

## Introduction

Chlorate acts as an herbicide and as such has been part of pesticide formulations in the past. In Europe its use as herbicide has been banned in 2008. The default maximum residue limit of chlorate in foodstuffs is 0.01 mg/kg but only recent improvements in the analytical methodology allowed the analysis of chlorate together with other polar residues down to the low µg/kg range. Subsequently chlorate was included in screening programs and was found in food samples in concentrations up to several mg/kg. Questions arise about the source of chlorate residues in food samples. Sources for chlorate other than pesticide formulations are natural contaminations in mineral fertilizers like Chile saltpeter, and its formation as disinfection byproduct in water chlorination. Chlorination is common practice in drinking water production and is also used in southern Europe for the disinfection of rainwater or surface water used for irrigation.

In this poster we show the development and validation of an accurate analytical method for the determination of chlorate, bromate and perchlorate in water and foodstuff using <sup>18</sup>O stable isotope labelled internal standards. The method was applied to drinking water, irrigation water and food samples. It has also been applied to washing experiments to identify the main source of chlorate residues in food.

## Conclusions

An analytical method for the determination of chlorate, bromate and perchlorate in water and foodstuffs was developed and validated. A 1290 Infinity UHPLC system was coupled to the highly sensitive Agilent 6490 triple quadrupole mass spectrometer equipped with an Agilent Jet Stream electrospray ionization source and operated in negative multiple reaction monitoring mode. Separation was achieved using a mixed mode HILIC separation column and acidified ammonium formate buffer and acetonitrile as mobile phase. Limits of quantitation (LOQ) for chlorate and perchlorate in food were 1.1 µg/kg and 0.6 µg/kg, respectively. Direct injection of drinking water samples diluted 1 in 10 with acetonitrile LOQ for chlorate and perchlorate in drinking water were 0.055 µg/L and 0.03 µg/L.

Drinking water samples from Italy used for irrigation showed chlorate concentrations up to 640 µg/L. The irrigation of plants with water containing chlorate was identified as source of chlorate in food samples. There is an active uptake of chlorate by the plant via the roots which is then deposited in the generative parts used as food products. This deposition can be up to 5 to 10% of the total absorbed chlorate, which was confirmed by pre-harvest analysis.

