

Functionalized surfaces as a tool for virus sensing: a demonstration of *Human mastadenovirus* detection in environmental waters.

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Table S1 (Supplementary material). qPCR results to HAdV standard analyses in unconcentrated and concentrated samples by IMS method

HAdV-5 Serial Dilutions “	UNCONCENTRATED					IMS - MONOCLONAL					IMS - POLYCLONAL				
	1° replica	2° replica	3° replica	Mean	RE (%)	1° replica	2° replica	3° replica	Mean	RE (%)	1° replica	2° replica	3° replica	Mean	RE (%)
-1	1.31E+06	9.58E+05	1.90E+06	1.39E+06	12.30	2.89E+06	6.81E+05	1.63E+06	1.74E+06	15.35	2.09E+06	5.90E+06	1.84E+06	3.28E+06	29.00
-2	6.95E+04	3.64E+04	3.22E+04	4.60E+04	4.07	1.36E+05	2.88E+04	9.40E+04	8.64E+04	7.65	9.08E+04	7.66E+04	5.14E+04	7.30E+04	6.46
-3	4.95E+03	3.22E+03	6.27E+02	2.93E+03	2.59	2.14E+04	3.01E+03	2.31E+04	1.59E+04	14.03	4.94E+03	3.13E+04	4.69E+04	2.77E+04	24.51
-4	2.37E+02	2.04E+02	1.56E+02	1.99E+02	1.76	2.31E+02	3.13E+02	1.28E+03	6.09E+02	5.39	4.68E+02	1.59E+03	1.92E+02	7.49E+02	6.63
-5	2.84E+01	0.00E+00	1.35E+01	1.40E+01*		3.62E+01	3.57E+01	5.26E+01	4.15E+01*		1.29E+02	4.69E+01	5.28E+01	7.63E+01	6.75
-6	ND	ND	ND			4.60E+00	9.87E+00	1.41E+01	9.51E+00*		1.79E+02	1.84E+01	2.18E+01	7.31E+01	64.71
-7	ND	ND	ND			5.02E+00	5.56E+01	3.54E+00	2.14E+01*		4.70E+01	9.83E+00	2.02E+01	2.57E+01*	
-8	ND	ND	ND			9.70E+00	5.40E+00	4.80E+01	2.10E+01*		2.05E+01	9.31E+00	2.23E+01	1.74E+01*	
-9	ND	ND	ND			1.27E+01	1.83E+01	1.34E+01	1.48E+01*		3.91E+01	5.49E+00	7.80E+01	4.09E+01*	
-10	ND	ND	ND			2.92E+00	1.87E+01	1.65E+01	1.27E+01*		4.21E+01	7.81E+00	7.53E+00	1.91E+01*	
-11	ND	ND	ND			-	1.31E+01	7.94E+00	1.05E+01*		-	1.79E+01	1.30E+01	1.55E+01*	
-12	ND	ND	ND			-	1.04E+01	9.82E+00	1.01E+01*		-	1.88E+01	2.15E+01	2.01E+01*	
-13	ND	ND	ND			-	5.31E+01	1.37E+01	3.34E+01*		-	7.37E+00	1.59E+01	1.16E+01*	

Results are presented in genomic copies/milliliter (GC/mL)

RE = viral recovery

ND = not detected

“Initial quantification of HAdV-5 used to dilute was 1.13E+08 GC/mL

* Mean results lower than the value used as the sensitivity of qPCR reactions therefore were not considered as positive.

