Kinetically Tunable, Subzero-Active, Visual Time-Temperature Indicators Based on the Permanganate/Oxalate Reaction

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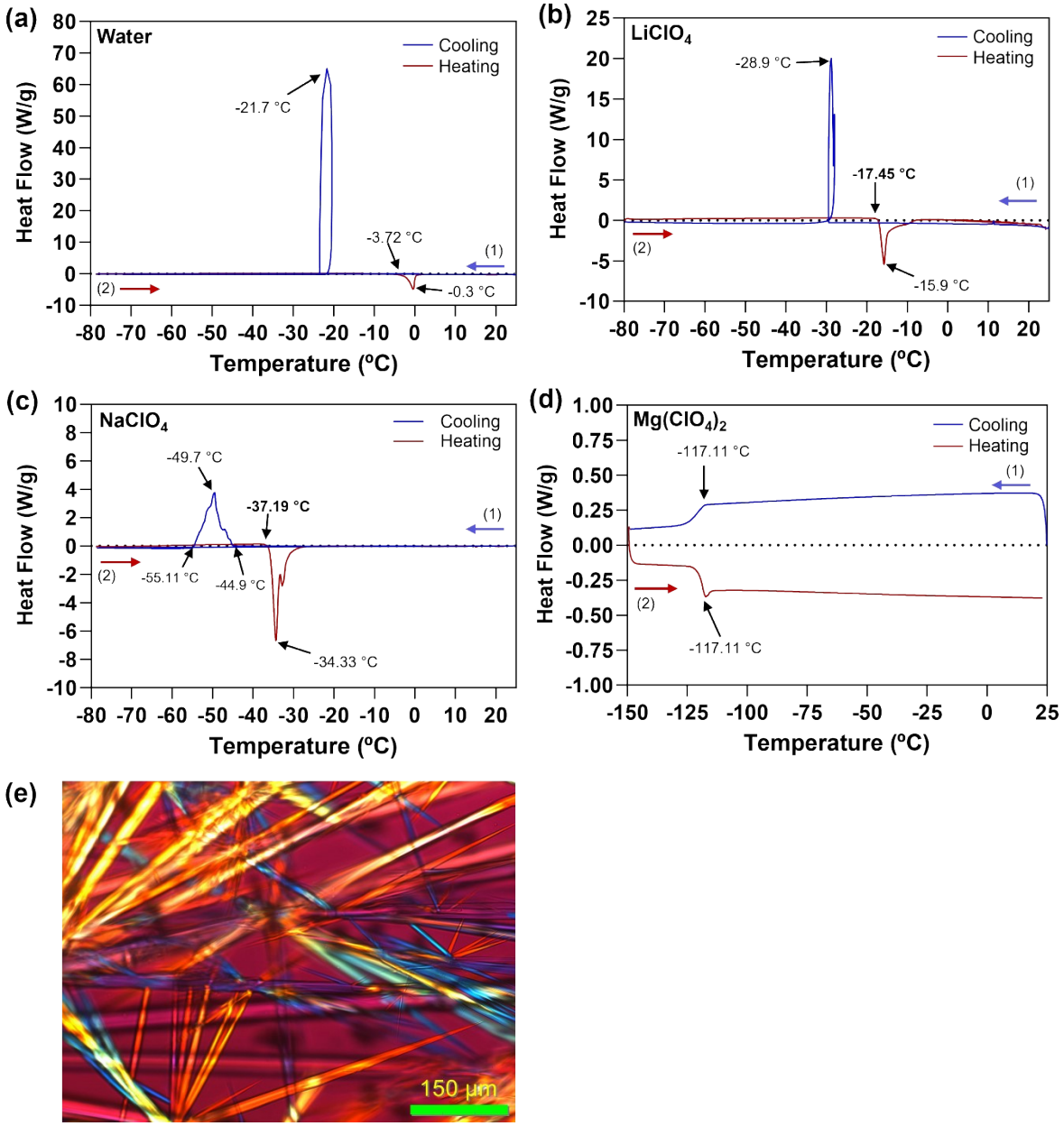
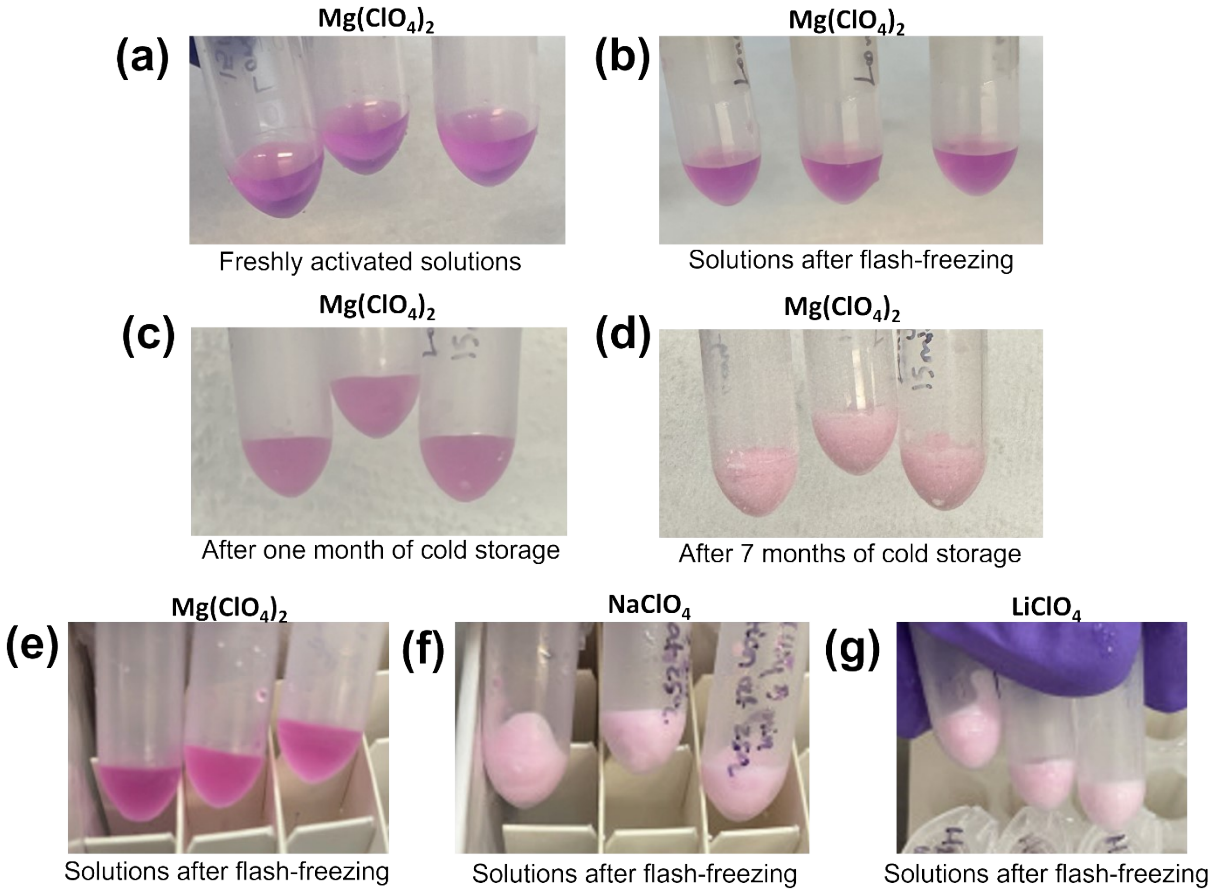
Differential Scanning Microscopy

DI water and eutectic solutions of $LiClO_4$, $NaClO_4$, and $Mg(ClO_4)_2$ were analyzed by differential scanning microscopy (DSC) as described in section 2.3 of main manuscript.

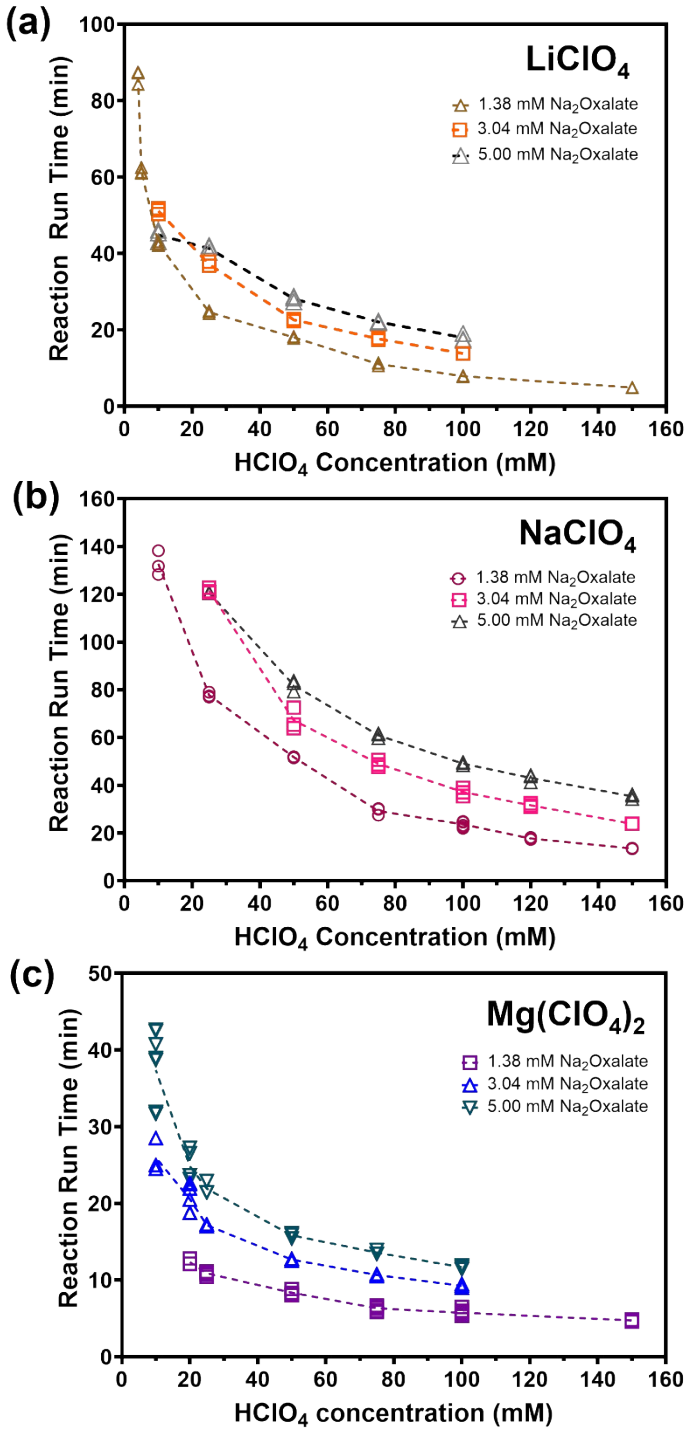


SI Figure S1. Thermographs of (a) 18 MΩ-cm deionized water, (b) 25% (*w*/*w*) $LiClO_4$, (c) 52% (*w*/*w*) $NaClO_4$ and (d) 44% (*w*/*w*) $Mg(ClO_4)_2$. Cooling step (1) was a decrease of 10 °C/min from 25 °C down to -80 °C (for a-c) and down to -150 °C (for d). The temperature was held for one minute and then the heating step was initiated. Heating step (2) was an increase of 10 °C/min up to 25 °C. Peaks in the positive direction are exothermic, representing solidification. The peak in the negative or endothermic direction represents the melting point. A cold-stage microscopy image (e) revealed the presence of crystals in the $Mg(ClO_4)_2$ solution at -68 °C, formed in 2 out of 6 trials.