## Results and Discussion

### Sample preparation and LC/MS/MS parameters

5 to 10 g homogenized sample

Addition of isotope labelled ISTDs

Extraction with acidified methanol

Centrifugation

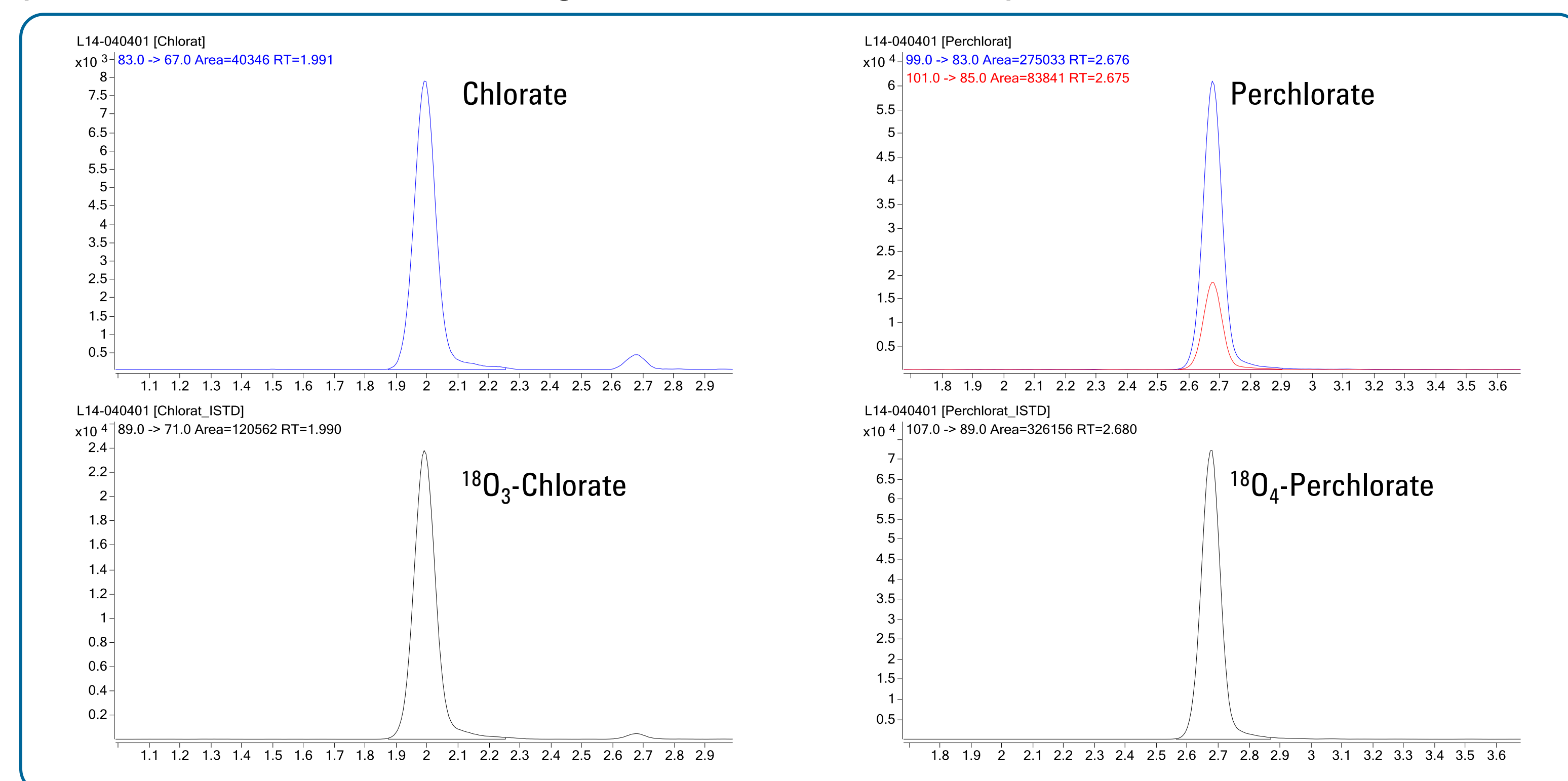
LC/MS/MS

- Mixed mode HILIC separation column (Obelisc R)
- Mobile phase A: acidified ammonium formate in water
- Mobile phase B: acetonitrile
- Total runtime: 7 min

