Reviewer’s report

Title: Comparative measurement of CNP and NT-proCNP in human blood samples: a methodological evaluation.

Version: 1 Date: 21 December 2012

Reviewer number: 3

Reviewer’s report:

To the Authors

General Considerations

The aim of this study was to investigate whether there are some differences of CNP and NT-proCNP levels between serum and plasma samples. In particular, this study was focused on the stability of CNP and NT-proCNP in full blood samples stored at room temperature. Authors found that levels of CNP and NT-proCNP are stable for at least two hours, even when sample processing is delayed or blood probes are stored at room temperature. Furthermore, NT-proCNP assay demonstrated more consistent and reliable data than CNP assay. As a result, Authors suggest that NT-proCNP assay should be preferably used in clinical applications.

The manuscript is concise and well written. The results are interesting, although the data reported, in particular those regarding the stability, are strictly method dependent. Authors are troubled about the great difference in results among different RIA and EIA methods (Discussion, page 6), but this discrepancy is theoretically conceivable, and so largely expected. It is well known that similar results have been reported also for the immunoassays of other natriuretic peptides, such as BNP and ANP related peptides (for a review on this important topic see: Clerico A. et al. Clin Chem 2000; 46: 1529-34; Clerico A. et al. Clin Chem 2004; 50: 33-50; Clerico A. et al. Clin Chim Acta 2012; 414: 112-9; Clerico A. et al. Adv Clin Chem 2012; 58: 31-44). For this reason a standardization/harmonization for ANP and BNP immunoassays is recommended since the dawn of this century (Clerico A. et al. Clin Chem 2000).

This large difference in results depends on the specificity of monoclonal or polyclonal antibodies used, the design of immunoassay system (competitive versus non-competitive assay), the analytical matrix (serum versus EDTA or heparinized plasma) used, and the huge number of circulating forms of CNP, as previously reported for ANP and BNP immunoassays (Clerico A. et al. Clin Chem 2000; Clerico A. et al. Adv Clin Chem 2012; 58: 31-44; Emdin M. et al. J Am Coll Cardiol 2011; 57: 1396-8). Authors should better discuss this important point in the revised version (this point should be considered as a minor essential revision).

Level of interest: An article of importance in its field
Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.