HLT07 MeDD

D 1.4.2
Characterisation of the flow generator NEXUS 3000 (syringe pump)

Author: Bissig Hugo, Federal Institute of Metrology METAS, Switzerland

Date 28.01.2014

The research leading to the results discussed in this report has received funding from the European Metrology Research Programme (EMRP). The EMRP is jointly funded by the EMRP participating countries within Euramet and the European Union.

Table of Contents

1 Introduction

1.1 Background

In task 1.4.2 the primary standards are subject to an inter comparison by means of a flow meter and a flow generator. This report covers the part of the characterization of the flow generator, in this case a syringe pump NEXUS 3000.

1.2 Motivation

The reason for the inter comparison using a flow generator as a transfer standard is the fact that in WP3 several drug delivery devices will be calibrated by different laboratories. In order to compare the influences of physical parameters on the accuracy, repeatability and reproducibility of these flow generators performed by different laboratories, the knowledge about the validated testing facilities is of crucial importance.

1.3 Goal

Characterization of the syringe pump in the flow range of 2 μl/min to 333 μl/min including repeatability and reproducibility.

2 Set up

The syringe pump is characterized by means of the METAS micro flow facility. The flow rate is determined dynamically.

The ambient temperature is around 22 °C and the temperature and the relative humidity are recorded for each measurement.

3 Device under Test

3.1 Syringe pump

NEXUS 3000
Manufacturer: Chemyx Inc.
Supplier: KR Analytical LTD, UK
Serial number: 2172176

Owner: Dutch Metrology Institute VSL, Thijsseweg 11, 2629 JA Delft, NL

3.2 Syringe used

Manufacturer

ILS Innovative Labor Systeme GmbH, 98714 Stützerbach, Germany

Type of syringes used

H-TLL, PTFE-Seal

volume syringe (μl)	flow rates (μl/min)	possible measurement time of 80 % of volume (min)	Inner Diameter (mm)	Article number	Internal number of set
2‘500	2	1000	7.28	2607031	1 (2 backup)
2'500	10	200	7.28	2607031	1 (2 backup)
5'000	33	121	10.30	2607051	1 (2 backup)
25'000	100	200	23.03	2607091	1 (2 backup)
25'000	333	60	23.03	2607091	1 (2 backup)

Table 1: Relevant information of the syringes

Note: Experiments using a 500 μl-syringe for the flow rate of 2 μl/min were carried out. However, only a bad repeatability was obtained and the reason might be due to the fact that the plunger has a thin rod. As the head of plunger can’t be fixed correctly to the syringe pump (perfectly in line with the body of the syringe), distortion of the plunger might occur and the flow rate would therefore not be reproducible enough. Mounting and dismounting the syringe would heavily influence the measurement results.

3.3 Characteristic spindle rotation time

The syringe pump NEXUS 3000 has a spindle increment per rotation of 1.5 mm.

This gives us the following characteristic spindle rotation time τ_{sp} depending on the syringe and the flow rate selected (see Table 2), which should cause visible oscillations in the flow.

volume syringe (μl)	flow rates (μl/min)	Inner Diameter (mm)	Spindle rotation time τ_{sp} (min)	Spindle rotation time τ_{sp} (sec)
2‘500	2	7.28	31.219	1'873.1
2'500	10	7.28	6.244	374.6
5'000	33	10.30	3.787	227.2
25'000	100	23.03	6.248	374.9
25'000	333	23.03	1.876	112.6

Table 2: characteristic spindle rotation time τ_{sp} depending on the syringe and the flow rate selected

3.4 Operation of NEXUS 3000

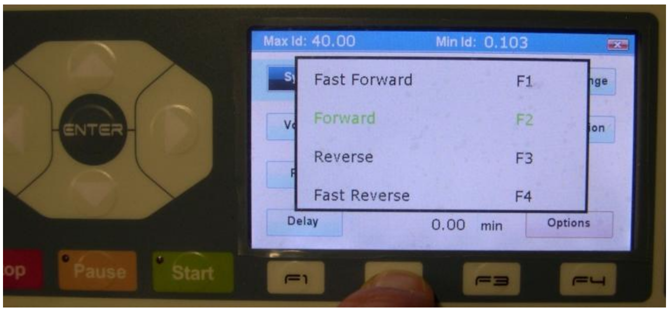
The operation of the syringe pump is straightforward and self-explanatory. Nevertheless, the different steps are quickly described.

1) First install the syringe on the pump after the syringe is properly filled with water and connected to the facility.

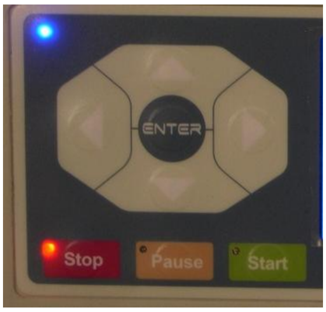
2) On the front panel, choose “Basic” (1 syringe) and press “Enter”

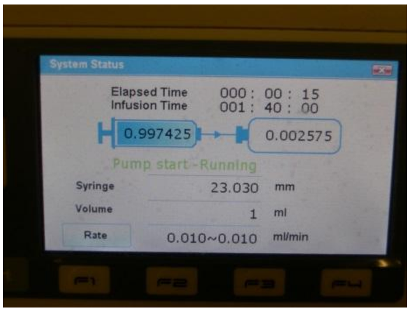
3) Set the syringe diameter (mm), the volume to be dispensed (ml) and the flow rate (ml/min). The operation direction is «Infusion».

