Frequently Asked Questions

Elecsys® β-Amyloid (1-42) CSF II
Elecsys Phospho-Tau (181P) CSF
Elecsys Total Tau CSF (tTau) Assays

Q What platforms are the Elecsys β-Amyloid (1-42) CSF II, Phospho-Tau (181P) CSF and Total Tau CSF (tTau) assays available on?

The Elecsys β-Amyloid (1-42) CSF II, Phospho-Tau (181P) CSF and Total Tau CSF (tTau) assays are available on the **cobas® e** 411 analyzer and **cobas e** 601/602, 801, and 402 modules.

Q Can the Elecsys β-Amyloid (1-42) CSF II, Phospho-Tau (181P) CSF and Total Tau CSF (tTau) assays be run (and reported) individually?

While the Abeta42, pTau181 and tTau assays are run individually, the individual results must be used to calculate the ratio between pTau181 and Abeta42 (pTau181/Abeta42 ratio) and/or the tTau and Abeta42 (tTau/Abeta42 ratio) to be considered on-label use of the assays, which were cleared by the FDA as the pTau181/Abeta42 and tTau/Abeta42 ratios only. Results of the Abeta42, pTau181 and tTau assays are reported separately by the instrument and the pTau181/Abeta42 and/or tTau/Abeta42 ratios must be calculated by the middleware or LIS system.[1]

Q Since the Elecsys β-Amyloid (1-42) CSF II, Phospho-Tau (181P) CSF and Total Tau CSF (tTau) assays must be run together and reported as the pTau181/Abeta4 and/or tTau/Abeta42 ratios, are reagent, calibrators, controls, etc. ordered together?

Materials for the β-Amyloid (1-42) CSF II, Phospho-Tau (181P) CSF and Total Tau CSF (tTau) assays must be ordered separately. Additionally, customers must order the proper 2.5 mL CSF tube directly from Sarstedt (2.5 mL Low bind False bottom tube, Order No. 63.614.625).[1]

Q Why do our pTau181/Abeta42 and tTau/Abeta42 ratios have a single cutoff (positive, negative) and our competitor's (Fujirebio Abeta42/40 ratio) has a "gray zone" (negative, positive, likely positive)?

During validation, the performance of the Elecsys CSF pTau181/Abeta42 and tTau/Abeta42 ratios were found to be optimal with the use of one cutoff, therefore, a gray zone was not required. Having two cutoffs and a gray zone for an assay is not the typical approach and it is not required by regulatory bodies. In addition, having two cutoffs for one assay and one cutoff for another assay can make the comparison between them challenging. As it relates to the Elecsys CSF pTau181/Abeta 42 ratios and Lumipulse Abeta42/40 ratios specifically, a head-to-head comparison using a single cutoff for each of the ratios demonstrated similar clinical performance.[5]

Q What is the sample stability?

Sample stability claims were established through experimental data by Roche or based on reference literature and only for the temperatures/time frames as stated in the method sheets. If samples are transported/stored at room temperature (15-25°C), measurement is to be performed within 5 days after sample collection. If transported/stored refrigerated at 2-8°C up to the time of measurement. Samples can be stored at 2-8°C for up to 14 days. If transported/stored frozen at -20°C ± 5°C up to the time of measurement, samples can be stored at -20°C ± 5°C for up to 8 weeks with one freeze/thaw cycle.[1]

Q Are these assays subject to biotin interference?

The pTau181, tTau CSF on E2G platforms (402 and 801) and Abeta42 assays have no biotin interference at concentrations up to 1200 ng/mL. Due to technical limitations, the level of biotin interference on the E1G platforms is 600 ng/mL. Pharmacokinetic studies have shown that serum concentrations of biotin can reach up to 355 ng/mL within the first hour after biotin ingestion for subjects consuming supplements of 20 mg biotin per day and up to 1160 ng/mL for subjects after a single dose of 300 mg biotin.[1, 8, 9]

Q What is the pre-analytical protocol for these assays?

Abeta42 is a sticky protein that is prone to aggregation and adhering to the surface of the specimen container.[2,3] Knowing this, manufacturers of assays that measure Abeta42 outline strict specimen collection and preparation instructions. The validated preanalytical protocol described by Roche was established to minimize variability in Abeta42 measurement.[4] Due to the sticky properties of the Abeta42 protein, the cut-off for the pTau181/Abeta42 and tTau/Abeta42 ratios provided in the method sheets are only valid if the required pre-analytical handling procedure is strictly followed.

Protocols: https://go.roche.com/order-csf

Q Lumbar puncture (LP) procedures happen daily in the hospital and physician's office settings. Will there be a different collection process required for the Elecsys AD CSF ratio? If so, is training needed?

