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### Glyphosate Contamination of Well Water from Various Agricultural Areas – A Case Study in Morocco

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### ABSTRACT

Glyphosate is a non-selective broad-spectrum herbicide widely used for weed control. It is currently one of the most important and widely sold herbicides in the world. Due to uncontrolled use and poor waste disposal, this herbicide has the potential to reach aquatic ecosystems, either surface water or groundwater, such as well water. The objective of this study was to show the degree of contamination of well water in 7 different agricultural areas of the Rabat-Sale-Kenitra region in Morocco. This is a prospective study carried out on 82 samples collected. The determination of glyphosate concentrations in these well waters was performed using the Enzyme-Linked Immunosorbent Assay (ELISA) technique. The results reveal that 83% of the samples collected showed traces of glyphosate (>0.075  $\mu$ g/l) with concentrations ranging from 0.075  $\mu$ g/l to 3.828  $\mu$ g/l. The highest glyphosate contamination of well water in Morocco. These results demonstrated glyphosate contamination of well water in Morocco. These results demonstrated glyphosate contamination of collected well water and the requirement to implement concrete sanitary measures (control, awareness campaigns...) to better manage the use of glyphosate and limit water contamination, the human health risks and environmental impact of this herbicide.

Keywords: glyphosate, well water, water contamination, ELISA, agricultural area, Morocco.

### INTRODUCTION

The modernization of agriculture relies on the use of chemicals such as fertilizers and pesticides to improve yields and crop quality. It has been widely demonstrated that these products are necessary to increase global agricultural productivity, but on the other hand, they have a negative impact on the biodiversity of ecosystems and can contaminate different water resources, thus posing a serious threat to the environment and human health. One of these phytosanitary products, glyphosate (N-(phosphonomethyl)glycine,  $C_3H_8NO_5P$ ) is a post-emergence, non-selective and broad-spectrum herbicide widely used worldwide to control

weeds in agriculture, forestry, along roadsides, railroads, utility corridors, in urban areas (roads, sidewalks, paved areas, gardens) and industrial areas (Duke and Powles, 2008; Madani and Carpenter, 2022). It was initially discovered in 1950 by Henri Martin, a Swiss researcher working at Cilag (a pharmaceutical company) (Franz et al., 1997), but was not tested or at least patented for herbicidal use. It was only in the 1970s that the chemist John Franz, recognized the herbicidal properties of glyphosate, synthesized and first tested the molecule as a herbicide (Franz et al., 1997). The molecule was patented shortly thereafter and first sold as a non-selective post-emergence herbicide by Monsanto beginning in 1974

under the name "Roundup" (Duke and Powles, 2008). Glyphosate, which is commonly administered to weed foliar parts, can enter plants via four different pathways: trunk, leaves or other green tissue, shoots, or roots (Sharma and Singh, 2001). Following application and penetration, it moves to the plant's active growth areas (Kanissery et al., 2019). The supposed mode of action of glyphosate in plants is the destruction of the shikimate pathway through inhibition of the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) that catalyzes the sixth step of the shikimic acid pathway (Kanissery et al., 2019; Gandhi et al., 2021). This alters the production of 3 essential aromatic amino acids (phenylamine, tyrosine, and tryptophan) produced by the shikimate pathway and required for plant growth (Matozzo et al., 2020). Indeed, this process impairs protein synthesis and growth, and ultimately leading to cell death. The plants treated with glyphosate normally die within 1-3 weeks, and no part of the plant can survive due to its homogeneous distribution throughout the plant (Chang and Liao, 2002).

Furthermore, glyphosate represents the most popular and widely sold herbicide in the world, and the development of genetically modified "Roundup<sup>TM</sup>-ready" cultures has only further increased its use to kill weeds and preserve cultures (Madani and Carpenter, 2022). At the same time, since 1996 and with the ongoing glyphosate use, 38 glyphosate-resistant weeds emerged in the croplands treated with the herbicide in 37 countries and 34 different cultures. (Heap and Duke, 2018). However, there is no evidence on the increase of glyphosate application rates to control resistance in glyphosate-resistant monocultures, but this approach is likely to be ineffective in the long term and may even lead to increased risks of soil and water contamination by the molecule (Maggi et al., 2020). The toxicity of glyphosate has long been a matter of controversy. An International Agency for Research on Cancer (IARC) study classified glyphosate and glyphosate-based herbicides (GBHs) as probably carcinogenic to humans (Group 2A) (International Agency for Research on Cancer (IARC), 2015). Much controversy has ensued regarding the carcinogenic nature of glyphosate. Other studies have also linked glyphosate exposure to the development of several complications, including adverse effects on the endocrine system and thyroid cells (Coperchini et al., 2023). In addition, a meta-analysis published in 2017 supported the hypothesis that the