4) Fill the syringe and bring the plunger of the syringe pump into position. Use the buttons F1 «Fast Forward», F2 «Forward», F3 «Reverse» and F4 «Fast Reserve». Play with the buttons prior of mounting any syringe as the direction is reversed due to the way we use the syringe pump. Otherwise in the worst case, you might break a syringe.

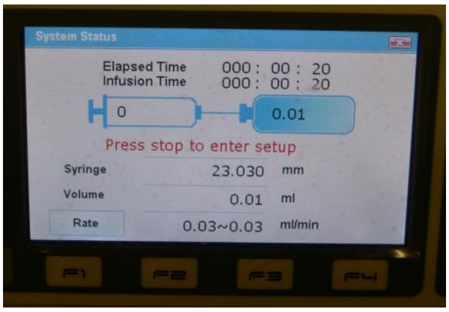


5) If the syringe is ready to be used, press «Start». You will get the following information displayed.





6) To stop the delivery you can press «Stop» or if the delivery is finished press «Stop» to enter the setup menu.



4 Measurement uncertainty

The reported measurement uncertainty is stated as the combined standard uncertainty multiplied by a coverage factor $k = 2$. The measured value (y) and the associated expanded uncertainty (U) represent the interval ($y \pm U$) which contains the value of the measured quantity with a probability of approximately 95 %. The uncertainty was estimated following the guidelines of the ISO (GUM:1995).

The measurement uncertainty contains contributions originating from the measurement standard, from the measurement method, from the environmental conditions and from the object being measured. The long-term characteristic of the object being measured is not included.

It is worth mentioning at this point that the stated measurement uncertainties are the uncertainties established for steady flow. As we average over several cycles of the spindle rotation time and do not state an instantaneous flow rate dependent on the syringe plunger position, the stated uncertainties are valid for the averaged flow, taking into account an averaging time that is an integer multiplication of the spindle rotation time.

5 Characterization Results and Discussion

5.1 Flow rate determination

As we use the Orthogonal Distance Regression to determine the slope of a segment of data which are the weighing and the corresponding time values, we need to make sure that the determined deviation of the syringe pump flow rate compared to the reference flow rate is not affected by the flow rate determination procedure.

First of all, the chosen fit window of data will give a single flow rate point and the corresponding time has to be chosen as the center time of the chosen fit window as can be seen in Figure 1.

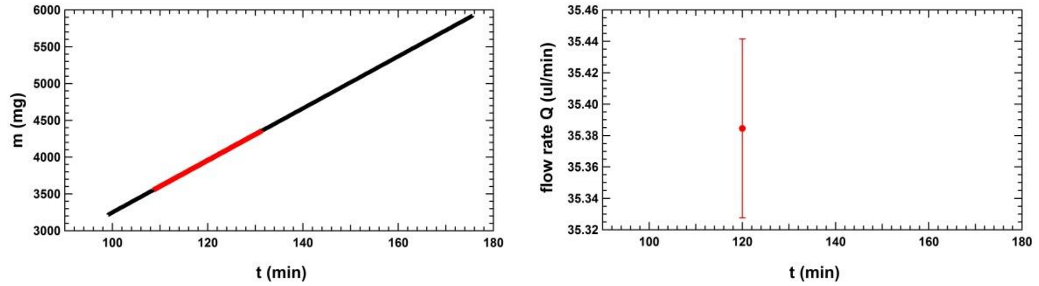


Figure 1: The chosen fit window of data to determine the flow rate will give a single flow rate point and the corresponding time has to be chosen as the center time of the chosen fit window.

Additionally, any averaging of flow rate data will also be displayed with the center value of these ranges of times or positions of the syringe plunger.

5.2 Flow periodicity due to the characteristic spindle rotation time

Independent of the fit window used, the averaging of the data has to be done with a time window which is an integer multiple of the characteristic spindle rotation time to avoid aliasing.

In the case of the gravimetric standing start-stop method or the gravimetric flying start-stop method the time window has to be an integer multiple of the spindle rotation time as well.

We will show this artefact with an experiment.

Flow rate 10 μl/min with the syringe of 2500 μl (Experiment 2013/10/16 09h15)

We use the collected data from the syringe plunger positions 800 μl to 200 μl, which corresponds to a time window of 3600 s.

In Figure 2, we show the determined flow rate with a fit window of 10 s. We see strong fluctuations caused by the syringe pump, which are due to the rotation of the spindle and introduce a periodicity in the data.

The calculated characteristic time τ_{sp} of the spindle is 6.244 min or 374.6 s for this flow rate and this syringe diameter.

