ADNI Alzheimer's Disease Neuroimaging Initiative

ADNI3 first BATCH CSF analyses of $A\beta_{1-42}$, $A\beta_{1-40}$, t-tau and p-tau_{181} using the automated Roche Elecsys and cobas e 601 immunoassay analyzer system

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Introduction

The first batch analysis of ADNI3 CSF samples included a total of 498 aliquot samples from ADNI3 rollover and new participants together with longitudinal samples.

Summary

A total of 498 pristine aliquots of CSF collected from ADNI3 rollover at initial visit (n=82), new participants at BASELINE visit (n=276) and longitudinal collection (n=140 were analyzed by the electrochemiluminescence immunoassays (ECLIA) Elecsys β-Amyloid(1-42) CSF, β-Amyloid(1-40) CSF, Phospho-Tau(181P) CSF, and Total-Tau CSF on a fully automated Elecsys **cobas e** 601 instrument and a single lot of reagents for each of the 4 measured biomarkers (provided in "UPENNBIOMK10" .CSV file). These immunoassays are for investigational use only. They are currently under development by Roche Diagnostics and not commercially available yet. Included in this report are summaries for precision performance and lot-to-lot performance for these analytes (Table 1 and Figure 1).

Method

The Roche Elecsys β-Amyloid(1-42) CSF, Elecsys β-Amyloid(1-40) CSF, Elecsys Total-Tau CSF, and Elecsys Phospho-Tau(181P) CSF immunoassays were used following a Roche Study Protocol at the UPenn/ADNI Biomarker Laboratory, according to the preliminary kit manufacturer's instructions and as described in previous studies (1-3). Analyses were performed in a series of runs, each sample run one time (in singlicate) for each of the 4 biomarker tests, over the time period of February 28, 2019 through March 15, 2019 following a standard new lot rollover protocol from the manufacturer over a 10 working day timeline that involved repeated analyses of quality control samples. Acceptance criteria as documented according to the Roche Protocol in the UPenn/ADNI Biomarker Laboratory were followed for these analyses.

In each of the 12 days of performing analyses, quality control results were within stated limits to meet acceptance criteria for precision and accuracy. A lot control was performed on the first day of the lot rollover on both measuring cells for the Roche Elecsys β-Amyloid(1-42) CSF, Elecsys Total-Tau CSF, and Elecsys Phospho-Tau(181P) CSF immunoassays. The Elecsys β-Amyloid(1-40) CSF immunoassay was not included in the lot rollover as the same lot from previous studies was in use. After verifying that calibration was successful, a QC set of Level 1 and Level 2 were prepared for each of the 3 assays according to the package insert and then aliquotted into two 500 uL tubes. For the first repetition, one aliquot of Level 1 and Level 2 for each assay were ran on the analyzer. After verifying the control results were within range, the second aliquot of each level for the three assays were ran on the analyzer for repetition two. These two repetitions made up the first run. This process was repeated two hours later for run 2, again verifying the QC was within range for each assay, level, and measuring cell for all repetitions. If for any reason a control result was out of range, another reconstituted control aliquot was used to repeat measurement for both levels of QC on the affected measuring cell. These two runs were performed each day, with a rack pack calibration also being performed on days 4 and 7, within 24 hours of a new reagent rack pack being loaded onto the analyzer. The analyte measuring ranges were, lower technical limit to upper technical limit for each biomarker: 200 to 1700 pg/mL for the Elecsys β-Amyloid (1-42) CSF immunoassay, 11 to 40,300 pg/mL for the Elecsys β-Amyloid (1-40) CSF immunoassay, 80 to 1300 pg/mL for the Elecsys Total-Tau CSF immunoassay and 8 to 120 pg/mL for the Elecsys Phospho-Tau (181P) CSF immunoassay. For results that are above the upper technical limit, the result is stated as ">" the respective upper technical limit values or if below the lower technical limit, the result is stated as "<" the respective lower technical limit value in the .CSV file "UPENNBIOMK10".

