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## FEATURED ARTICLE

# Clinical reporting following the quantification of cerebrospinal fluid biomarkers in Alzheimer's disease: An international overview

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#### Abstract

**Introduction:** The current practice of quantifying cerebrospinal fluid (CSF) biomarkers as an aid in the diagnosis of Alzheimer's disease (AD) varies from center to center. For a same biochemical profile, interpretation and reporting of results may differ, which can lead to misunderstandings and raises questions about the commutability of tests.

**Methods:** We obtained a description of (pre-)analytical protocols and sample reports from 40 centers worldwide. A consensus approach allowed us to propose harmonized comments corresponding to the different CSF biomarker profiles observed in patients.

**Results:** The (pre-)analytical procedures were similar between centers. There was considerable heterogeneity in cutoff definitions and report comments. We therefore identified and selected by consensus the most accurate and informative comments regarding the interpretation of CSF biomarkers in the context of AD diagnosis.

**Discussion:** This is the first time that harmonized reports are proposed across worldwide specialized laboratories involved in the biochemical diagnosis of AD.

#### KEYWORDS

Alzheimer's disease, cerebrospinal fluid biomarkers, clinical report, consensus approach, harmonization

## 1 | INTRODUCTION

Alzheimer's disease (AD) has gradually become one of the major global public health issues due to its prevalence, which increases with age and life expectancy, and the economic cost of caring for patients whose cognitive decline progressively leads to loss of functional autonomy.<sup>1</sup>

The diagnosis of AD is based on a multidisciplinary approach, involving, among other things, evaluation of the medical history together with clinical symptoms and signs, neuropsychological tests, and neuroimaging. The quantification of cerebrospinal fluid (CSF) core biomarkers (amyloid beta peptides [ $A\beta_{1-40}$  and  $A\beta_{1-42}$ ], total tau [t-tau] and its phosphorylated form on threonine 181 [p-tau(181)]) has progressively proven useful for the diagnosis of AD and its prodromal forms.<sup>1</sup> CSF biomarkers are now included in international guidelines for the diagnosis of AD in research settings and clinical practice<sup>2,3</sup> and the Alzheimer's Association appropriate use criteria for the use of lumbar puncture and CSF testing in the diagnosis of AD have been published.<sup>4</sup> Such biochemical diagnostics are currently implemented in many specialized centers around the world. Different methods of analysis have

been developed over the last decade and each laboratory has implemented the one best suited to its own practice. Related to this diversity there are also variations in pre-analytical and analytical conditions (such as sample tubes, storage, dilution of the biological sample, definition of cut-off values) between centers. The subsequent interpretation of the analytical results may depend on the calculation of ratios (such as t-tau/ $A\beta_{1-42}$  or  $A\beta_{1-42}/A\beta_{1-40}$ <sup>5-7</sup>), the use of scales (PLM,<sup>8</sup> Erlangen<sup>9</sup> scores), or on additional experiments (eg, dilution if t-tau is above the limit for detection<sup>10</sup>). Some laboratories mentioned the use of the A/T/N<sup>11</sup> classification, which is, however, based on data additional to CSF biomarkers, and is used more in the research setting than in the clinic. Depending on the laboratory, the type of report sent back to physicians (prescribing or referring physicians, and general practitioners) varies greatly, which may raise questions about the commutability of the tests and cause misunderstanding. It is therefore very important to harmonize comments on the reporting of results, so that the conclusions are similar regardless of where the analysis is performed.

Our work provides an overview of the procedures used in 40 centers worldwide performing CSF analysis to support AD diagnosis. For

each clinical laboratory participating in this work, we report the pre-analytical (eg, type of sample tubes, storage conditions, potential non-compliance with pre-specified local protocols) and analytical (quantified biomarkers, methods) conditions. We also detail each partner's post-analytical procedures, such as cutoff values, and use of ratios or scales for the interpretation of the results. Then, we list the clinical reports (for each biochemical profile) sent to the physicians responsible for the prescriptions. On the basis of the most frequently used reports and in-depth exchange and discussion between the participants, we propose harmonized reports adapted to each biochemical CSF profile.