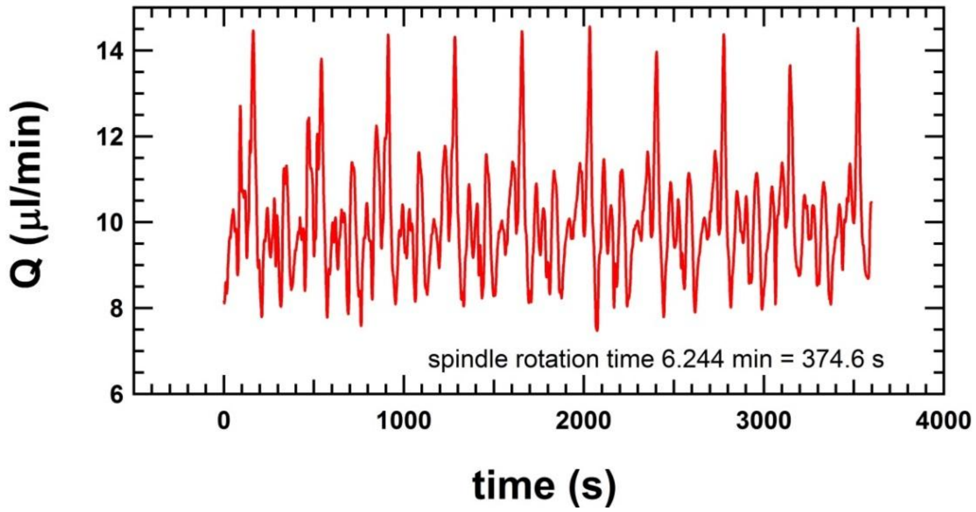


Figure 2: Flow rate determination with a fit window of 10 s.

By means of the Fast Fourier Transformation (Figure 3) we can extract the time periodicity of the flow rate signal shown in Figure 2. The calculated characteristic time τ_{sp} of the spindle, which is 374.6 s for this flow rate and this syringe diameter, corresponds to the time periodicity extracted from the experiment as can be seen in Table 3, where the peaks of the Fast Fourier Transformation are reported.

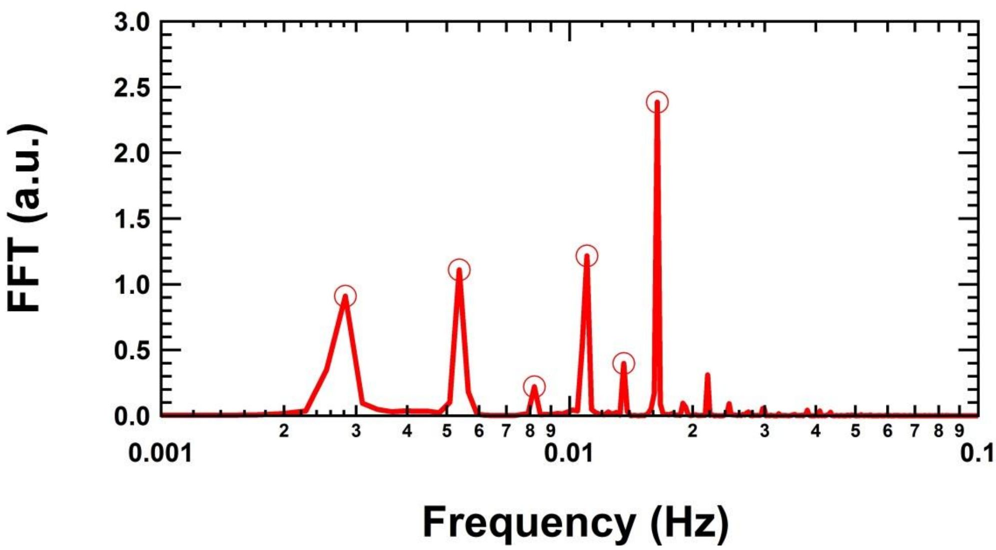


Figure 3: Time periodicity of the flow rate signal shown in Figure 2. Results (open circles) are reported in Table 3.

Peak frequency (Hz)	Order of peaks	Time periodicity (s)
0.002825	1	354.0
0.005367	2	372.6
0.008192	3	366.2
0.011017	4	363.1
0.013559	5	368.8
0.016384	6	366.2

Table 3: Peak frequencies from the Fast Fourier Transformation to extract the time periodicity of the flow rate signal shown in Figure 2

In Figure 4, we show 3 different ways of determining the average flow rate for this experiment.

(1) The first one is the integration of the flow rate determined with the fit window of 10 s. We have determined the flow rate as a function of time by incrementing the position of the fit window by the time step Δt_i, which is 0.1 s in our case due to the read out frequency of the balance. The integration time $\sum \Delta t_i$ can now be chosen between the minimum of 0.1 s or the maximum, which is the time frame of our data (3600 s for this experiment) Therefore we get

$$\bar{Q} = \sum Q_i \cdot \Delta t_i \, / \sum \Delta t_i$$

The integration time $\sum \Delta t_i$ is plotted on the x-axis «averaging time» in Figure 4 and 5 and the integrated flow rate $\bar{Q}$ on the y-axis (blue curve).

(2) The second one is the integration of the flow rate determined with the fit window of 1526 s. We have determined the flow rate as a function of time by incrementing the position of the fit window by the time step Δt_i, which is 0.1 s in our case due to the read out frequency of the balance. The integration time $\sum \Delta t_i$ can now be chosen between the minimum of 0.1 s or the maximum, which is the time frame of our data (3600 s for this experiment) Therefore we get

$$\bar{Q} = \sum Q_i \cdot \Delta t_i \, / \sum \Delta t_i$$

The integration time $\sum \Delta t_i$ is plotted on the x-axis «averaging time» in Figure 4 and 5 and the integrated flow rate $\bar{Q}$ on the y-axis (black curve).

(3) The 3rd one is the flow rate determination by means of the ratio between the increase in mass and the increase in time, where

$$Q = \Delta m / \Delta t$$

The increase in time Δt is plotted on the x-axis «averaging time» in Figure 4 and 5 and the flow rate Q on the y-axis (green curve).