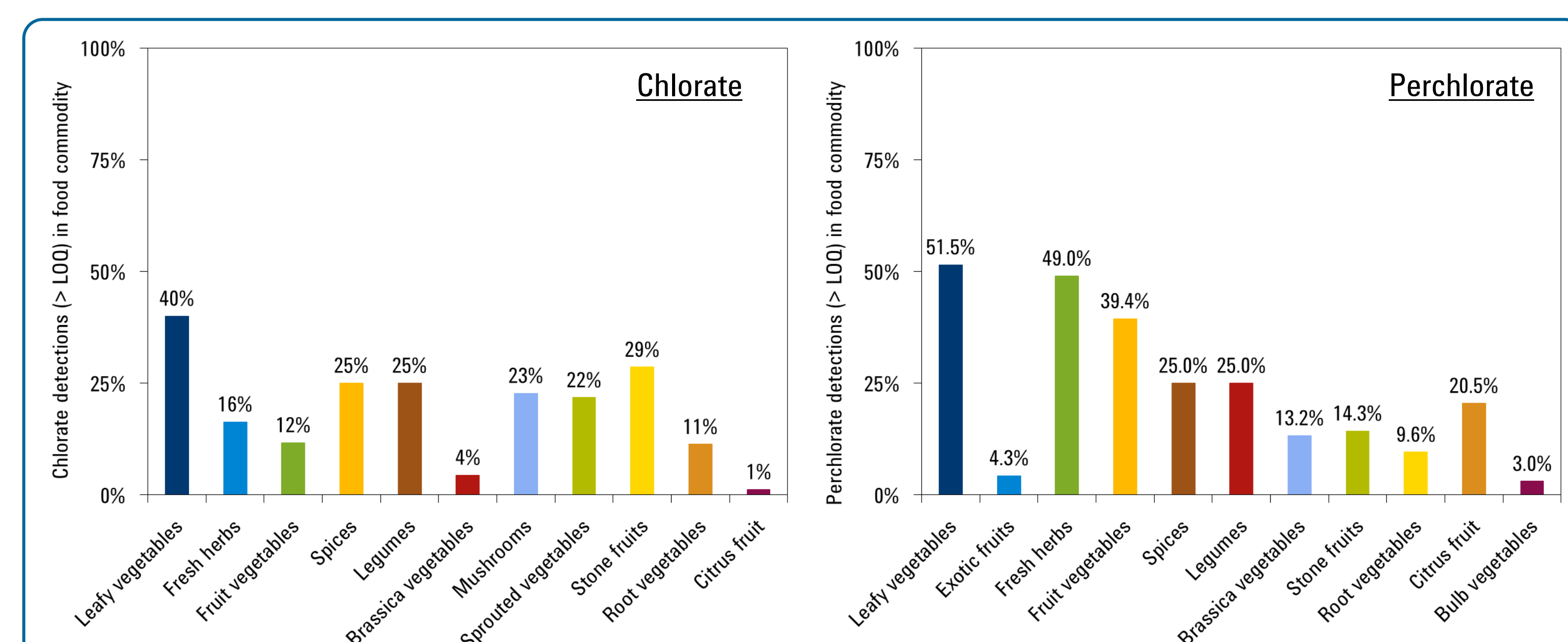
Compound	ISTD	Precursor	Product	CE
Bromate	☐	127	111	24
<sup>18</sup> O <sub>2</sub> -Perchlorate	☑	107	89	26
Perchlorate	☐	99	83	26
Perchlorate	☐	101	85	26
<sup>18</sup> O <sub>3</sub> -Chlorate	☑	89	71	22
Chlorate	☐	83	67	22



Food samples were extracted using the Quick Polar Pesticides method (QuPPE). Water samples were diluted 1:10 with acetonitrile prior to injection. The method was validated for food and water samples and limits of quantitation (LOQ) for chlorate and perchlorate in food were 1.1 µg/kg and 0.6 µg/kg, respectively. LOQs for chlorate and perchlorate in drinking water were 0.055 µg/L and 0.03 µg/L. Figure 1 shows the chromatograms of chlorate and perchlorate for a contaminated green oak leaf lettuce sample.

