

Table S1: Ct values obtained for each virus under each condition

Virus	Sample	Cepheid T0	E swab T0	Virocult T0	Cepheid H4	E swab H4	Virocult H4	Cepheid H24 [4°]	E swab H24 [4°]	Virocult H24 [4°]	Cepheid H72 [AT]	E swab H72 [AT]	Virocult H72 [AT]	Cepheid H72 [4°]	E swab H72 [4°]	Virocult H72 [4°]
FluA	FA1	27.8	27.5	26.6	28.4	28.6	27.4	28.6	28.7	27.2	30.1	30.1	28.9	28.9	28.9	27.5
	FA2	24.8	25.5	24.6	24.5	24.8	24.2	24.3	25.1	24.9	26.2	27	25.7	24.4	24.5	25.1
	FA3	23.9	21.5	24.2	24.2	23.4	24.3	24.3	23.2	23.8	24.9	25.2	24.8	24.5	23.2	24.3
	FA4	26.1	24.1	24.5	25.6	24.2	22.7	25.7	25.8	25.5	25.9	27.1	19.4	25.5	26.4	26.5
	FA5	26.7	26	25.5	28.6	27.2	27.3	28.9	26.7	26.4	32	29.9	34.9	29.9	30.1	27.4
FluB	FB1	21.5	21.9	22.3	21.8	21.9	22.3	22.1	21.6	23.6	22	21.9	24.8	21.4	21.6	22.2
	FB2	22.4	21.9	22.2	22.2	22.1	22.3	23	21.9	22.1	22.2	21.8	23.3	22	22.1	22.7
	FB3	28.4	27.9	28.8	28	28.4	28.5	28.7	28.2	28.4	28.9	28.1	27.3	28.5	28.8	29
	FB4	22	21.8	21.8	21.6	21.7	22.1	22	21.6	21.6	22	22.1	22.5	21.5	22	22.2
	FB5	25.1	24.8	24.9	25.6	24.7	25.1	25.2	24.3	25.4	25	25.7	25.4	24.8	24.7	25
RSV	VA1	24.6	25.7	24.6	24.3	24.1	25.1	24.2	26.8	24.6	26.9	24.2	24.3	24.1	27.1	24.7
	VA2	21	20.3	20.9	20.5	20.4	20.4	20.3	20.3	20.5	20.4	20.4	20.5	20.3	20.2	20.8
	VA3	23.4	23.3	23.3	23.5	23.7	23.7	23.4	23.3	23.3	23.2	22.3	23.4	24.3	23.3	23.7
	VB1	27.2	27.7	27.2	27.4	27.3	27.5	27.4	27.1	27.7	27.4	27.4	28	27.6	26.9	28.1
	VB2	25.7	25.5	25.4	25.7	25.8	26.1	25.2	25.3	24.9	25.4	25.4	25.7	25.5	25.5	24.9

Figure S1: Comparison of obtained Ct for each sample stored for different periods of time, under different temperature conditions and gathered according to VTM: A: Cepheid®; B: Virocult®; C: E-swab®. Each virus family is represented by a different symbol: FluA, open squares; FluB, closed squares; RSV, open circles