We clearly see in Figure 4 and 5 that all 3 methods lead to approximately the same result **if the averaging time is chosen as an integer multiple of the spindle rotation time**, which is indicated by the blue/magenta circles for the blue curve and the light green circles for the green curve.

If the averaging time is not chosen as an integer multiple of the spindle rotation time, then in the case of the blue and the green curve large deviations can be implemented as systematic errors – as large as the measurement uncertainty or even larger for shorter averaging times!

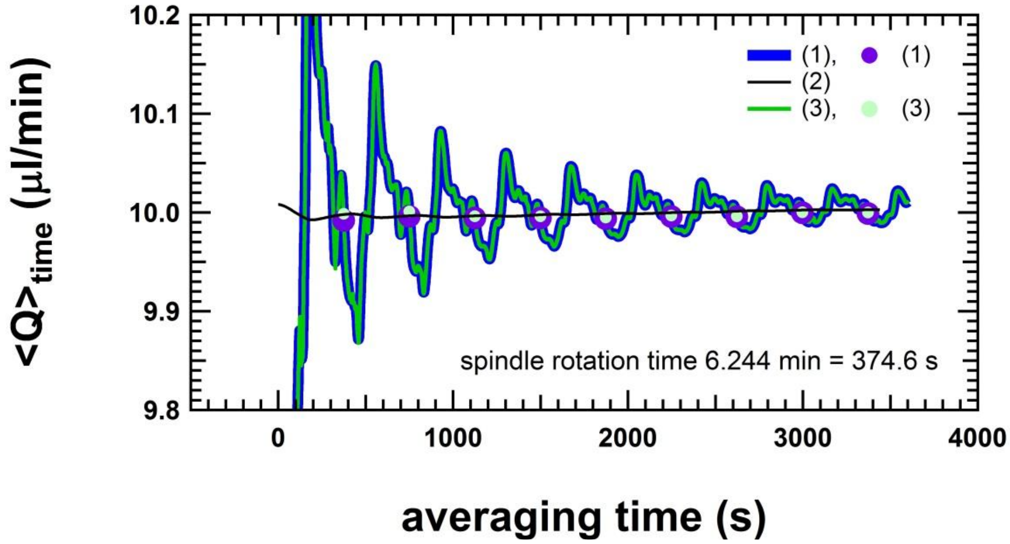


Figure 4: Average flow rate by using either the integration method (1, blue curve) and (2, black curve) or the ratio between the increase in mass and the increase in time (3, green curve). The integer multiples of the spindle rotation time are indicated by the blue/magenta circles for the blue curve and the light green circles for the green curve.

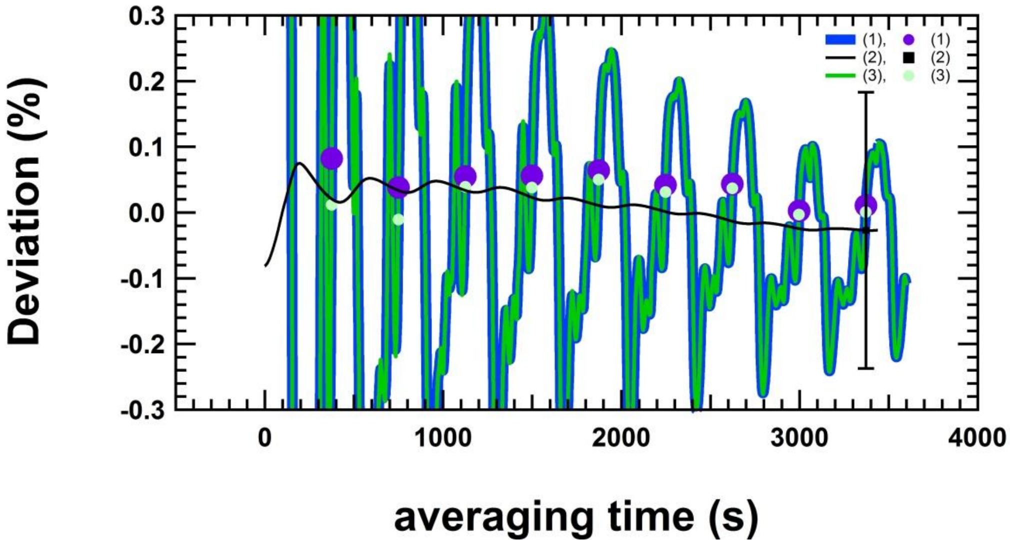


Figure 5: Deviations of the average flow rate by either using the integration method (1, blue curve) or (2, black curve) or the ratio between the increase in mass and the increase in time (3, green curve). The integer multiples of the spindle rotation time are indicated by the blue/magenta circles for the blue curve and the light green circles for the green curve. The black square is the result of the flow rate determination for steady flow with the corresponding uncertainty.

In conclusion, the time window is important and has to be chosen to be an integer multiple of the characteristic spindle rotation time!

5.3 Flow rate depending on plunger position

We performed an experiment where we used the syringe of a volume of 2500 μl. We set the plunger at the starting position being at the marking of 2300 μl. The following parameters were set at the syringe pump: the volume to be dispensed was set to 2200 μl, the flow rate was set to 50 μl/min and the inner diameter of the syringe body was set to 7.28 mm. Then the pump was started and the weighing data were collected.