This work is an essential step towards a consensual harmonization of clinical reporting after CSF analysis in the context of AD diagnosis, as advocated by the Biofluid Based Biomarkers Professional Interest Area (BBB-PIA) working group of the Alzheimer's Association. This harmonization is of great importance given the prevalence of AD and the increasing number of laboratories performing these diagnostic assays worldwide.

## 2 | METHODS

### 2.1 | Partners involved

Centers and laboratories specialized in AD diagnosis were contacted through the French Society of Clinical Biology (SFBC, <https://www.sfbc-asso.fr/>), the International Society to Advance Alzheimer's Research and Treatment (ISTAART) BBB-PIA, or the Society for Neurochemistry and Clinical CSF analysis (<http://www.neurochem.info/>). A total of 40 centers (17 French and 23 from 15 different countries, see authors' affiliations and Supplementary Figure S1 in the Supporting Information) provided different levels of information regarding their practice. For the interpretation of the surveys, each laboratory was anonymized. No personal or clinical patient data were used for this project, which therefore did not require ethical clearance. Data were collected between June and December 2020.

### 2.2 | Inquiries

Clinical laboratories performing CSF testing were asked to provide information on the pre-analytical and analytical protocols in their clinical practice (eg, type of tubes used, centrifugation or storage protocol, type of kit) and their criteria for non-conformity with local protocols. Post-analytical information was also requested, such as the cutoff values of the analytes, and the use of ratios or scales. All potential combinations of amyloid  $\beta$ , t-tau and p-tau(181) were then regrouped in eight different profiles labeled as follows: (1) "all normal," in which amyloid ( $A\beta_{1-42}$  or ratio  $A\beta_{1-42}/A\beta_{1-40}$ ), t-tau, and p-tau(181) are within reference range values; (2) "all pathological," in which all biomarkers show pathological values; (3) "amyloid," in which only amyloid values are pathological; (4) "t-tau," in which only t-tau values are pathological; (5) "amyloid & t-tau," in which amyloid and t-tau values are patholog-

### HIGHLIGHTS

- Diversity of global practices in cerebrospinal fluid biomarker quantification
- Definition of a common terminology for biochemical biomarkers' reporting
- Harmonization of clinical reporting for Alzheimer's disease diagnosis

### RESEARCH IN CONTEXT

1. **Systematic review:** The quantification of cerebrospinal fluid (CSF) biomarkers (beta-amyloid peptides, total tau, and phosphor-tau proteins) is gradually being implemented in specialized clinical laboratories as an aid in the diagnosis of Alzheimer's disease (AD). However, for the same biochemical profile, the interpretation and reporting of results may differ from one center to another, which can lead to misunderstandings and raises questions about the commutability of tests.
2. **Interpretation:** Consensus approach to provide the most accurate and informative comments regarding the interpretation of CSF biomarkers will benefit AD care, prevention, and treatment strategies.
3. **Future direction:** This work is an essential first step towards harmonization of the clinical reporting of CSF biomarkers panel for the diagnosis of AD. We also consider this work useful to new CSF biomarkers and from the perspective of defining reporting comments for emerging blood biomarkers of AD.

ical; (6) "amyloid & p-tau(181)," in which amyloid and p-tau(181) values are pathological; (7) "t-tau & p-tau(181)," in which values for both tau biomarkers are pathological; and (8) "p-tau(181)," in which only p-tau(181) values are pathological. These profiles were also associated with their corresponding values of the scales and ratios (see Supplementary Table S3). We asked the participants to provide the different reporting texts they used according to the most common biochemical profiles they encountered.

### 2.3 | Data processing and decision making

Clinical comments were compiled into a single table and returned to participants for selection. The percentage of similar reports and an initial vote to identify the two most relevant comments resulted in a short list of comments for each biochemical profile. This list was then commented on electronically and a series of video conferences was held to reach a consensus on the proposed comments for different profiles.

**TABLE 1** Pre-analytical and analytical features used by the different centers (in %) for the measurement of AD biomarkers in CSF

	Participant centers (in %)
Use of similar collection tube	94.0
Measurement on frozen sample	97.0
Automated immunoassay analyzer	88.2
Cutoff based solely on manufacturer's information	16.6
Dilution of t-tau if above limits	59.4
Systematic measurement of Ab <sub>1-40</sub>	58.1
Use of the Ab <sub>1-42</sub> /Ab <sub>1-40</sub> ratio	82.3
Use of other ratios than Ab <sub>1-42</sub> /Ab <sub>1-40</sub>	57.6
Use of scales	38.2
Turnover <= 1 week	34.5

The various comments were then translated into the different national languages of the participants. The laboratory of Prof. Sylvain Lehmann (France) in Montpellier was in charge of piloting the study and preparing, collecting, and analyzing all the survey responses.