Figure S1A

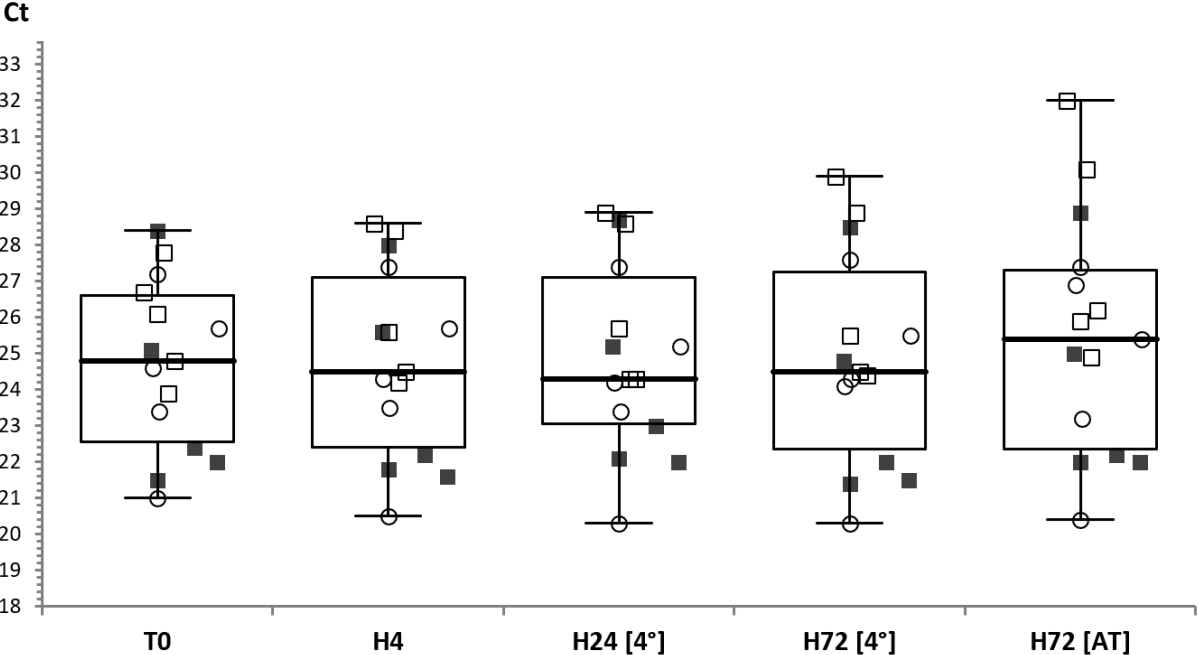


Figure S1B

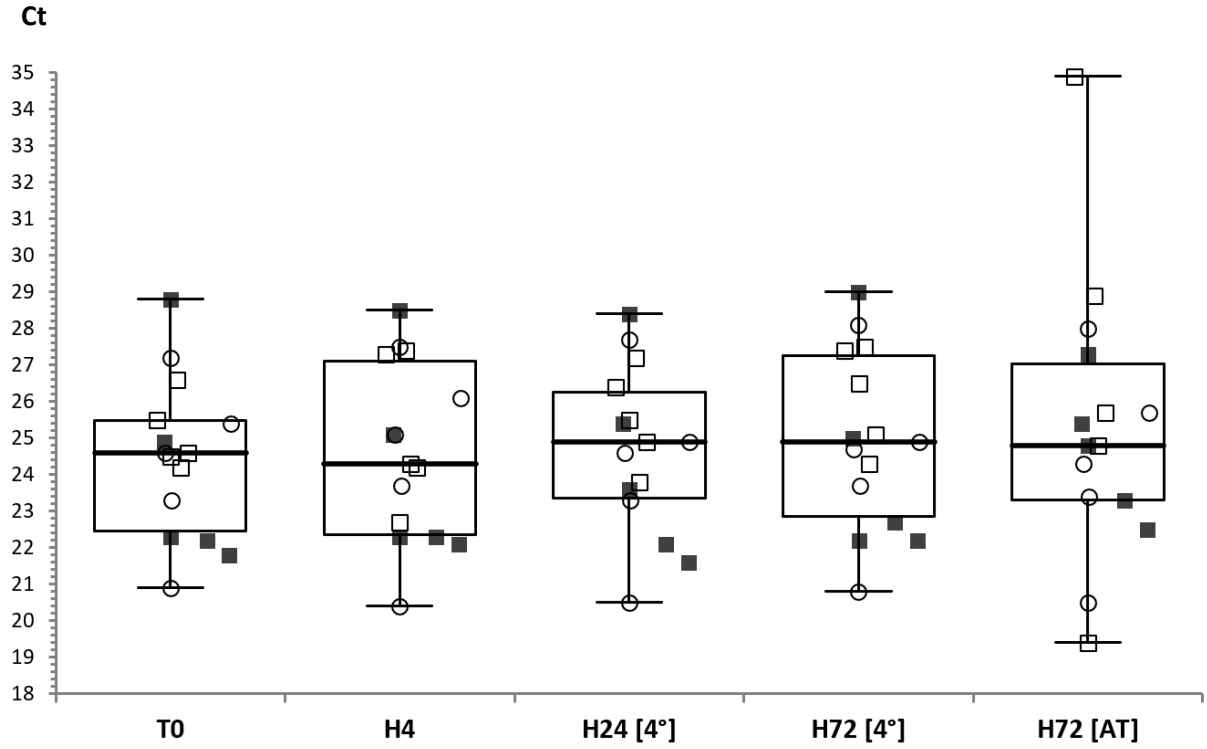


Figure S1C

Ct

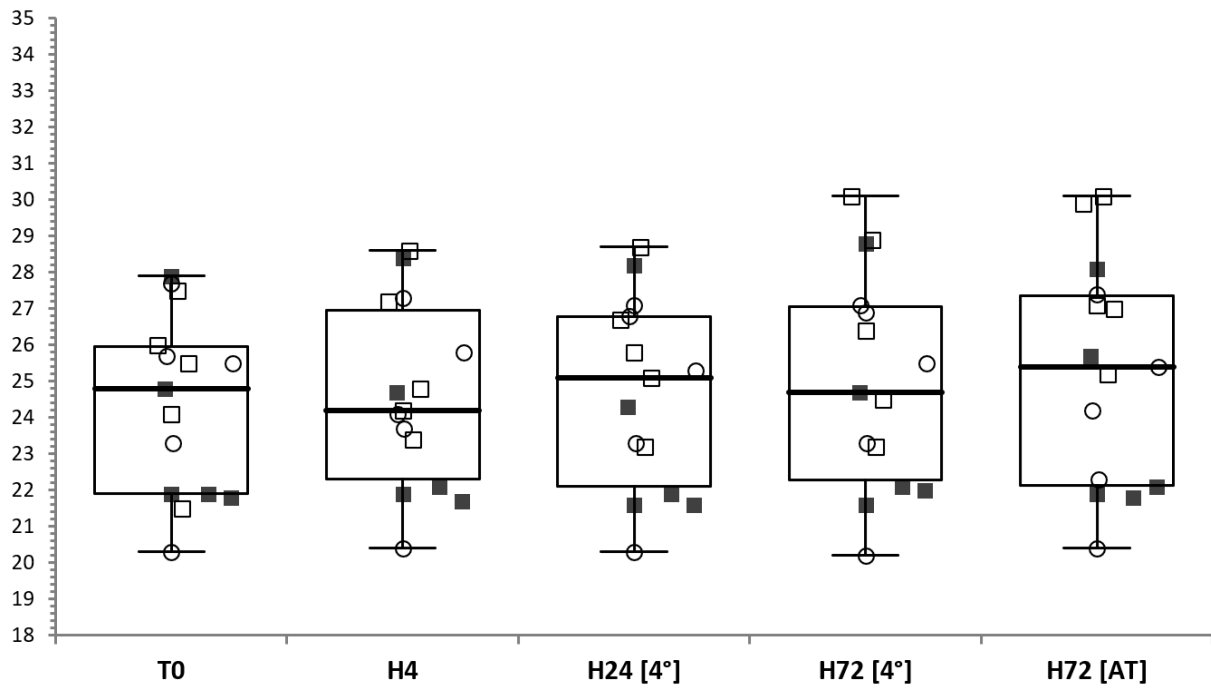


figure S2A-2C: Representation of dCt for each virus under different conditions in different VTM. Fig. S2A: FluA, S2B, FluB; S2C RSV. On each figure, each sample (n=5) is represented by a different symbol. The dotted horizontal lines represent the cut-off dCt of 1.3 Ct.

