Application Note

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Email Notifications from Your Online LC

Messages about instrumental and experimental status sent by Agilent Online LC Monitoring Software for time-saving decisions and unattended operation

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Abstract

This application note demonstrates the functionality provided by the Agilent Online LC Monitoring Software to send email notifications about different instrument and experimental states. The online LC experiment can be tracked remotely, saving time through unattended experiment monitoring.

Introduction

In modern pharmaceutical and chemical production environments, it is of crucial importance that processes can be conducted in an unattended and remotely controlled manner. Critical instrument issues such as errors and interruptions, or important events such as finished reactions or changed reaction behavior, must be reported immediately and automatically. This reporting is possible through emails sent from the monitoring instrumentation, such as reaction-controlling online LC instruments.

This application note shows how the Online LC Monitoring Software can be set up and used to generate email notifications about critical instrument states and reaction events in near real time. These alerts allow a remote user to react faster to instrument and reaction issues for more safety, and to save money and time. In addition, in the case of repeated experiments, an experiment comparison will be shown within the Online LC Monitoring Software by a comparative visualization. This enables fast decisions towards the experimental success of identical experiments.

Experimental

The instrumentation used in this study is detailed in Table 1, and the method parameters are outlined in Table 2.

Table 1. Instrumentation.

Product Type	Agilent Product Description
Instrument	– Agilent 1290 Infinity II High Speed Pump (G7120A) – Agilent 1260 Infinity II Online Sample Manager Set (G3167AA): – Agilent 1260 Infinity II Online Sample Manager (G3167A) clustered with external valve (p/n 5067-6680) located at the Agilent 1290 Infinity Valve Drive (G1170A), and Agilent Online LC Monitoring Software – Agilent 1290 Infinity II Multicolumn Thermostat (G7116B) – Agilent 1290 Infinity II Diode Array Detector (G7117B) with Agilent InfinityLab Max-Light Cartridge Cell (10 mm, G4212-60008)
Additional Pump	Agilent 1260 Infinity II Isocratic Pump (G7110B)
Column	Agilent InfinityLab Poroshell 120 EC-C18, 2.1 × 30 mm, 1.9 µm (p/n 695775-302)
Software	– Agilent OpenLab CDS, version 2.6 – Agilent Online LC Monitoring Software, version 1.2

Table 2. Method parameters.

Parameter	Value
Analytical Method Conditions	
Solvents	A) Water + 0.1% formic acid (FA) B) Acetonitrile (ACN) + 0.1% FA
Analytical Flow Rate	1.3 mL/min
Gradient	40% B to 90% B in 0.85 min, stop time: 1.0 min
Column Temperature	45 °C
Feed Speed	80% of analytical flow rate
Flush-Out Solvent	Water:ACN 9:1 + 0.1% FA (S2)
Flush-Out Volume	Automatic
Injection Volume	1 µL
Needle Wash	3 s, water:ACN 1:1 + 0.1% FA (S1)
Sampling	See sampling methods for sampling to vial
Diode Array Detector	290 ±4 nm, reference: off, 40 Hz data rate
Sampling (with Dilution)	
Sampling	Sampling from reactor into deep-well plate sealed with silicon mats
Target Volume	600 µL
Dilution Factor	100
Sample Volume	6 µL
Draw Speed	Setting 2 – Draw speed: 100 µL/min – Wait time: 3.6 s – Dispense speed: 130 µL/min (Ejection of sample into well before dilution)
Dilution Solvent	S2
Dilution Eject Speed	10,000 µL/min (after sample ejection for mixing)
Schedule	Interval: 4 min Experiment run time: 120 min

Table 3. Reaction conditions.

Reaction Conditions	
Reactor	Mettler-Toledo EasyMax 102 equipped with 50 mL reaction vessel; temperature controlled by a connected JULABO thermostat
Additional Pump	Continuously running at 5 mL/min; connecting the reactor with the Online Sample Manager interface and back to reactor
Solvent	50 mL acetone:water 2:1 (v/v)
Educt	*p*-Anisaldehyde, 0.5 mL
Stirring	At 25 °C
Reaction Start	Add 100 µL NaOH 50% in water (w/w)

Setup of the Online LC Monitoring Software for email notifications

In the configuration settings of the Online LC Monitoring Software, a user or group of users can be defined by their email address to receive general notifications about the instrument or sample status (Figure 1). The generation of more reaction-specific emails will be discussed in the "Results and discussion" section. In addition, an electrical output signal via ERI can be defined. This can be used to generate, for example, optical or acoustical signals at an external device.