## 2.4 | Role of the funding/sponsoring source

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors. The study supporters had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; or decision to submit the manuscript for publication.

## 3 | RESULTS

### 3.1 | Pre-analytical and analytical conditions overview

Table 1 summarizes the current practice across participating centers and Supplementary Table S1 provides detailed data on the analytical and pre-analytical procedures of each participating laboratory. The majority of centers (94%) used very similar polypropylene (PP) tubes for sample collection, and storage conditions at  $-20^{\circ}\text{C}$  for short term and  $-80^{\circ}\text{C}$  for long term. This is in line with standard operating procedures (SOPs) and the various experimental works in the field.<sup>12-15</sup> The use of collection tubes different from those recommended by each laboratory was identified as a non-conformity by 97% of centers. For secondary tubes that have a lower impact on pre-analytical variability,<sup>13</sup> we observed a greater diversity of origin, but these were PP microtubes from 0.5 to 2 mL in most cases.

For the analytical part, Fujirebio immunoassays (Lumipulse) were used in 76.5% of the centers for the quantification of the four analytes (A $\beta_{1-42}$ , A $\beta_{1-40}$ , t-tau, and p-tau[181]). Other laboratories measured the four analytes using Roche (Elecys), Euroimmun (ELISA), Fujirebio (ELISA), IBL (ELISA), or MSD (V-Plex), either alone or in combination. One center used a liquid chromatography mass spectrometry (LC-MS) approach for A $\beta_{1-42}$ . Overall, 88.2% used automated immunoassay analyzers. We observed an important heterogeneity of the cutoffs selected for the different analytes, not only for A $\beta_{1-42}$ , but also for t-tau and p-tau(181).

The selection of cutoffs by the centers was based on information given solely by manufacturers in 16.6% of cases, on literature (4.2%), on other laboratories/colleagues (12.5%), on internal data (45.8%), or on a combination of these approaches (25%). Cutoffs for A $\beta_{1-40}$  have generally not been defined, as this analyte is mainly used to calculate the A $\beta_{1-42}$ /A $\beta_{1-40}$  ratio, which has its own cutoff. A $\beta_{1-40}$  was systematically quantified in 58.1% of laboratories or added in case of discordance (ie, when A $\beta_{1-42}$  was normal but tau biomarkers were abnormal, or vice versa) in 22.6% of laboratories (Table 1 and Supplementary Table S1). Seventeen percent of the laboratories mentioned the use of a "grey zone," which corresponds to profiles where the values, often close to the cutoffs, correspond to a situation that remains undetermined.

Dilution of samples, when the upper limit of detection was exceeded, was only performed for t-tau and only in 59.4% of the laboratories. The A $\beta_{1-42}$ /p-tau(181) and t-tau/p-tau(181) ratios were computed in 39.4% and 30.3% of the laboratories, respectively. The validated PLM<sup>8</sup> and Erlangen<sup>9</sup> scales combining biomarkers are used by only 17.6% of the participating laboratories, a low percentage that is certainly an underestimate depending on the country, the Erlangen score being widely used in Germany for example. CSF biomarkers were also used by seven centers to establish the ATN<sup>11</sup> research classification that also includes imaging information.

### 3.2 | Clinical reports according to CSF biomarker profiles

Supplementary Table S3 (row 6) shows the mean and minimum/maximum frequency of the eight biochemical profiles, observed in consecutive series of samples (55 to 3000 samples) in 15 of the participating laboratories. Supplementary Table S2 lists the comments initially provided by participants for the biochemical profiles: the most frequent ones ("all normal," "all pathological," "amyloid," "t-tau") and the less frequent ("amyloid & t-tau or amyloid & p-tau[181]," "t-tau & p-tau[181]" and "p-tau[181]"). We observed very similar comments provided for "all normal," "all pathological," and "amyloid" profiles in 30%, 26%, and 36.6% of the centers, respectively. The comments for the other profiles showed more heterogeneity. In a second step, each laboratory was asked to select, from among all the reports previously listed, the two that best reflected their own current clinical practice. They were also given the opportunity to add additional comments based on available clinical information.



