For research article:
"When Brownian Motion Meets Clinical Laboratory Automation: A DLS-Inspired Autocorrelation Function for Characterizing Workflow Performance in Sample Processing"

Response to Reviewer 1 Comments

1. Summary

Thank you very much for taking the time to review our manuscript. Please find below our detailed, point-by-point responses to the comments received. All revisions, including those made in response to the comments and suggestions of the other reviewers, have been incorporated and are clearly indicated (using track changes) in the resubmitted manuscript.

We are grateful for the insightful and constructive feedback, which has contributed to improving the clarity and quality of our work, and we remain at your disposal for any further revisions or clarifications.

2. Questions for General Evaluation	**Reviewer's Evaluation**	**Response and Revisions**
Does the introduction provide sufficient background and include all relevant references?	Yes/**Can be improved**/Must be improved/Not applicable	*Revisions have been made to improve clarity and completeness.*
Is the research design appropriate?	Yes/**Can be improved**/Must be improved/Not applicable	*Revisions have been made to further clarify the study design and its rationale.*
Are the methods adequately described?	Yes/Can be improved/**Must be improved**/Not applicable	*The section has been revised to improve clarity and transparency. In addition, the proposed approach has been applied to real laboratory data.*
Are the results clearly presented?	Yes/**Can be improved**/Must be improved/Not applicable	*Revisions have been made to improve clarity and readability.*

Are the conclusions supported by the results?	Yes/**Can be improved**/Must be improved/Not applicable	*Improvements have been made to enhance consistency.*
Are all figures and tables clear and well-presented?	Yes/**Can be improved**/Must be improved/Not applicable	*Figures and tables have been revised for improved clarity and readability. Two additional figures have been added to the Results section.*

3. Point-by-point response to Comments and Suggestions for Authors

Comments 1: The authors propose a method, inspired by the autocorrelation function used in Dynamic Light Scattering (DLS), to characterise sample turnaround time (TAT) and workflow performance in highly automated clinical laboratories. A function $G_{exp}(t)$ is defined such that each sample contributes a value of 1 until its TAT is reached and 0 thereafter, with the normalised sum across samples yielding a decaying curve. A broken-line fitting function $G_{fit}(t)$ is introduced, from which the authors derive descriptive parameters, principally TL50 (the midpoint of the constraint range) and TLV50 (the fraction of samples completed at TL50). Seven simulated scenarios, generated in Microsoft Excel, are presented (unimodal Gaussian, unimodal random with and without delay, and bimodal random variants), together with a simulated time course of TLV50 used to distinguish buffering from non-buffering systems.
The conceptual analogy is creative, the writing is generally clear, and the topic (real-time monitoring of laboratory automation) is relevant to the readership. However, several substantive issues should be addressed before the manuscript can be considered for publication.

Response 1: *We sincerely thank the Reviewer for the careful evaluation of our manuscript and for the constructive comments. We appreciate the recognition of the originality of the proposed DLS-inspired approach, the clarity of the manuscript, and the relevance of the topic for highly automated clinical laboratories. Specifically, we have clarified the methodological description, expanded the Results and Discussion sections, included the application of the proposed method to real laboratory data to demonstrate its practical applicability, added two new figures, and strengthened the interpretation of the proposed parameters. We believe that these revisions have substantially improved the clarity, methodological rigor, and overall quality of the manuscript.*

Comments 2: Relationship to the survival function and the question of novelty. On inspection, the term inside Equation (1), namely $[|t_i - \tau_x| - (t_i - \tau_x)] / [2(|t_i - \tau_x| + \varepsilon)]$, reduces to an indicator that returns approximately 1 when t is below the sample TAT and 0 when t is above it. Consequently, $G_{exp}(t)$ is the fraction of samples not yet completed at time t, which is the empirical survival function $S(t) = P(TAT > t)$, equivalently one minus the empirical cumulative distribution function. The authors themselves invoke the survival-analysis analogy in the Introduction (Kaplan-Meier, log-rank, Cox). The manuscript would be considerably strengthened by stating this equivalence explicitly and early, then clarifying what the DLS framing and the proposed parameters add beyond a standard empirical survival or complementary cumulative distribution analysis. As written, the central object risks being a re-derivation of the survival function under new terminology. Please articulate the specific,

demonstrable advantage of TLV50 over established tail metrics such as the 90% completion time, the median, or the survival function evaluated at a clinically chosen threshold.

