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Chloronitramide Anion: A New DBP Detected in Drinking Water

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Acknowledgments

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Presentation Outline

- Introduction
- Method Development and Validation
- Matrix Interferences
- Field Sample Analysis and Results
- Conclusions

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Drinking Water D/DBPs

Haloacetic acids (HAA5) MCL = 60 µg/L
Total trihalomethanes (TTHMs) MCL = 80 µg/L
Bromate MCL = 10 µg/L
Chlorite MCL = 1.0 mg/L
Chloramines as Cl_2 MRDL = 4.0 mg/L
Chlorine as Cl_2 MRDL = 4.0 mg/L
Chlorine dioxide as ClO_2 MRDL = 0.8 mg/L

Unregulated haloacetic acids & halomethanes
Haloacetonitriles & halonitromethanes
Haloketones & haloamides
Halofuranones such as MX
Aldehydes & ketones
Nitrosamines, etc.

Unregulated Oxyhalides
Perchlorate
Chlorate
Iodate
Cyanogen chloride, etc.

Halocyclopentadienes
Chloronitramide anion
?

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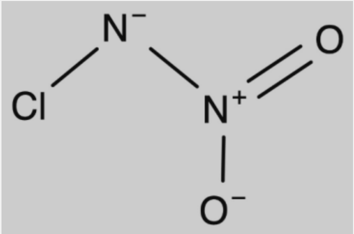
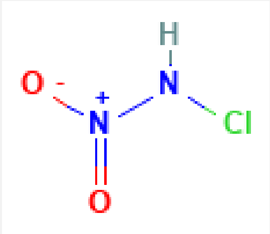
What is chloronitramide anion?

- Chloronitramide anion is a previously-unknown chemical compound in drinking water, which was recently identified as a decomposition product of inorganic chloramines by Fairey, et al. Science, 386(6724):882-887, (2024). https://doi.org/10.1126/science.adk6749

Name	Chloronitramide anion	Chloronitramide
Chemical formula	$ClN_2O_2^-$	$ClHN_2O_2$
Molecular weight (MW)	95.46 g/mol	96.47 g/mol
CAS No.	None	58999-86-3
Chemical structure		
Chemical property	Oxidant	Oxidant

Concern about chloronitramide anion?

- It was detected in all US municipal systems using chloramines to kill bacteria and viruses. The concentrations were greater than the MCLs of HAAs and TTHMs in a significant fraction of the samples studied.
- While the toxicity of chloronitramide anion is currently unknown, its structural similarity to other potentially toxic compounds raise concerns about potential health risks.
- The discovery of this previously-unknown DBP highlights the need for ongoing research into the safety of water treatment methods and the potential for unintended consequences.
- Researchers are calling for immediate toxicological evaluation, new analytical method development, and quantification of chloronitramide anion in source waters, finished drinking waters, and wastewater effluents as well.
- Future research may also advance in modeling water treatment and understanding DBP formation from the use of chloramines in drinking water disinfection.

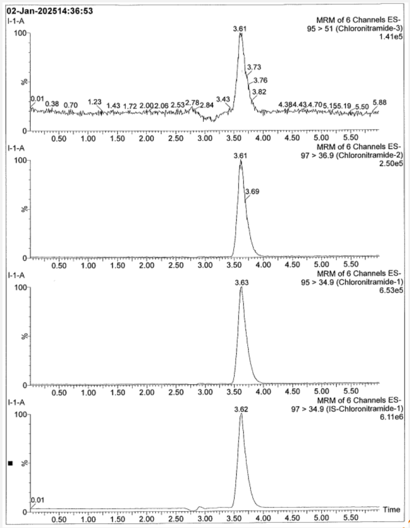
Suitable Analytical Techniques

- Ion chromatography (IC) with electrical conductivity detection (ECD) or UV detection at 243 nm.
- IC–electrospray ionization–ultrahigh resolution mass spectrometry (IC–ESI–UHRMS).
- Hydrophilic interaction liquid chromatography–ultrahigh-resolution tandem mass spectrometry (HILIC–UHRMS).
- Hydrophilic interaction liquid chromatography–tandem mass spectrometry (HILIC–MS/MS).
- Reversed-phase liquid chromatography–tandem mass spectrometry (RPLC–MS/MS).

Method Development & Validation

- RPLC–MS/MS
- Direct aqueous injection (DAI)
- Internal standard calibration
- Dark color containers & vials
- No sample preservation
- Fast analysis

