
Injection Vol.:	15 µL
Column Temp.:	50 °C
Interface Voltage	4 kV
DL temperature	120 ° C
Block Heater	400 ° C
Interface	400 °C
Nebulizer gas	3 L/min
Drying gas	17 L/min
Heating gas	3 L/min

3. Results

- Calibration curves were prepared in duplicate using surface water, subterranean, and effluent samples.
- The samples showed good agreement with the calibration curves, with $r^2 \geq 0.99$ for all tested water matrices, as shown in **Table 2.**.
- To improve method reliability, a repeatability study was conducted using four replicates at 7.5 ng/mL, evaluating the mean, relative standard deviation (RSD), and accuracy, with an acceptable variation range of 75–125%. Only permethrin in the surface water matrix exceeded the allowed deviation, possibly due to losses during the filtration step. More complex matrices were found to increase RSD and reduce precision, as observed for ametryn, metolachlor, and simazine in the effluent water matrix.

Table 2. Calibration range used for each analyte in different types of water matrices (continue).

Analyte	Surface water		Subterranean		Effluent	
	Calibration range (µg/L)	r^2	Calibration range (µg/L)	r^2	Calibration range (µg/L)	r^2
2,4,5 T	0.5-10	0.9929	0.5-10	0.9979	0.5-10	0.9977
2,4,5 TP	0.5-10	0.9985	0.5-10	0.9947	0.5-10	0.9998
2,4-D	1-20	0.9935	1-20	0.9986	1-20	0.9999
Acephate	0.5-25	0.9988	0.5-10	0.9927	2-50	0.9873
Alachlor	0.02-2	0.9931	0.02-2	0.9971	0.1-5	0.9994
Aldicarbe	0.5-10	0.9919	0.5-10	0.9965	0.5-10	0.9833
Aldicarb Sulfone	0.5-10	0.9963	0.5-10	0.9993	0.5-10	0.9940
Aldicarb Sulfoxide	0.5-10	0.9985	0.5-10	0.9951	0.5-10	0.9998
Ametrine	0.02-2	0.9985	0.02-2	0.9906	0.1-5	0.9997
Atrazine	0.1-5	0.9928	0.1-5	0.9927	0.1-5	0.9999
2-Hydroxyatrazine	0.1-5	0.9972	0.1-5	0.9990	0.5-10	0.9998
Desethylisopropylatrazine	0.5-25	0.9936	0.5-10	0.9972	2-50	0.9964
Desethylatrazine	0.1-5	0.9956	0.1-5	0.9999	0.5-10	0.9867
Desisopropylatrazine	0.1-10	0.9960	0.1-5	0.9932	0.5-10	0.9896
Bentazone	0.1-10	0.9924	0.1-5	0.9977	0.1-5	0.9906
Carbaryl	0.02-2	0.9952	0.02-5	0.9976	0.02-2	0.9923
Carbendazim	0.02-2	0.9973	0.02-2	0.9996	0.1-5	0.9985
Carbofurane	0.1-10	0.9987	0.1-5	0.9987	0.5-10	0.9993
Ciproconazole	0.1-10	0.9903	0.1-5	0.9964	0.1-5	0.9997
Chlorpyrifos	0.5-25	0.9978	0.5-25	0.9987	0.5-10	0.9984
Chlorpyrifos Oxon	0.1-5	0.9929	0.1-5	0.9956	0.1-5	0.9967
Demeton-O+S	0.02-4	0.9896	0.02-4	0.9998	0.04-4	0.9995
Difenconazole	0.02-2	0.9933	0.1-5	0.9939	0.1-5	0.9969

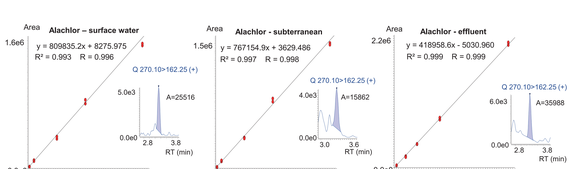

Fig 2. Linearity obtained for Alachlor in surface water, groudwater and effluent samples (0.02 – 2 ppb).

Table 2. Calibration range used for each analyte in different types of water matrices.

Analyte	Surface water		Subterranean		Effluent	
	Calibration range (µg/L)	r^2	Calibration range (µg/L)	r^2	Calibration range (µg/L)	r^2
Dimetoate	0.02-2	0.9955	0.1-5	0.9936	0.1-5	0.9991
Diuron	0.1-10	0.9980	0.1-5	0.9929	0.5-10	0.9999
Epoxiconazole	0.5-10	0.9912	0.1-5	0.9916	0.1-5	0.9975
Fipronil	0.1-10	0.9933	0.1-10	0.9977	0.1-5	0.9990
Flutriafol	0.5-10	0.9989	0.5-10	0.9976	1-25	0.9992
Malathion	0.02-2	0.9984	0.1-5	0.9842	0.02-2	0.9983
Metamidophos	1-25	0.9979	1-10	0.9982	1-25	0.9908
Metolachlor	0.1-5	0.9932	0.1-5	0.9969	0.1-5	0.9994
Metribuzin	1-25	0.9948	1-50	0.9952	2-50	0.9992
Molinate	1-25	0.9999	1-50	0.9999	1-25	0.9999
Ometoate	0.1-5	0.9891	0.1-5	0.9988	0.5-10	0.9996
Permethrin	1-50	0.9866	1-50	0.9991	1-25	0.9991
Picloram	2-50	0.9993	2-50	0.9988	2-50	0.9910
Profenofos	0.1-5	0.9904	0.1-10	0.9988	0.1-10	0.9996
Propanyl	0.5-10	0.9972	1-50	0.9874	1-25	0.9984
Propargite	0.1-10	0.9995	0.1-10	0.9906	1-25	0.9985
Protioconazole	0.5-10	0.9995	1-50	0.9915	1-25	0.9977
Protioconazole Destio	0.5-10	0.9987	0.5-10	0.9987	1-25	0.9995
Simazine	0.5-10	0.9917	0.5-10	0.9977	0.5-10	0.9998
Tebuconazole	0.5-10	0.9956	0.5-10	0.9946	0.5-10	0.9994
Terbufos	0.5-10	0.9966	0.5-50	0.9952	1-25	0.9949
Thiamethoxam	0.1-10	0.9902	0.1-5	0.9972	0.5-10	0.9990
Thiodicarb	0.1-10	0.9984	0.1-5	0.9966	0.1-5	0.9978

4. Conclusion

- The LCMS-8050 accurately quantified 46 analytes regulated by drinking water and water quality standards set by the Brazilian Ministry of Health and the National Environment Council, using a rapid method with analysis time under 6 minutes. The method proved to be robust and reliable, showing good reproducibility, negligible carryover, and a consistent linear working range across all water matrices.

Reference

1) CONSELHO NACIONAL DO MEIO AMBIENTE. Resolução CONAMA nº 357. Brasília, 2005.
2) CONSELHO NACIONAL DO MEIO AMBIENTE. Resolução CONAMA nº 396. Brasília, 2008. 3)
3) CONSELHO NACIONAL DO MEIO AMBIENTE. Resolução CONAMA nº 430. Brasília, 2011.
4) MINISTÉRIO DA SAÚDE. Portaria GM/MS nº 888. Brasília, 2021.