Response 2: *We thank the Reviewer for this important observation. We agree that the kernel used in Eq. (1) behaves as an indicator function, returning approximately 1 when the elapsed time t is lower than the TAT of sample x and approximately 0 when t exceeds it. Consequently, $G_{exp}(t)$ is mathematically equivalent to the empirical survival function of the TAT distribution, i.e., S(t) = P(TAT > t), or, equivalently, to the complementary empirical cumulative distribution function. We have now stated this equivalence explicitly in the revised manuscript [Section 2.2., Lines 184–193]. The aim of the proposed method is not to replace survival analysis, but rather to recast the empirical survival function in a discrete-time operational framework suitable for continuous monitoring of laboratory workflow. In this context, the DLS-inspired approach provides a direct description of the progressive depletion of the sample workload over time and allows a simple graphical and quantitative description of the entire analytical process, specifically of the dynamic clearance process. We revised the manuscript to be more clear on this aspect [Introduction, Lines 139–147].*

The main novelty of the proposed approach does not lie in the mathematical form of Gexp(t), but in the definition and interpretation of the associated operational parameters. In particular, TL50 is a user-defined reference time representing the expected midpoint of the analytical process, whereas TLV50 is the value of Gexp(t) evaluated at TL50, i.e., the fraction of samples that have not yet completed processing at the expected halfway point. Unlike conventional metrics such as the median TAT or the 90% completion time, TLV50 is intended as an in-process performance indicator. It allows deviations from the expected workflow to be detected before the target TAT is reached, thereby providing an early warning of delays or congestion while the analytical process is still ongoing. It is intended to summarize the central position of the dynamic clearance curve, while the upper tail can still be characterized by TLV90, for example. The same framework can naturally be extended to other user-defined checkpoints (e.g., TLV25 or TLV75) according to the operational requirements of individual laboratories. Although this concept was already discussed in the original manuscript, we have further clarified and emphasized its practical implications in the revised Methods and Discussion sections [Section 2.3., Lines 210–255; Discussion, Lines 624–641].

Accordingly, we have revised the manuscript to clarify that the proposed methodology should be interpreted as an operational implementation and parameterization of the empirical survival (or complementary cumulative distribution) function rather than as a fundamentally new probabilistic function [Section 2.2., Lines 194–198]. Its added value lies in providing a compact and routinely usable framework for comparing sample-flow dynamics between analytical systems, time periods, or sample categories.

Comments 3: Absence of real-world data. All results, including the buffering and non-buffering comparison in Figure 4, are generated from Excel RAND() values. The manuscript is therefore a proof of concept with no empirical demonstration on actual laboratory automation data. For a claim of a "novel tool for evaluating, monitoring, and comparing the performance of laboratory automation systems," at least one worked example using real intra-laboratory TAT records (for instance, a single analyser line or a defined laboratory section over a defined period) is needed to show that the parameters behave as claimed and reveal something not visible from conventional statistics. Without this, the practical contribution

remains unproven.

Response 3: *We thank the reviewer for this valuable comment. We emphasize that the manuscript is primarily methodological and that its main objective is to introduce and mathematically describe an innovative framework for representing and analyzing TAT dynamics. For this reason, the examples based on randomly generated datasets were intentionally used to illustrate the theoretical behaviour of the proposed parameters under controlled conditions, allowing their properties to be demonstrated without the confounding factors typically present in real laboratory data. To address the Reviewer's concern and further demonstrate the practical applicability of the proposed approach, we have added new sections in which our method is applied to real laboratory data [Section 2.6., Lines 300–316; Section 3.4., Lines 546–606; Discussion, Lines 672–680]. Specifically, we analyzed the turnaround times of routine complete blood count (CBC) samples processed by a fully automated clinical laboratory during a representative working day (08:00–17:00). The analysis was performed at hourly intervals to monitor the temporal evolution of laboratory workflow, and the corresponding overall daily parameters were also calculated. This application demonstrates that the proposed methodology provides meaningful operational insights into workflow dynamics and enables the identification of performance variations throughout the working day. We emphasize that the temporal sampling interval is entirely user-defined and can be adapted to the intended application. In the present example, an hourly interval was selected for clarity of presentation. However, if implemented within appropriate laboratory software, such as laboratory middleware or a laboratory information system (LIS), the proposed methodology could be applied in real-time by automatically updating the analysis at much higher temporal resolution (e.g., every minute). This would enable continuous monitoring of sample flow, providing an early warning of emerging workflow inefficiencies and supporting continuous assessment of TAT performance. We have revised the manuscript accordingly to clarify this point in the Discussion and Conclusion sections [Lines 624–641 and 705–723]. Broader validation across different laboratories and automation platforms will be the subject of future studies.*