DAI–RPLC–MS/MS Process



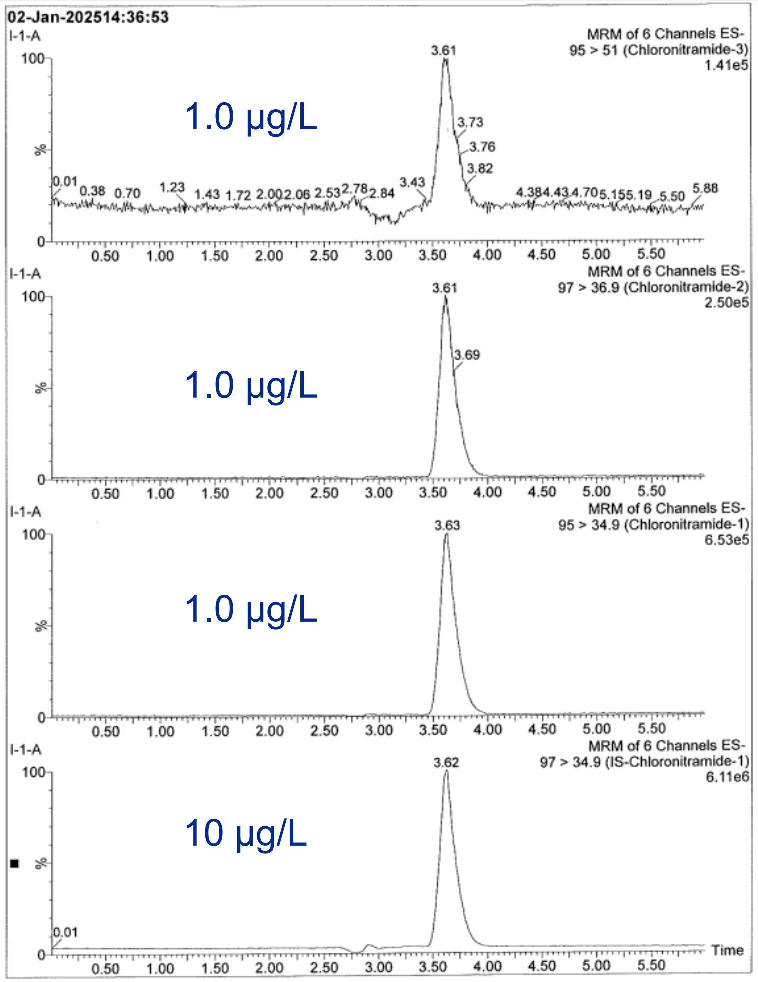
Chromatograms

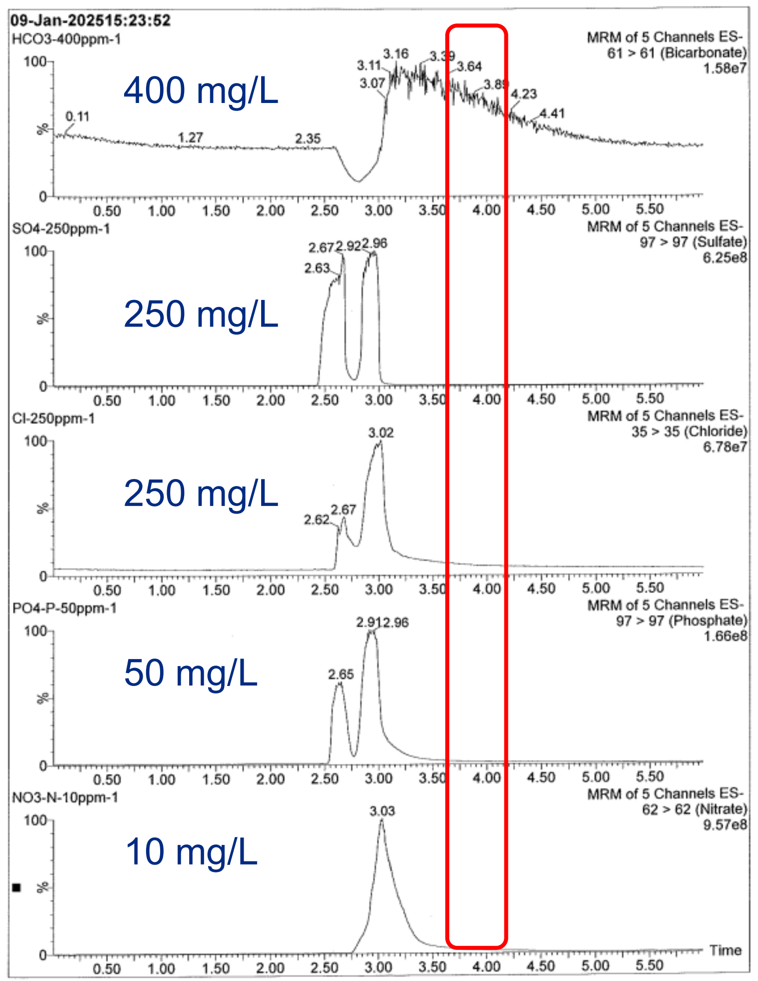
m/z 95 > 51
$^{35}Cl^{14}N_2{}^{16}O_2^- > {}^{35}Cl^{16}O$

m/z 97 > 36.9
$^{37}Cl^{14}N_2{}^{16}O_2^- > {}^{37}Cl$

m/z 95 > 34.9
$^{35}Cl^{14}N_2{}^{16}O_2^- > {}^{35}Cl$

m/z 97 > 34.9
$^{35}Cl^{15}N_2{}^{16}O_2^- > {}^{35}Cl$





$HCO_3^- > HCO_3^-$
m/z 61 > 61

$HSO_4^- > HSO_4^-$
m/z 97 > 97

$Cl^- > Cl^-$
m/z 35 > 35

$H_2PO_4^- > H_2PO_4^-$
m/z 97 > 97

$NO_3^- > NO_3^-$
m/z 62 > 62

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IDC — Sensitivity

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Analyte Mass (m/z)	Spike Conc (µg/L) n ≥ 4	LCMRL (µg/L)	DL (µg/L)	MDL (µg/L)
95>34.9	0, 0.025,0.005, 0.01, 0.02, 0.03, 0.04, 0.05, 0.1	0.0085	0.0030	0.0069
97>36.9	0, 0.005, 0.01, 0.02, 0.03, 0.04, 0.05, 0.1	0.0079	0.0045	0.0096
95>51	0, 0.04, 0.05, 0.1, 0.2, 0.3, 0.4, 0.5	0.049	0.0076	0.049