exposure to glyphosate has a toxic effect on the reproductive system of male rodents by reducing their level of sperm (Cai et al., 2017). Similarly, another study suggested that glyphosate is a sexspecific endocrine disruptor with androgenic effects in humans (Lesseur et al., 2021). A review of several studies aimed at determining the impact of glyphosate and GBHs on the nervous system showed that, in addition to cancer, reproductive, and endocrine effects, these herbicides have significant brain and behavioral adverse effects, as well as increase the risk of some severe neurological diseases such as Parkinson's (Madani and Carpenter, 2022). The most exposed population are farmers and green space professionals (landscapers, gardeners of private properties, public parks, lawns of sports fields as well as road maintenance and municipalities) (De Graaf et al., 2020). The absence of hygiene and safety measures during application increases the risks associated with this exposure. Indeed, a study conducted in Azaguié in the south of Côte d'Ivoire, for example, showed that during the application of plant protection products in this region, no hygiene and safety measures were observed (Soro et al., 2019). In the same study, about 88% of the horticulturalists responding to the survey did not wear gloves, 71% did not use nose covers, and 94% remained without special clothing during the spraying process (Soro et al., 2019).

According to Directive 98/83/EC, the maximum contamination level (MCL) for glyphosate residues set by the European Union in drinking water samples is 0.1 µg/l (Council Directive 1998; Cengiz et al., 2017). Glyphosate and its metabolites (AMPA) have been detected in several surface and groundwater samples, including in Europe (Sanchís et al., 2012; Poiger et al., 2017; Suciu et al., 2023), China (Geng et al., 2021), Canada (Van Stempvoort et al., 2016), the United States (Battaglin et al., 2014), Brazil (Olivo et al., 2015) and Argentina (Demonte et al., 2018; Okada et al., 2018). Therefore, the study of water contamination, and more specifically, well water near agricultural areas, by glyphosate represents a major interest for scientific research, control and food safety. It is in the same register, and with the few studies carried out in Morocco that the presented study is registered. Its objective was to determine the degree of contamination of well waters in different agricultural rural areas of the region of Rabat-Sale-Kenitra in Morocco.

### MATERIALS AND METHODS

#### Materials

#### Sampling

A total of 82 well water samples were collected during the period from September 2022 to October 2022 from 7 different agricultural areas (Bouchouk, Bouknadel, Oulja (Onk Jmel) - Bribri, Kenitra, Skhirat, Tiflet and Khemissat) in the Rabat-Sale-Kenitra region of Morocco (Figure 1). The region is delimited to the north by the Tangier-Tetouan-Al Hoceima region, to the southeast by the Fez-Meknes region, to the south by the two regions of Beni Mellal-Khenifra and Casablanca-Settat, and to the west by the Atlantic Ocean.

### Glyphosate content determination

The glyphosate assay kit is from the company "Novakits France". This glyphosate ELISA microtiter plate kit from Eurofins Abraxis is an enzyme-linked immunosorbent assay for the quantitative/qualitative sensitive detection of glyphosate in water samples (groundwater, surface water, well and tap water). Glyphosate concentrations less than standard 1 (0.075 µg/l) are reported as containing <0.075 µg/l of glyphosate. Previous studies have already demonstrated the validity (even in comparison with HPLC-based method) and efficiency of this kit for the determination of glyphosate (Rendon-von Osten and Dzul-Caamal, 2017; Reynoso et al., 2020).

#### Measuring instrument

The Diareader ELX800G is an automated microplate reader designed by DiaLab. The instrument is a general photometric, microprocessor-controlled system designed to read and calculate the result of enzyme-linked immunosorbent assay (ELISA) tests. It is capable of reading a variety of plate formats (such as 96-well plates). It provides high precision and accuracy, with a wide dynamic range to accommodate samples of varying concentrations.