Table S2 (Supplementary material). Results of HAdV-5 infectious particles concentrated by IMS and detected by Viral isolation and subsequent qPCR

Inoculated samples	1° passage				2° passage			
	-1		-2		-1		-2	
	Monoclonal	Polyclonal	Monoclonal	Polyclonal	Monoclonal	Polyclonal	Monoclonal	Polyclonal
Direct inoculation	2.04E+06	1.36E+06	1.98E+05	2.55E+05	1.42E+07	1.46E+07	8.55E+06	1.46E+07
freeze and thaw -20°C	1.17E+06	2.10E+06	6.71E+03	3.50E+04	1.56E+07	1.35E+07	4.49E+06	6.17E+06
freeze and thaw -20°C filtered	1.19E+06	1.16E+06	7.29E+04	4.94E+04	1.72E+07	1.60E+07	8.79E+06	6.86E+06
freeze and thaw -80°C	2.51E+05	6.85E+05	6.61E+03	1.08E+05	8.78E+06	1.04E+07	4.31E+06	1.70E+07
freeze and thaw -80°C filtered	6.70E+03	1.34E+05	1.24E+02	1.77E+03	6.04E+06	1.37E+07	9.66E+05	3.78E+06

Results are presented in genomic copies / 5 microliter (GC/5µL)

Initial quantification of HAdV-5 used to dilute was 1.13E+08 GC/mL

Table S3 (Supplementary material). HAdV in water samples comparing ultracentrifugation and IMS methods. The results of HAdV-F and C were established by qPCR and AdV by Nested PCR

Sites	HAdV-F			HAdV-C			AdV		
	Ultra.	IMS – M.	IMS – P.	Ultra.	IMS – M.	IMS – P.	Ultra.	IMS – M.	IMS – P.
		GC/L			GC/L				
LMM-5685	2.22E+02	-	-	-	-	-	-	-	-
LMM-5686	-	-	-	-	3.09E+05	1.59E+05	-	-	-
LMM-5687	-	-	2.25E+04	-	-	1.25E+05	-	-	-
LMM-5688	-	-	2.28E+04	-	5.83E+06	-	-	-	-
LMM-5689	-	5.68E+04	1.94E+04	-	2.84E+05	-	-	-	-
LMM-5690	-	1.79E+03	-	-	3.50E+05	-	-	-	-
LMM-5691	-	-	-	-	2.23E+05	3.31E+05	-	-	-
LMM-5692	-	-	-	-	9.67E+04	2.47E+05	-	-	-
LMM-5693	1.04E+03	-	-	2.11E+03	-	2.92E+05	-	+	-
LMM-5694	-	-	-	-	-	1.99E+05	-	+	-
LMM-5695	-	-	-	-	1.56E+05	-	-	-	-
LMM-5696	-	4.55E+02	3.35E+04	-	-	4.09E+05	-	-	-
LMM-5697	2.00E+02	-	-	1.18E+03	1.31E+05	-	-	-	-
LMM-5698	7.91E+02	-	-	-	-	-	-	-	+
LMM-5699	-	-	-	-	2.26E+05	-	-	-	-