Freezing of Antifreeze-based TTIs

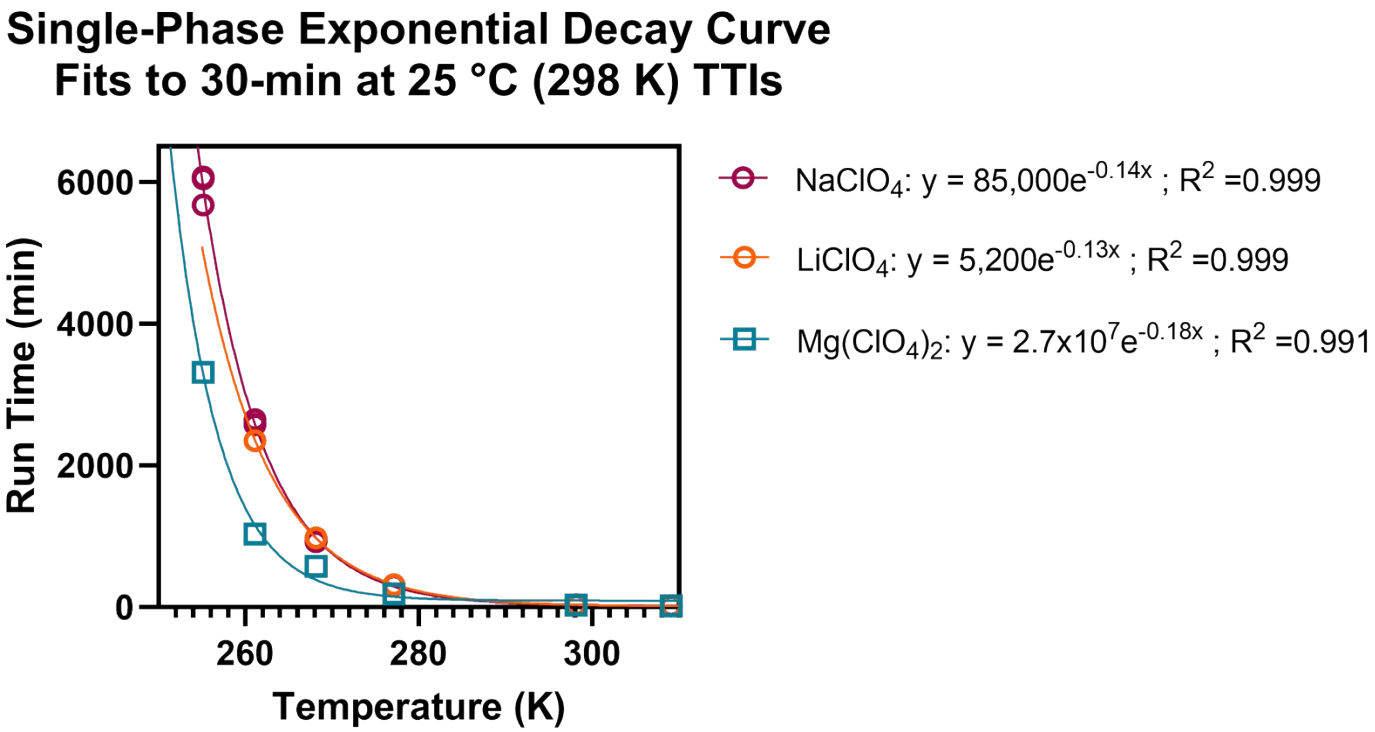


SI Figure S2. Freezing of Antifreeze-based TTIs: (a-e) $Mg(ClO_4)_2$, (f) $NaClO_4$, and (g) $LiClO_4$. (A) Freshly activated 15-minutes-at-25°C $Mg(ClO_4)_2$ system, each in triplicate. Solutions are in the liquid state. (b) Same TTI as in (a) after flash-freezing in dry ice and ethanol bath for 10 minutes. There is not an obvious (visual) crystallization or solidification of the solutions, but they seemed to be in a solid state based on the lack of movement when shaken. (c) After a month of storage in a LN_2 freezer (-140 °C) of the flash-frozen TTIs from (b). The temperature of the freezer is considerably below the reported freezing point of $Mg(ClO_4)_2$, however, the decrease in temperature did not cause the crystallization of the solutions. (d) After seven months stored in a LN_2 freezer (-140 °C). The solutions crystallized, changing their physical appearance, this changed took place after 5 months. However, similar experiments with $Mg(ClO_4)_2$ systems have shown crystallization as soon as one month of storage at -80 °C or have not shown crystallization yet after 26 months of storage. The 30-minutes-at-25°C $Mg(ClO_4)_2$ system showed crystallization after 8 months of storage at -140 °C. For comparison, (e) 5-minutes-at-25°C $Mg(ClO_4)_2$ system, (f) 5-minutes-at-25°C $NaClO_4$ system, and (g) 5-minutes-at-25°C $LiClO_4$ system right after flash-freezing in a dry ice and ethanol bath for 10 minutes. $NaClO_4$ and $LiClO_4$ show crystallization as soon as they pass their freezing point.



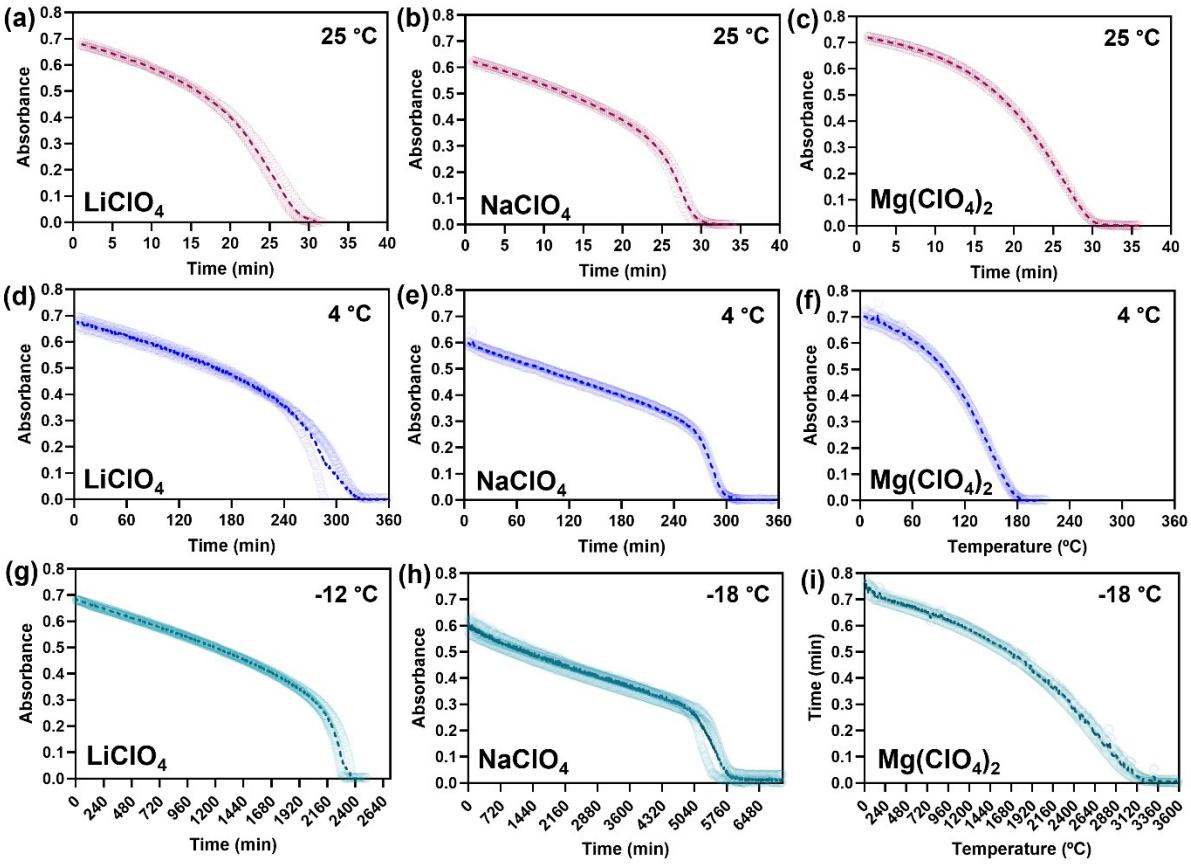
SI Figure S3. Interplay of $HClO_4$ and $Na_2C_2O_4$ in the three antifreeze-containing systems: (a) $LiClO_4$, (b) $NaClO_4$, and (c) $Mg(ClO_4)_2$. The $HClO_4$ is varied between 10 mM and 150 mM (x-axis), while the $Na_2C_2O_4$ is tested at 1.38 mM, 3.04 mM and 5.0 mM. No Mn(II) (as $Mn(ClO_4)_2$) was added in these experiments. For all experiments, n ≥ 3. All replicates are represented by the symbols, while the dashed line connect the mean values.

Fitting to the 30-min at 25 °C (298 K) TTIs



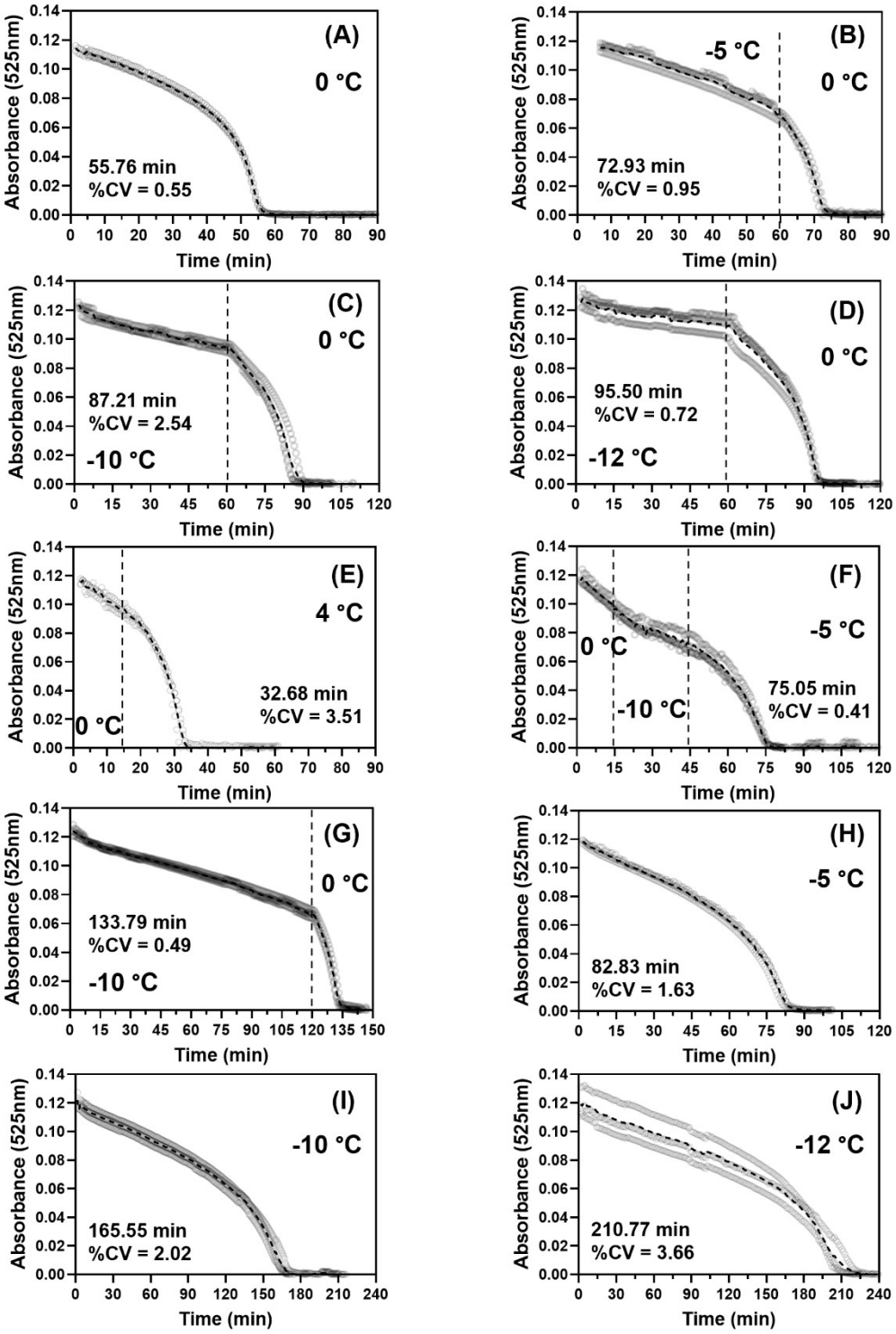
SI Figure S4. Single-phase exponential decay curve fits to the 30-min at 25 °C (298 K) TTIs run in the 3 different antifreeze salt systems. Fitted equations and their R^2 values are shown. The pre-exponential terms correspond to the approximated run times (in min) at the melting point of the antifreeze system.