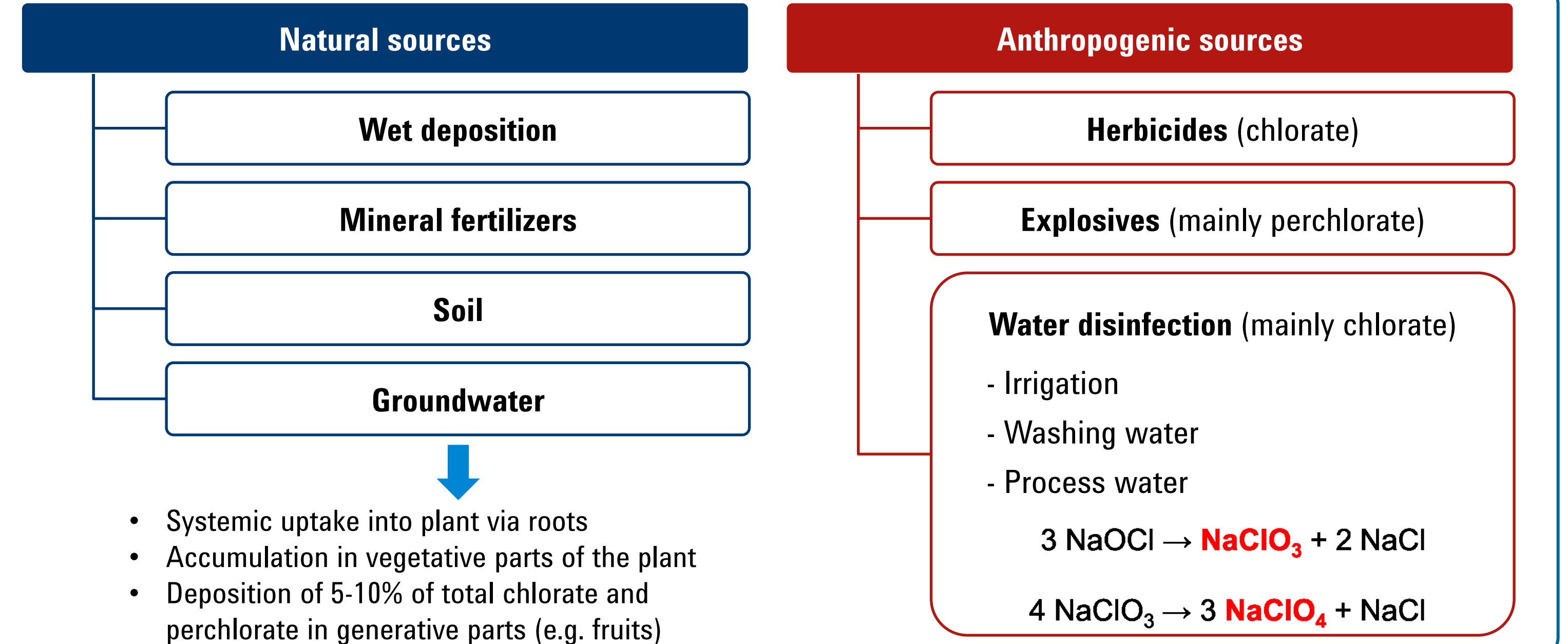


**Figure 1.** Chromatograms of chlorate, perchlorate and their internal standards in a contaminated green oak leaf lettuce sample (chlorate: 0.040 mg/kg; perchlorate: 0.076 mg/kg).



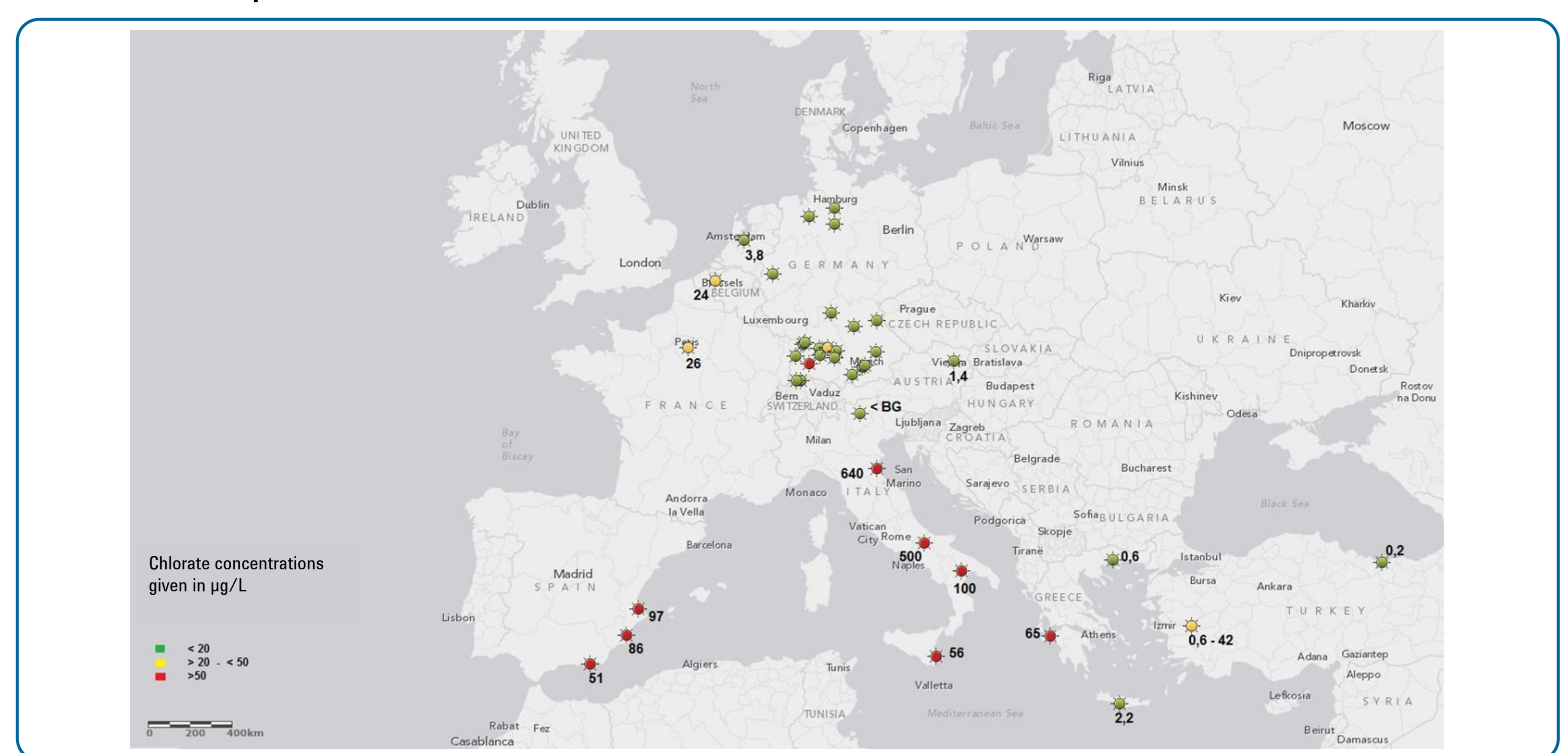
**Figure 2.** Positive chlorate and perchlorate detection (> reporting limit of 0.01 mg/kg) in different food commodities relative to number of conventionally grown samples of particular commodity (total 1,138 samples).