**TABLE 2** Summary of consensus comments for interpretation of biochemical profiles of AD biomarkers in CSF

amyloid	t-tau	p-tau(181)	Consensus comments
N	N	N	Biochemical profile not consistent with Alzheimer's disease.
P	P	P	Biochemical profile consistent with Alzheimer's disease.
P	N	N	Biochemical profile consistent with an amyloidopathy.
N	P	N	Biochemical profile not consistent with Alzheimer's disease; may be consistent with other neurodegenerative disease and/or neuronal damage. (If t-tau is close to/above upper limit of detection with a high t-tau/p-tau[181] ratio, the profile may indicate Creutzfeldt-Jakob disease)
P	P	N	Atypical biochemical profile; may be consistent with Alzheimer's disease.
P	N	P	Atypical biochemical profile; consistent with Alzheimer's disease.
N	P	P	Atypical biochemical profile; not consistent with Alzheimer's disease.
N	N	P	Atypical biochemical profile; not consistent with Alzheimer's disease.

Note to be added to all comments: This biochemical profile must be interpreted in its clinical context and in conjunction with a physician.

Abbreviations: N, normal; P, pathological; p-tau(181), tau phosphorylated at threonine 181; t-tau, total tau.

On the basis of these proposals, and after several rounds of exchanges (electronically and by videoconference) with all the partners, we generated harmonized comments for each profile, associated in some cases with additional information (Table 2 and Supplementary Table S3). We also translated these comments into 11 languages corresponding to the different countries of the participating laboratories (Supplementary Table S4). It should be noted that our survey found that less than 5% of centers use plots/graphs in addition to numerical values. In our group discussion, the consensus was not to use additional plots/graphs that might interfere or make the commentary be returned with the numeric values less clear.

## 4 | DISCUSSION

In this work, we collected information from 40 centers located in 15 different countries (Supplementary Figure S1) that measure CSF amyloid and tau biomarkers in the clinical setting. Moving from the measurement of biomarkers for clinical research, which is mainly performed on retrospective cohorts, to routine clinical measurement is a real challenge.<sup>4</sup> Even with established SOPs<sup>12</sup> and international guidelines for the handling of CSF,<sup>15</sup> pre-analytical and analytical deviations may be present in real world settings, affecting the result, and some-

times going unreported. To provide high-quality results, it is important to ensure that tests achieve sufficient levels of performance to make a meaningful contribution to diagnosis and ultimately to patient care. Finally, a critical step in the medical use of CSF testing is how the results are communicated to clinicians and, when appropriate, to patients themselves. The harmonization of reporting is in this regard essential to avoid misunderstandings while comparing results between centers and to provide accurate, informative, and harmonized information that will have an impact on prevention, care and treatment strategies.<sup>1,16</sup>

We have focused our work only on CSF biomarkers currently used in clinical practice, which are part of official guidelines<sup>2,3</sup> and measured using IVD (In Vitro Diagnostics) assays. The main context of use (COU) of these tests is the diagnosis of AD. It is particularly important to keep this COU in mind because it influences the choice of pathological cutoffs (which may vary depending on the clinical question). This also explains why comments in our proposed consensus clinical reports focus on the diagnosis of AD rather than dementia or neurodegenerative diseases in general.

The methodology to be used in the development of clinical practice guidelines is well established.<sup>17</sup> The first phase generally involves conducting a systematic review and synthesis of the literature. Different works focus on the interpretation of biochemical profiles,<sup>8,9,15</sup> but we could not find previous publications dealing with the clinical reporting of CSF results for AD diagnosis. This observation is not surprising since these tests have only recently been widely used in clinical routine. Therefore, we employed a "consensus" methodological approach directly based on the agreement among experts through iterative ratings with feedback.