FA1 and FB1 white square, FA2 and FB2 black square, FA3 and FB3 white diamond, FA4 and FB4 black diamond, FA5 and FB5 white circle.

Figure 2C: VA1 white square, VA2 black square, VA3 white diamond, VB1 black diamond, VB2 white circle. (\*p value <0,005)

Fig. S2A

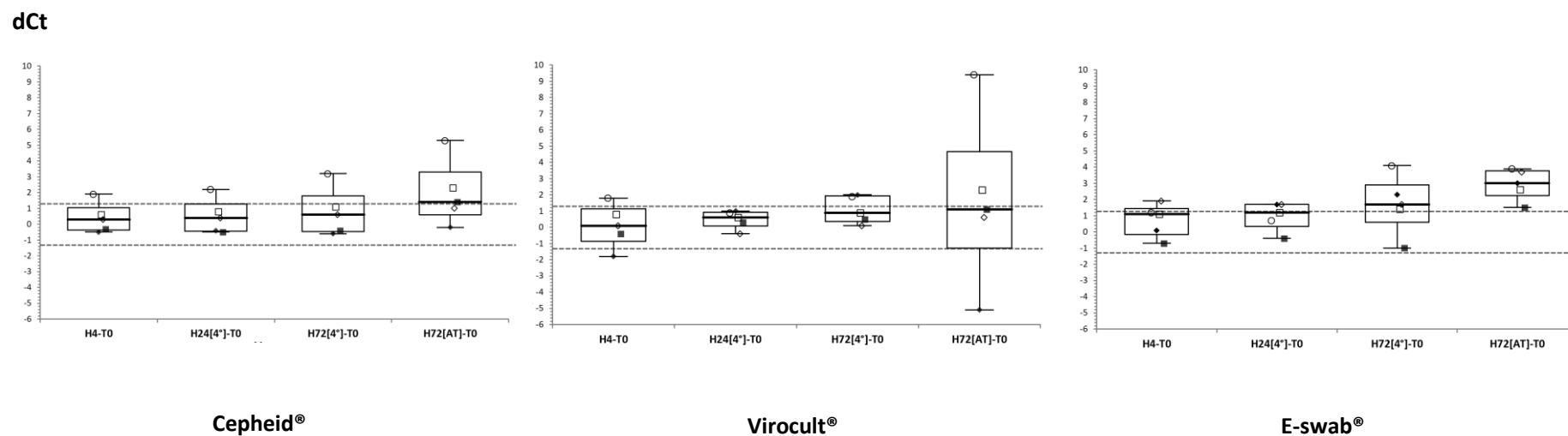


Fig. S2B

dCt

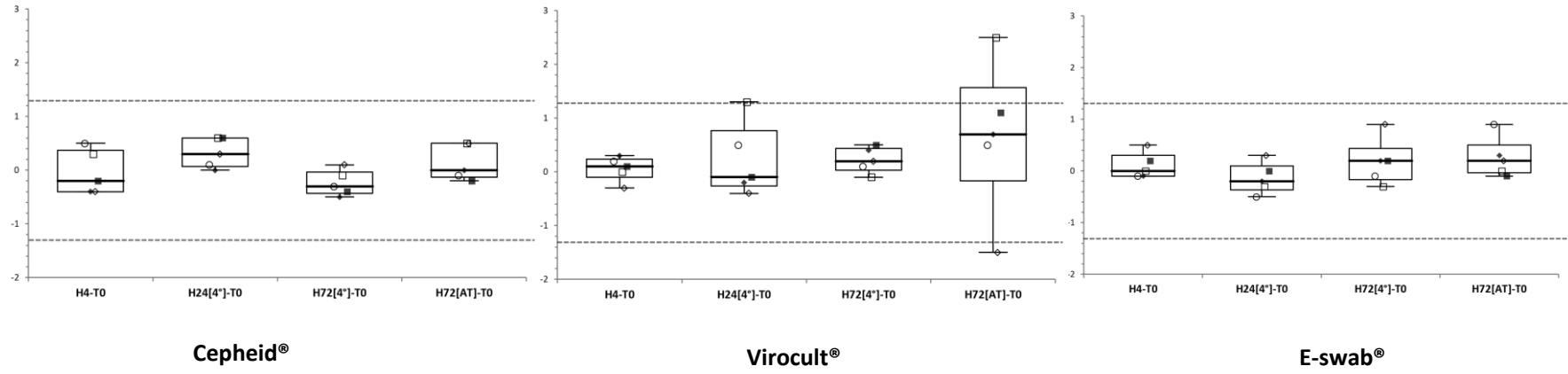
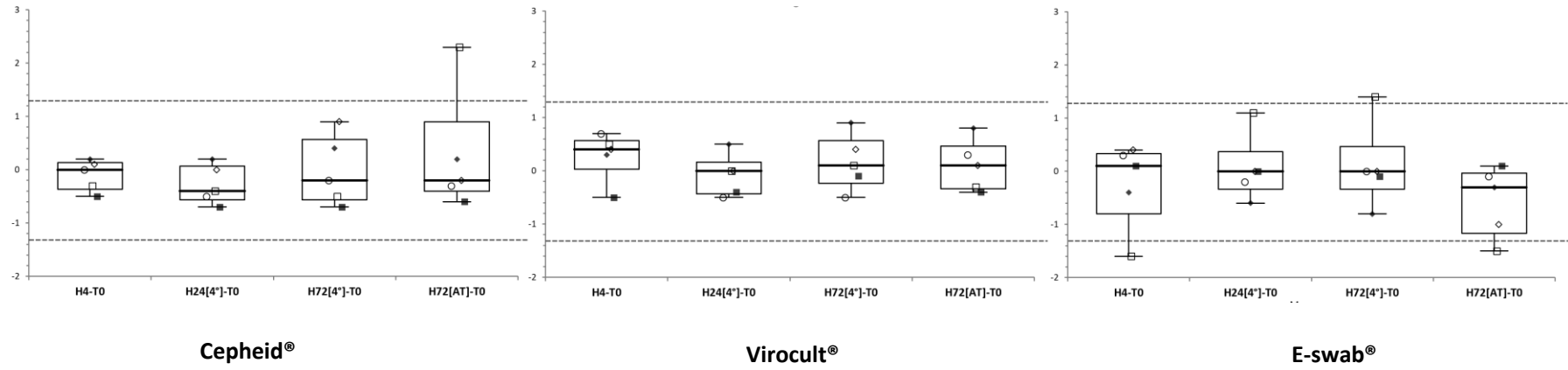


Fig. S2C

dCt



### **Supplementary details on the methodological approach.**

This study should be considered as a proof of concept study as few samples for each virus were tested under each condition. Feasibility constraints were the limited availability of assays, the cost and the time to perform the study.

This study was designed to mimic encountered situations from real life. One retained scenario was the conservation of a sample over the week-end. Even in these conditions, the relative loss of sensitivity was considered as minor at 4°C and we did not consider pertinent to test shorter times.

In order to be homogeneous whatever the virus, the choice of relatively late amplification signals with Ct between 21 and 27 for each virus was made. Indeed, these samples are probably those most sensitive to the storage conditions and could lead to false negative results under harsh storage conditions.

As the used assay is only qualitative, we performed two types of analyses; one purely qualitative and one taking into account the Ct values. This last analysis was done to get a surrogate appraisal of the possible loss of sensitivity under different storage conditions. To objectivize a possible loss of target, the delta Ct was calculated. To fit with the Quamic 2019 recommendations (the French recommendation to comply with the normative NF EN ISO 15189), we have set the threshold at 2 Ct for significant difference. Obviously, for diagnosis purpose, the most important remains the qualitative analysis.