In Figure 6, we clearly see a flow rate dependency of the plunger position of the syringe. The plunger position of the syringe is calculated according the starting conditions:

$$position\ plunger\ (\mu l) = starting\ position\ plunger\ (\mu l) - flow\ rate\ (\mu l/min)/60 \cdot t_{syringe\ pump\ working}\ .$$

To get a better impression we analysed the flow rates over 3 cycles at 4 different plunger position ranges (1600 μl – 1412.65 μl, 1300 μl – 1112.65 μl, 1000 μl – 812.65 μl, 500 μl – 312.65 μl) for 4 measurements. The data points are shown in Figure 7 and in Table 4.

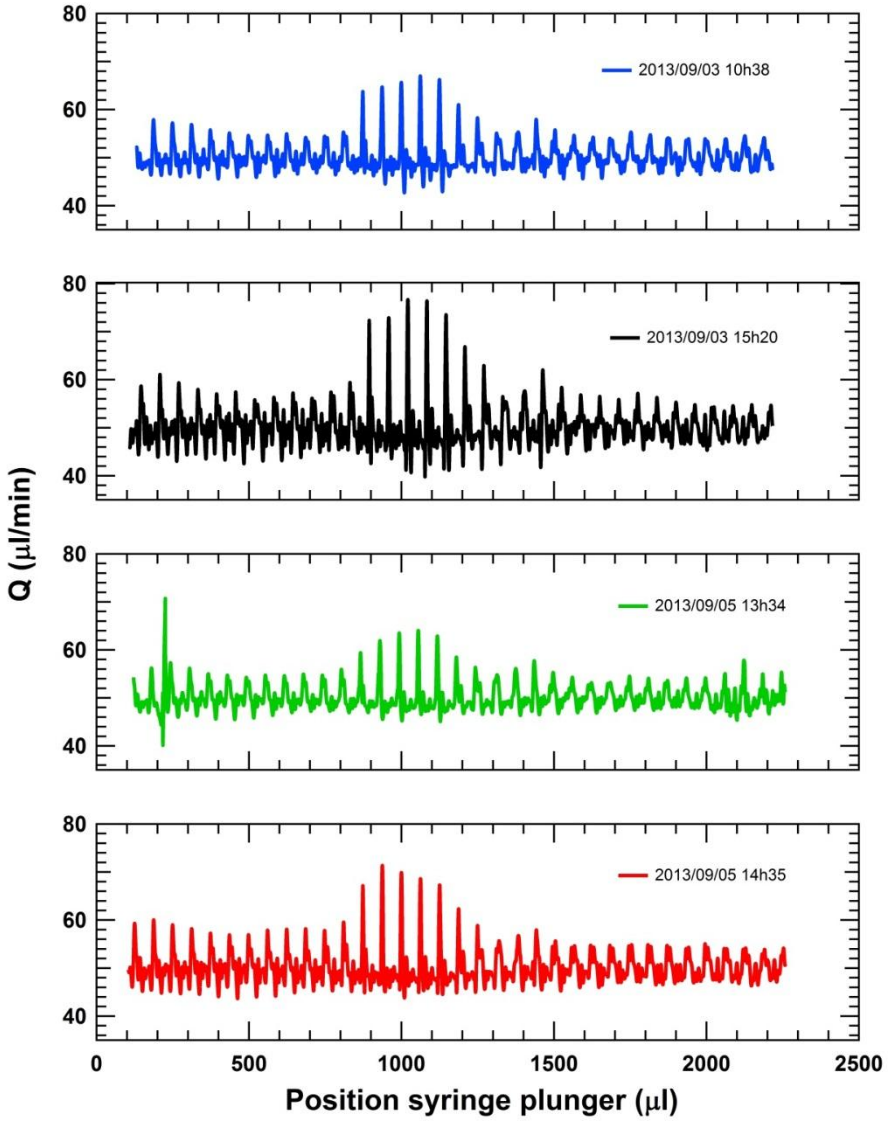


Figure 6: Flow rate dependency of the plunger position measured with a syringe of a volume of 2500 μl and a diameter of 7.28 mm.