The following is a brief description of the analytical performance results (Table 1 and Figure1): %CVs based on a normal and an abnormal CSF pool run throughout the 12 analytical runs ranged from 1.32 to 1.47% for t-tau and p-tau181 and from 2.97 to 3.76% for Aβ42 and Aβ40 (Table 1). These precision results are consistent with our expectation using this highly automated system. We evaluated lot-to-lot performances for Aβ42, t-tau and p-tau181 and determined acceptable results for each of these 3 analytes, all well within 10% between-lot variance. The linear regression analyses in the form of equations {Y(2019 results) = m*X(2016/2017 results on pristine replicate aliquots) + Y intercept}(see Figure 1 for Aβ42) were: Y=1.066X-105 and R^2=0.978 for Aβ42; Y=0.954X + 8.37 and R^2=0.99 for t-tau; Y=0.988X-0.22 and R^2=0.993 for p-tau181.
Exploratory Elecsys β-Amyloid(1-42) CSF immunoassay measurement results above the technical limit of 1700 pg/mL have been provided by Roche Diagnostics based on an extrapolation of the calibration curve. Thus all results above 1700 pg/mL for A42 in the .CSV file "UPENNBIOMK10" were obtained by extrapolation.

Please note:

The Elecsys β-Amyloid(1-42) CSF immunoassay in use is not a commercially available IVD assay. It is an assay that is currently under development and for investigational use only. The measuring range of the assay is 200 (lower technical limit) – 1700 pg/mL (upper technical limit). The performance of the assay beyond the upper technical limit has not been formally established. Therefore use of values above the upper technical limit, which are provided based on an extrapolation of the calibration curve, is restricted to exploratory research purposes and is excluded for clinical decision making or for the derivation of medical decision points.

Investigators should include the above disclaimer in any publication using Elecsys β-Amyloid(1-42) CSF immunoassay values above the upper technical limit.

It should also be noted that values above the measuring range for a particular sample may differ from concentration values measured by any potential future Elecsys β-Amyloid (1-42) CSF immunoassay assay.

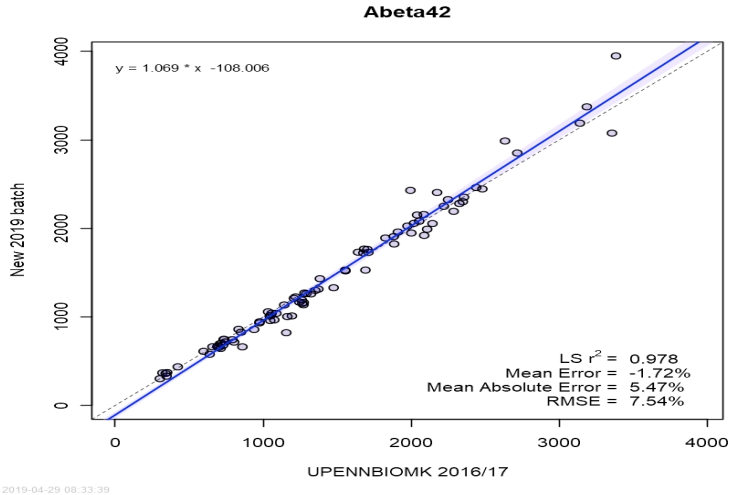
As part of the validation process for the $A\beta_{1-42}$ test method, Roche conducted collaborative studies of comparisons between the Elecsys β-Amyloid (1-42) CSF immunoassay and two reference methods (4,5) certified by the Joint Committee for Traceability in Laboratory Medicine (JCTLM) (1-5).

Ongoing analyses will further describe the technical and clinical performance characteristics for the Roche Elecsys immunoassays for detection of AD pathology.

Table 1. Precision performance using abnormal and normal CSF pools.

biomarker	Pool	N	Mean	SD	CV
Abeta40	Abnormal	12	13440	505.5	3.76
Abeta40	Normal	12	12253	364.2	2.97
Abeta42	Abnormal	12	511.3	16.22	3.17
Abeta42	Normal	12	982.5	35	3.56
Tau	Abnormal	12	554.6	7.837	1.41
Tau	Normal	12	188.2	2.761	1.47
PTau	Abnormal	12	42.18	0.5581	1.32
PTau	Normal	12	15.77	0.2277	1.44
Abeta42/Abeta40	Abnormal	12	0.03806	0.0006651	1.75
Abeta42/Abeta40	Normal	12	0.08018	0.001891	2.36

Figure1. Lot-to-lot performance for Roche Elecsys immunoassays for Aβ42, t-tau and p-tau181 (see text for more details).



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