LCMRL = Lowest Concentration Minimum Reporting Level

MRL Confirmation (n = 7)

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Analyte Mass (m/z)	Spike Conc (µg/L)	Upper PIR (%)	Lower PIR (%)	Mean Rec (%)	RSD (%)
95>34.9	0.005	130 ≤ 150	83 ≥ 50	106.2	5.6
97>36.9	0.01	133 ≤ 150	58 ≥ 50	96.0	9.9
95>51	0.05	145 ≤ 150	54 ≥ 50	99.4	11.6

PIR = Prediction Interval of Results

IDC — Accuracy & Precision

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QC Sample	Spike Conc (µg/L)	Analyte/IS Mean Rec ± RSD (%), n = 7			
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51	* IS Mass m/z 97>34.9
LCS	25.0	100.6 ± 0.6	100.3 ± 0.3	100.7 ± 0.9	98.3 ± 0.6
LCS	5.0	100.2 ± 1.1	100.9 ± 1.1	104.5 ± 2.0	92.0 ± 4.3
LCS	0.1	100.5 ± 1.7	104.4 ± 0.8	92.8 ± 7.0	97.2 ± 0.4

Impacts of pH Values

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DI-Water pH Value	Spike Conc (µg/L)	Analyte/IS Mean Rec ± RSD (%), n = 4			
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51	* IS Mass m/z 97>34.9
pH 10	5.0	100.1 ± 3.0	100.8 ± 3.1	100.8 ± 3.0	78.9 ± 3.0
pH 9	5.0	98.9 ± 4.8	99.7 ± 4.7	100.3 ± 4.6	79.7 ± 4.7
pH 8	5.0	108.3 ± 11.2	109.0 ± 11.0	109.3 ± 11.5	79.3 ± 2.4
pH 7	5.0	102.0 ± 4.1	102.6 ± 4.2	103.4 ± 4.4	79.6 ± 1.3
pH 6	5.0	102.1 ± 1.8	102.9 ± 1.8	103.3 ± 2.0	78.2 ± 2.3
pH 5	5.0	106.8 ± 3.8	107.3 ± 3.8	107.4 ± 3.6	75.0 ± 3.2
pH 4	5.0	103.5 ± 1.5	103.9 ± 1.8	104.6 ± 1.7	80.1 ± 1.5

Impacts of Free Chlorine Levels

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Free Chlorine Level (as Cl_2)	Spike Conc (µg/L)	Analyte/IS Mean Rec ± RSD (%), n = 4			
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51	* IS Mass m/z 97>34.9
0.5 mg/L	5.0	97.6 ± 0.7	97.2 ± 0.6	98.6 ± 0.8	108.2 ± 0.9
1.0 mg/L	5.0	97.9 ± 0.2	97.5 ± 0.2	98.4 ± 0.4	105.6 ± 0.3
2.0 mg/L	5.0	97.1 ± 0.7	97.1 ± 0.6	98.1 ± 0.9	103.6 ± 0.1
3.0 mg/L	5.0	96.3 ± 0.5	95.9 ± 0.6	97.3 ± 0.7	101.9 ± 0.3
4.0 mg/L	5.0	96.4 ± 0.9	95.8 ± 0.6	97.0 ± 1.0	99.8 ± 0.4

Impacts of Chloramine Concentrations

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4:1 Cl_2:N Concentration	Analyte/IS Mean Conc ± STD (µg/L), n = 4			
	Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51	* IS Mass m/z 97>34.9
0.5 ppm Cl_2:0.125 ppm N	1.20 ± 0.01	1.22 ± 0.02	1.22 ± 0.01	10.0 ± 0.2
1 ppm Cl_2:0.25 ppm N	2.40 ± 0.06	2.45 ± 0.06	2.48 ± 0.07	9.8 ± 0.2
2 ppm Cl_2:0.5 ppm N	2.15 ± 0.78	2.15 ± 0.79	2.21 ± 0.80	10.1 ± 0.2
4 ppm Cl_2:1 ppm N	3.88 ± 1.16	3.80 ± 1.20	3.94 ± 1.18	9.9 ± 0.1
6 ppm Cl_2:1.5 ppm N	6.88 ± 0.24	6.86 ± 0.25	6.96 ± 0.26	9.6 ± 0.3
8 ppm Cl_2:2 ppm N	9.22 ± 0.03	9.16 ± 0.03	9.28 ± 0.03	9.7 ± 0.1
10 ppm Cl_2:2.5 ppm N	9.37 ± 0.04	9.37 ± 0.08	9.46 ± 0.05	9.6 ± 0.1