Comments 4: The "fitting" procedure. Equation (2) is described as fitting $n_{X(min)}$ and $n_{X(max)}$. However, these quantities appear to be simply the counts of samples with TAT below and above TL50, which can be read directly from the data without any optimisation. Please specify precisely what is being fitted, by what algorithm, with what objective function, and over what free parameters. If no genuine fit is performed, the language of "fitted parameters" should be revised to "computed quantities," and the value added by $G_{fit}(t)$ over $G_{exp}(t)$ should be justified.

Response 4: *We thank the reviewer for this important comment. We agree that the description of the fitting procedure in the original manuscript was not sufficiently clear and may have led to misunderstanding. Equation (1) defines the experimental curve, $G_{exp}(t)$, directly from the observed TAT values, whereas Equation (2) describes the theoretical DLS-inspired model. The latter was fitted to the experimental data using the Microsoft Excel Solver tool by iteratively adjusting the sample fractions, $n_{X(min)}/n$ and $n_{X(max)}/n$. The optimization minimized the objective function (QSUMDIFF), defined as the sum of the squared differences between the experimental curve described by Equation (1) and the theoretical model described by Equation (2) over the entire observation interval. Therefore, $n_{X(min)}$ and $n_{X(max)}$ are not simply counts read directly from the experimental data but optimized weights associated with the two representative TAT values that best describe each subgroup. The purpose of the fitting*

procedure is not merely to reproduce the experimental DLS curve but to generate a compact analytical representation of the complete TAT distribution while preserving its overall dynamic behaviour. This reduced parameterization enables straightforward quantitative comparison of different laboratory workflows using a limited number of descriptive parameters. To clarify this point, we have revised the manuscript by explicitly describing the optimization procedure performed with Microsoft Excel Solver, the objective function used, and the role of Equation (2) as the fitted theoretical DLS-inspired model corresponding to the experimental curve defined by Equation (1) [Section 2.3., 210–223].

Comments 5: Definition and operational determination of TL50, $\tau_{X(min)}$, and $\tau_{X(max)}$. TL50 is defined as the midpoint of the range $[\tau_{X(min)} + \tau_{X(max)}]/2$ rather than the median of the observed distribution. As a result, TLV50 equals 0.5 only under symmetric distributions, and its interpretation depends entirely on how the two constraints are set. In the simulations these are fixed by design. In a real laboratory, how are $\tau_{X(min)}$ (theoretical minimum) and $\tau_{X(max)}$ (maximum admissible TAT) to be determined, and how sensitive are TLV50 and the reported intersections to these choices? A sensitivity analysis is warranted, since the headline metric is anchored to operator-selected constraints.

Response 5: *We thank the reviewer for this comment. The previous sentence "The $\tau_{X(max)}$ and $\tau_{X(min)}$ can be determined experimentally or estimated on the basis of track time course and analytical determination time for each sample" has been now clarified. We have revised the manuscript to explicitly state that $\tau_{X(min)}$ is experimentally determined as the minimum achievable TAT imposed by the analytical workflow (reaction time plus technical processing time), whereas $\tau_{X(max)}$ is a predefined upper admissible TAT established according to laboratory operational criteria, beyond which the sample is considered to exhibit an abnormal delay. Both limits are fixed before the fitting procedure, remain constant throughout the analysis, and are not fitting parameters. Accordingly, only the fractions $n_{X(min)}$ and $n_{X(max)}$ are optimized during fitting. We have clarified this point in Section 2.3 to avoid any ambiguity regarding the operational determination of these parameters and also corrected several inaccuracies throughout the manuscript [Section 2.3., Lines 224–233]. By definition [Section 2.3., Lines 234–255], TL50 is the time corresponding to 50% of completed sample and is defined from the predetermined values of $\tau_{X(min)}$ and $\tau_{X(max}$ for symmetrical distributions and could be different for other distributions. In Section 2.4. lines 268–281, the definition of TLV50 is reported, and is defined as the y-coordinate of the intersection between the experimental curve $G_{exp}(t)$ and TL50, indicating the fraction (or percentage) of samples (ranging from 0 (0%) to 1 (100%)) that have not completed their TAT at TL50 time. Therefore, TLV50 is 50% for symmetrical distribution and could be different for other distributions. Since TL50 is defined from the predetermined values of $\tau_{X(min)}$ and $\tau_{X(max)}$, TLV50 is inherently dependent on these operational constraints.*