Figure 2 shows the ratio of positive samples for chlorate and perchlorate of different food commodities. Of the 156 positive samples for chlorate 102 (corresponding to 8.9%) were above the reporting limit for chlorate (0.01 mg/kg) whereas of the 302 positive samples for perchlorate only 1 sample exceeded the preliminary reference value of the *Standing Committee on the Food Chain and Animal Health (SCoFCAH)*.



**Figure 3.** Natural and anthropogenic sources of chlorate and perchlorate in agriculture.

Figure 3 shows the natural and anthropogenic sources of chlorate and perchlorate in the environment and agriculture. Due to the multiple use of water in food production and processing, chlorate as byproduct from water disinfection might play an important role in the overall exposure to chlorate. During chlorination chlorate is formed from hypochlorite by disproportionation. Figure 4 shows a map for Europe with chlorate concentrations in disinfected drinking water. High chlorate concentrations were observed especially in southern Europe.



**Figure 4.** Chlorate concentrations in disinfected drinking waters across Europe.

### Irrigation of lettuce with disinfected drinking water

- Water:** chlorate concentration 43 µg/L; perchlorate concentration 1.2 µg/L
- Irrigation with 50 L/kg lettuce; assuming 10% systemic uptake
- Lettuce:** calculated concentration: chlorate 0.22 mg/kg; perchlorate < 0.01 mg/kg  
measured concentration: chlorate 0.06 mg/kg; perchlorate < 0.01 mg/kg  
**Measured uptake equivalent to 28% of theoretical concentration**

### Washing of lettuce with disinfected drinking water

- Water:** chlorate concentration 700 µg/L; perchlorate concentration < 0.1 µg/L
- Washing for 1 minute, draining for 5 minutes → homogenization and analysis
- Lettuce:** measured concentration without washing: < 0.01 mg/kg  
measured concentration with washing: 0.072 ± 0.004 mg/kg (n = 4)  
**Absorption of water equivalent to about 10% of product mass**



**Figure 5.** Estimation of the contribution of chlorate in disinfected irrigation and process water to the overall chlorate concentration in lettuce.

Figure 5 shows that both, irrigation and washing can contribute significantly to the overall chlorate concentration in lettuce. The results illustrate that chlorate should be considered as a contaminant rather than a pesticide residue.