Chemicals

- *p*-Anisaldehyde
- Acetone
- NaOH
- Formic acid

Additional materials

- Agilent 96-deep-well plates, 1 mL, polypropylene (part number 5043-9305)
- Agilent sealing mat, 96 wells, round, preslitted, silicone (part number 5043-9317)
- Agilent 0.8 mm id PTFE tubing (part number 5041-2191), ferrule (part number 5022-2154), PTFE nuts (part number 50222158), fittings, and ferrule (part number 5065-4454)

Solvents and chemicals

- All solvents were purchased from Merck, Germany.
- Chemicals were purchased from VWR, Germany.
- Fresh, ultrapure water was obtained from a Milli-Q integral system equipped with LC-Pak polisher and a 0.22 µm membrane point-of-use cartridge (Millipak).

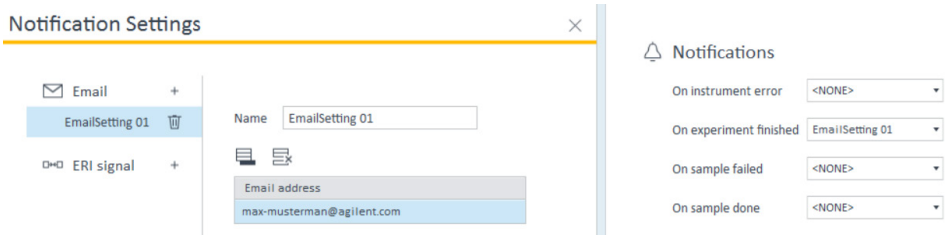


Figure 1. Settings to generate email notifications about general instrument and sample events for a user or group of users defined by their email address.

Results and discussion

For the transition of a small-molecule chemical reaction from the development phase to a scale-up plant, or later to a production plant, all reaction parameters must be well understood. This understanding enables the user to conduct the reaction in a controlled and safe manner. Reaction control is possible with the Compound Limit control and notification function provided in the Online LC Monitoring Software (Figure 2).

This function allows the construction of a trending band for all involved compounds (i.e., educts, products, and intermediates). In the simplest case, this band is simply a limit line for a minimum and maximum for, for example, area%, area, or concentration. For a well-known reaction, a more complex trending band can be constructed with a deviation for each sampling point, as outlined in Figure 2 for the educt of the reaction.[1] If the measured value is above or below these limits, an email notification can be requested.