Impacts of Common Anions

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DI-Water pH Value	Spike Conc (µg/L)	Analyte/IS Mean Rec ± RSD (%), n = 4 or 7			
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51	* IS Mass m/z 97>34.9
Chloride at 250 mg/L	5.0	101.4 ± 1.5	101.5 ± 0.7	103.4 ± 0.8	63.8 ± 1.3
Sulfate at 250 mg/L	5.0	93.3 ± 0.7	93.5 ± 0.6	95.5 ± 0.6	78.3 ± 0.3
Nitrate at 10 mg N/L	5.0	92.5 ± 0.4	93.2 ± 0.5	94.5 ± 0.5	72.6 ± 0.2
Phosphate at 50 mg P/L	5.0	95.8 ± 0.2	96.1 ± 0.3	97.3 ± 0.5	78.3 ± 0.3
Bicarbonate at 400 mg/L	5.0	95.8 ± 3.1	97.9 ± 2.8	98.4 ± 2.4	7.8 ± 2.4
LFSSM	25.0	94.8 ± 0.2	94.3 ± 0.3	95.9 ± 0.2	99.6 ± 0.6
LFSSM	5.0	95.9 ± 0.4	95.9 ± 0.4	104.9 ± 0.5	104.7 ± 0.5
LFSSM	0.1	99.9 ± 6.1	105.2 ± 4.5	98.6 ± 19.4	103.8 ± 1.3

Initial Results of $Cl_2/UV/H_2O_2$ DW (1–6 °C)

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Sample Site	Holding Time (Day)	Chloronitramide Anion Mean Conc of FS & DUP (µg/L)		
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51
AZ-DS-1	3	2.26	2.28	2.33
AZ-DS-2	7	0.56	0.52	0.56
CA-DS-10	11	0.32	0.28	0.33
CA-DS-11	11	0.16	0.16	0.14
CA-DS-9	2	0.24	0.25	0.26
CO-DS-2	3	0.41	0.41	0.41
IN-DS-5	6	32.6	32.7	32.9
IN-DS-6	0	0.85	0.86	0.86
IN-DS-7	2	1.07	1.08	1.07
IN-DS-8	0	0.81	0.82	0.81
IN-DS-9	5	2.15	2.16	2.23
MI-DS-1	5	<0.1	<0.1	<0.1

Initial Results of Chloraminated DW (1–6 °C)

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Sample Site	Holding Time (Day)	Chloronitramide Anion Mean Conc of FS & DUP (µg/L)		
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51
CA-DS-1	1	111	111	112
CA-DS-2	1	28.2	28.3	28.5
CA-DS-3	7	19.5	19.52	19.8
CA-DS-4	7	22.1	22.2	22.5
CA-DS-5	6	28.2	28.3	28.7
CA-DS-6	6	21.8	21.8	22.1
CA-DS-7	6	11.8	11.9	12.2
CA-DS-8	15	0.85	0.85	0.83
CO-DS-1	2	28.1	28.2	28.3
FL-DS-1	1	55.4	55.4	55.6
GA-DS-1	4	1.10	1.08	1.17
IN-DS-1	6	31.7	31.8	32.1
IN-DS-2	4	39.7	39.7	40.1

Initial Results of Chloraminated DW (1–6 °C)

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Sample Site	Holding Time (Day)	Chloronitramide Anion Mean Conc of FS & DUP (µg/L)		
		Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51
IN-DS-3	6	3.41	3.54	3.47
IN-DS-4	3	7.65	7.72	7.77
KY-EP-1	2	5.34	5.35	5.46
KY-DS-1	2	8.12	8.17	8.29
KY-EP-2	2	3.59	3.59	3.61
KY-DS-2	2	7.02	7.03	7.17
MA-DS-1	4	18.4	18.5	18.5
MN-DS-1	2	12.0	12.0	12.3
MN-DS-2	2	7.67	7.75	7.94
MO-DS-1	7	14.2	14.1	14.7
TX-DS-1	3	91.0	90.5	92.0
TX-DS-2	7	83.1	82.5	82.9
TX-DS-3	5	145	145	146

MS Recoveries (stored at 1-6 °C)

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Sample Site	Native Conc (µg/L)	Spike Conc (µg/L)	Analyte Mean Rec ± RSD (%), n = 4		
			Mass m/z 95>34.9	Mass m/z 97>36.9	Mass m/z 95>51
AZ-DS-1	2.13	5.0	103.3 ± 1.2	104.0 ± 1.0	104.7 ± 1.8
CA-DS-1	122	100	98.5 ± 1.2	97.0 ± 1.0	99.8 ± 1.0
CA-DS-4	41.6	20.0	105.5 ± 0.8	104.7 ± 0.9	105.6 ± 0.7
CO-DS-2	0.51	5.0	97.4 ± 0.8	97.8 ± 0.6	98.0 ± 1.0
IN-DS-2	68.2	50.0	104.5 ± 2.7	103.5 ± 2.8	104.2 ± 2.7
MN-DS-1	21.2	10.0	99.6 ± 0.6	98.8 ± 0.6	97.6 ± 0.6

Impacts of Holding Time (1-6 °C)