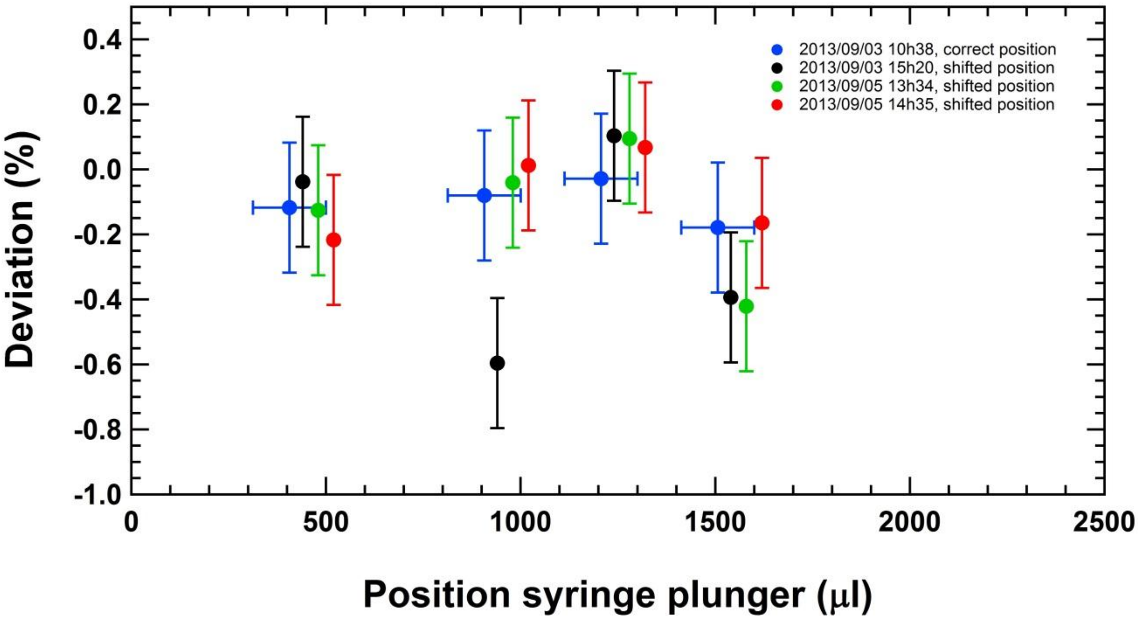


Figure 7: Flow rates determined over 3 cycles at 4 different syringe plunger positions for 4 measurements. The blue circles are plotted at the correct center position of the data used for the determination of the deviation. The other circles are shifted in order to avoid the overlap of the data points. The horizontal error bar indicates the range of data used to perform the average.

Q (µl/min)	Measurement	Section (µl)	Center position (µl)	State of center position	Deviation (%)	Uncertainty (%)	σ position (%)
50	2013/09/03 10h38	1600 – 1412.65	1506.33	Correct	-0.179	0.2	0.063
	2013/09/03 10h38	1300 – 1112.65	1206.33	Correct	-0.028	0.2	
	2013/09/03 10h38	1000 – 812.65	906.33	Correct	-0.081	0.2	
	2013/09/03 10h38	500 – 312.65	406.33	Correct	-0.117	0.2	
	2013/09/03 15h20	1600 – 1412.65	1540	Shifted	-0.394	0.2	0.320
	2013/09/03 15h20	1300 – 1112.65	1240	Shifted	0.103	0.2	
	2013/09/03 15h20	1000 – 812.65	940	Shifted	-0.596	0.2	
	2013/09/03 15h20	500 – 312.65	540	Shifted	-0.038	0.2	
	2013/09/05 13h34	1600 – 1412.65	1580	Shifted	-0.421	0.2	0.220
	2013/09/05 13h34	1300 – 1112.65	1280	Shifted	0.094	0.2	
	2013/09/05 13h34	1000 – 812.65	980	Shifted	-0.041	0.2	
	2013/09/05 13h34	500 – 312.65	580	Shifted	-0.126	0.2	
	2013/09/05 14h35	1600 – 1412.65	1620	Shifted	-0.165	0.2	0.140
	2013/09/05 14h35	1300 – 1112.65	1320	Shifted	0.068	0.2	
	2013/09/05 14h35	1000 – 812.65	1020	Shifted	0.012	0.2	
	2013/09/05 14h35	500 – 312.65	620	Shifted	-0.217	0.2	

Table 4: Flow rates determined over 3 cycles at 4 different syringe plunger positions for 4 measurements. The results are shown in Figure 7.

The origin of this dependency on the syringe plunger position is not due to inner diameter variations of the syringe body as we can see in Figure 8. The inner diameter of the syringe is constant over the whole volume and the small deviations are too small to change the cross section sufficiently to cause this variation in the flow rate fluctuations.

Deviation from the mean inner diameter: 0.2 μm.

D_1 = 7.2850 mm, S_1 = 41.682 mm^2

D_2 = 7.2852 mm, S_2 = 41.684 mm^2

$\Delta S = S_2 - S_1$ = 0.002 mm^2

Relative change in surface $\Delta S/S = 5\ 10^{-5}$, which is equal to the relative change in flow rate.

However, it is probably more a combination of the mechanical properties of the motion and the geometry of the spindle along the axis.

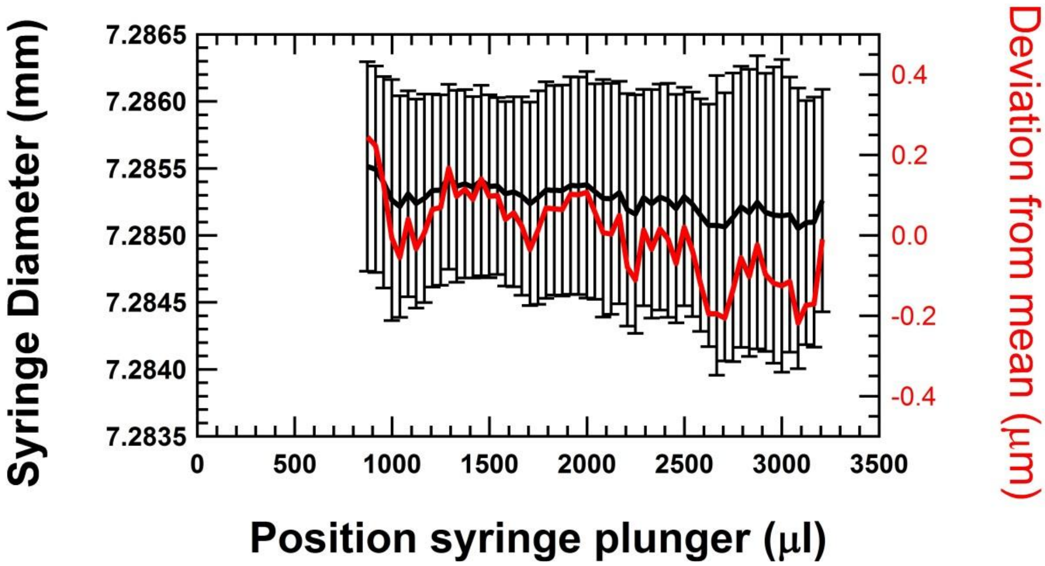


Figure 8: Measured inner diameter of the syringe body along the plunger position of the syringe. The variations in the diameter are negligible.