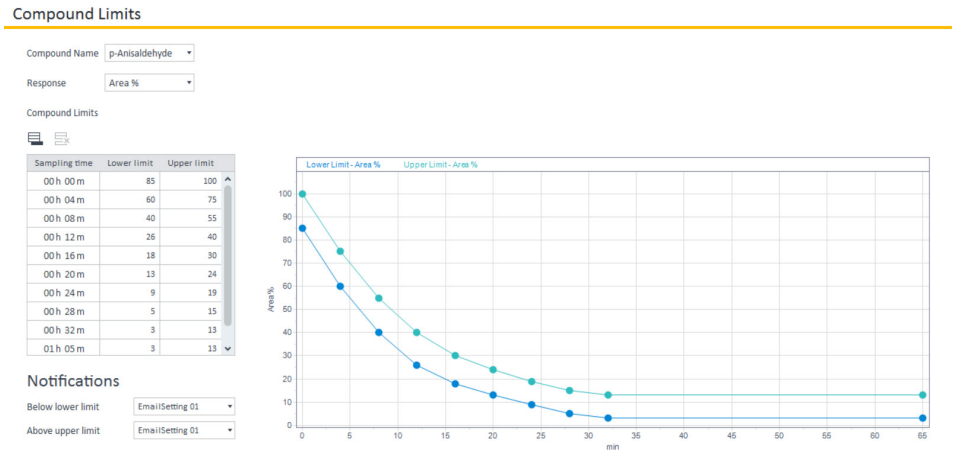
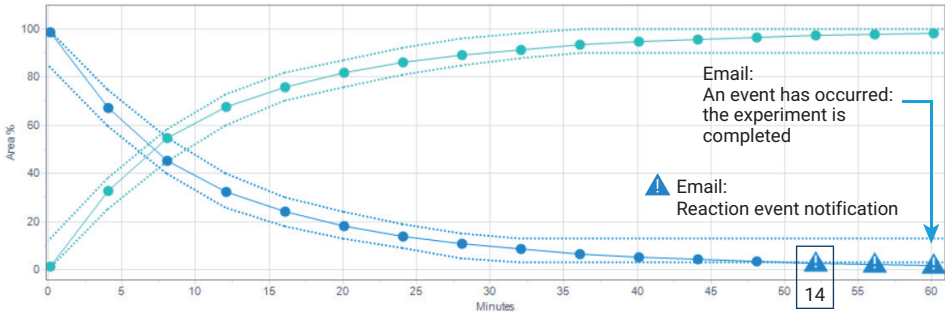


Figure 2. Compound Limits and email notifications with the capability to construct a trending band to guide a reaction (blue: lower limit, green: upper limit).

For a real, well-known, small-molecule chemical reaction, the trending plot was created as the data from the individual sampling points were made available. The predefined trending bands were already displayed, and the appearing data were automatically compared with the individual lower and upper limits for the educt and the product. Figure 3 displays the sampling data points and limit bands as available for the finished experiment. If the data resulting from a sampling point were out of the predefined limit bands, they were marked with a triangle in the trending plot and a notification was sent automatically. The text message generated for the email notification for sampling point 14 is displayed in the text box in Figure 3. Another notification was sent when the experiment was finished at 60 minutes. The results of initial and final sampling points are outlined in Table 4.



Online LC Monitoring Experiment: An event has occurred
For the experiment, an event has occurred: a measured sample is outside the limits defined in the experiment setup
Details:
Sample Name: Sample-14
Error text: The compound p-Anisaldehyde with 'Area %'-Value '2.745' at sampling time 00:52:06 is below of the defined limit value '3.000'

Figure 3. Trending plot of a finished experiment with sampling data points and notifications for sample results outside the limit band.

Table 4. Result data of initial and final data points where notifications were sent out. A lower limit of 3 area% educt was chosen as the reporting level.

Sample	Compound	RT (min)	Area%	Area	Height
	<ALL>				
Sample-1	p-Anisaldehyde	0.241	98.512	355.424	831.715
	Anisylidene acetone	0.296	1.488	5.368	10.151
Sample-3	p-Anisaldehyde	0.241	45.400	116.972	274.891
	Anisylidene acetone	0.296	54.600	140.675	303.738
Sample-14	p-Anisaldehyde	0.241	2.745	10.646	24.881
	Anisylidene acetone	0.296	97.255	377.234	807.673
Sample-15	p-Anisaldehyde	0.241	2.229	8.604	20.114
	Anisylidene acetone	0.296	97.771	377.415	810.987
Sample-16	p-Anisaldehyde	0.241	1.802	7.033	16.514
	Anisylidene acetone	0.296	98.198	383.206	827.731

The trending plot shown in Figure 3 displays the expected behavior of the monitored reaction with desired notifications towards the end of the reaction. However, if an undesired event occurs, the notification function can also be very helpful. For instance, this event could be a malfunction in heating, cooling, or reagent dosing. In these cases, the sampling points will occur outside the limits defined by the trending band and trigger a notification event. Figure 4 displays an example of a simulated malfunction in the heating unit with an uncontrolled up-heating after the third sampling point. Due to increasing reaction speed, notifications for the educt, which exceeded its trending band, and for the product, which fell below its band, were sent. Each sampling point outside its band will be marked with a triangle. The text of the respective messages is shown in Figure 3. Table 5 summarizes the data occurring after the undesired up-heating.