### Method

#### Preparation of the dosage

Before use, the samples, microtiter plate, and reagents are left at room temperature. The concentrated wash buffer (5X) is diluted at a ratio of 1:5. The required amount of microtiter plate strips is removed from the foil bag. Standards, controls and samples are derivatized before each run:

- derivatization reagent is diluted with 3.5 ml of derivatization reagent diluent and vortexed;
- a 250 µl volume of control, standard or sample is pipetted into a labeled glass test tube;
- 1 ml of assay buffer is added to each test tube and vortexed to mix;
- 100 µl of diluted derivatization reagent is added to each test tube and vortexed;
- Everything is incubated at room temperature for 10 minutes.

### Dosage procedure

A volume of 50  $\mu$ L of the standards (0, 0.075, 0.20, 0.5, 1.0, and 4.0 µg/l), control, or derivatized samples are added to the test strip wells. Then, 50 µl of the antibody solution is successively added to each well. The wells are then covered with parafilm, mixed for 60 seconds in a circular motion, and then left to incubate for 30 minutes at room temperature. Afterwards, 50 µl of the enzyme conjugate solution is successively added to each well. The wells are again covered, mixed for 60 seconds and then incubated for 60 minutes at room temperature. After the second incubation, the parafilm is removed, the contents of wells are decanted and the plate is inverted and blotted on paper towels. The wells are then washed three times with the diluted wash buffer (at least 250 µl for each well and wash step). Afterwards, 150 µl of substrate solution (color) is added successively to each well. These wells are subsequently covered with parafilm and mixed by moving the wells in a circular motion for 30 seconds. Thereafter, the wells are incubated for 30 minutes at room temperature and out of sunlight. Finally, 100 µl of stop solution is added to the wells in the same order as the substrate solution (color) and the absorbance is observed at 450 nm using a microtiter plate ELISA photometer.

### RESULTS

### Glyphosate content in $\mu$ g/l of well water samples collected in the Bouchouk region

A total of 11 well water samples were collected in the Bouchouk region. The concentration of glyphosate in the well water collected in the region varied between 0.075  $\mu$ g/l and 0.451  $\mu$ g/l (Table 1).



Figure 1. Mapping of water sampling sites in the Rabat-Sale-Kenitra region

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Samples	Location	Coordinates	Glyphosate in µg/l
S1	Bouchouk	NL : 34°5'58" WL : 6°46'8"	0.109
S2	Bouchouk	NL : 34°5'36" WL : 6°46'37"	0.086
S3	Bouchouk	NL : 34°5'36" WL : 6°46'8"	0.103
S4	Bouchouk	NL : 34°6'7" WL : 6°46'4"	0.143
S5	Bouchouk	NL : 34°7'8" WL : 6°45'7"	0.075
S6	Bouchouk	NL : 34°6'50" WL : 6°44'56"	0.321
S7	Bouchouk	NL : 34°6'50" WL : 6°44'56"	0.451
S8	Bouchouk	NL : 34°6'29" WL : 6°45'15"	0.381
S9	Bouchouk	NL : 34°6'36" WL : 6°45'10"	0.367
S10	Bouchouk	NL : 34°6'36" WL : 6°45'10"	0.342
S11	Bouchouk	NL : 34°7'35" WL : 6°44'41"	0.287

Table 1. Glyphosate content in µg/l and geographic data of the well water samples collected in the Bouchouk region

# Glyphosate content in $\mu$ g/l of well water samples collected in the Bouknadel region

A total of 10 well water samples were collected in the Bouknadel region. The concentration of glyphosate in the well waters collected in the region varied between  $<0.075 \mu g/l$  and  $0.196 \mu g/l$  (Table 2).

## Glyphosate content in $\mu g/l$ of well water samples collected in the Oulja (Onk Jmel) - Bribri region

A total of 11 well water samples were collected from the Oulja (Onk Jmel) - Bribri region. The concentration of glyphosate in the well waters collected in the region varied between <0.075  $\mu$ g/l and 0.088  $\mu$ g/l (Table 3).