Absorbance Profiles of Antifreeze-based TTIs



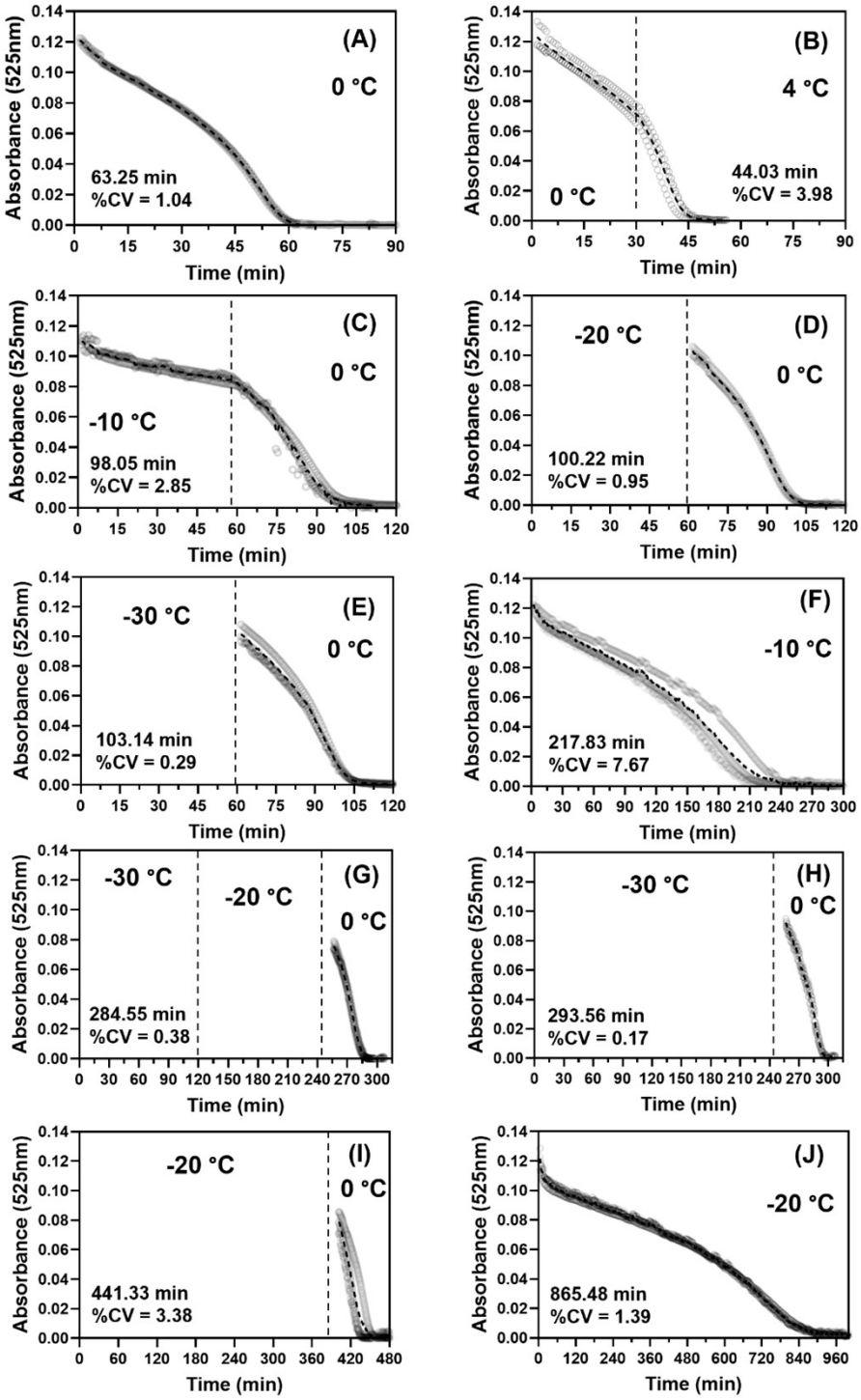
SI Figure S5. Absorbance profiles of the 30-min at-25 °C TTIs using (a) $LiClO_4$, (b) $NaClO_4$, and (c) $Mg(ClO_4)_2$ antifreeze salts. For comparison, the three systems were also run at 4 °C (d-f). For subzero temperatures, (g) $LiClO_4$ was run at -12 °C, while (h) $NaClO_4$ and (i) $Mg(ClO_4)_2$ were run at -18 °C. For all cases, triplicates were performed (data points shown with circles). The dotted line represents the mean.

Temperature Variation in $LiClO_4$ System



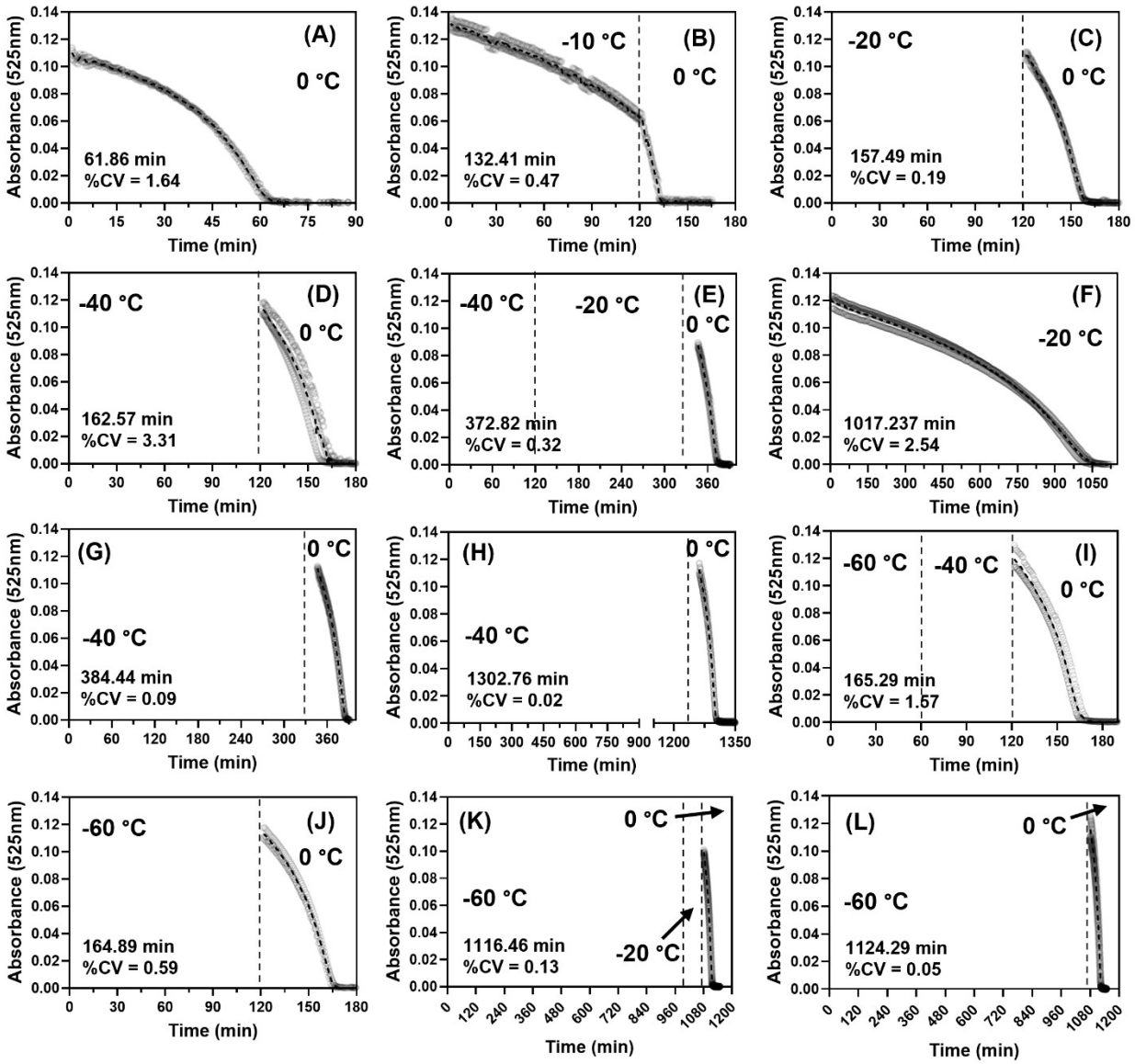
SI Figure S6. Absorbance profiles of the ten scenarios presented in Fig. 3A for the $LiClO_4$ system that runs 60 min at 0 °C. Temperature is varied while the reaction is running. Temperatures are shown in each panel, with the vertical line showing when there was a temperature change. All scenarios were run in triplicates (circles), the dotted line represent the mean value. Each panel also includes the average run time and %CV. Absorbance values ranged between 0 and 0.14 as a light path of 0.1 cm was used, differing from other absorbance data from experimented that used a path light of 0.555 cm (or the values were adjusted for direct comparison).

Temperature Variation in $NaClO_4$ System



SI Figure S7. Absorbance profiles of the ten scenarios presented in Fig. 3B for the $NaClO_4$ system that runs 60 min at 0 °C. Temperature is varied while the reaction is running. Temperatures are shown in each panel, with the vertical line showing when there was a temperature change. If absorbance data are missing, the storage was performed in the messenger freezer without data collection. All scenarios were run in triplicates (circles), the dotted line represent the mean value. Each panel also includes the average run time and %CV. Absorbance values ranged between 0 and 0.14 as a light path of 0.1 cm was used, differing from other absorbance data from experimented that used a path light of 0.555 cm (or the values were adjusted for direct comparison).

Temperature Variation in $Mg(ClO_4)_2$ System



SI Figure S8. Absorbance profiles of the twelve scenarios presented in Fig. 3C for the $Mg(ClO_4)_2$ system that runs 60 min at 0 °C. Temperature is varied while the reaction is running. Temperatures are shown in each panel, with the vertical line showing when there was a temperature change. If absorbance data are missing, the storage was performed in the messenger freezer without data collection. All scenarios were run in triplicates (circles), the dotted line represent the mean value. Each panel also includes the average run time and %CV. Absorbance values ranged between 0 and 0.14 as a light path of 0.1 cm was used, differing from other absorbance data from experimented that used a path light of 0.555 cm (or the values were adjusted for direct comparison).

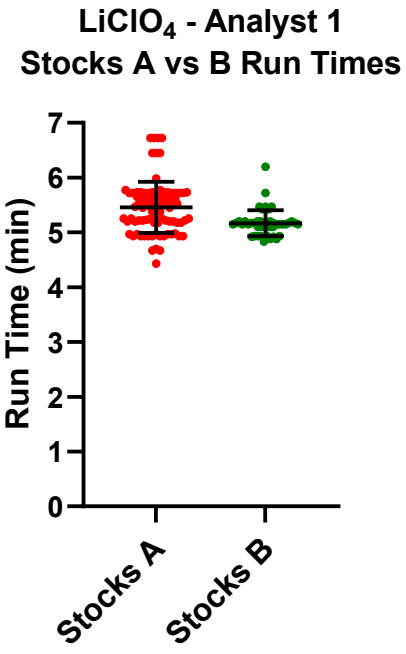
SI Figure S9. Robustness of run time and reaction trajectory consistency under conditions of variable stock solutions (Stocks A vs. Stocks B), reagent lots (Stocks A vs. Stocks C), or Analysts. (Pages 10-29)

Analyst 1's LiClO4 Stocks

$LiClO_4$ - Analyst 1 Stocks A vs Stocks B

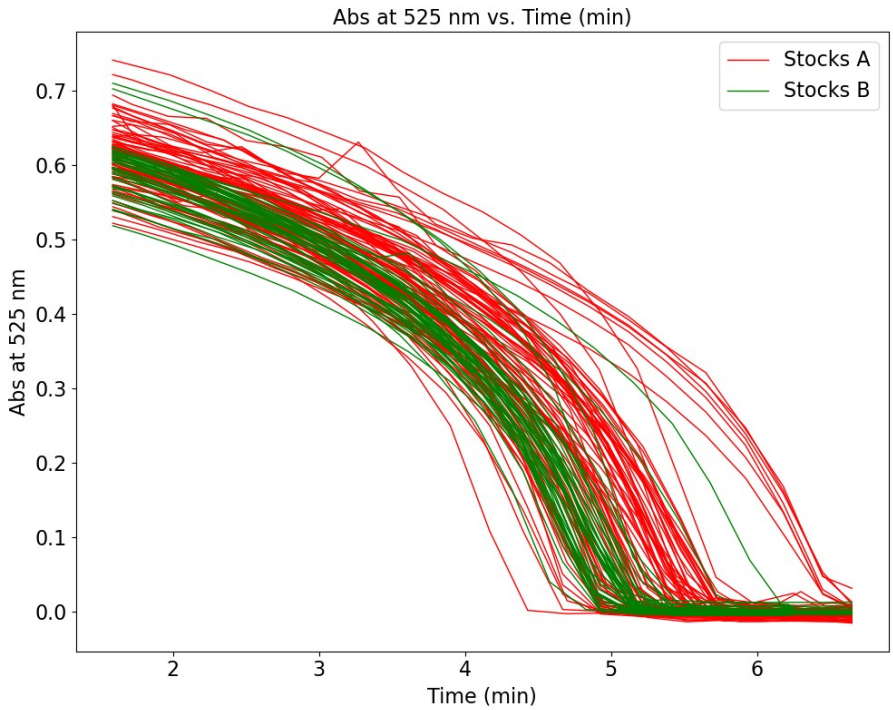
Run Times

ROPE as % of Combined Mean: 8%



Trajectory Consistency

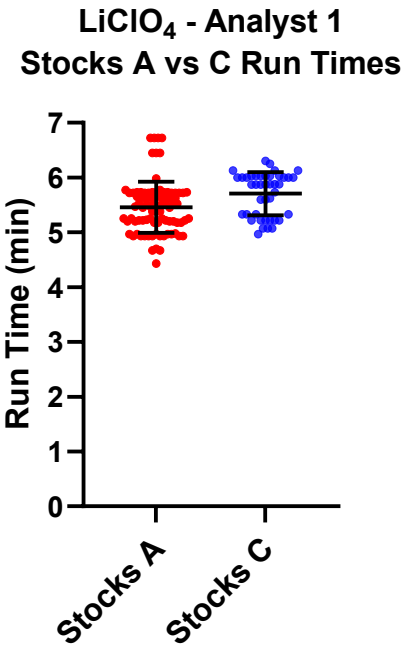
Analyst 1 Stocks A vs Stocks B: 6.3% RMSD