Regarding the requested sensitivity analysis, we believe that such an analysis is not required within the proposed framework. Since $\tau_{X(min)}$ and $\tau_{X(max)}$ are established a priori (predefined operational constraints) according to the analytical workflow and laboratory-defined acceptable TAT and are kept constant throughout the analysis, they are not free parameters that are varied during model fitting. Their role is to define a fixed reference interval for a given analytical process. Consequently, TL50 and the corresponding TLV50 are intended to be interpreted and compared only under the same predefined operational conditions, rather than across different $\tau_{X(min)}$–$\tau_{X(max)}$ settings. We have clarified this aspect in the revised manuscript to avoid ambiguity regarding the interpretation of TL50 and TLV50 [Section

2.3., Lines 243–249].

Comments 6: Figure 4 demonstrates a narrative rather than a result. The buffering and non-buffering trajectories are random TLV50 values arranged into labelled regions (A, B, C, C1, C2). Without a generative model of buffering dynamics or empirical observation, this figure illustrates hoped-for behaviour rather than demonstrating that the metric discriminates real buffering capacity. Please either provide a defined dynamic model from which these trajectories emerge, or replace the simulation with real monitoring data showing recovery and non-recovery episodes.

Response 6: *We thank the reviewer for this observation. Figure 4 is not intended to represent experimental evidence or the output of a mechanistic model. Rather, as reported in Figure 4 caption [Section 3.3., Lines 521–525] it represents the "Time course of TLV50 under simulated non-buffering (left) and buffering (right) operating conditions". Thus it is purely illustrative, namely to demonstrate how the proposed TLV50 framework can be used to visualize different theoretical monitoring scenarios, including buffering and non-buffering behaviors. The trajectories were intentionally generated as representative examples and are not derived from experimental measurements or from a dynamic mathematical model. We have revised the text to explicitly state that Figure 4 is a conceptual simulation intended only to illustrate the interpretation of TLV50 trajectories [Section 3.3., Lines 517 and 543–545]. To address the Reviewer's concern, the revised manuscript now includes a real-world application [Section 2.6., Lines 300–316; Section 3.4., Lines 546–606], and in Figure 6 [Lines 569–578] we present the hourly evolution of TLV50 over a complete laboratory working day, together with the corresponding percentage of delayed samples for each hourly time interval. These real laboratory data demonstrate that the proposed framework is capable of monitoring workflow dynamics also under routine operating conditions, thereby addressing the Reviewer's request for empirical evidence of the practical applicability of the method [Discussion, Lines 672–680]. We acknowledge that validation of these scenarios with longitudinal experimental datasets represents an important direction for future work.*

Comments 7: Claim of predictive, real-time, early-warning capability. The Discussion states that the method enables detection of deviations "even before the estimated time is reached." At any time t, $G_{exp}(t)$ reports only the fraction already completed and the fraction already overdue; it is descriptive of the current state. Please clarify the mechanism by which the approach is predictive rather than contemporaneous, and distinguish it from a simple running count of in-process and overdue samples.