Samples	Location	Coordinates Glyphosate in µg/l	
S12	Bouknadel	NL : 34°6'41" WL : 6°44'29"	0.155
S13	Bouknadel	NL : 34°6'41" WL : 6°44'29"	0.187
S14	Bouknadel	NL : 34°6'41" WL : 6°44'17"	0.196
S15	Bouknadel	NL : 34°6'34" WL : 6°44'5"	0.170
S16	Bouknadel	NL : 34°6'34" WL : 6°44'5"	<0.075
S17	Bouknadel	NL : 34°7'18" WL : 6°43'51"	<0.075
S18	Bouknadel	NL : 34°7'8" WL : 6°43'2"	0.080
S19	Bouknadel	NL : 34°6'55" WL : 6°43'19"	<0.075
S20	Bouknadel	NL : 34°6'50" WL : 6°43'20"	0.091
S21	Bouknadel	NL : 34°6'45" WL : 6°43'26"	0.130

Table 2. Glyphosate content in  $\mu$ g/l and geographic data of the well water samples collected in the Bouknadel region

Table 3. Glyphosate content in  $\mu g/l$  and geographic data of the well water samples collected in the Oulja (Onk Jmel) - Bribri region

Samples	Location	Coordinates	Glyphosate in µg/l
S22	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S23	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	0.088
S24	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S25	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S26	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S27	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S28	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S29	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	<0.075
S30	Oulja – Onk Jmel	NL : 33°59'17" WL : 6°48'23"	0.087
S31	Bribri	NL : 33°59'13" WL : 6°45'43"	<0.075
S32	Bribri	NL : 33°59'55" WL : 6°45'6"	<0.075

## Glyphosate content in $\mu g/l$ of well water samples collected in the Kenitra region

A total of 11 well water samples were collected in the Kenitra region. The concentration of glyphosate in the well waters collected in the region varied between 0.134  $\mu$ g/l and 3.828  $\mu$ g/l (Table 4).

### Glyphosate content in $\mu$ g/l of well water samples collected in the Skhirat region

A total of 14 well water samples were collected in the Skhirat region. The concentration of glyphosate in the well waters collected in the region varied between 0.152  $\mu$ g/l and 0.372  $\mu$ g/l (Table 5).

## Glyphosate content in $\mu g/l$ of well water samples collected in the Tiflet region

A total of 14 well water samples were collected in the Tiflet region. The concentration of glyphosate in well water collected in the region varied between <0.075  $\mu$ g/l and 0.467  $\mu$ g/l (Table 6).

# Glyphosate content in µg/l of well water samples collected in the Khemissat region

A total of 11 well water samples were collected from the Khemissat region. The concentration of glyphosate in well water collected in the region varied between  $< 0.075 \mu g/l$  and  $0.204 \mu g/l$  (Table 7).

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Samples	Location	Coordinates	Glyphosate in µg/l
S33	Laababda - Kenitra	NL : 34°19'25" WL : 6°30'37"	0.245
S34	Laababda – Kenitra	NL : 34°18'30" WL : 6°30'23"	0.149
S35	Laababda – Kenitra	NL : 34°19'25" WL :6°30'37"	0.554
S36	Laababda – Kenitra	NL : 34°19'25'' WL :6°30'37''	3.828
S37	Laababda – Kenitra	NL : 34°19'25" WL : 6°30'37"	0.189
S38	Laababda – Kenitra	NL : 34°19'25" WL : 6°30'37"	0.363
S39	Laababda – Kenitra	NL : 34°19'25" WL : 6°30'37"	0.221
S40	Laababda – Kenitra	NL : 34°19'25'' WL : 6°30'37''	0.283
S41	Laababda – Kenitra	NL : 34°18'57'' WL : 6°30'37''	0.144
S42	Kenitra	NL : 34°19'06'' WL : 6°30'08''	0.194
S43	Kenitra	NL : 34°19'33'' WL : 6°29'08''	0.134

Table 5. Glyphosate cont	tent in µg/l and g	geographic data of the w	vell water samples c	collected in the Sk	hirat region
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Samples	Location	Coordinates	Glyphosate in µg/l
S44	Skhirat	NL : 33°48'01" WL : 7°01'05"	0.158
S45	Skhirat	NL : 33°47'32" WL : 6°59'57"	0.163
S46	Skhirat	NL : 33°47'32" WL : 6°59'59"	0.175
S47	Skhirat	NL : 33°47'41" WL : 7°00'12"	0.264
S48	Skhirat	NL : 33°47'43" WL : 7°00'13"	0.187
S49	Skhirat	NL : 33°47'43" WL : 7°00'05"	0.208
S50	Skhirat	NL : 33°47'54" WL : 7°00'08"	0.154
S51	Skhirat	NL: 33°47'54" WL: 7°00'08"	0.165
S52	Skhirat	NL : 33°47'59" WL : 7°00'14"	0.152
S53	Skhirat	NL : 33°47'59" WL : 7°00'14"	0.237
S54	Skhirat	NL : 33°47'56" WL : 7°00'18"	0.372
S55	Skhirat	NL : 33°47'56" WL :7°00'18"	0.206
S56	Skhirat	NL : 33°47'56" WL : 7°00'18"	0.325
S57	Skhirat	NL : 33°47'53" WL : 7°00'11"	0.274