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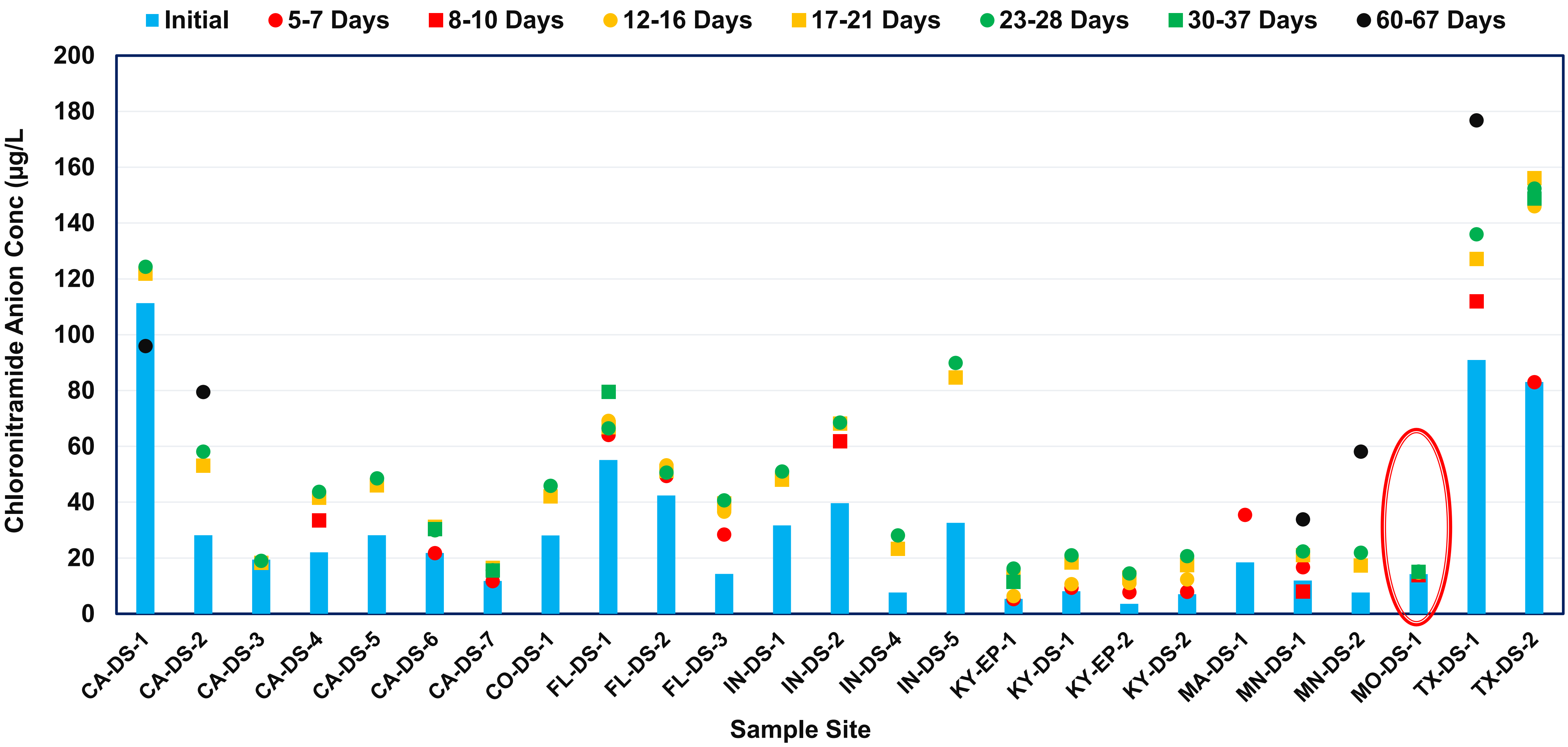

Impacts of Storage Temperature (1)