$LiClO_4$ - Analyst 1 Stocks A vs Stocks C

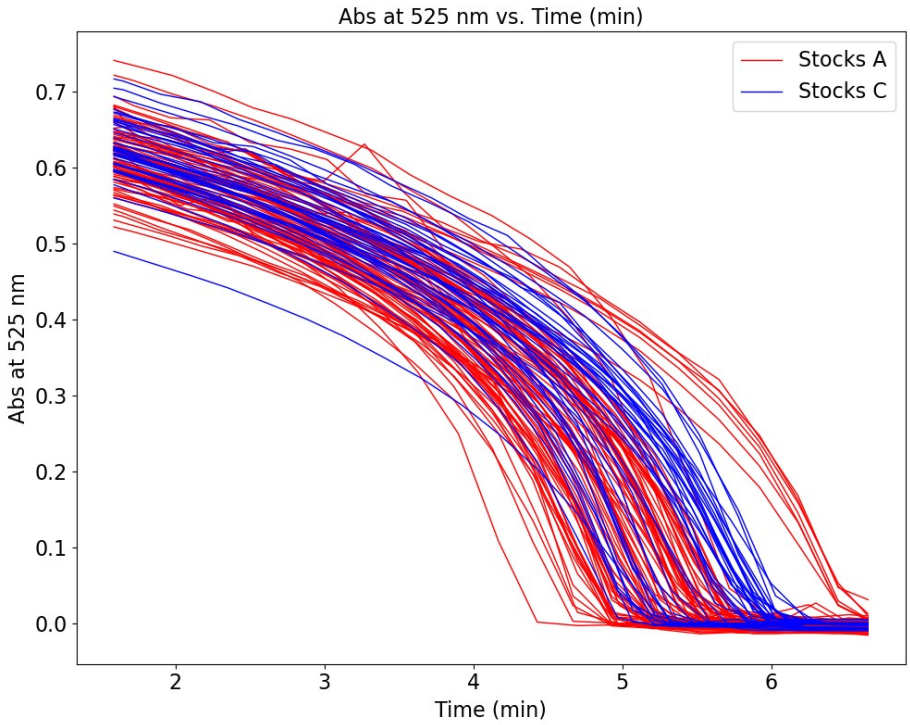
Run Times

ROPE as % of Combined Mean: 7%



Trajectory Consistency

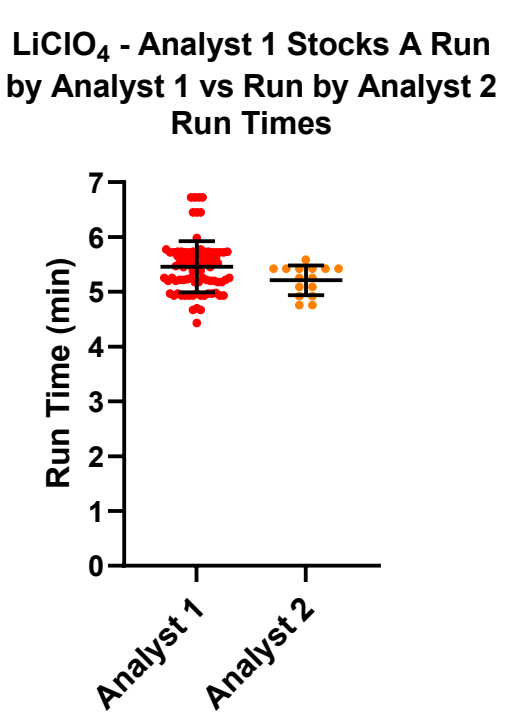
Analyst 1 Stocks A vs Stocks C: 5.4% RMSD



$LiClO_4$ - Analyst 1 Stocks A Run by Analyst 1 vs. Run by Analyst 2

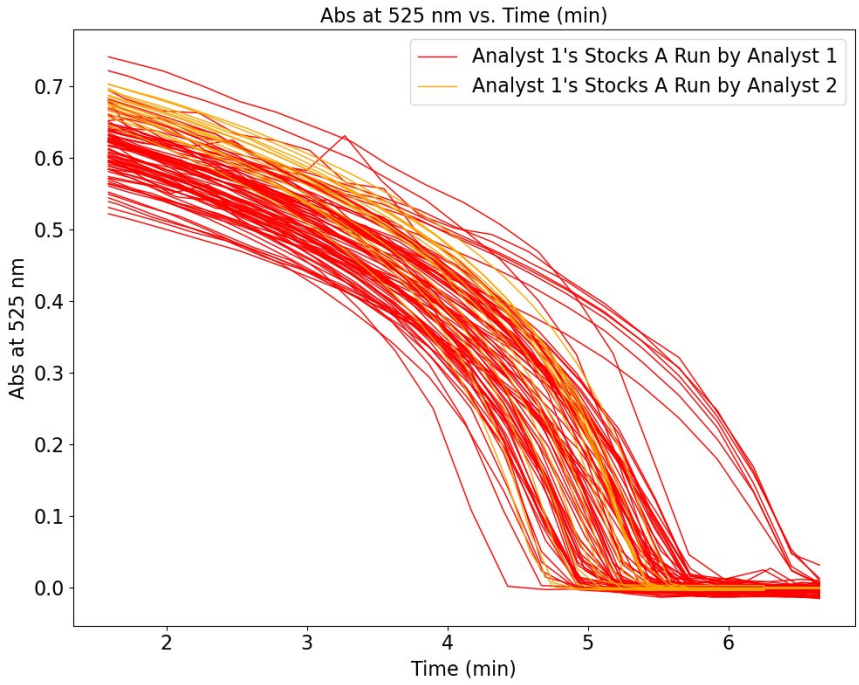
Run Times

ROPE as % of Combined Mean: 8%



Trajectory Consistency

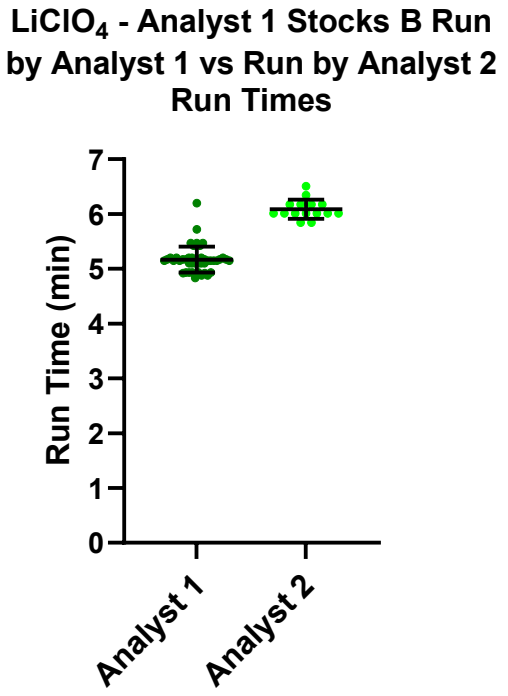
Analyst 1 Stocks A Run by: Analyst 1 vs Analyst 2: 6.5% RMSD