5.4 Measurement procedure

For each experiment of repeatability measurements, we filled the syringe with degassed water and applied our filling procedure to avoid air bubbles to stick at the wall or at the plunger in the glass syringe.

The filling procedure consists of:

- Connect the syringe to the filling piping
- Flush the piping and the connected syringe with CO_2 and hold the syringe in order that the CO_2 remains in the syringe body (CO_2 is heavier than air)
- Fill the piping and the connected syringe with degassed water.
- Remove the syringe from the «luer-lock» adapter and try to keep as much water as possible inside the syringe body
- Immerse the plunger into water (separate beaker) in order that the sealing part is wet
- Push the plunger into the syringe body and turn the syringe around in order to get the water close to the plunger and the CO_2-air mixture close to the outlet
- If gas bubbles stick to the plunger, shake them away by turning around the syringe (close the outlet with your thumb)
- If all the gas is above the water, push the gas out of the syringe
- To reconnect the syringe to the «luer-lock» adapter water has to flow out of the «luer-lock» adapter and the outlet of the syringe has to be full of water.

We mounted the syringe on the syringe pump and set the plunger at the plunger starting position. Then the following parameters were set at the syringe pump: the volume to be dispensed, the flow rate and the diameter of the glass syringe used (information from the manufacturer). Then the pump was started and the weighing data were collected.

To repeat the measurement, we simply filled the syringe again, set the plunger at the plunger starting position and started the syringe pump.

For the reproducibility measurements, the syringe was first emptied and dried and refilled only several days later to perform a second round of repeatability measurements.

5.5 Flow rate Q ~2 µl/min, 1st set of syringes

Measurement time: 3 h

Syringe used: 2500 µl volume, 1st set of syringes

In Figure 9, we can see the syringe mounted in the syringe pump and connected via a Luer-Lock connector to the 1/8” tubing.

Figure 9: syringe (2500 µl volume) mounted in the syringe pump.

We perform the flow rate determination with fit windows of 3600 s and 10 s (only for the second round of measurements). The deviations of the flow rates are plotted against the position of the syringe plunger (see Figure 10).

We want to determine the deviation of the syringe pump at the syringe plunger positions between 800 µl and 500 µl.

As the characteristic spindle rotation time is 31.219 min for this flow rate, we can average over 4 cycles which represents a volume of 249.76 µl: from 800 µl to 550.24 µl. The corresponding average values are added as full circles in Figure 10.

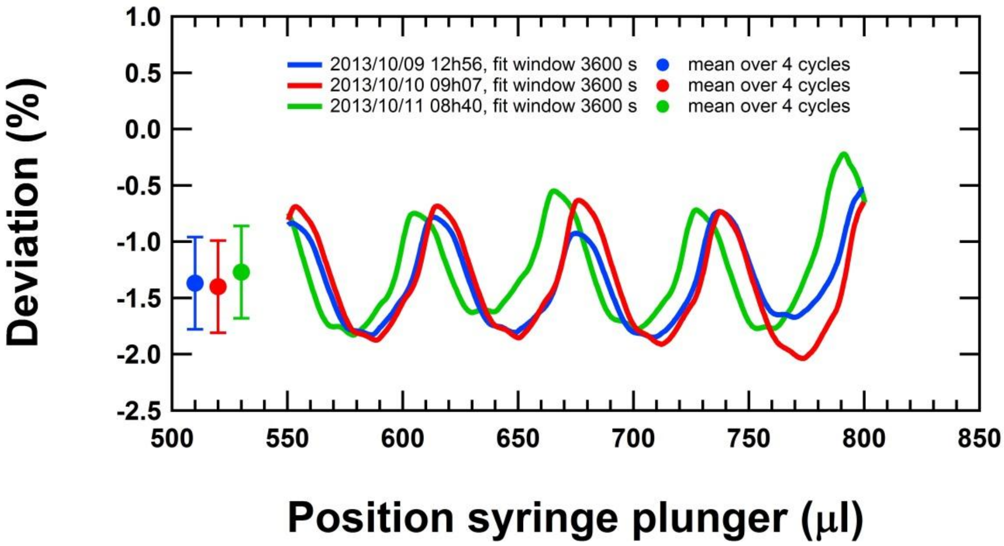


Figure 10: The determined deviations of the flow rates with a fit window of 3600 s are plotted against the position of the syringe plunger. The averages over 4 cycles from 800 µl to 550.24 µl which represents a volume of 249.76 µl are added as full circles.

