Detection of viral genomes and inflammatory markers in endomyocardial tissues of patients with idiopathic and explained dilated cardiomyopathy and healthy heart and surgery control groups

EV 1079

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Introduction and purpose

- Cardiotropic viruses are now suspected to be etiological causes or cofactors in the development of idiopathic dilated cardiomyopathy (iDCM) [1].
- However, common viruses could be considered as innocent bystanders when they are detected in dilated cardiomyopathy (DCM) of other etiologies [2].
- Here, we analysed Endomyocardial Biopsy samples (EMBs) of iDCM patients for the presence of cardiac viral infection and inflammatory markers, comparatively to explained DCM and to control groups.

Patients & Methods

- Between 2008 and 2014, EMBs were performed in Reims University Hospital according to AHA&ESC guidelines in 13 patients (sex ratio M/F=2.5, mean age 47.5±11 years) suffering from idiopathic DCM (iDCM) and 10 suffering from explained DCM (eDCM) (sex ratio M/F=2.33, median age 47.3±15.9 years) [3,4]. For each of these patients, 3-6 fragments were sampled and 2 were flash frozen for virological analyses. Human enteroviruses (EV), human parvovirus B19 (PVB19) and all Human Herpes virus (HHV) were detected by specific real–time PCR assays (Argene Biomérieux®). CD3, CD68 and HLA DR immuno-staining were performed for cardiac inflammatory marker detection.
- All results were compared to those obtained from large cardiac samples of 11 healthy heart controls (sex ratio M/F=4.5, mean age 36.4+/-11.1 years) who died from suicide, intoxication or traumatic accident and were autopsied at Reims University Hospital.
- Only EV and PVB19 genomic detection results were compared to those obtained from 47 right atrium tissues sampled in patients during extracorporeal circulation (sex ratio M/F=4.22, mean age 68.2±10.2 years) (surgery controls). Surgery controls were classified as heart valve surgery controls or aorto-coronary bypass surgery controls
- Qualitative variables were compared using Fischer exact test. Quantitative variables were compared using Mann Whitney U test. A p value <0.05 was considered as significant.

Results

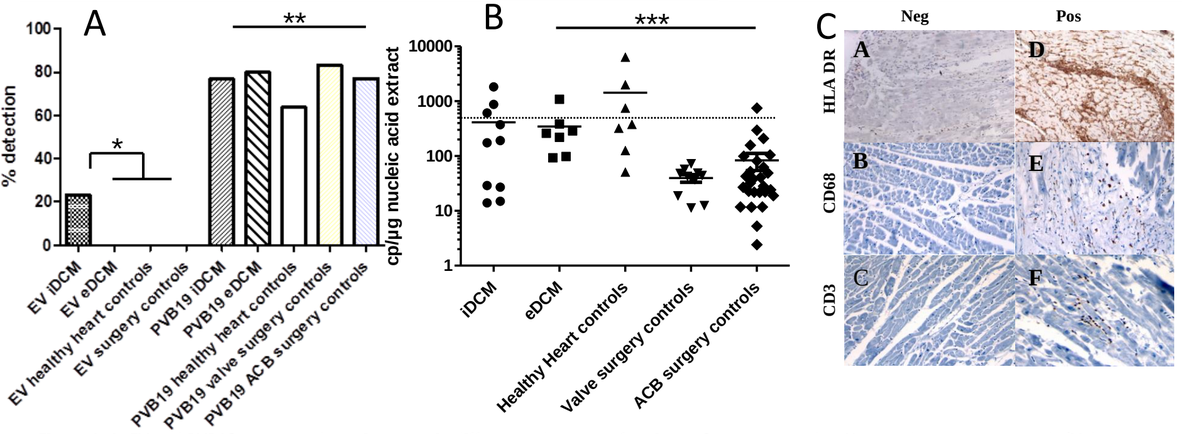

Figure 1: A. EV and PVB19 genomes detection rates in different subgroups of patients. Surgery controls = Heart valve surgery controls (Valve surgery controls) and Aorto-coronary bypass surgery controls (ACB surgery controls); *p=0.003, according to Fischer exact test; ** p=0.99, according to Fischer exact test, for all inter group comparisons performed. B. PVB19 viral loads in different subgroups of patients; Plain horizontal line corresponds to mean of each group, whereas dashed line corresponds to clinically relevant threshold for the maintenance of myocardial inflammation due to PVB19 [5]; *** p<0.005, according to Mann Whitney U test when comparing PVB19 viral load in eDCM patients versus PVB19 viral load in all controls groups; all others inter group comparisons gave non significant results. C. CD3, CD68 and HLA-DR immunohistochemical assays . A&D : negative and positive HLA DR assay; B&E negative and positive CD68 assay; C&F negative and positive CD3 assays.

Table 1:
Viral genome & inflammatory markers detection in iDCM patients group.

	Viral genome detected	Inflammatory marker detected
1	EV + PVB19 + HHV6	CD68
2	EV + PVB19	HLA-DR + CD68
3	EV + HHV6	HLA-DR + CD68
4	.	.
5	.	.
6	PVB19	.
7	PVB19 + EBV + HHV6	.
8	PVB19	CD68
9	PVB19	HLA-DR
10	PVB19	.
11	PVB19	CD68
12	PVB19	.
13	PVB19 + HHV4	CD68

- EV, PVB19, HHV6, HHV4, HHV7 or HHV1 genomes were identified in 84.6% (11/13), 80% (8/10), 100% (11/11) and 78.2% (37/47) of iDCM, eDCM, healthy heart and surgery control groups, respectively (*P*>0.05 for any intergroup comparison)
- EV RNA was detected in 23.1% (3/13) of iDCM patients but not in eDCM, healthy heart and surgery controls (*P*=0.003) (Figure 1A). Mean EV viral load was 803 copies/µg in iDCM patients.
- PVB19 DNA was detected in 76.9% (10/13), 80% (8/10), 63.6% (7/11) and 78.2% (37/47) of iDCM, eDCM, healthy heart and surgery control groups (*P*=0.99) (Figure 1A). Mean PVB19 viral loads were: 413, 346, 1428 and 71 copies/µg in iDCM, eDCM healthy heart and surgery control groups (Figure 1B).
- CD3, CD68 & HLA-DR immunohistochemical assays (Figure 1C) were positive in 53.8% (7/13), 60.0% (6/10) and 18.2% (2/11) of iDCM, eDCM and healthy heart control groups. Cardiac Inflammatory markers were detected in 100% (3/3) of EV positive iDCM and only in 60% (6/10) of PVB19 positive iDCM patients. (Table 1)

Conclusions

- EV genome detection was positive and was always associated with cardiac inflammatory markers in only 23% of iDCM patients, whereas PVB19 genome detection was detected in approximatively 70% of iDCM, eDCM healthy heart and surgery control groups and was not always associated with the presence of cardiac inflammation markers. These findings suggested that only a persistent EV infection could be an etiological cause or a cofactor in the development of a subset of iDCM cases.
- The statistically significant difference of PVB19 viral load between eDCM patients and controls could be interpreted as a alpha type 1 error due to a population bias (absence of very low PVB19 viral loads <100 copies/µg in eDCM patients group comparatively to surgery controls.)

References : 1. Kühl, Circulation 2005. 2.Andreoletti JID 2000. 3.Dickstein Eur J Heart Fail 2008. 4.Cooper J Am Coll Cardiol 2007. 5.Bock CT, NEJM 2010.

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