The LP can be done both in an outpatient and inpatient setting by qualified healthcare providers trained in the procedure. The collection of CSF has specific requirements meant to minimize the impact of amyloid beta adherence and adsorption on plastic (polystyrene) materials. The details of the CSF collection are included in the method sheet. The reasons for specific requirements for this collection are not Elecsys AD CSF ratio assay specific or unique, but are driven by the Abeta42 analyte characteristics. The person(s) performing the LP should refer to the method sheets for guidance and the laboratory should ensure that the information is provided and emphasized to the collection sites.[1] Customers must order the proper 2.5 mL CSF tube directly from Sarstedt (2.5 mL Low bind False bottom tube, Order No. 63.614.625).[1]

Q What is the traceability of our assays?

The Abeta42 assay has been standardized against the three certified reference materials (CRMs) ERM®-DA480/IFCC, ERM-DA481/IFCC and ERM-DA482/IFCC. These CRMs were developed by an International Federation of Clinical Chemistry (IFCC) and Laboratory Medicine working group for CSF proteins.[1,6,7]

The pTau181 assay has been standardized against a purified reference material Tau (172-205) [pThr181] amide, absolutely quantified via amino acid analysis. Calibrator values are based on weighted pTau reference material, traceable to the National Institute of Standards and Technology (NIST) amino acid reference calibrators.[1]

The tTau assay has been standardized against a reference method. Calibrator values are based on weighted purified reference tTau material, traceable to NIST amino acid reference calibrators.[1]

Q What's the difference between the pTau181/Abeta42 and tTau/Abeta42 ratios?

While the amyloid beta 42 is the biomarker common to both ratios, the pTau181 and the Total-Tau are two different biomarkers.

pTau 181 is a highly specific biomarker of AD tau pathology. It reflects the formation of hyperphosphorylated tau at position 181 and represents the accumulation in neurofibrillary tangles. It can be detected in MCI, which is the earliest symptomatic Alzheimer's disease stage.

Total-Tau biomarker mirrors the intensity of neuronal damage. It also increases in the early symptomatic disease stage (MCI) and is more indicative of the axonal degeneration that occurs in AD.[25]

Q How does the Roche pTau181/Abeta42 ratio fit into diagnostic criteria for Alzheimer's disease (AD)?

The use of AD biomarkers has been included in the new consensus research diagnostic criteria for Alzheimer's disease (AD) and mild cognitive impairment (MCI), and preclinical AD, proposed by the National Institute on Aging (NIA) and the Alzheimer's Association. These new criteria take into account that AD dementia is part of a continuum of clinical and biological phenomena.[10,11]

The NIA and Alzheimer's Association workgroups agree that both biomarkers are important for AD: "Only biomarkers that are specific for hallmark AD proteinopathies (i.e., Abeta and pathologic tau) should be considered as potential biomarker definitions of the disease" and that "evidence of abnormalities in both Abeta and pathologic tau biomarkers should be present. Furthermore, the International Working Group 2 (IWG2) criteria recommend the use of either CSF amyloid beta and tau biomarkers or PET imaging for evaluation of AD patients."[12,19]

The Appropriate Use Recommendations for Lecanemab indicate that "A positive amyloid biomarker – either elevated amyloid on PET or elevated phosphorylated tau and low Aβ42 level (increased pTau/ Aβ42 ratio) in the CSF – is required prior to initiating treatment with Lecanemab to establish that abnormal amyloid, the target of anti-amyloid MABs, is present.[13]

Q Why does Roche include pTau181/Abeta42 and tTau/Abeta42 in its ratios?

Abeta42, pTau181 and tTau are core validated biomarkers of Alzheimer's hallmark pathology that are detectable before clinical symptoms emerge.[16-18]

The IWG criteria recommend both tau and amyloid biomarkers to increase specificity for AD pathology.[12, 19]

The core CSF biomarkers (tTau, pTau, and Aβ42) are strongly associated with Alzheimer's disease and with mild cognitive impairment due to Alzheimer's disease.[20]

Extracellular plaque deposits of Aβ peptides (leading to low Abeta42 in CSF) and intraneuronal tau-containing neurofibrillary tangles (NFTs) caused by increased levels of phosphorylated Tau are the defining neuropathological features of AD.[21]

The pTau181/Abeta42 and tTau/Abeta42 ratios have been shown to have high concordance with amyloid PET and thus are preferred over single biomarkers.[1,14,15]

Elecsys CSF pTau181/Aβ42 and tTau/Abeta42 ratios are highly concordant with amyloid PET across the disease spectrum.[1,22]

Q Is it recommended that clinicians use one ratio over another? Or should they both be ordered?

Both ratios have been validated against amyloid PET and demonstrate similar performance, so both can be used for amyloid pathology detection. pTau181 is a more specific biomarker for neurofibrillary tangles than tTau,[25] thus offering an advantage to the pTau181/abeta42 ratio. In addition, the ptau181/Abeta42 ratio is specifically called out in the Appropriate Use Recommendations for Lecanemab.[13]

Q What ratios qualify patients for new amyloid-targeting therapies?