# Comparison of maximum glyphosate content between different sampling regions

The highest levels of glyphosate were observed in the region of Kenitra, followed by Tiflet, Bouchouk, Skhirat, Khemissat, Bouknadel and Oulja (Onk Jmel) - Bribri with concentrations of 3.828  $\mu$ g/l, 0.467  $\mu$ g/l, 0.451  $\mu$ g/l, 0.372  $\mu$ g/l, 0.204  $\mu$ g/l, 0.196  $\mu$ g/l and 0.088  $\mu$ g/l, respectively. The area of Oulja (Onk Jmel) - Bribri had the lowest levels of glyphosate with concentrations ranging from <0.075  $\mu$ g/l to 0.088  $\mu$ g/l (Figure 2).

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Samples	Location	Coordinates	Glyphosate in ug/l
S58	Ain Johra - Tiflet	NL : 33°55'03" WL : 6°24'51"	0.197
S59	Ain johra -Tiflet	NL : 33°53'41" WL : 6°22'5"	0.148
S60	Tiflet	NL : 33°53'35" WL : 6°16'46"	0.276
S61	Tiflet	NL : 33°53'34" WL : 6°18'26"	<0.075
S62	Tiflet	NL : 33°53'47" WL : 6°16'32"	0.205
S63	Tiflet	NL : 33°55'54" WL : 6°15'21"	0.209
S64	Tiflet	NL : 33°53'55" WL : 6°15'17"	0.151
S65	Tiflet	NL : 33°53'57" WL : 6°14'43"	0.467
S66	Tiflet	NL : 33°53'57" WL : 6°14'43"	0.201
S67	Tiflet	NL : 33°53'57" WL : 6°14'43"	0.416
S68	Tiflet	NL : 33°53'58" WL : 6°14'42"	0.392
S69	Tiflet	NL : 33°54'03" WL : 6°14'18"	0.191
S70	Tiflet	NL : 33°54'06" WL : 6°14'55"	0.209
S71	Tiflet	NL : 33°53'50" WL : 6°14'26"	0.103

Table 7. Glyphosate content in µg/l and geographic data of the well water samples collected in the Khemissat region

Samples	Location	Coordinates	Glyphosate in µg/l
S72	Kemouni (sidi Allal El Bahraoui) - Khemissat	NL : 33°58'46" WL : 6°30'18"	<0.075
S73	Kemouni (sidi Allal El Bahraoui) - Khemissat	NL : 33°56'51" WL : 6°27'57"	0.115
S74	Sidi Allal Msedder - Khemissat	NL : 33°53'38" WL : 6°13'09"	0.116
S75	Sidi Allal Msedder - Khemissat	NL : 33°53'26" WL : 6°12'03"	0.160
S76	Ait Abbou — Khemissa <b>t</b>	NL : 33°53'24" WL : 6°11'56"	0.172
S77	Ait Abbou – Khemissat	NL : 33°51'26" WL : 6°07'48"	0.119
S78	Ait Abbou – Khemissat	NL : 33°51'08" WL : 6°07'44"	0.092
S79	Ait Ouribel – Khemissat	NL : 33°46'39" WL : 6°08'10"	0.204
S80	Hajama – Khemissat	NL : 33°49'12" WL : 6°06'44"	0.189
S81	Hajama – Khemissat	NL : 33°51'59" WL :6°14'47"	0.127
S82	Hajama – Khemissat	NL : 33°51'57" WL : 6°14'49"	0.095



Figure 2. Minimum and maximum Glyphosate content in different sampling regions in µg/l

