PERITONEAL CELL-FREE DNA:
AN INNOVATIVE METHOD FOR DETERMINING ACUTE CELL DAMAGE IN PERITONEAL MEMBRANE AND FOR MONITORING THE RECOVERY PROCESS AFTER PERITONITIS

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INTRODUCTION and AIMS

Peritonitis and exit site infections are the major complications of Peritoneal Dialysis (PD) and account for 25% of hospital admissions of PD patients. Cell-free DNA (cfDNA) is composed of circulating extracellular DNA fragment that originates from necrotic and apoptotic cells.CfDNA is present in the peritoneal effluent of stable PD patients, but there is no data on cfDNA in PD patients with peritonitis. In this study, we investigated the variation of peritoneal cfDNA levels subsequent to peritonitis in PD patients.

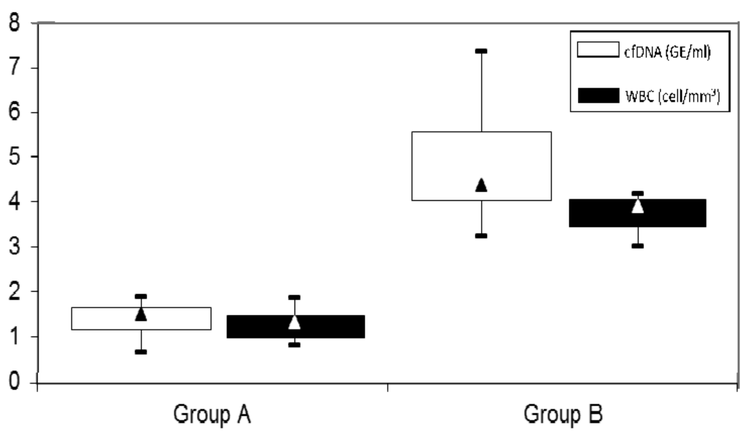
METHODS

We enrolled 53 PD patients: 30 without any history of systemic inflammation and peritonitis in the last 3 months (group A) and 23 with acute peritonitis (group B). CfDNA were quantified in peritoneal effluent by Real Time PCR. Peritoneal samples on day 1, 3, 10, 30 and until the 120th from the start of peritonitis were collected for WBC counts and cfDNA evaluation in group B.

RESULTS

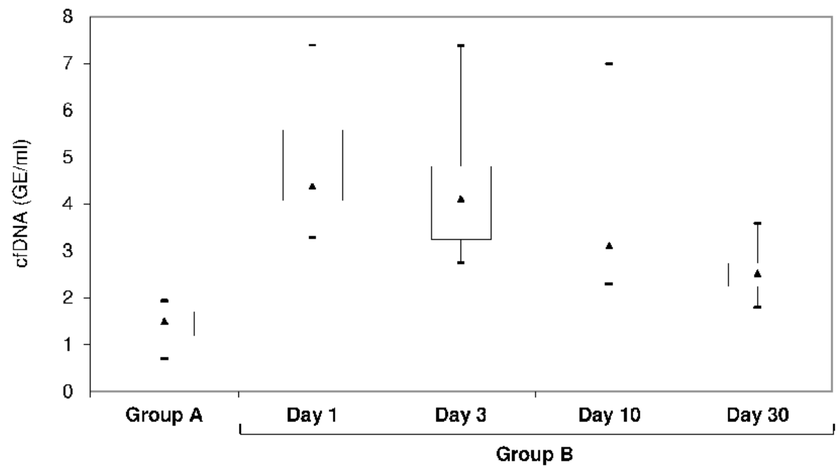
A total of 53 chronic PD patients were enrolled in this study. 73% patients were treated with continuous ambulatory PD (CAPD) and 26% with automated PD (APD). The average length of PD treatment was 21 months and the range was: minimum: 3.6 - maximum: 132.9 months. All 23 PD patients with acute episode of peritonitis were treated and clinically recovered from peritonitis after 13.5±5.4 days. 18/23 patients had a first episode of peritonitis and responded to first-line antibiotics, whereas 5/23 had a relapsing episode of peritonitis, but subsequently responded to another course of intra-peritoneal antibiotics. 3 patients required catheter removal with need to switch to hemodialysis and 4 patients dead during the study period.

Fig.1



- Quantitative analysis of cfDNA showed significantly higher levels in PD patients with peritonitis compared with patients without peritonitis ($p<0.01$), similarly as White Blood Cells (WBC) increasing during acute episode of peritonitis (**Figure 1**).
- Quantitative analysis of cfDNA showed significantly higher levels in group B on day 1, 3, 10 and 30 compared with group A ($p<0.05$) (**Figure 2**). In particular, peritoneal cfDNA level on day 30 is still significantly elevated when compared with group A.