Online LC Monitoring Experiment: An event has occurred
For the experiment, an event has occurred: a measured sample is outside the limits defined in the experiment setup.
Details:
Sample Name: Sample-4
Error text: The compound p-Anisaldehyde with 'Area %'-Value '12.725' at sampling time 00:11:59 is below of the defined limit value '26.039'.

Online LC Monitoring Experiment: An event has occurred
For the experiment, an event has occurred: a measured sample is outside the limits defined in the experiment setup.
Details:
Sample Name: Sample-4
Error text: The compound Anisylidene acetone with 'Area %'-Value '87.275' at sampling time 00:11:59 exceeds the defined limit value '72.958'.

Figure 4. Example for an undesired up-heating event and corresponding notifications.

Table 5. Result data of trending plot points where the educt and product left their trending bands and respective notifications were sent out.

Sample	Compound	RT (min)	Area%	Area	Height
	<ALL>				
Sample-4	p-Anisaldehyde	0.241	12.725	52.745	124.314
	Anisylidene acetone	0.296	87.275	361.740	785.489
Sample-5	p-Anisaldehyde	0.241	2.556	11.009	25.448
	Anisylidene acetone	0.296	97.444	419.631	912.098
Sample-6	p-Anisaldehyde	0.241	0.804	3.477	7.727
	Anisylidene acetone	0.296	99.196	428.789	928.553

If a reaction is conducted multiple times in a production process, it is important to ensure that the reaction always follows the same course. This can be confirmed by the comparison functionality of the Online LC Monitoring Software, as already shown for the comparison of different reaction parameters.[2] For this comparison, the trending plots of the peak area of educt and product from three reaction replicates were overlaid (Figure 5). The overlay shows that two of the reactions followed the same course, while in the third reaction, with the simulated temperature malfunction, the educt and product take a different course.

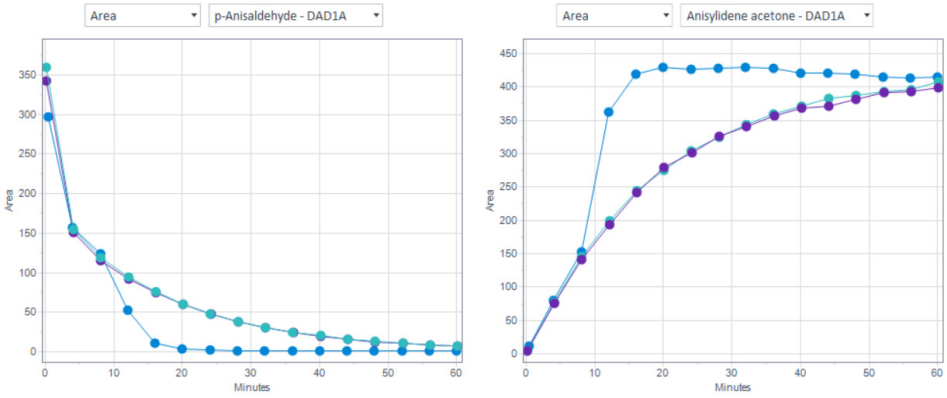


Figure 5. Comparison of peak area of educt and product from three replicate reactions. The purple and turquoise trending plots show the same reaction course. The blue trending plot displays the reaction course with the simulated temperature malfunction.

Conclusion

This application note demonstrates the use of the email notification feature built into the Agilent Online LC Monitoring Software. This function allows immediate notification of general events from the unattended working system. For the individually monitored reaction, specific details, such as sampling points occurring outside a predefined trending band, could be applied for notifications to the remotely located user. Finally, a given reaction performed multiple times can be compared to ensure the same performance. This functionality allows the user to run the system unattended to save time and costs.

References

1. Lautz, C. *et al.* Automated Reaction Monitoring by the Agilent 1260 Infinity II Prime Online LC. *Agilent Technologies application note*, publication number 5994-3980EN, **2021**.
2. Naegele, E. *et al.* Which Reaction Shows the Best Performance? *Agilent Technologies application note*, publication number 5994-5922EN, **2023**.

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