$LiClO_4$ - Analyst 1 Stocks B Run by Analyst 1 vs. Run by Analyst 2

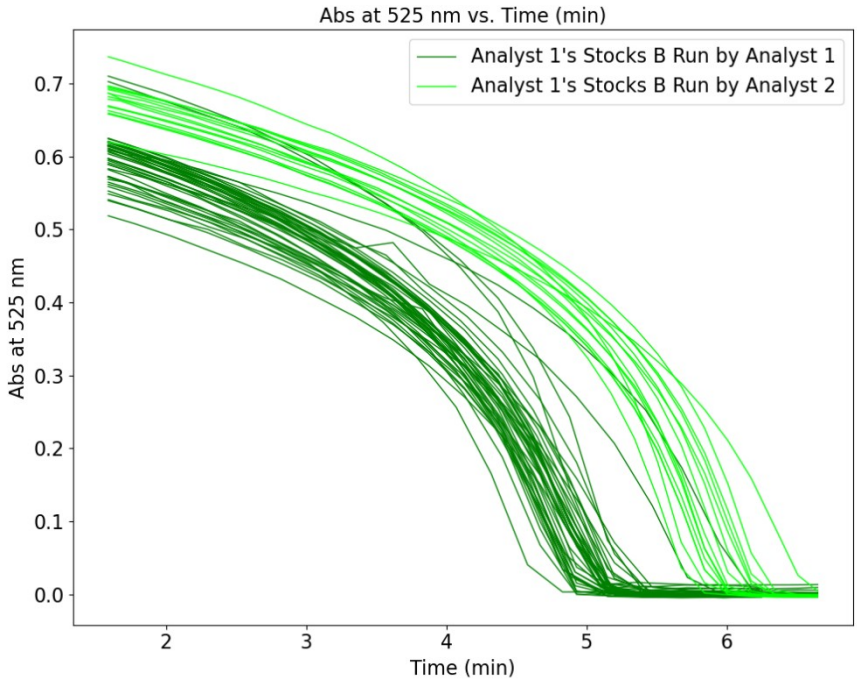
Run Times

ROPE as % of Combined Mean: 19%



Trajectory Consistency

Analyst 1 Stocks B Run by: Analyst 1 vs Analyst 2: 25% RMSD

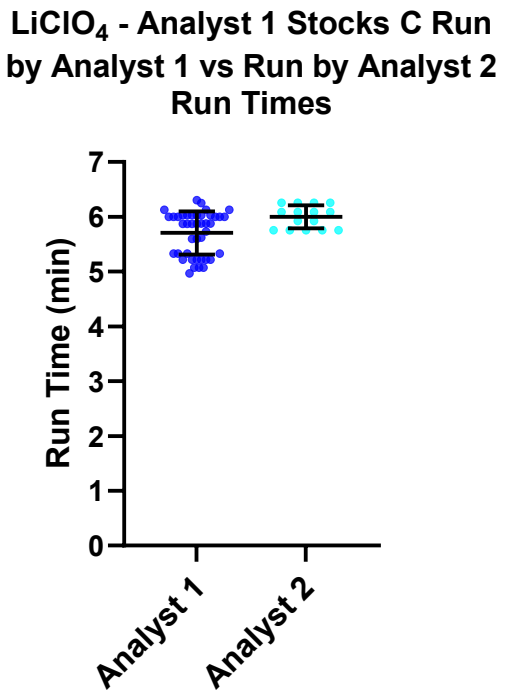


$LiClO_4$ - Analyst 1 Stocks C Run by Analyst 1 vs. Run by Analyst 2

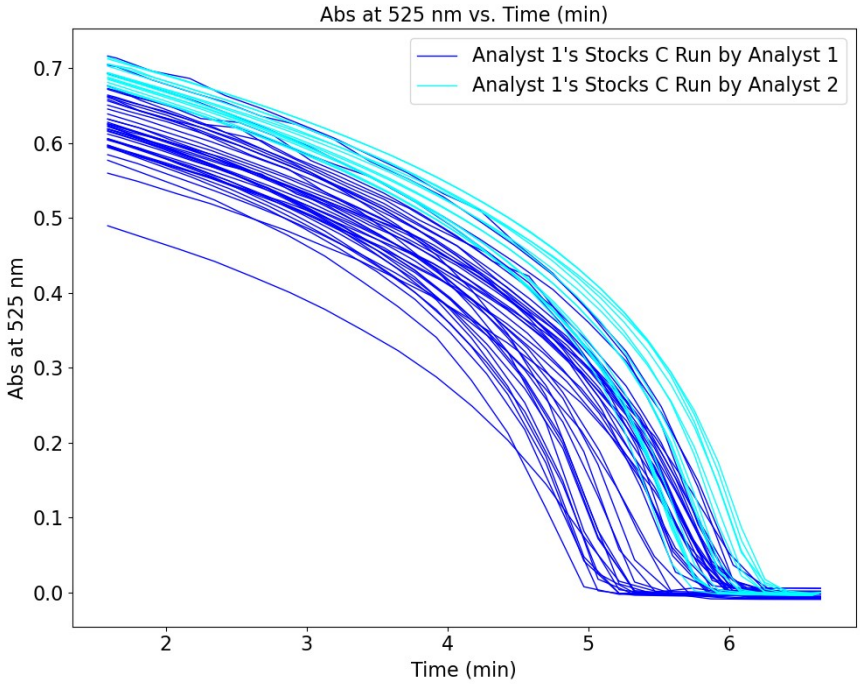
Run Times

ROPE as % of Combined Mean: 8%

Analyst 1 Stocks C Run by: Analyst 1 vs Analyst 2: 12% RMSD



Trajectory Consistency

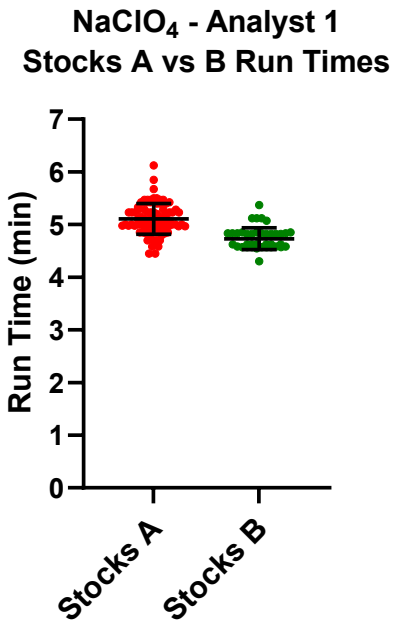


Analyst 1's NaClO4 Stocks

$NaClO_4$ - Analyst 1 Stocks A vs Stocks B

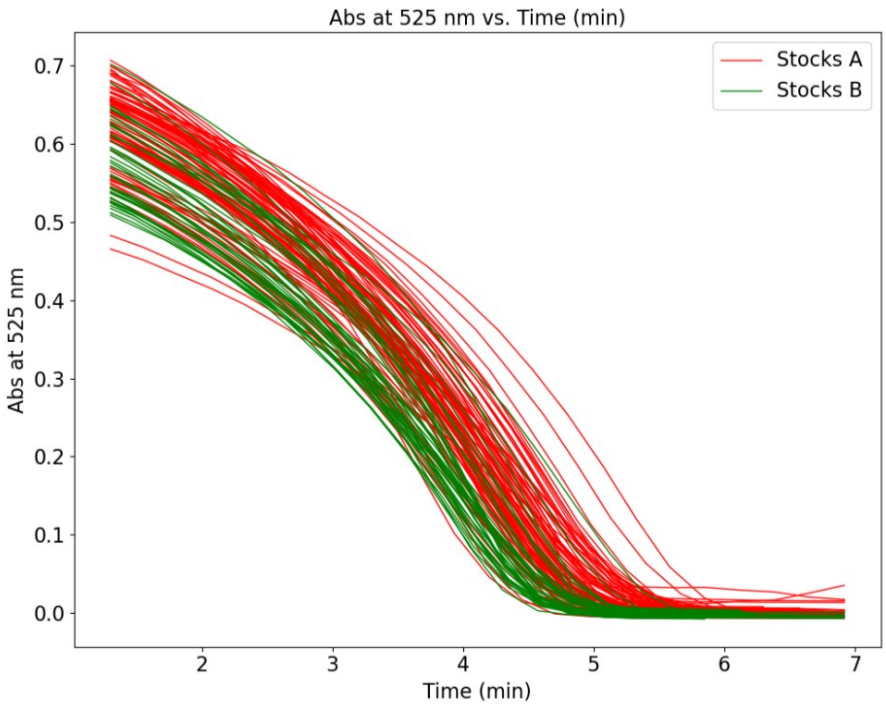
Run Times

ROPE as % of Combined Mean: 9%



Trajectory Consistency

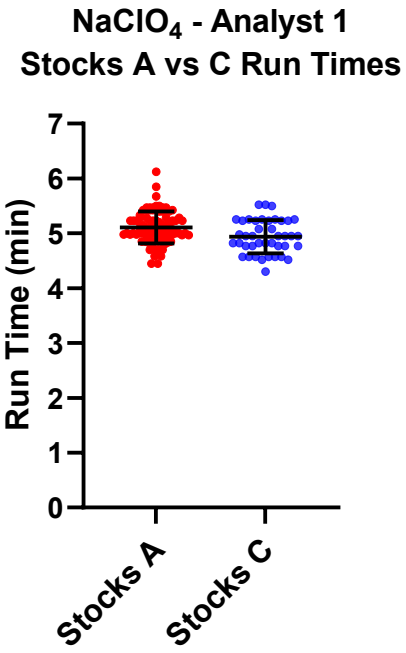
Analyst 1 Stocks A vs Stocks B: 9.2% RMSD



$NaClO_4$ - Analyst 1 Stocks A vs Stocks C

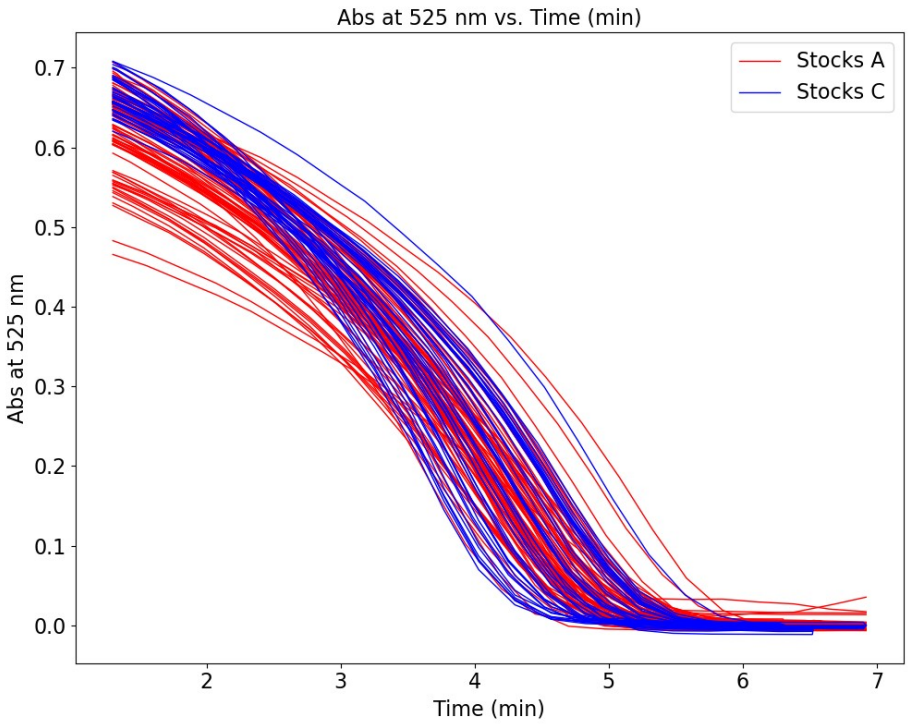
Run Times

ROPE as % of Combined Mean: 6%



Trajectory Consistency

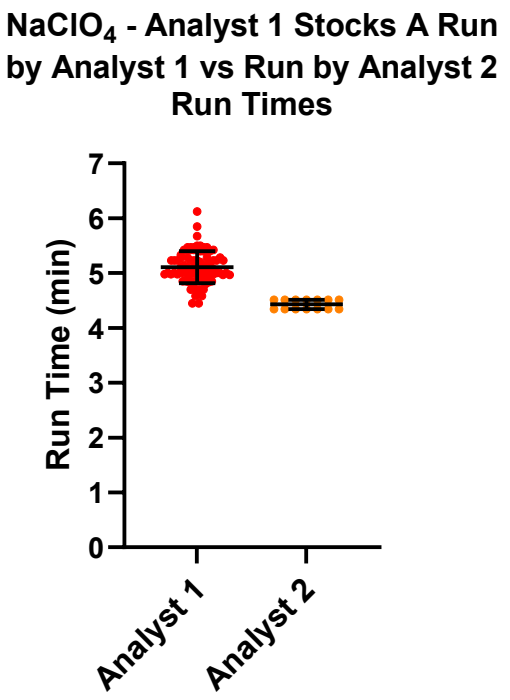
Analyst 1 Stocks A vs Stocks C: 4.1% RMSD



$NaClO_4$ - Analyst 1 Stocks A Run by Analyst 1 vs. Run by Analyst 2

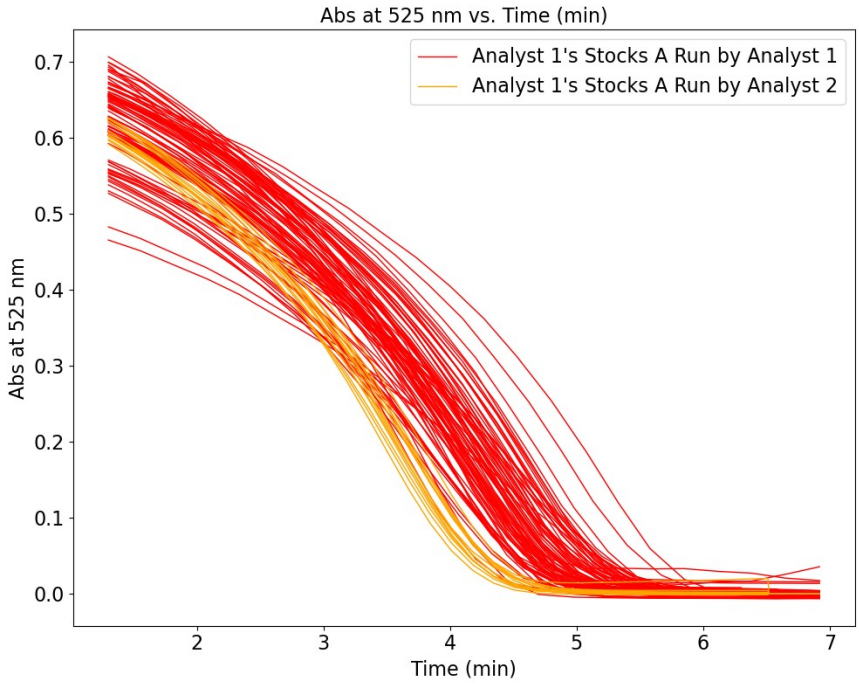
Run Times

ROPE as % of Combined Mean: 15%



Trajectory Consistency

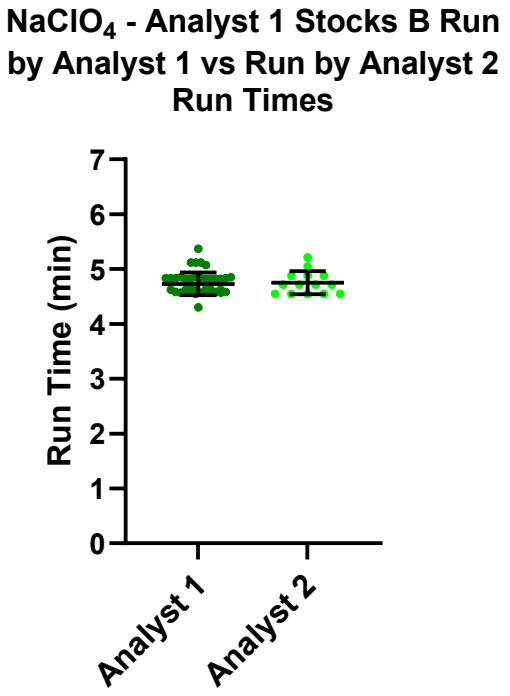
Analyst 1 Stocks A Run by: Analyst 1 vs Analyst 2: 14% RMSD