#### DISCUSSION

In this study, glyphosate concentration was measured in micrograms per liter ( $\mu g/l$ ) in a total of 82 well water samples. The samples were collected from 7 different agricultural areas (Bouchouk, Bouknadel, Oulja (Onk Jmel) - Bribri, Kenitra, Skhirat, Tiflet, and Khemissat) in the Rabat-Sale-Kenitra region of Morocco. According to the 2014 census, the Rabat-Sale-Kenitra region has a population of 4,580,866, representing 13.5 % of the Moroccan population. It is the second largest region (in terms of population) in the country after the Casablanca-Settat region. In addition, as the area is known for its agricultural production, it is necessary to comprehend the glyphosate concentrations in well water in the region. Also, it is important to note that the study was conducted in several different agricultural areas (7 in total), which may allow obtaining a more complete representation of glyphosate concentrations in well water in the study area.

Out of 82 samples collected in the Rabat-Sale-Kenitra region, 83% or 68 samples showed traces of glyphosate (>0.075  $\mu$ g/l) with concentrations ranging from 0.075 µg/l to 3.828 µg/l. Depending on the area, the highest glyphosate levels were observed in sample S36 from the Kenitra agricultural area with a concentration of 3.828  $\mu$ g/l, followed by the Tiflet area with sample S65=0. 467  $\mu$ g/l, Bouchouk with sample S7=0.451  $\mu$ g/l, Skhirat with sample S54=0.372 µg/l, Ait Ouribel - Khemissat with sample S79=0.204  $\mu$ g/l and the Bouknadel area with sample S14=0.196 µg/l. In the area of Oulja (Onk Jmel) - Bribri, the well water samples collected did not show any trace of glyphosate contamination ( $<0.075 \mu g/l$ ) except for samples S23=0.088 µg/l and S30=0.087 µg/l. This

S36=3.828  $\mu$ g/l from the area of Kenitra which was collected in an area of industrial agriculture (large farm and intensive agriculture). In 2020, after conducting a comprehensive global analysis of the potential risks of environmental contamination by glyphosate and its major metabolite AMPA worldwide, a study by Maggi et al. (2020) revealed that low contamination occurs in almost all cultivated lands where glyphosate is used (Maggi et al., 2020). Several parameters, including glyphosate use, soil type and PH, glyphosate level in the soil, leaching, and weather conditions may explain the glyphosate contamination of well water. Indeed, a study by Suciu et al. (2023) reported that the use of water to prepare the glyphosate mixture, the use of glyphosate on the farm and the cleaning of spray equipment after application in the vicinity of wells would make them more vulnerable to contamination from local point sources (Suciu et al., 2023). Meanwhile, Masiá et al. (2014) showed the glyphosate contamination of groundwater in the Lombardy region, stating that point source contamination was due to losses and use of herbicides near farms, or from sprayers and trucks cleaning near wells (Masiá et al., 2014). In addition, it has been shown that the presence of these compounds in groundwater depends mainly on vertical transport processes in combination with additional variables such as land management, hydrology, and herbicide use (Van Stempvoort et al., 2016). The obtained findings are consistent with the results presented in a study carried out in Mexico, which found glyphosate in 90% of the assessed groundwater samples, with a maximum

can be explained by the fact that the samples col-

lected in the region were harvested in a traditional

and domestic agricultural area, unlike the samples

collected in the other areas studied, such as sample

concentration of 1.42 µg/l (Rendon-von Osten and Dzul-Caamal, 2017). Similarly, a study in Catalonia observed concentrations of 2.5 µg/l in groundwater (Sanchís et al., 2012). A study conducted in Argentina by Lutri et al. (2020) reported that of all the water samples collected, glyphosate was found in 66% of the surface water samples with concentrations ranging from 0.2 to 167.4  $\mu$ g/l, in 15.8% of the groundwater samples with concentrations of 1.3 to 2  $\mu$ g/l, and in the precipitation sample collected with a concentration of 0.2  $\mu$ g/l (Lutri et al., 2020). Other studies, however, have reported concentrations well above those obtained in the presented study. Notably, in Australia where Davis et al. (2013) reported concentrations of 54 µg/l of glyphosate (Davis et al., 2013). Vera-Candioti et al. (2021) found a maximum glyphosate concentration of 111 µg/l in agricultural areas in the Pampa region of Argentina (Vera-Candioti et al., 2021). Another study by Peruzzo et al. (2008) even revealed the glyphosate concentrations in water up to 700 µg/l in Argentina (Peruzzo et al., 2008).