Response 7: *We thank the reviewer for this important comment. We agree that the proposed approach is not predictive in the sense of forecasting the final TAT of individual samples. Rather, its predictive capability is limited to the interval between TL50 and $\tau_{X(max)}$. Specifically, if, at TL50, the fraction of samples that have completed the analytical process is lower than expected (i.e., an abnormally high TLV50), this indicates that more samples than the expected 50% are still being processed. Although these samples have not yet reached $\tau_{X(max)}$, their persistence beyond TL50 suggests that the analytical workflow is slowing down and that a proportion of them will likely exceed the predefined maximum admissible TAT if the current trend continues. Therefore, the proposed approach provides an early operational warning of emerging workflow deterioration before delayed samples actually reach $\tau_{X(max)}$. We have revised the Discussion to clarify that the forecasting capability refers to this short-term*

anticipation of delay development rather than prediction of the final TAT of individual samples [Discussion, Lines 624–641].

Comments 8: Statistical reliability threshold. Table 1 states that more than 30 samples suffice for statistical reliability, without justification. For characterising the tail of a skewed TAT distribution and for stable estimation of survival at a chosen time point, 30 observations may be inadequate. Please provide the basis for this threshold, ideally with a short analysis of estimator variance as a function of n.

Response 8: *We thank the Reviewer for this comment. In the present work, the threshold of n > 30 was intended only as a conventional reference for the theoretical simulations rather than as a strict criterion for accurate estimation of the tails of the TAT distribution. It was selected because n > 30 is widely adopted as a practical minimum sample size in statistical analyses, based on the Central Limit Theorem. According to this point, for sufficiently large sample sizes (commonly approximated as n > 30), the sampling distribution of the sample mean approaches a normal distribution regardless of the underlying population distribution. Therefore, n > 30 was considered an appropriate reference to represent a typical statistical scenario in the theoretical simulations. It is recognized, however, that more complex, highly skewed, or heterogeneous distributions may require substantially larger sample sizes to ensure reliable characterization, particularly of the distribution tails. Since the present work introduces a theoretical framework through representative simulated scenarios, rather than establishing minimum sample size requirements for practical applications, this conventional threshold was considered appropriate. To avoid any misleading interpretation, we have now revised the manuscript to clarify that > 30 does not represent an intrinsic requirement of the proposed methodology and should not be interpreted as sufficient for all real and practical applications [Section 3.1., Table 1 and Lines 326–333].*

Comments 9: Multiclass applicability. The requirement that TAT values of different classes not overlap is a strong constraint, as real laboratory TAT distributions for serum, plasma, and other matrices typically overlap substantially. Please discuss how the method performs under overlap, and temper the claims regarding routine multiclass deployment accordingly.

Response 9: *We thank the reviewer for this important comment. We agree that overlap between TAT distributions may occur in routine laboratory practice. However, the statement in the manuscript refers to a mathematical property of the fitting procedure rather than to a practical characteristic of laboratory workflows, regardless of the mathematical model employed. Simultaneous multiclass fitting is possible only when the different sample classes exhibit sufficiently distinct TAT ranges, allowing the corresponding parameters to be resolved independently. If two or more classes have substantially overlapping TAT distributions, the fitting procedure cannot uniquely discriminate their individual contributions, resulting in parameter overlap and reduced parameter identifiability. Therefore, multiclass analysis is intrinsically limited to sample classes characterized by distinct TAT distributions. When substantial overlap exists, separate correlation functions should be generated and analyzed independently for each class. We have revised the manuscript to clarify that this limitation is inherent to the fitting procedure itself and is not specific to the proposed methodology [Section 2.5., Lines 288–298].*

Comments 10: Terminology. "Correlation function" and "autocorrelation function" are,

strictly, misnomers in this context, since no correlation between two signals is computed; the quantity is a completion or survival decay. Retaining the DLS analogy is reasonable for exposition, but the precise mathematical meaning should be stated so that readers from laboratory medicine are not misled. Relatedly, the title refers to "particle physics," whereas DLS and Brownian motion belong to soft-matter and physical chemistry rather than to particle physics; please reconsider the title for accuracy.

Response 10: *We thank the reviewer for this comment. We agree that the proposed function is not an autocorrelation function in the strict classical sense. The terminology was intentionally adopted by analogy with the autocorrelation function used in Dynamic Light Scattering (DLS), from which the mathematical formulation of the proposed framework was inspired. To avoid possible misunderstanding, we have revised the manuscript to explicitly state that the proposed function is inspired by the autocorrelation function used in DLS analysis and describe sample completion dynamics [Introduction, Lines 142–146].*
We have revised the title to avoid the potentially misleading expression "Particle Physics" and to more accurately reflect the physical principles underlying the proposed approach. The new title is: "When Brownian Motion Meets Clinical Laboratory Automation: A DLS-Inspired Autocorrelation Function for Characterizing Workflow Performance in Sample Processing."