GC/L = Genomic copies /Liter

Ultra. = Ultracentrifugation method

IMS - M. = Immunomagnetic separation method using Monoclonal Antibody

IMS - P. = Immunomagnetic separation method using Polyclonal Antibody

+ = positive sample

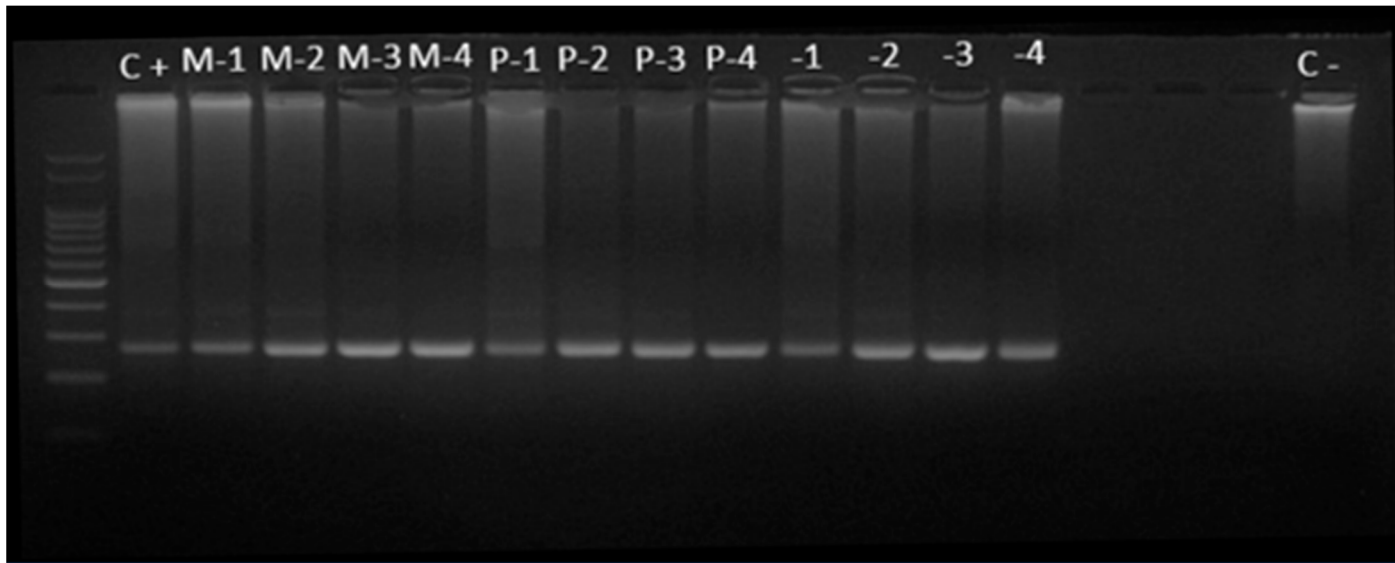


Figure S1 (Supplementary material). Nested PCR results with the unconcentrated HAdV standard samples and concentrated by the IMS technique

M (-1 to -4) = Immunomagnetic separation method using Monoclonal Antibody

P (-1 to -4) = Immunomagnetic separation method using Polyclonal Antibody

-1 to -4 = Unconcentrated samples

C+ = positive control

C- = negative control

Test Statistics ^{a,b}	
N	6
Chi-Square	9,091
Df	2
Asymp. Sig.	,011
a. GRUP = 1	
b. Friedman Test	

Each node shows the sample average rank.

Sample1-Sample2	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
NCONC-IMSMON	-,833	,577	-1,443	,149	,447
NCONC-IMSPOL	-1,667	,577	-2,887	,004	,012
IMSMON-IMSPOL	-,833	,577	-1,443	,149	,447

Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same.
Asymptotic significances (2-sided tests) are displayed. The significance level is ,05.
Significance values have been adjusted by the Bonferroni correction for multiple tests.

Figure S2 (Supplementary material). Statistical analysis of results found in HAdV standard analyses of unconcentrated and concentrated samples by IMS method