$NaClO_4$ - Analyst 1 Stocks B Run by Analyst 1 vs. Run by Analyst 2

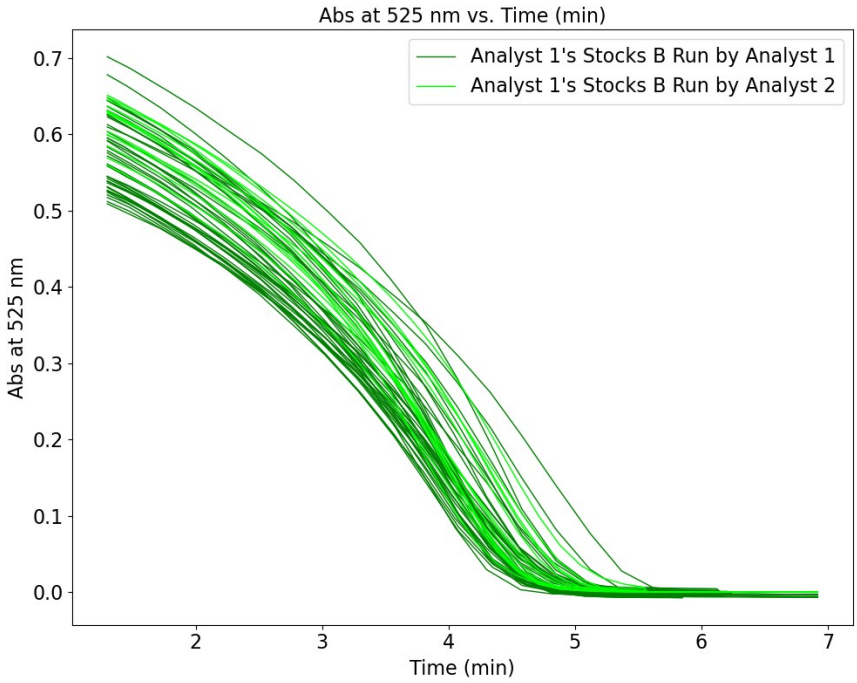
Run Times

ROPE as % of Combined Mean: 3%



Trajectory Consistency

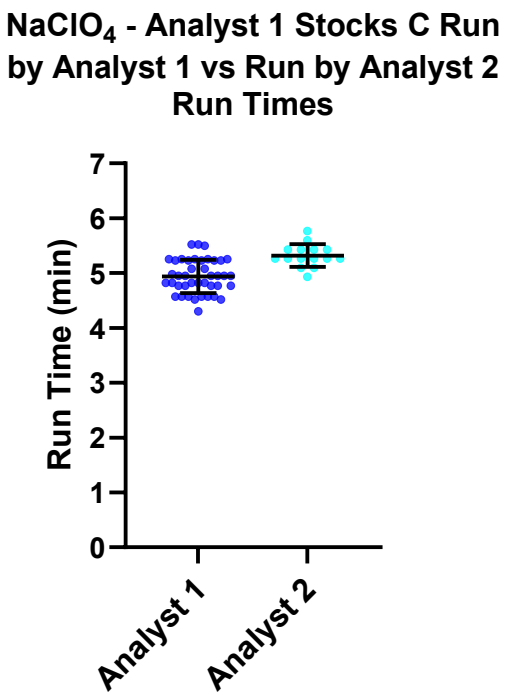
Analyst 1 Stocks B Run by: Analyst 1 vs Analyst 2: 6.2% RMSD



$NaClO_4$ - Analyst 1 Stocks C Run by Analyst 1 vs. Run by Analyst 2

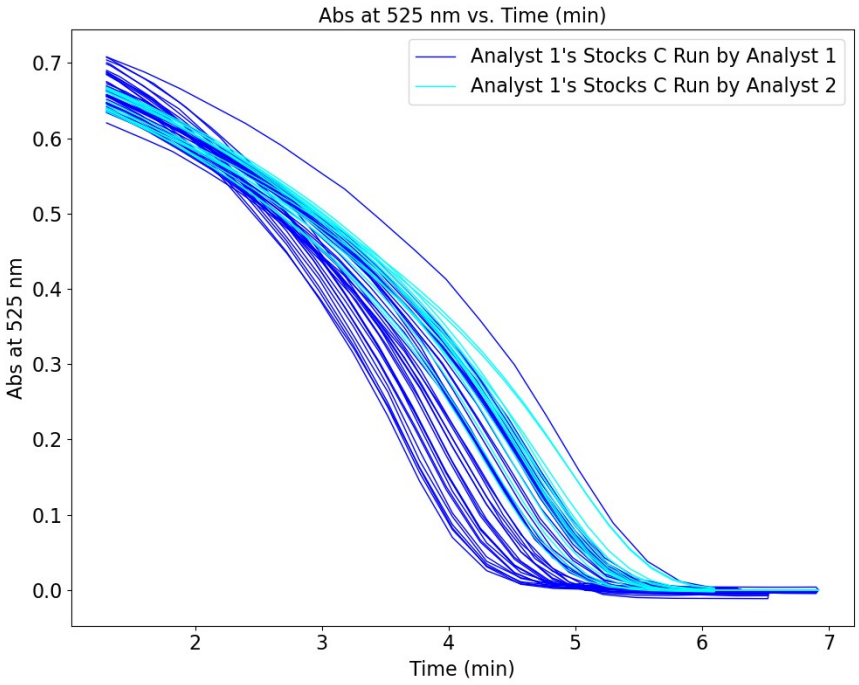
Run Times

ROPE as % of Combined Mean: 10%



Trajectory Consistency

Analyst 1 Stocks C Run by: Analyst 1 vs Analyst 2: 8.3% RMSD

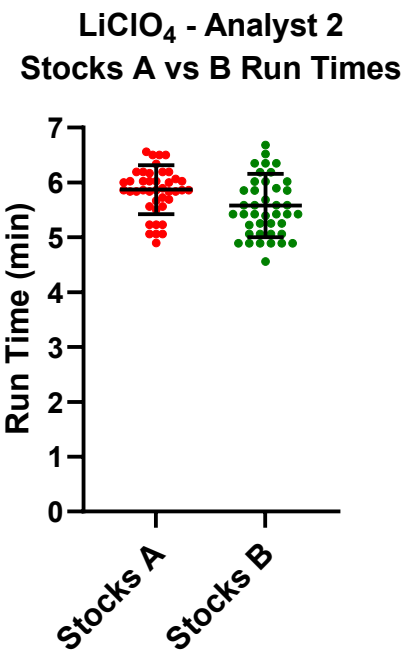


Analyst 2's $LiClO_4$ Stocks

$LiClO_4$ - Analyst 2 Stocks A vs Stocks B

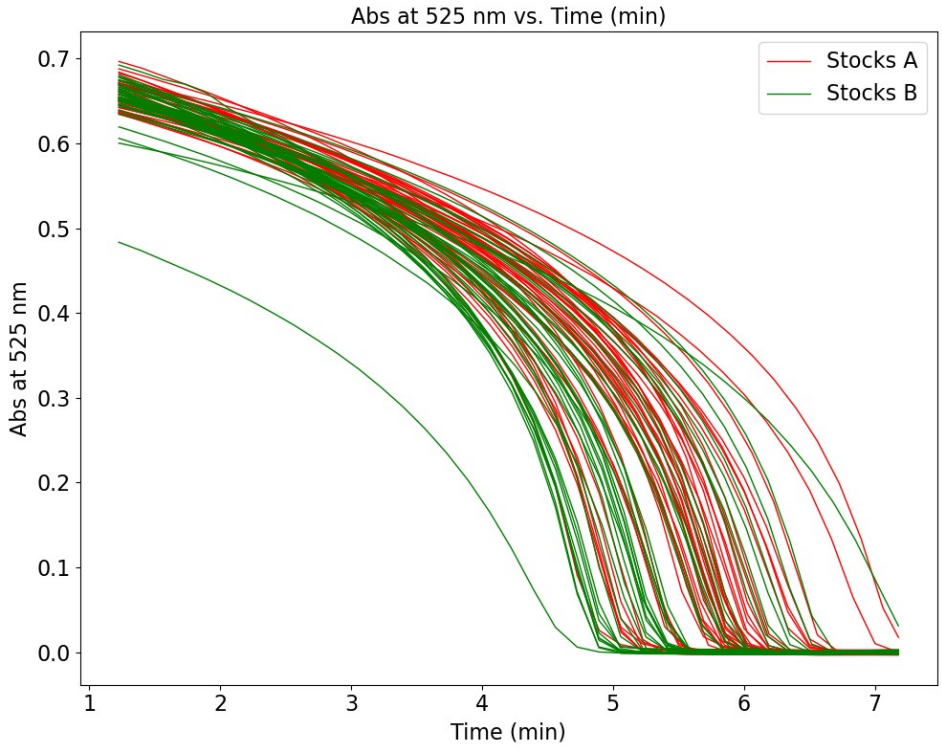
Run Times

ROPE as % of Combined Mean: 9%



Trajectory Consistency

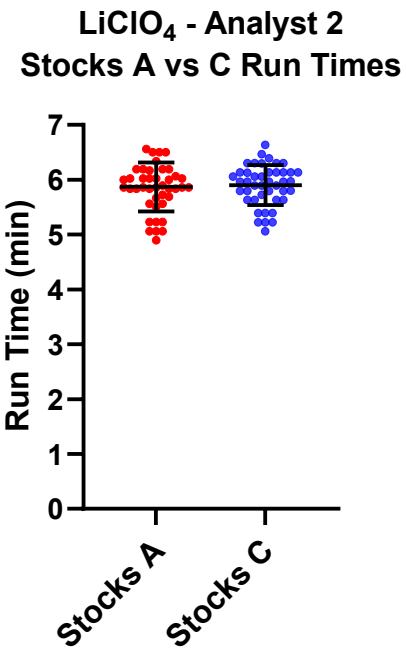
Analyst 1 Stocks A vs Stocks B: 6.1% RMSD



$LiClO_4$ - Analyst 2 Stocks A vs Stocks C

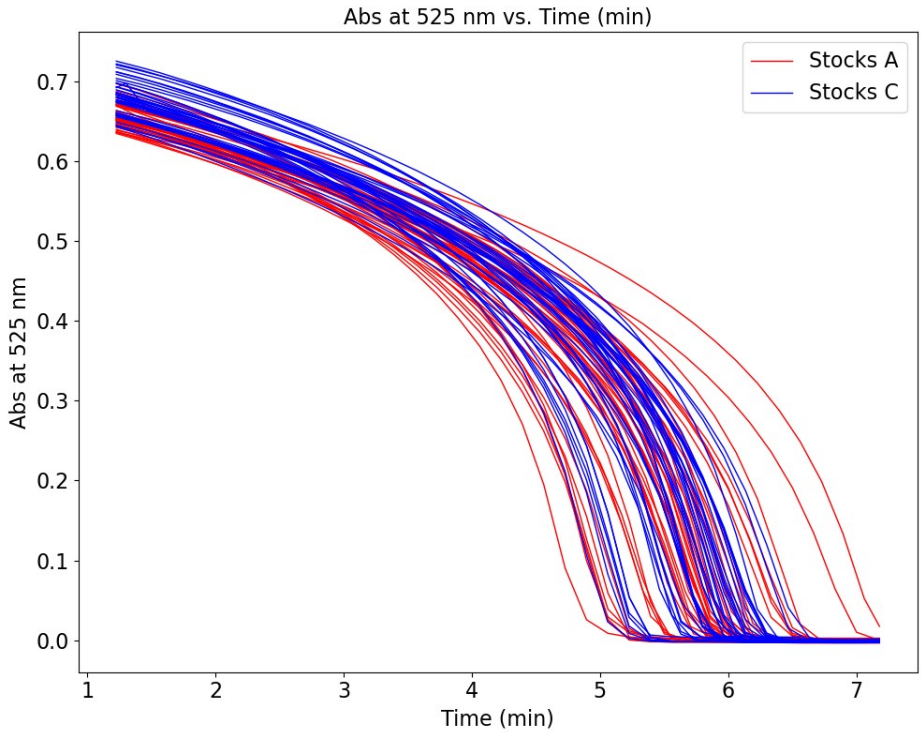
Run Times

ROPE as % of Combined Mean: 4%



Trajectory Consistency

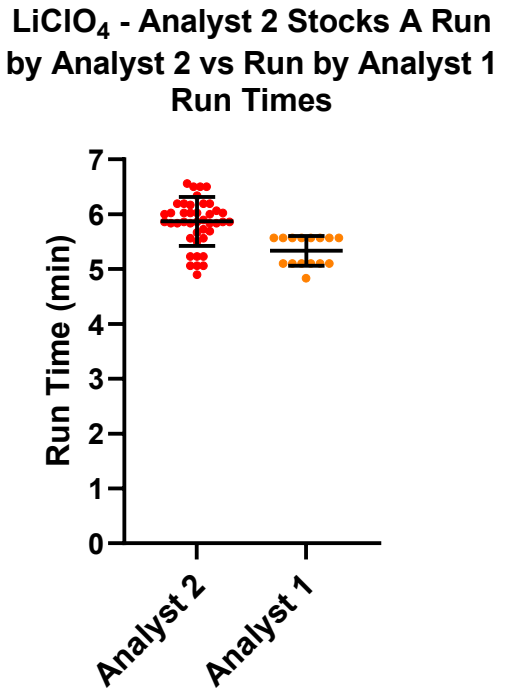
Analyst 1 Stocks A vs Stocks B: 2.5% RMSD