In this study, 73% or 60 of 82 well water samples collected in the Rabat-Sale-Kenitra region, had the glyphosate levels above the 0.1  $\mu$ g/l standard described by Directive 98/83/EC (Council Directive 1998; Cengiz et al. 2017). For the case of each area, the standard was exceeded in 82% or 9 of the 11 samples collected in the Bouchouk area  $[0.075-0.451 \ \mu g/l]$ , 50% or 5 of the 10 samples collected in the Bouknadel area [<0.075-0.196  $\mu$ g/l], no samples of the 11 collected in the Oulja (Onk Jmel) - Bribri area [<0.075–0. 088 µg/l], 100% of the 11 samples collected in the Kenitra area [0.134-3.828 µg/l], 100% of the 14 samples collected in the Skhirat area  $[0.152-0.372 \ \mu g/l]$ , 93% or 13 of the 14 samples collected in the Tiflet area [ $<0.075-0.467 \mu g/l$ ] and 73% or 8 of the 11 samples collected in the Khemissat area [<0.075–  $0.204 \,\mu g/l$ ]. However, all of the obtained results remain below the American standard established by the U.S. Environmental Protection Agency which limited the maximum concentration in drinking water to 700 µg/L, and the Canadian standard established by the Ministry of the Environment set at 280 µg/L (Álvarez Bayona et al., 2022). These results suggest the importance of establishing an international standard, or at least, in the case of Morocco, a Moroccan national standard defining the maximum allowable concentration of glyphosate in water, in order to better manage the control and use of glyphosate as well as limit contamination and related human health risks. Despite the

controversy about the toxicity and carcinogenicity of glyphosate and GBHs, several previous studies have demonstrated that the exposure to this molecule can be linked to the development of several complications, including reproductive (Walsh et al., 2000), genotoxic, hormonal, enzymatic (Walsh et al., 2000; Daruich et al., 2001; Richard et al., 2005), carcinogenic (Guyton et al., 2015), and neurological (Barbosa et al., 2001) complications for human health (Rendon-von Osten and Dzul-Caamal, 2017). The results of this study show the presence and contamination of well water collected in the region of Rabat-Sale-Kenitra by glyphosate warning of the importance of the implementation of control, monitoring and awareness campaigns for the reasoned and safe use of glyphosate. This is necessary, in order to limit any potential risk to the environment and human health, especially with the significant risks of consumption of these waters by the population living near these contaminated wells.

### CONCLUSIONS

The analyses carried out in this study determined the concentrations of glyphosate in 82 well water samples collected from 7 agricultural areas in the Rabat-Sale-Kenitra region of Morocco: Bouchouk, Bouknadel, Oulja (Onk Jmel) - Bribri, Kenitra, Skhirat, Tiflet and Khemissat. This is one of the first studies carried out in the framework of the determination of glyphosate levels in well water in Morocco. The results of the analyses revealed that out of 82 well water samples collected, 83% or 68 samples showed traces of glyphosate  $(>0.075 \mu g/l)$  with concentrations ranging from 0.075  $\mu$ g/l to 3.828  $\mu$ g/l. The highest glyphosate concentrations were observed in the agricultural area of Kenitra with a concentration of 3.828 µg/l. The agricultural area of Oulja (Onk Jmel) - Bribri, did not show any trace of glyphosate contamination ( $<0.075 \mu g/l$ ) in the well water samples collected except for 2 samples which showed concentrations of 0.087 µg/l and 0.088 µg/l. This study demonstrated the degree of contamination of well water collected in different agricultural areas of the Rabat-Sale-Kenitra region in Morocco and highlighted the need for continuous monitoring and awareness campaigns to better manage the use of glyphosate and limit water contamination by this herbicide. Awareness campaigns for the population living in these areas and consuming the water from these wells can also be conducted to avoid all potential risks to human health related to the exposure to glyphosate. Similarly, raising awareness of good agricultural practices among farmers could greatly contribute to the promotion of sustainable agriculture in the area. In addition, it is important to undertake studies on the longterm effects of drinking this water on producers in this locality. These results may also indicate the importance of establishing an international standard, or at least, in the case of Morocco, a national standard setting out the maximum concentration allowed of glyphosate in water for human consumption, in order to facilitate water management and control. Additional studies should be conducted in order to expand the knowledge on water contamination and environmental impact and human health risks of glyphosate.

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