Background

One of the main issues in biobanking is the establishment of standard operating procedures for specimen collection, preparation and storage, to control for pre-analytical variation. For biological fluids such as serum or plasma, there is currently a lack of sensitive biomarkers for the quality control of cryopreservation conditions. In order for a serum biomarker to be used for QC, it should be ubiquitous and show 100% loss of activity upon inadequate storage conditions and temperature variations.

Methods

Immunoenzymatic assays were used to assess the stability of the following serum candidate quality control biomarkers, belonging to the coagulation super-family:

- C5a
- CD40L
- G-CSF
- CMP-12
- CXCL1
- CCL5
- IL-1a
- IL-1b
- IL-1ra
- IL-1b
- IL-2
- IL-4
- IL-5
- IL-6
- IL-8
- IL-12
- IL-23
- IL-17
- MIF
- MIP-1a
- MIP-1b
- Serpin E1
- Serpin E2
- CCL1
- CCL2
- CCL5
- CCL10
- CXCL1
- CXCL10
- CXCL11
- CXCL12
- TNF-a
- TREM-1

Chromometric assays were used to assess the stability of the following plasma candidate quality control biomarkers, belonging to the coagulation factors family:

- TP
- TCA
- Fibrinogen
- Protein S
- Protein C
- Factor II
- Factor VII
- Factor VIII

Results

Only CD40 Ligand in serum showed an on-off response to 24h storage at 37°C and protein S in plasma showed a degradation response to 9 years storage at -80°C.

Conclusions

Biomarkers that have an on-off response to temperature variation could serve as quality indicators for the core processes of biobanking, which are the preparation and storage of biological fluids. The identification of such biomarkers in serum samples is needed. This is the first time, to our knowledge, that such a biomarker is described in serum.

Figure 1

Pixel densities for ubiquitous cytokines revealed by the cytokine array after 3min (C5a, CD40L, G-CSF, CXCL1, CCL5, IL-1a, IL-1b, IL-2, IL-5, IL-6, IL-8) exposures to X-ray film. Medians, and 25th and 75th percentiles are shown. 1, 2, 3 and 4 correspond to P0 (baseline, 4h at 20°C), P5 (5 freeze-thaw cycles), P10 (10 freeze-thaw cycles), P37/24 (24h at 37°C) conditions respectively.

Figure 2

Mean values and standard deviations of sCD40L in serum, assessed by ELISA. Serum was subjected to 4h at 20°C (P0), 24h at 16°C (P16/24), 12h at 20°C (P20/12), 24h at 20°C (P20/24), 48h at 20°C (P20/48), 12h at 37°C (P37/24), or 24h at 37°C (P37/24).

Figure 3

Median values of coagulation parameters at P0 (day of collection) and P9 (after 9-year storage at -80°C); 25th and 75th percentiles are shown.