The Alzheimer’s Association QC program for CSF biomarkers

Background

The objective of the QC program is to standardize CSF biomarker measurements between Labs. The program consists of two parts, a standardized protocol for lumbar puncture (LP) and CSF sample processing, and an external QC program for comparison of CSF biomarkers between Labs.

A first meeting was chaired by Drs. Blennow and Zetterberg (Göteborgs universitet) together with Maria Carrillo from the Alzheimer’s Association. Representatives from the ADNI, Biotech companies, Clinical and Research CSF Labs and Pharma Companies participated in the meeting. This protocol is based on the discussions held at the ICAD meeting in Vienna, and following comments on the protocol by the representatives.

1) A standardized protocol for LP and CSF processing.

A protocol for standardization of LP and CSF processing is attached. This protocol can be implemented by the participating centers. The protocol can be referred to in future publications as “The Alzheimer’s Association flow chart for LP and CSF processing”. For the CSF processing and analysis part, it is recommended that the clinical laboratories in this program adhere to national quality guidelines and have received a national (or international) accreditation for medical laboratories.

2) The CSF QC program

The QC program will is open for Clinical Labs, Research Labs and Pharma Company Labs. The QC program allows a comparison of biomarker levels between Labs (and thus also publications), and also allow evaluation of the longitudinal stability for CSF biomarker levels for a Lab. Note that the aim is to standardize biomarkers between Labs, not to identify the “true” biomarker level or similar. The QC program is open for the generally (commercially) available assay formats (Innogenetics ELISA and Luminex assays, Meso-Scale assays) that are used on a more or less routine (clinical or research) basis. The program can thus be extended to other assays in the future. Since the principle is to compare CSF biomarker levels between Labs using the same assay, in-house assays or assays where samples have to be sent to a commercial Lab cannot be part of the program.

a) Between-Lab comparisons

In the QC program, 2 QC samples (aliquots of pooled CSF) are sent out to the participating Labs for each round (3 rounds per year). The QC samples will have different biomarker levels, with “normal” CSF biomarkers or with “AD pattern” (high T-tau, P-tau and low Aβ42). For each round, different QC samples will be distributed.

b) Longitudinal evaluations

In addition, one QC sample will be analyzed each round (an aliquot of the same pool), to evaluate the longitudinal stability.
The QC samples should be analyzed as part of the participants ordinary lab activities (clinical routine or research series), with the 3 extra samples added to ELISA, Luminex, or Meso-Scale plates that should have been analyzed anyway. The QC samples have been prepared from fresh CSF, and will all have gone through one freeze-thaw cycle. The QC samples have been analyzed by five Labs (Göteborg, Amsterdam, Erlange, US-ADNI/Penn and Zwijnaarde) to “set the levels”. This is done by analyzing the samples on 6 different occasions (in total 18 runs), using ELISA (Innogenetics T-tau, P-tau181 and Abeta1-42), Luminex (InnoBia AlzBio3) and Meso-Scale (Abeta triplex).

Shipment

• The samples are distributed by the Göteborg Lab, and will be sent by courier on dry ice.
• The samples are stored at the participating Labs at minus 70 or 80ºC.
• The participating Labs should make a note that the samples arrived frozen, with dry ice left, and the date of arrival.
• The QC samples for the program will be sent out together once a year, to be analyzed at three time points (February, May and October).

QC samples and analyses

• The volume of a QC CSF sample is 500 µl.
This will be enough for:
Innogenetics ELISAs (T-tau 25 µlx2, Abeta42 25µlx2 and P-tau 75 µlx2) = 250 µl
Innogenetics Luminex (75 µlx2) = 150 µl
Meso-Scale Abeta triplex (25 µlx2) and T-tau (25 µlx2) = 100 µl, or different combinations.

• The samples are labeled:
yyyy-xA and yyyy-xB (e.g. 2011-5A and 2011-5B) where yyyy equals year for analysis and x round since the beginning of the program i.e. sample 2011-5A should be analyzed in 2011 as a part of fifth round of the program.
In addition there is one sample labeled QC-Long, that should be analyzed on all occasions.

• The samples should be analyzed in duplicate.
• For reporting data, please enter the results into our report form, and send to the Göteborg Lab, by clicking the send button or by fax or e-mail, (see the report form). Please note that data on the report form from each lab should include:
  - Lab ID (preferable the number you have been assigned).
  - Condition of the samples at arrival, and storage.
  - Method used (different report from for each method).
  - Batch or lot number for the kit and calibrator (standards).
  - Expire date of the kit used.
  - Date of analysis.
  - Mean of measured levels for T-tau, P-tau and Abeta(x) in pg/ml.
  - CV of measured samples
  - Max CV for standard duplicates.
  - Internal QC samples within ±2SD from mean value.
- OD (Innogenetics ELISAs), FI (Innogenetics Luminex method) and ECL (Meso-Scale Abeta triplex) values for Blank, Lowest standard and Highest standard.

• The checklist should be filled in by the Lab and sent to the Göteborg Lab, by clicking the send button or by fax or e-mail, (see the checklists).

Data reporting for the QC program

• Results are entered into a computer program, and will be reported;
  a) as compared with mean level for all other Labs (mean, SD)
  b) as the longitudinal stability for the individual Lab (% deviation over time)
- see attached files with examples (fictive data) of a report for the between-Lab and longitudinal comparisons.

Publications

The outline of the program, the protocol for standardization of LP and CSF processing, and data/results from the program will be summarized in a joint publication with the participants as co-authors.

Information

Further information on this QC program will be posted on the programs webpage (www.neurochem.gu.se/theAlzAssQCprogram) where you also will find all protocols, checklists, report forms and some of the results from the finished rounds.

Contact

All contact regarding the program will go via our E-mail address neurochem@neuro.gu.se.

Göteborg 2010-09-30

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