Fig. 2



cfDNA: boxplots per day

- A significant positive correlation was observed between cfDNA concentration and WBC on day 1 (rho=0.89) and day 3 (rho=0.5)(both, $p<0.05$). However, no statistically significant correlation was observed between cfDNA and WBC on day 10 and 30.
- In group B, peritoneal cfDNA level tends to progressively decrease during follow up of peritonitis. From this decreasing curve, we estimated that 49 days are necessary to reach the value of 51 GE/ml (75percentile in controls) and 63 days to reach 31 GE/ml (median) (**Figure 3**).
- We observed a new rapid increase of cfDNA level (consistent with WBC counts) in 5 relapsing patients, at the first day of relapsing peritonitis. We reported 2 patients trends to explain these observations (**Figure 4**).

Fig. 3

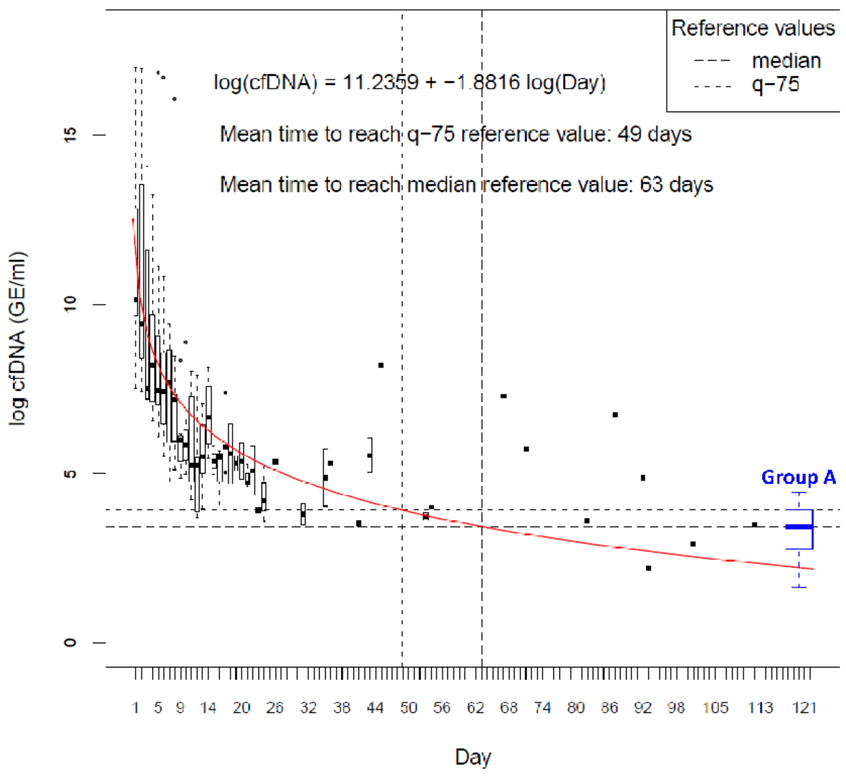
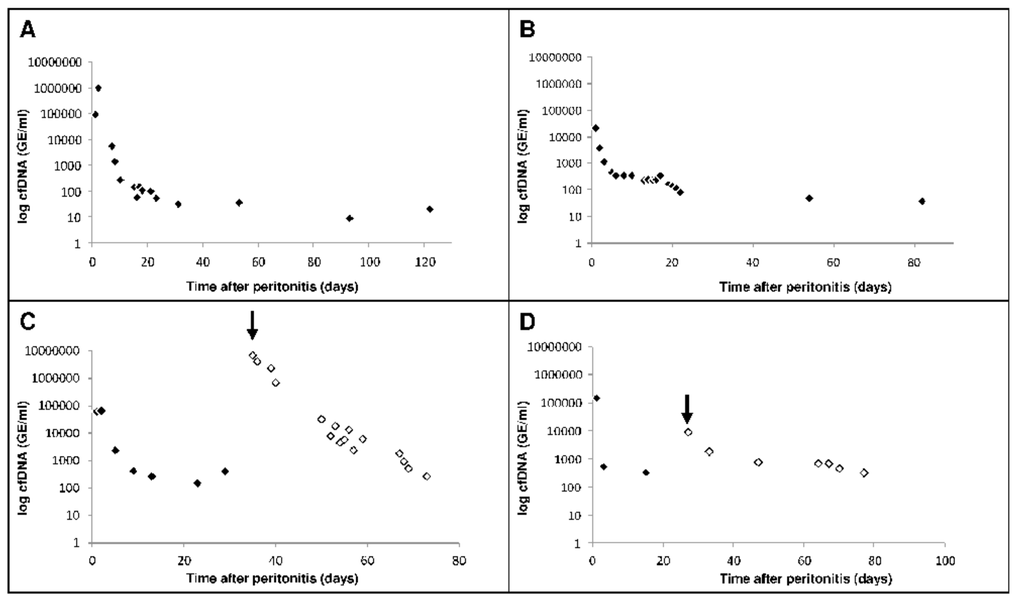


Fig. 4



CONCLUSIONS

Our results demonstrated that cfDNA increased in peritoneal effluent of PD patients with peritonitis and tended to progressively decrease in relation with membrane repair process. Peritoneal cfDNA quantification could be an innovative method to determine acute damage and an inverse index of repair process.

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L2) Dialysis. Peritoneal dialysis.
Grazia Maria Virzì
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