Our review of the pre-analytical protocols of the different centers firstly shows that study over the last ten years of the confounding factors related to this phase<sup>12</sup> and the definition and harmonization of SOPs<sup>14,15</sup> for CSF AD biomarkers have been successfully implemented. Indeed, most of the centers use very similar PP tubes (only differing in their size and shape), which have previously shown low adsorption with amyloid  $\beta$  peptides.<sup>18</sup> However, the volume of stored samples differs among centers, and this may still affect amyloid  $\beta$  quantification. Pre-analytical procedures, including centrifugation, secondary aliquots in microtubes and freezing at  $-20^{\circ}\text{C}$  for a few days or at  $-80^{\circ}\text{C}$  were also very similar, with the exception of the secondary tubes. Rather than using fresh samples, use of frozen secondary samples may represent a more easy-to-use protocol,<sup>19</sup> which probably adapts to current numbers of tests requested.

Regarding the analytical part, in addition to the three core biomarkers  $A\beta_{1-42}$ , t-tau and p-tau(181), we observed that more than 83% of the laboratories also measure  $A\beta_{1-40}$  and thereafter compute the  $A\beta_{1-42}/A\beta_{1-40}$  ratio. The rationale is likely related to the fact that the ratio improves diagnostic performance<sup>6,20</sup> and reduces biases linked to collection tube, volume of sample, and storage. In terms of detection method, it is also unsurprising that more than 88% of the laboratories are using automated chemiluminescence immunoassays that have a reduced analytical variability<sup>21,22</sup> and offer a more flexible test throughput. Despite this, one striking finding was the wide dispersion

of the cutoff values used by the different laboratories, even for  $A\beta_{1-42}$ , which will benefit from a metrological harmonization as a result of the development of both mass spectrometry reference methods and certified reference materials.<sup>23</sup> This diversity is linked on the one hand to the different assays/protocols used, but also to the fact that most centers adapt the values proposed by the test providers using results from their own cohort (16.6% vs 45.8%, Supplementary Table S1). In addition, there is a highly variable distribution of the percentages of CSF profiles between centers (Supplementary Table S3). This is consistent with the prevalence of AD, which varies widely between centers, from a low of 25% to a high of 53%. This indirectly leads to different optimal threshold definitions; therefore, the positive and negative predictive values of the tests also vary considerably.

Regarding the interpretation and reporting of the CSF biomarker results, the inquiries from the different centers (Supplementary Table S2) showed first of all that the different comments involve "Alzheimer's disease," which is in line with the COU of the CSF biomarkers. We also note the term "biochemical profile." It should be kept in mind that if CSF biomarkers do mainly reflect the amyloid and tau pathologies of AD, they may show pathologic change without clinical symptoms and this may occur sometimes more than a decade before clinical manifestation of AD.<sup>1</sup> Nevertheless, in accordance with the National Institute on Aging and Alzheimer's Association Research Framework, AD can be defined as a biological construct,<sup>24</sup> which led us in the comments to refer to AD per se, rather than with its pathological changes.

There are several points to consider in the derivation of the consensus comments from the initial reporting of the biomarker profiles. First, when all biomarkers are pathological, most initial comments from the centers indicated that the profile is "suggestive" or "consistent" with AD, showing some caution in asserting the diagnosis. The performance of CSF biomarkers for AD is very good,<sup>1</sup> but there may still be room for improvement using other p-tau biomarkers such as p-tau(217)<sup>25</sup> or p-tau(231).<sup>26</sup> In addition, the physician's diagnosis of AD remains multidisciplinary, combining clinical history, symptomatology, neuropsychological testing, imaging, and biology,<sup>1</sup> and accordingly CSF biomarkers alone are not diagnostic of the disease. The consensus comment in this pathological situation was therefore "Biochemical profile consistent with Alzheimer's disease," and we have chosen to mention in all cases that "This biochemical profile must be interpreted in its clinical context and in conjunction with a physician" (Table 2).

Second, when all biomarkers are normal, one may clearly indicate that the profile is "not consistent" with AD. This makes sense because AD is intrinsically associated with amyloid and tau pathology and retrospective studies show that normal CSF profiles virtually rule out AD.<sup>27</sup>

Third, when only amyloid biomarkers are pathological there is an obvious consensus to indicate that the biochemical profile is consistent with an "amyloidopathy" (Table 2 and Supplementary Table S3). However, this pathological situation may also be considered indicative of the presence of AD in the disease continuum.<sup>24</sup>

Fourth, when  $A\beta_{1-40}$  is assessed, there is usually no defined cutoff value for this analyte alone. Variations of  $A\beta_{1-40}$  in frontotemporal dementia<sup>28</sup> or cerebral amyloid angiopathy<sup>29</sup> have been described but

they are minimal and therefore only useful when associated with clinical information in favor of these diagnoses. However, assessment of  $A\beta_{1-40}$  in combination with  $A\beta_{1-42}$  ( $A\beta_{1-42}/A\beta_{1-40}$  ratio) has proven to be highly informative for AD diagnosis in clinical routine<sup>6,7,20</sup> and this ratio is currently used in 88.2% of the centers.