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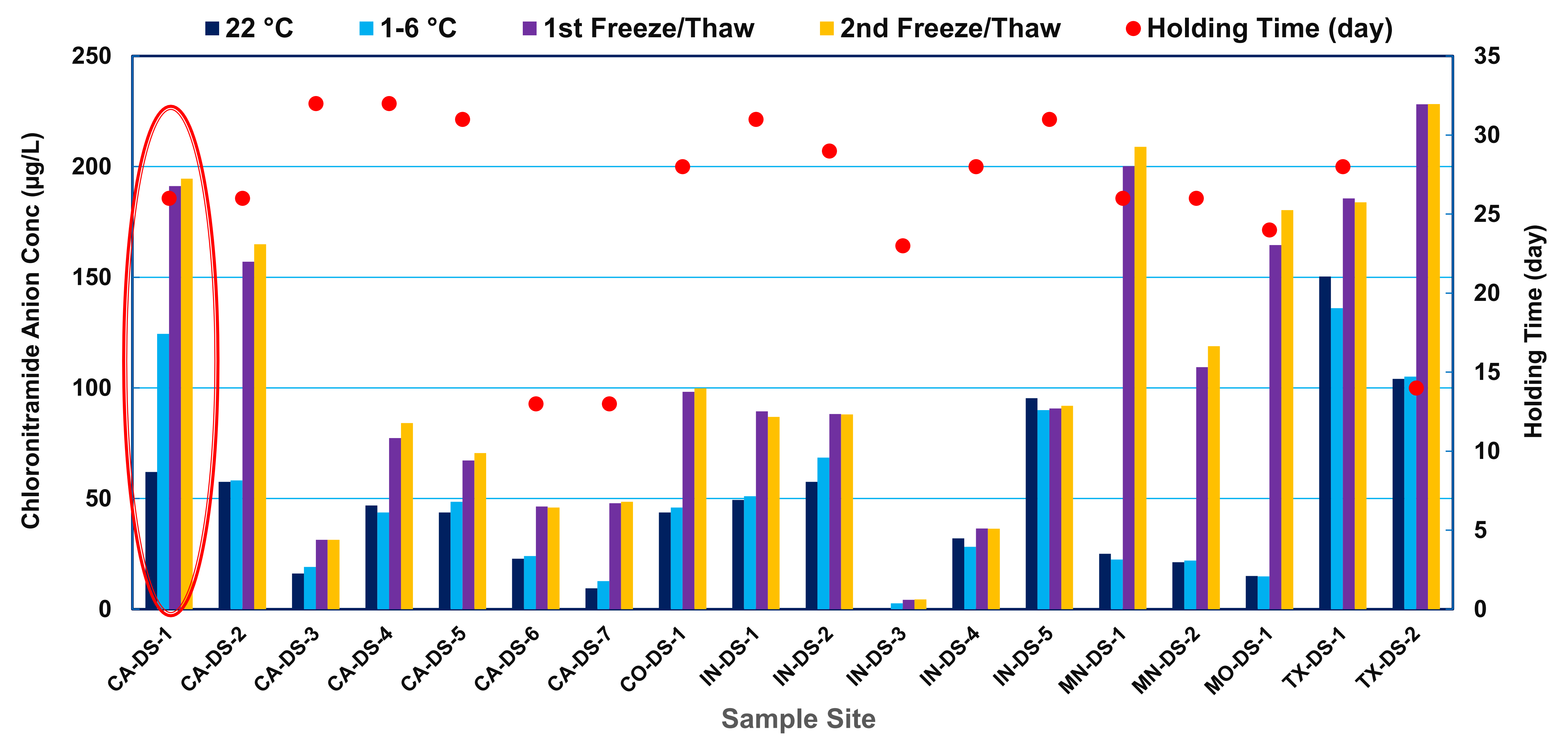
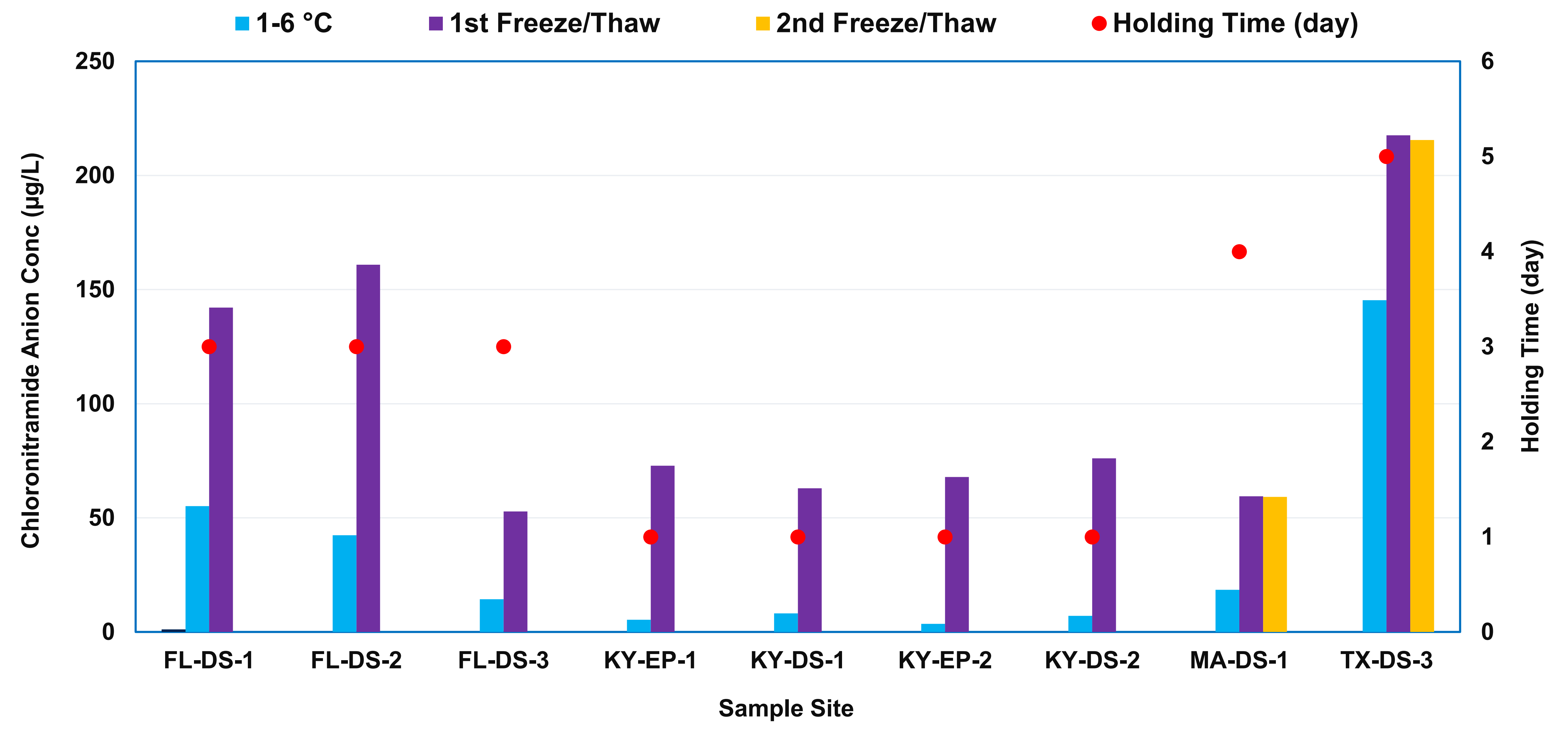