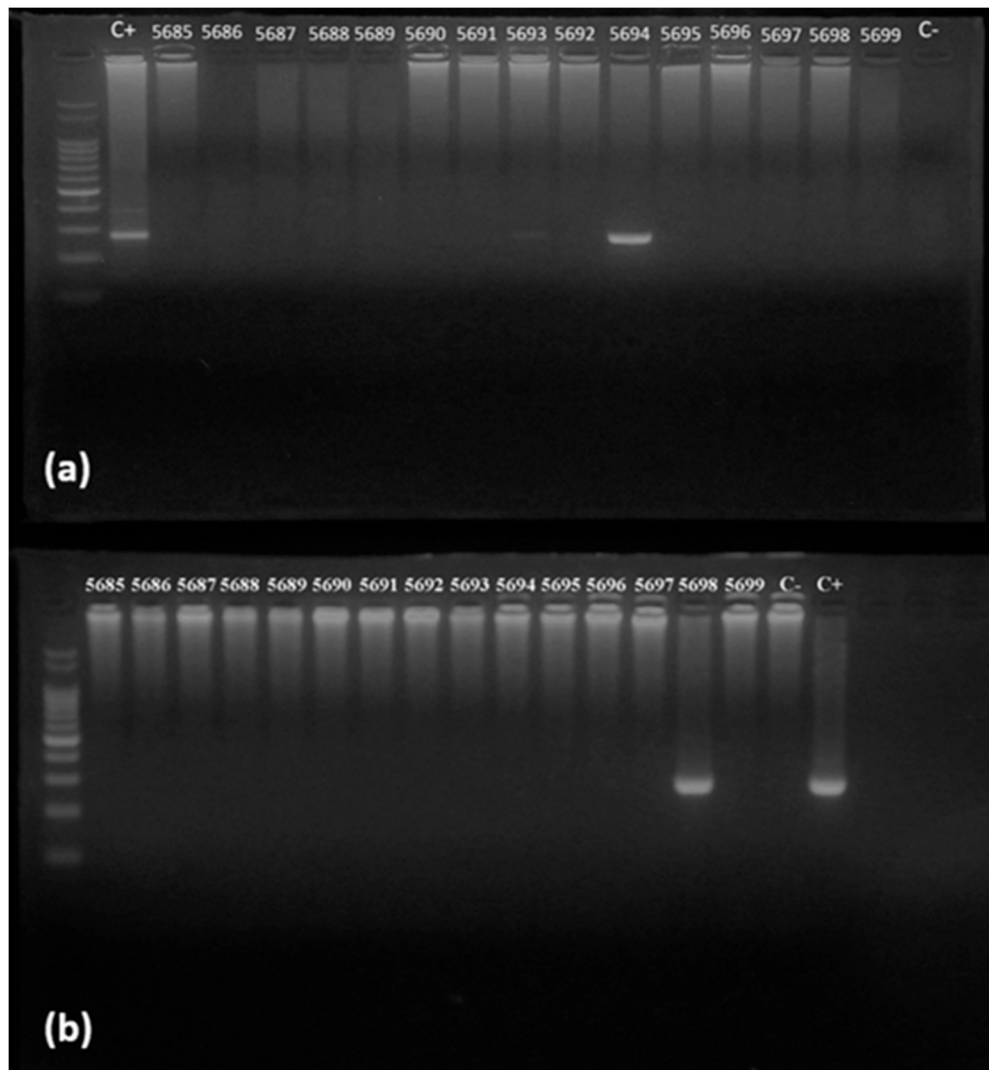
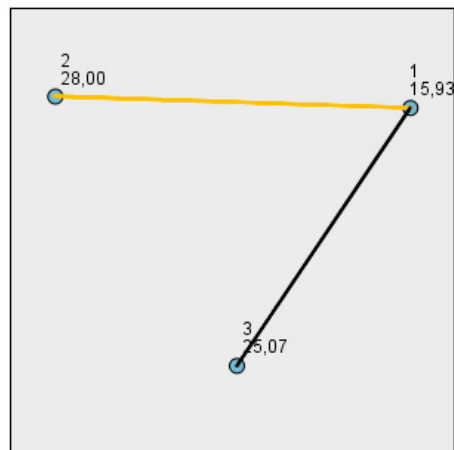


Figure S3 (Supplementary material). Nested PCR results in water samples that were carried out after IMS concentration using monoclonal antibody (a) and polyclonal antibody (b)

Test Statistics ^{a,b}		
	HAdV-F	HAdV-C
Chi-Square	,337	8,783
df	2	2
Asymp. Sig.	,845	0,012
a. Kruskal Wallis Test		
b. Grouping Variable: GRUPO		

Pairwise Comparisons of GRUPO



Each node shows the sample average rank of GRUPO.

Sample 1-Sam...	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
1-3	-9,133	4,247	-2,151	,032	,095
1-2	-12,067	4,247	-2,841	,004	,013
3-2	2,933	4,247	,691	,490	1,000

Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same. Asymptotic significances (2-sided tests) are displayed. The significance level is ,05. Significance values have been adjusted by the Bonferroni correction for multiple tests.

Figure S4 (Supplementary material). Statistical analysis of results found in naturally contaminated water samples concentrated by IMS and ultracentrifugation methods (1: Ultracentrifugation, 2: IMS – Monoclonal, 3: IMS – Polyclonal)