Fifth, the profile showing an isolated increase in t-tau is present in 2.7% to 15.7% of cases, depending on the center. Initial comments indicated that this profile was not consistent with AD but rather with other neurodegeneration and/or neuronal damage (such as cerebrovascular disease or—if strongly increased—Creutzfeldt-Jakob disease [CJD]). For the latter possibility, it should be noted that a very high level of t-tau (close to or above the high detection limit of the assays) associated with a high t-tau/p-tau(181) ratio is strongly in favor of this diagnosis, if other causes of major neuronal injury, for example, stroke and encephalitis, are excluded.<sup>10</sup> The consensus comment for this profile could therefore refer to this particular situation if clinical information or the diagnostic hypothesis suggests the presence of CJD (see Table 2).

Sixth, to cover all situations, we also defined consensus comments for the less frequent profiles (< 5% of cases). Thus, when amyloid biomarkers are pathological and associated with an increase in p-tau(181) but not t-tau, one may consider that this atypical profile "is consistent" with AD. When both amyloid and t-tau biomarkers are pathological (with p-tau[181] normal), the consensus is to consider that this atypical profile "may be consistent" with AD. On the other hand, profiles with normal amyloid but abnormal t-tau and/or p-tau(181) are considered as "not consistent" with AD (Table 2). This is an important consensus decision emphasizing that AD can exist only in the presence of amyloid pathology. This situation is also reminiscent of the "suspected non-Alzheimer's disease pathophysiology (SNAP)."<sup>30</sup> It should be noted that in these cases, special attention should be paid to the search for amyloidopathy using in particular the  $A\beta_{1-42}/A\beta_{1-40}$  ratio.

A limitation of this work is that we are not exhaustive in consulting laboratories using CSF biomarkers. Therefore, some results such as the percentage of use of different ratios or scales may be biased. In addition, CSF biomarkers are not always used for the diagnosis of AD and, therefore, many centers and countries are missing from this international study.

In conclusion, this work is an essential first step towards harmonization of the clinical reporting of the CSF biomarkers panel for the diagnosis of AD. The proposed framework is adaptable and applicable to new CSF biomarkers passing regulatory criteria and prospective validations for clinical application. We also consider this work useful from the perspective of defining reporting comments for emerging blood biomarkers of AD.

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Charlotte Teunissen's institution received consulting fees from Roche and received Kits for blood-based biomarkers from Quanterix and ADX Neurosciences. She leads the Global Biomarker Standardisation Consortium, Blood based Biomarker PIA, CSF Society.

Kaj Blennow has served as a consultant (and received fees), at advisory boards, or at data monitoring committees for Abcam, Axon, Biogen, JOMDD/Shimadzu, Julius Clinical, Lilly, MagQu, Novartis, Roche Diagnostics, and Siemens Healthineers, and is a co-founder of Brain Biomarker Solutions in Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program, all outside the present work.

Daniel Alcoléa participated in advisory boards from Fujirebio-Europe and Roche Diagnostics and received speaker honoraria from Fujirebio-Europe, Roche Diagnostics, Nutricia, Krka Farmacéutica S.L., and Esteve Pharmaceuticals S.A. Dr Alcoléa declares a filed patent application (WO2019175379 A1 Markers of synaptopathy in neurodegenerative disease).

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Melanie Hart is member of the Royal College of Pathologists Special Advisory Board for Immunology—clinical scientist representative.

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Léonor Nogueira is a co-author of a patent concerning the detection of antibodies toward citrullinated protein for the diagnosis of Rheumatoid Arthritis.

Adelina Orellana has submitted MMP-10 CSF biomarker in the European Patent Office (Patent number 21382305 7, priority date April 9th 2021)

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S. Lehmann and C. Delaby had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

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#### SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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