As both Elecsys pTau181/Abeta42 and tTau/Abeta42 CSF ratios have been validated against amyloid PET, both can be used for detecting amyloid pathology.[1]

Q Does Roche have a blood-based biomarker (BBBM) test for amyloid pathology?

There are currently no FDA-approved BBBMs on the market. However, the Roche BBBM pipeline continues to progress and we have received Breakthrough Device Designation (BDD) from the FDA in the future Elecsys Amyloid plasma panel (pTau181 and ApoE4). Currently, we offer some AD biomarkers as Research Use Only (RUO) assays: pTau181, ApoE4, NfL and GFAP. As RUOs, these are not for diagnostic procedures. When available, BBBMs are expected to be an initial test, followed by additional AD confirmatory testing required for those who meet certain criteria.

Q What is the difference between PET, MRI and CT imaging tools for Alzheimer's disease?

Positron Emission Tomography (PET) offers an in-vivo assessment of AD pathology, and it has been validated against autopsy. Amyloid PET is currently the only imaging tool that can confirm amyloid pathology. While MRI and CT scans are used in patients presenting with cognitive complaints, their main role is to rule out other conditions, not to detect amyloid pathology. These scans provide additional information to physicians regarding brain structure, infections, strokes, blood vessel abnormalities, normal pressure hydrocephalus, tumors, and aid in the differential diagnosis. CSF amyloid test would not reduce the need for CT or MRI as they are used at different points of the diagnostic journey for different purposes.[26,27]

Q What is the concordance between the Elecsys pTau181/Abeta42 ratio and amyloid PET?

The concordance between these two methods was found to be 90% in our validation study included in the method sheets[1]:

	Elecsys pTau181/ Abeta42 ratio	Elecsys tTau/ Abeta42 ratio
Cutoff (+)*	> 0.023	> 0.28
Cutoff (-)†	≤ 0.023	≤ 0.28
PPA%	88.2 (84.4-91.2)‡	85 (80.9 – 88.4)
NPA%	92.6 (89.1-95.1)‡	94 (90.7 – 96.2)
OPA/TPA%	90.2 (87.7-92.3)‡	89.2 (86.5 – 91.3)

PPA, positive percent agreement; NPA, negative percent agreement
OPA, overall percent agreement; TPA: total percent agreement
*Consistent with positive amyloid PET scan result.
†Consistent with negative amyloid PET scan result.
‡95% CI are calculated using a Wilson score method for binomial proportions.

Q Can our CSF biomarkers be used in conjunction with a PET scan? If so, how? Does PET enhance the specificity of CSF results or vice versa?

Yes, the Elecsys AD CSF ratio can be used in conjunction with a PET scan as well as an adjunct to other clinical diagnostic evaluations. Due to its capability to detect amyloid positivity, which was demonstrated by the high concordance with amyloid PET scan, adding this test to PET scan can increase AD diagnostic accuracy in patients with amyloid positivity.[1]

However, due to lower cost, better access, and insurance coverage, there is an increased use of CSF testing to the detriment of amyloid PET, meaning that CSF testing replaces amyloid PET for amyloid detection in patients for whom LP is not contraindicated or who are not anxious about having a LP.

The Appropriate Use Recommendations for Lecanemab indicate that "A positive amyloid biomarker – either elevated amyloid on PET or elevated phosphorylated tau and low Aβ42 level (increased p-tau/ Aβ42 ratio) in the CSF – is required prior to initiating treatment with Lecanemab to establish that abnormal amyloid, the target of anti-amyloid MABs, is present.[13]

Q What pushback should we expect from radiologists? How are radiologists compensated?

Access to PET is limited and its performance is low volume, i.e. not a major revenue stream for the radiologist. There is likely to be limited pushback from radiologists. The compensation of radiologists/ neuroradiologists depends on whether they have ownership in the practice, are employed full or part-time, independent contractor or if they are compensated on a production-based model (volume of images read). In some situations/locations, LPs are performed in the radiology departments and the procedure is reimbursed.

Q How do we anticipate imaging companies reacting to testing moving to the core lab?

Access to PET is limited and its performance is low volume. Pushback from imaging companies is not a major concern at this point. This objection will be addressed during future neurology offerings from Roche.

Q Will Roche be launching a β-Amyloid (1-40) CSF assay for customers who want to calculate the Abeta42/40 ratio?

An Elecsys assay to measure β-Amyloid (1-40) is not currently available. Abeta42, pTau181 and tTau were selected for use in two ratios: pTau181/Abeta42 and tTau/Abeta42 based on concordance predictions for amyloid status as determined by amyloid PET scan.[1,5,22]

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Roche Diagnostics
9115 Hague Road
Indianapolis, IN 46256

diagnostics.roche.com