$LiClO_4$ - Analyst 2 Stocks A Run by Analyst 2 vs. Run by Analyst 1

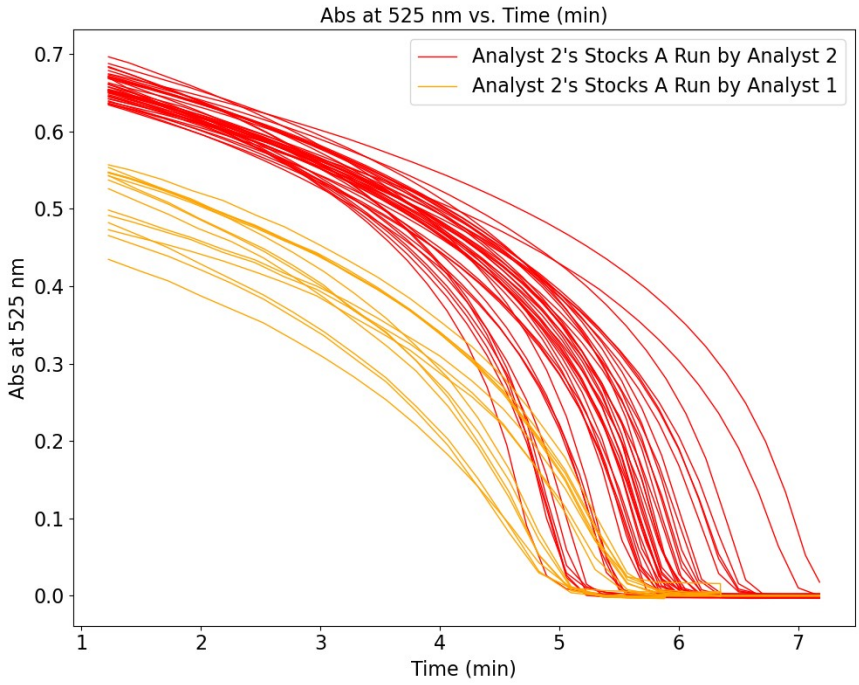
Run Times

ROPE as % of Combined Mean: 13%



Trajectory Consistency

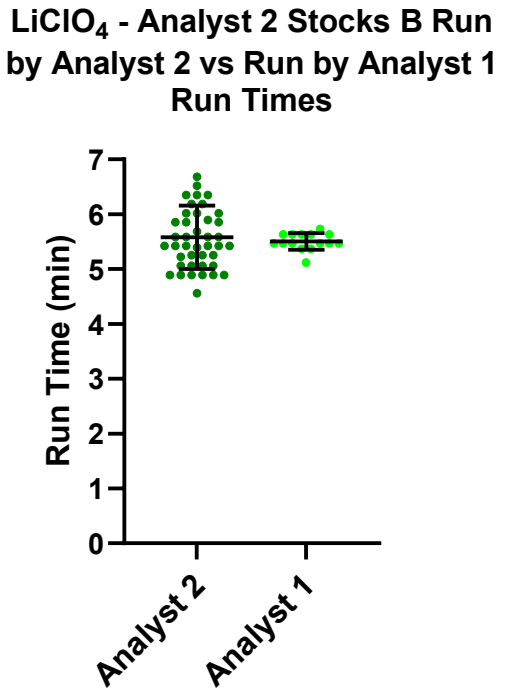
Analyst 2 Stocks A Run by: Analyst 2 vs Analyst 1: 22% RMSD



$LiClO_4$ - Analyst 2 Stocks B Run by Analyst 2 vs. Run by Analyst 1

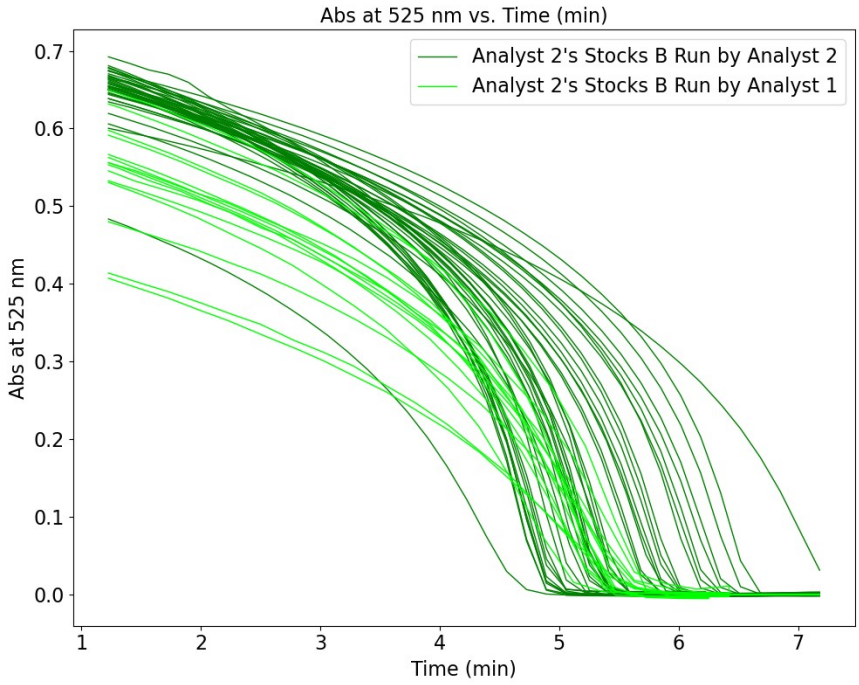
Run Times

ROPE as % of Combined Mean: 5%



Trajectory Consistency

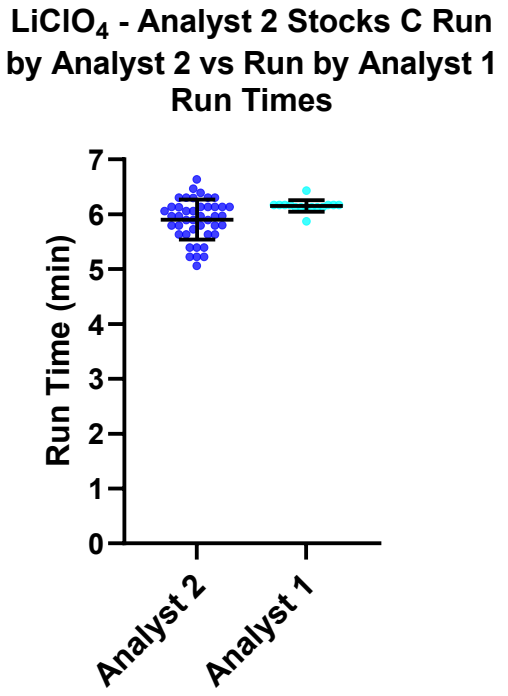
Analyst 2 Stocks B Run by: Analyst 2 vs Analyst 1: 13% RMSD



$LiClO_4$ - Analyst 2 Stocks C Run by Analyst 2 vs. Run by Analyst 1

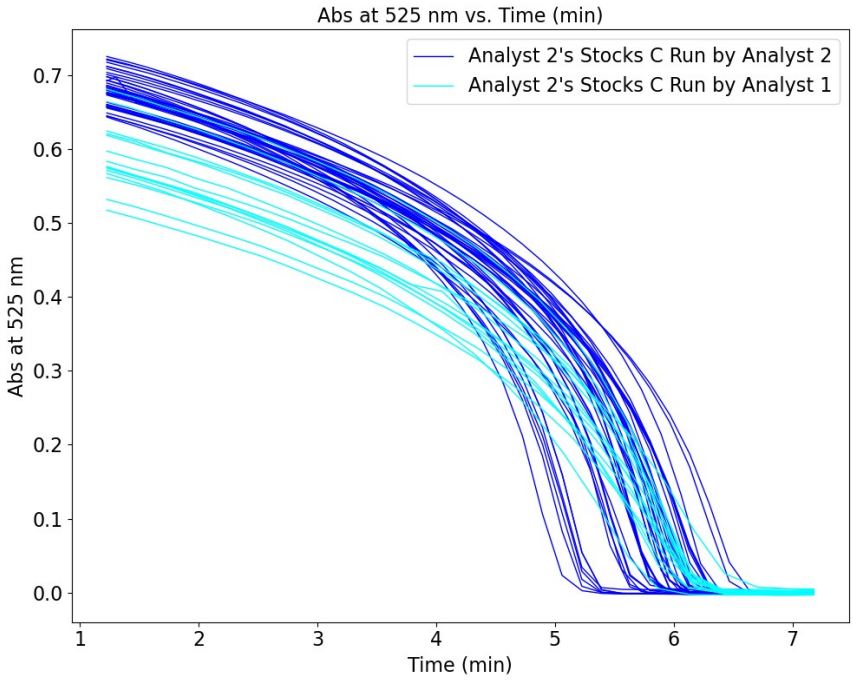
Run Times

ROPE as % of Combined Mean: 6%



Trajectory Consistency

Analyst 2 Stocks C Run by: Analyst 2 vs Analyst 1: 8.7% RMSD

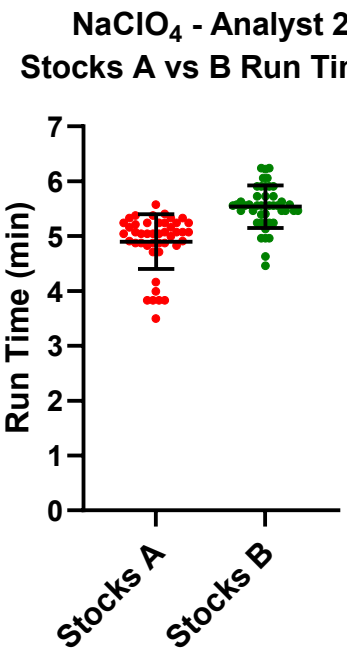


Analyst 2's $NaClO_4$ Stocks

$NaClO_4$ - Analyst 2 Stocks A vs Stocks B

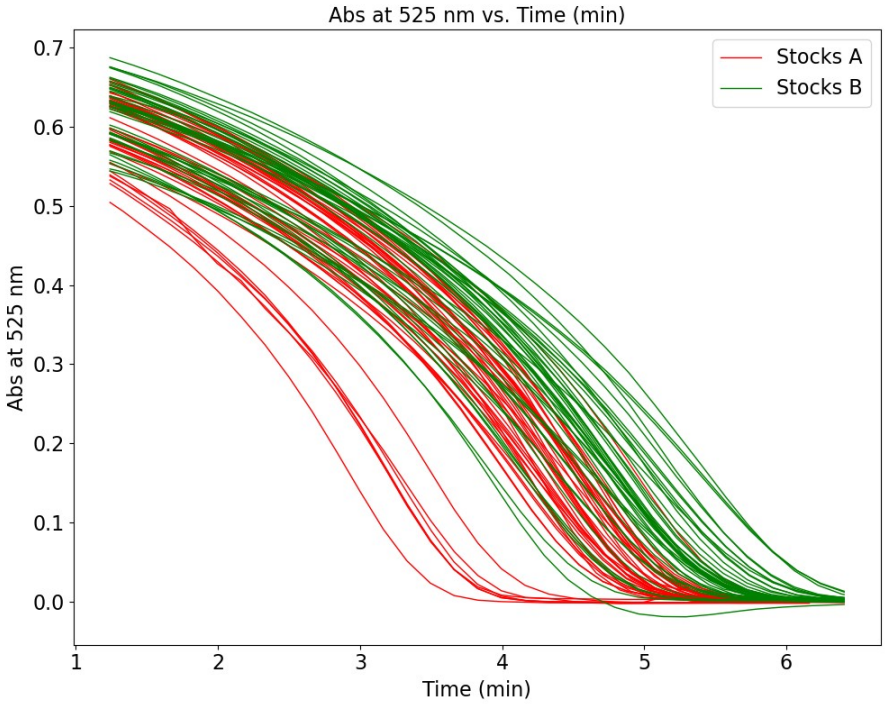
Run Times

ROPE as % of Combined Mean: 16%



Trajectory Consistency

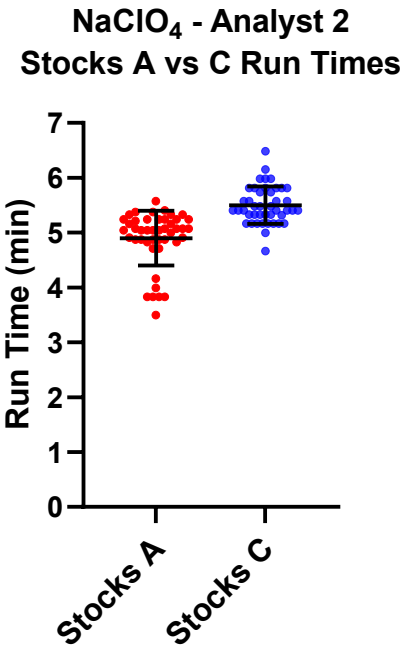
Analyst 2 Stocks A vs Stocks B: 11% RMSD