Impacts of Storage Temperature (2)



Impacts of Storage Temperature (3)

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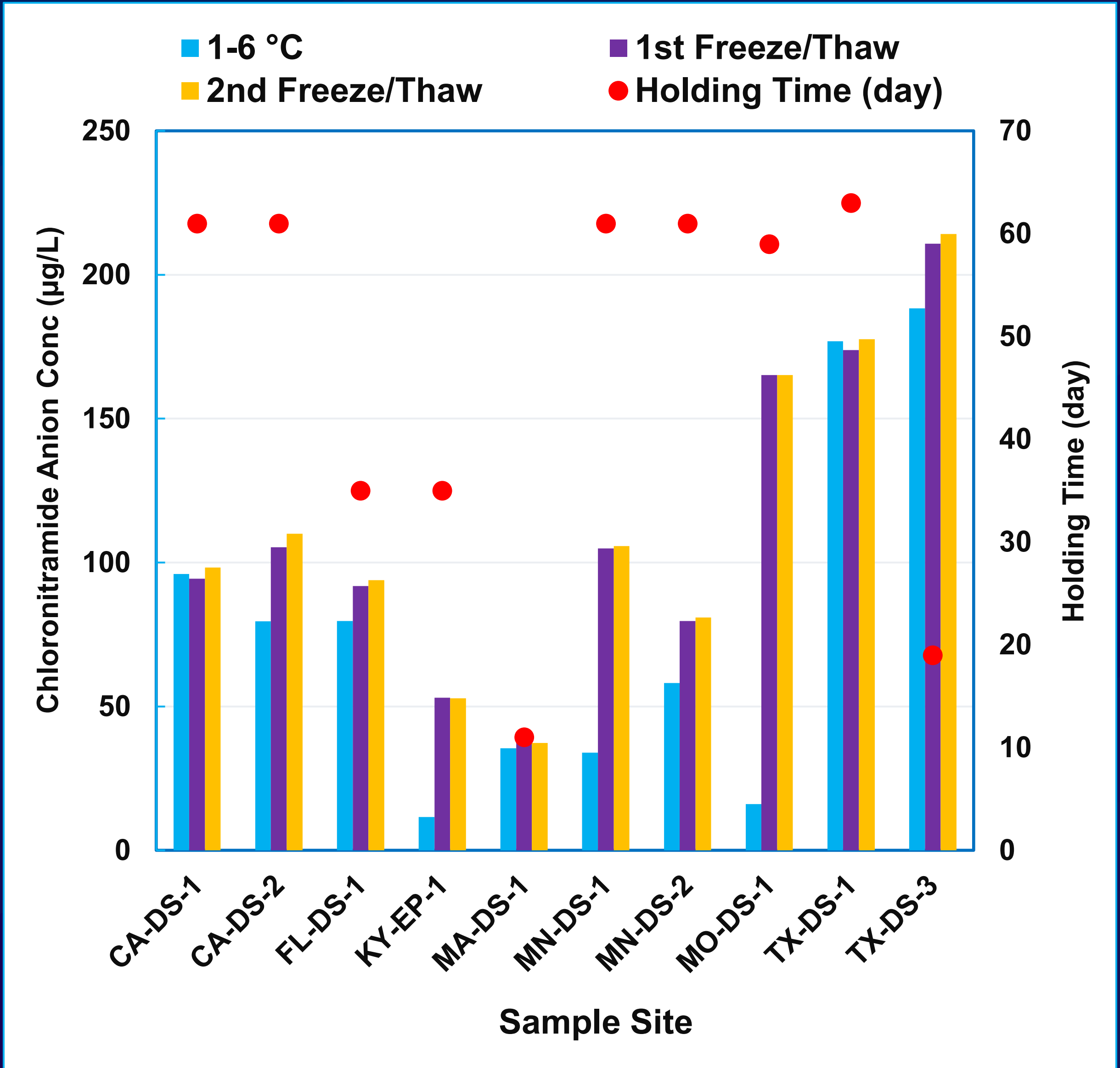