Comments 11: Situating the framework within healthcare workflow analytics. The Discussion advances claims about operational optimisation, performance monitoring, and cost-effectiveness in healthcare systems, but these claims are not anchored in the broader literature on healthcare workflow and operational analytics. The manuscript would benefit from connecting the proposed monitoring framework to prior work that characterises clinician and system workflow behaviour and operational performance in digital health settings. I recommend the authors cite the following relevant study, which examines workflow patterns, user engagement, and operational performance through mixed methods and provides a useful frame for interpreting performance-monitoring metrics in clinical operations: Sumner, J.; Shankar, R.; Bundele, A.; Yap, A.; Mohamed Ali, J.; Teng, G.G.; Phang, K.F.; Yip, A.W.; Lim, Y.W. Insights from high and low clinical users of telemedicine: A mixed-methods study of clinician workflows, sentiments, and user experiences. International Journal of Medical Informatics 2025, 184, 106044. https://doi.org/10.1016/j.ijmedinf.2025.106044. Incorporating this reference in the Discussion would strengthen the positioning of the proposed metric within established approaches to monitoring and interpreting workflow performance in healthcare systems.

Response 11: *We thank the reviewer for this suggestion. The study recommended by the reviewer addresses clinician workflows, user engagement, and telemedicine adoption, whereas the present work focuses on automated clinical laboratory workflows, specifically on the mathematical characterization and near real-time monitoring of analytical turnaround time (TAT) during sample processing. Although both studies concern healthcare operations, they investigate different types of workflows, with distinct objectives, methodologies, and operational contexts. The scientific background relevant to automated laboratory workflows and laboratory automation has already been discussed in the Introduction, while the Discussion was intentionally focused on the interpretation and potential*

applications of the proposed monitoring methodology. Therefore, we believe that the suggested reference falls outside the specific scope of the present study and have not included it.

Comments 12: Reproducibility. Given that the entire manuscript rests on simulation, the random seeds, the exact transformation functions applied to RAND() for each of the seven cases, and the code or spreadsheets used to generate Figure 4 should be provided as supplementary material. "Available on request" is insufficient for a methods paper whose contribution is the simulation itself.

Response 12: *We thank the reviewer for this comment. We would like to clarify that the proposed methodology is not based on a specific stochastic realization. The RAND() function was used only to generate representative TAT values following the predefined simulated distributions described in the Materials and Methods. The seven simulation scenarios are fully defined by the mathematical framework and are subsequently analyzed through the fitting procedure described by Equation (2). The parameters reported in the manuscript, including those used to generate Figure 4, are obtained from the fitted distributions (namely fitted parameters) rather than from the particular sequence of random numbers generated by RAND(). Consequently, different random realizations produce different individual simulated TAT values but lead to the same representative behavior of the fitted parameters. Since the mathematical equations governing both the simulation scenarios and the fitting procedure are fully described in the manuscript, we believe that the methodology is reproducible without requiring a specific random seed or spreadsheet. Since the objective was to illustrate the behavior of the proposed mathematical framework under predefined TAT distributions rather than to reproduce a specific random realization, the conclusions do not depend on a particular sequence of random numbers. Different realizations generated using the same equations produce different individual TAT values but preserve the same distribution characteristics and the corresponding behavior of the proposed method. Therefore, we believe that the methodological description provided in the manuscript is sufficient.*

Comments 13: Sensitivity to the smoothing constant ε. The constant ε is introduced to avoid indeterminate values at $t_i = \tau_x$, but its magnitude and its effect on the curve near the transition are not reported. Please state the value used and demonstrate that conclusions are insensitive to it.