$NaClO_4$ - Analyst 2 Stocks A vs Stocks C

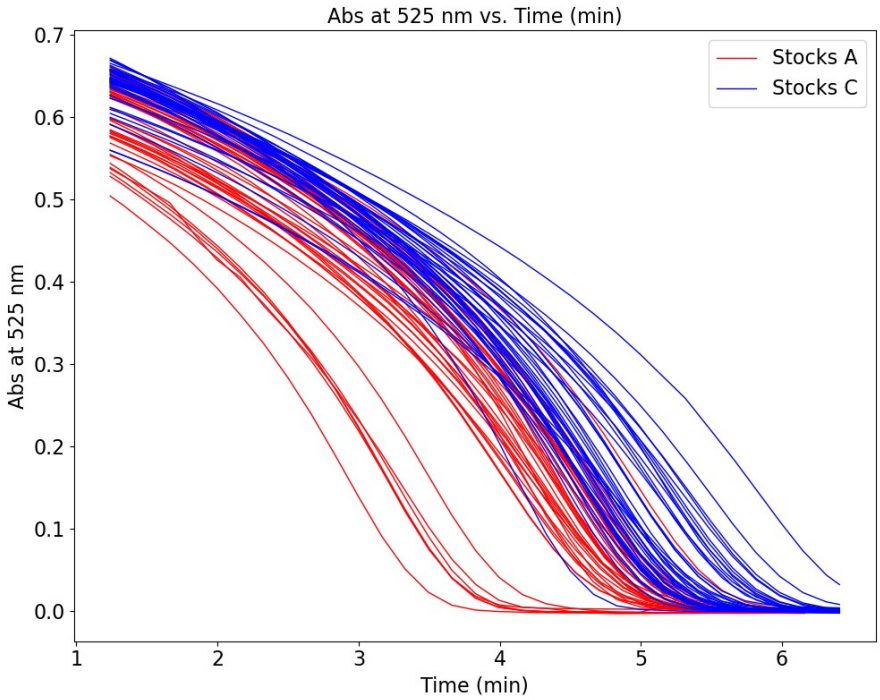
Run Times

ROPE as % of Combined Mean: 15%



Trajectory Consistency

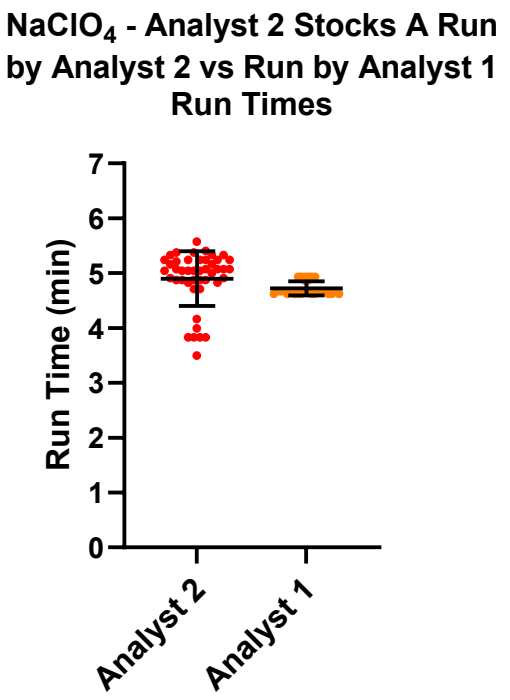
Analyst 2 Stocks A vs Stocks B: 13% RMSD



$NaClO_4$ - Analyst 2 Stocks A Run by Analyst 2 vs. Run by Analyst 1

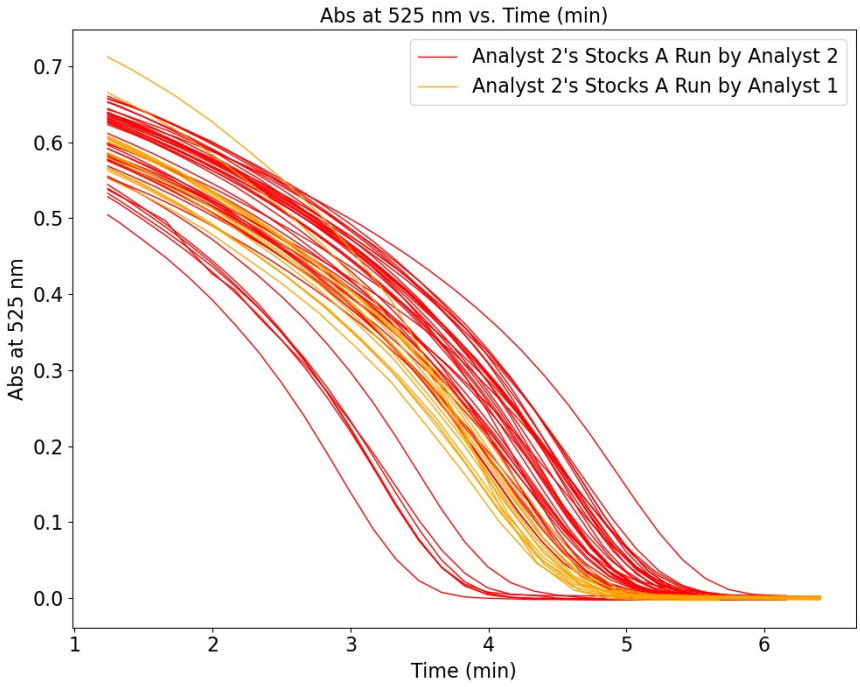
Run Times

ROPE as % of Combined Mean: 7%



Trajectory Consistency

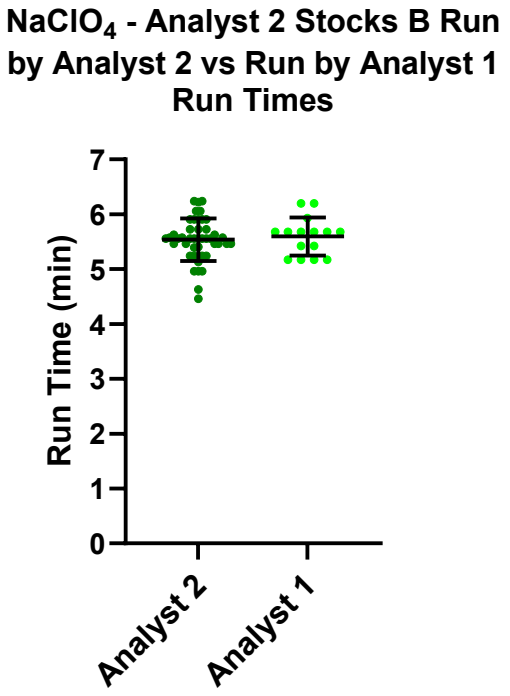
Analyst 2 Stocks A Run by: Analyst 2 vs Analyst 1: 6.3% RMSD



$NaClO_4$ - Analyst 2 Stocks B Run by Analyst 2 vs. Run by Analyst 1

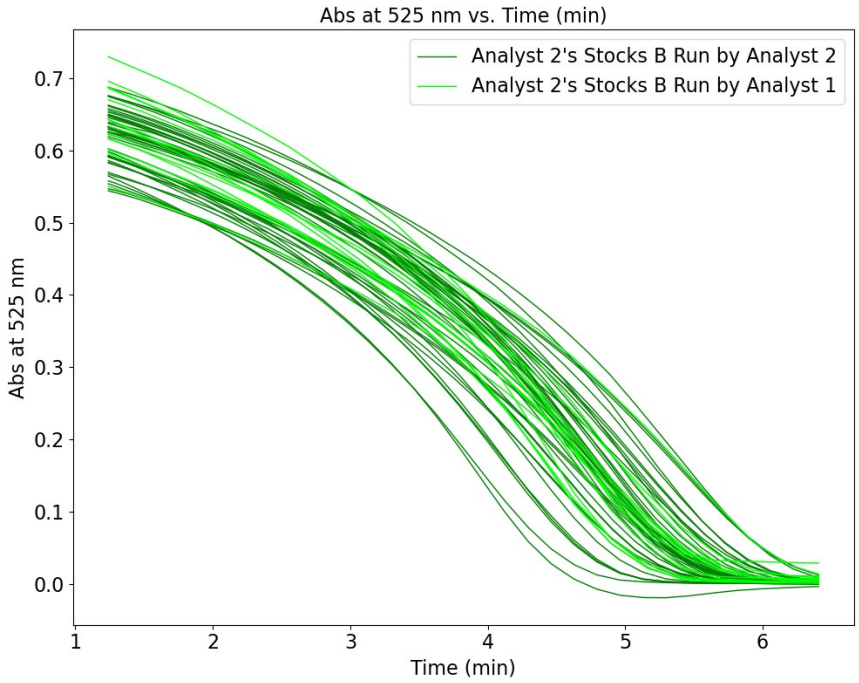
Run Times

ROPE as % of Combined Mean: 5%



Trajectory Consistency

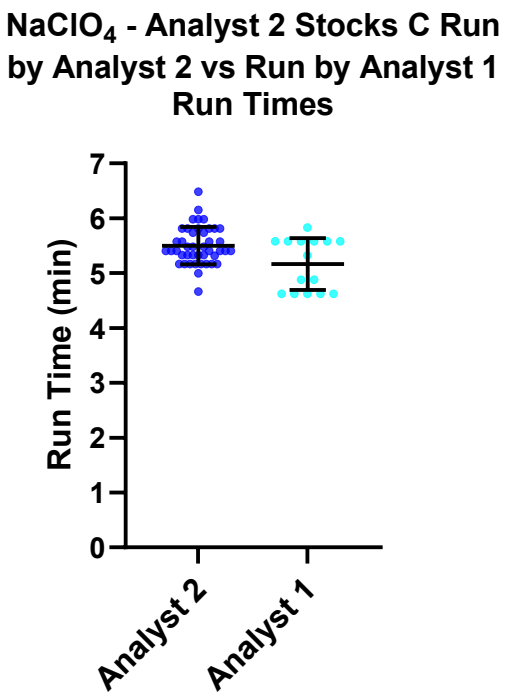
Analyst 2 Stocks B Run by: Analyst 2 vs Analyst 1: 1.5% RMSD



$NaClO_4$ - Analyst 2 Stocks C Run by Analyst 2 vs. Run by Analyst 1

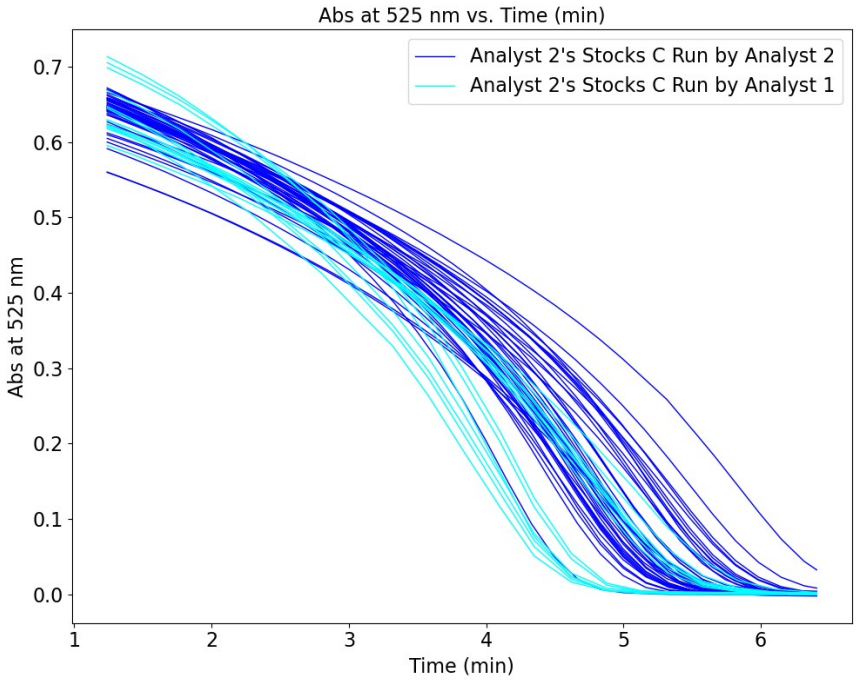
Run Times

ROPE as % of Combined Mean: 10%



Trajectory Consistency

Analyst 2 Stocks C Run by: Analyst 2 vs Analyst 1: 7.4% RMSD



Robustness of run time and reaction trajectory consistency under conditions of variable stock solutions (Stocks A vs. Stocks B), reagent lots (Stocks A vs. Stocks C), or Analysts.