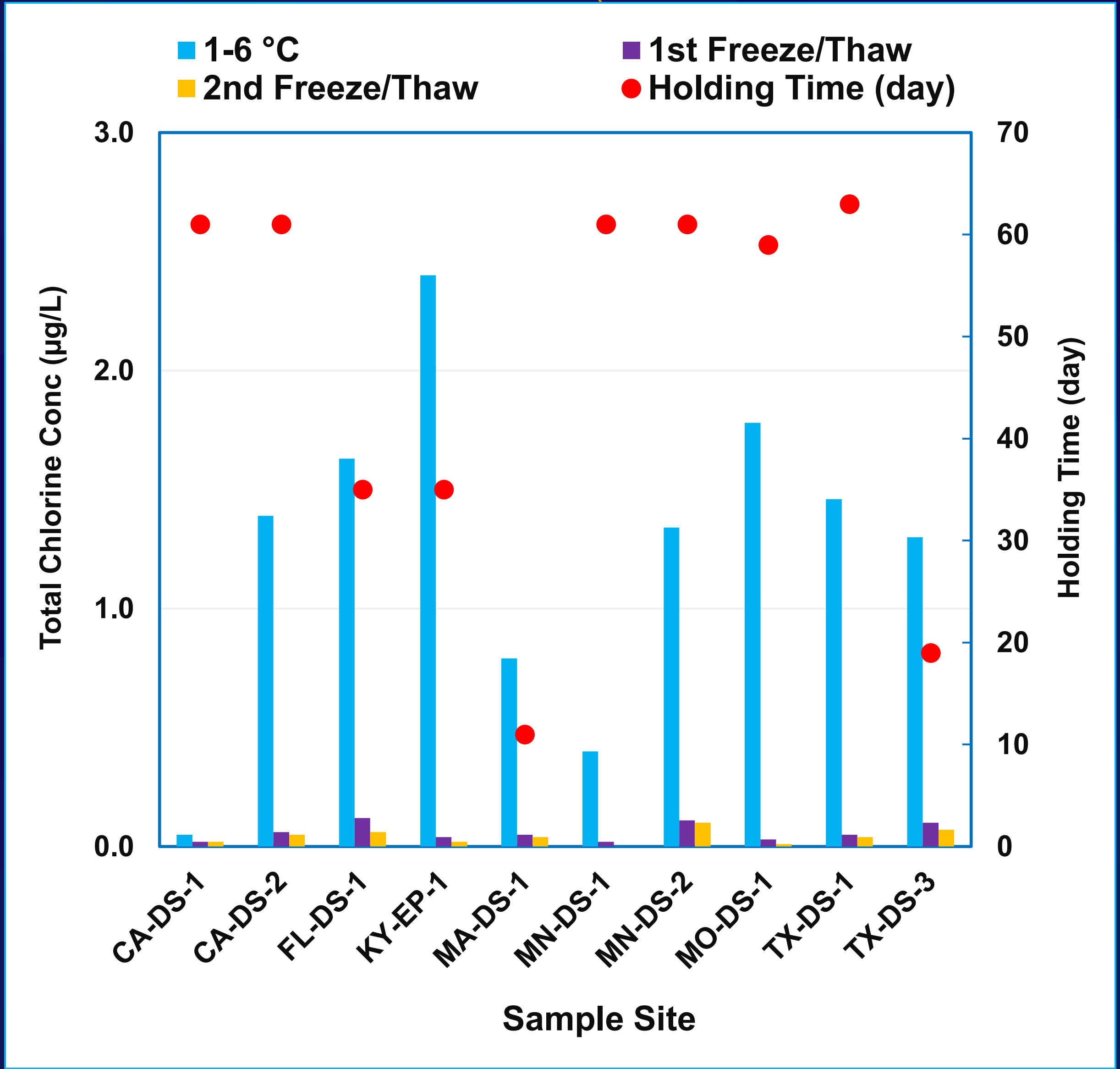

MS Recoveries (stored at -25 °C)

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Sample Site	Native Conc (µg/L)	Spike Conc (µg/L)	Mean MS/MSD Conc (ug/L)	Mean MS/MSD Rec (%)	MS/MSD RPD (%)
AZ-DS-1	2.26	5	8.1	117.3	0.7
CO-DS-2	0.41	5	5.6	104.4	1.5
FL-DS-1	52.8	25	76.6	95.2	4.5
CA-DS-1	111	100	224	112.6	1.8
CA-DS-4	77.3	100	168	90.6	2.1
CA-DS-6	46.4	100	139	92.8	0.2
IN-DS-1	89.5	100	192	102.0	1.3
IN-DS-2	88.3	100	168	79.4	2.4
MN-DS-1	200	100	297	96.4	4.7

Conclusions

- The DAI–LC–MS/MS method could determine chloronitramide anion in a typical alkalinity range of drinking water at an MRL of 0.1 µg/L.
- This method demonstrated satisfactory sensitivity, accuracy and precision. Common anions except bicarbonate, pH, free chlorine concentrations, and chloramine concentrations did not interfere with the quantitation of chloronitramide anion.
- Surprisingly, freezing samples caused chloramine decomposition and more chloronitramide anion formation.
- Chlorine/chloramine quenching agents could not be used to preserve drinking water samples. Storing samples in a freezer was not recommended.

Conclusions (Cont'd)

- Chloronitramide anion was detected in all studied chloraminated drinking water samples at the initial analysis concentrations of 0.85 to 111 µg/L.
- The concentrations substantially increased (up to 200 µg/L) with the increase in holding time due to the continuous decomposition of chloramines. Ideally, the samples should be analyzed as early as possible, within 7 days of holding time.
- 11 out of 12 chlorinated drinking water samples were also detected with chloronitramide anion at low µg/L with an exception for MI-DS-1.
- Chloronitramide anion was initially detected in chlorinated IN-DS-5 at 32.6 µg/L, which was increased to ~90 µg/L after two weeks of holding time or freeze/thaw.

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THANK YOU

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