Response 13: *We thank the reviewer for this comment. We clarify that ε is not a smoothing parameter, but a numerical regularization constant introduced only to avoid division by zero when $t_i=\tau_x$. In all analyses, ε was fixed at 10^{-12}, which is several orders of magnitude smaller than the time resolution of the analysis, i.e. 1 minute. Therefore, ε affects only the singular point at $t_i=\tau_x$ and has no practical influence on the shape of the curve, TLV values, or fitted parameters. We have now reported the value of ε in the revised manuscript and clarified its numerical role [Section 2.2., Lines 177–179].*

Comments 14: The keyword "statistical parametric mapping" appears in the abstract keyword list but the concept is never used in the manuscript. Please remove it or address it in the text.

Response 14: *We thank the reviewer for pointing this out. We agree that the keyword "statistical parametric mapping" was not discussed in the manuscript. Accordingly, it has been removed from the*

keyword list in the revised version.

Comments 15: Several typographical and grammatical corrections are needed, including: "for a for a single class" (repeated phrase in the captions of Figures 1, 2, and 3); "smaller particle diffuse more rapidly" (line 128) should read "particles diffuse"; "allow evaluation the system's" (line 143); "50% of sample will have complete their TAT" (line 187) should read "samples will have completed"; "total number of sample" (Table 2) should read "samples"; "hepararinized plasma" (line 217) should read "heparinized"; and "Total Automaton" in reference 3 should read "Total Automation."

Response 15: *We thank the reviewer for carefully identifying these typographical and grammatical issues. All the reported typographical and language errors have been corrected throughout the revised manuscript. Regarding Reference 3, we would like to clarify that "Total Automaton" is not a typographical error but is the original title of the published article, as reported by the publisher and indicated in the corresponding doi: 10.2147/JHL.S362614. Therefore, we have retained the original title to ensure consistency with the published reference and its DOI record.*

Comments 16: The symbol for the second sample class (β) does not render in several locations in Sections 3.2.3 and in Table 4, appearing as a blank. Please correct throughout.

Response 16: *We thank the reviewer for this comment. We carefully checked the manuscript and confirmed that the symbol β is correctly displayed throughout the document, including Sections 3.2.3 and Table 4. Therefore, no modifications were required.*

Comments 17: Subscript formatting for TAT_X, $\tau_{X(min)}$, and $\tau_{X(max)}$ is inconsistent across the text, tables, and figure captions. Please standardize.

Response 17: *We thank the reviewer for this comment. We carefully checked the manuscript, tables, and figure captions, and the subscript formatting for TAT_X, $\tau_{X(min)}$, and $\tau_{X(max)}$ appears to be displayed consistently in the submitted version. Therefore, no corrections were required.*

Comments 18: Equation (1) and Equation (2) would benefit from a worked numerical example for a small set of samples, which would help readers verify the reduction to a step function described in major comment 1.

Response 18: *We thank the reviewer for this suggestion. We believe that the requested worked example was already illustrated in the original manuscript in Figure 3 (panels b and c). However, the stepwise behavior of the experimental correlation function and its fitting is directly visible in the newly introduced Figure 5 [Section 3.4., Line 550], reporting representative real laboratory example. Since this behavior follows directly from the mathematical formulation of Equations (1) and (2), we believe that including an additional worked numerical example would be redundant and would not provide further methodological insight.*

Comments 19: In Table 1, "Number of objects required for statistical reliability" lists 10^3 to 10^{13} for DLS against more than 30 for the proposed method; please ensure the comparison is like

for like, since these refer to different statistical objects.

Response 19: *We thank the reviewer for this comment. We agree that the reported values refer to different statistical objects. However, this is precisely the purpose of Table 1, which is intended to highlight both the similarities and the fundamental differences between conventional Dynamic Light Scattering and the proposed laboratory automation approach. In DLS, statistical reliability requires a very large number of scattering particles because the measured signal arises from their collective Brownian motion. In contrast, the proposed method analyzes laboratory samples as the statistical units. Therefore, the comparison is not intended to imply equivalence between the two quantities, but rather to emphasize the different statistical nature and scale of the two systems. To avoid possible misunderstanding, we have clarified this point by revising Table 1 [Section 3.1., Table 1 and Lines 326–333].*

4. Response to Comments on the Quality of English Language

Point 1: The English is fine and does not require any improvement.
Response 1: We thank the reviewer for the positive evaluation of the English language.

5. Additional clarifications
We have no additional comments to provide.