We performed measurements to establish the reproducibility of the syringe pump with the chosen syringe and got the results number 4 and 5, as can be seen in Figure 11. The full

circles are the averages using a fit window of 3600 s and the open circles are the averages using a fit window of 10 s. It is worthwhile to mention that the first measurement of a new series always slightly differs from the following results. However, we keep this measurement of the second round to establish a reproducibility that reflects the use in general.

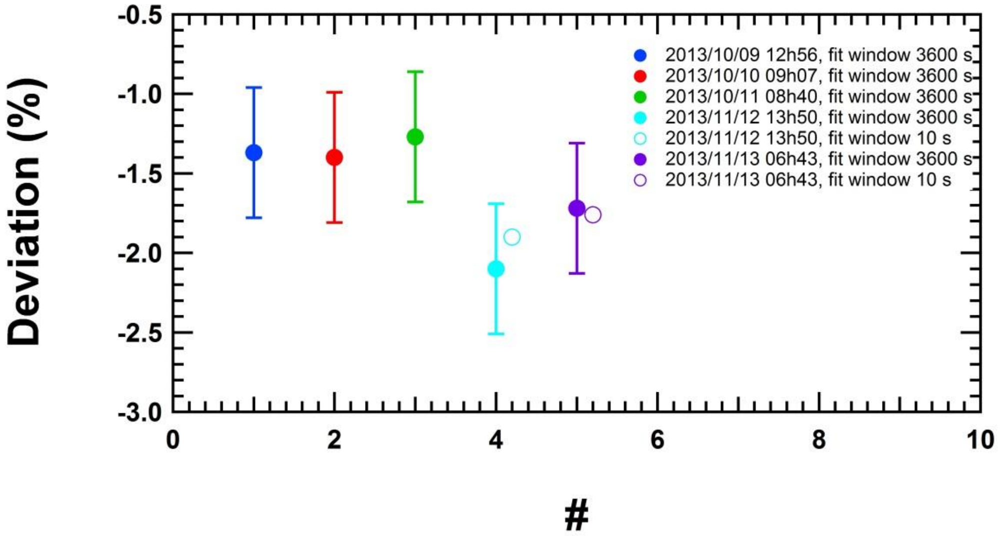


Figure 11: Repeatability and Reproducibility measurements for the flow rate of 2 μl/min using a syringe with a volume of 2500 μl. Measurements 1-3 are part of the first round and the measurements 4-5 are part of the second round. The standard deviation of the measurements 1 - 3 is 0.068 % (repeatability) and of the measurements 1-5 is 0.34 % (reproducibility).

The deviations from the selected flow rate of 2 μl/min are summarized in the table below as well as the measure for repeatability and reproducibility.

Q (μl/min)	Measurement #	Round	Fit window (s)	Deviation (%)	Uncertainty (%)	σ repeat (%)	σ reprod (%)
2	1	First	3600	-1.27	0.41	0.068	0.34
	2	First	3600	-1.40	0.41		
	3	First	3600	-1.37	0.41		
	4	Second	3600	-2.1	0.41		
	4	Second	10	-1.9	To be defined		
	5	Second	3600	-1.72	0.41		
	5	Second	10	-1.76	To be defined		

Table 5: The deviations from the selected flow rate of 2 μl/min presented in Figure 11.

Note: It is worthwhile to mention that for the last measurement we had air bubbles sticking at the syringe wall and the plunger as can be seen in Figure 12. However, they did not move and therefore not influence the delivered volume at this flow rate.

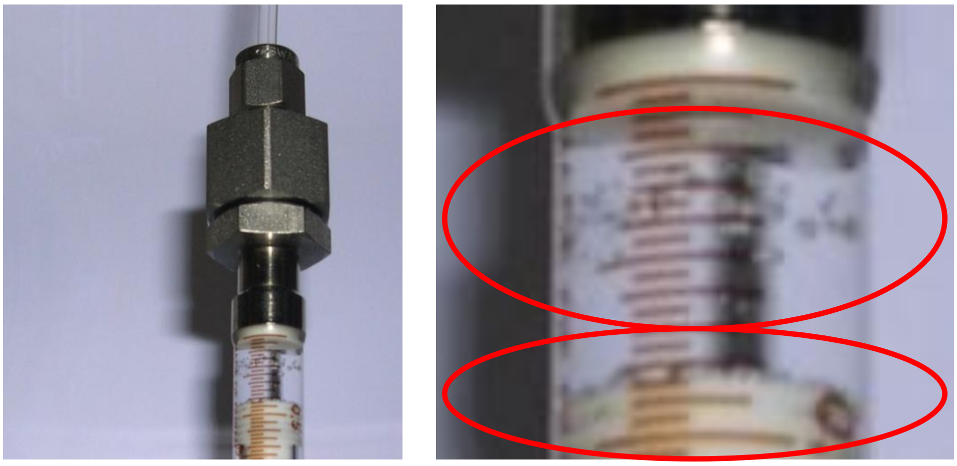


Figure 12: Air bubbles sticking at the syringe wall and the plunger.

We made also some measurements using syringes with a volume of 250 μl and 500 μl. The repeatability of the measurements was very poor which might be due to the thin plunger of the syringe. As the plunger is not fixed on the syringe pump, but only pushed by the moving block, the stability for good repeatability measurements is not given.

5.6 Flow rate Q ~10 μl/min, 1st set of syringes

Measurement time: 2 h

Syringe used: 2500 μl volume, 1st set of syringes

In Figure 13, we can see the syringe mounted in the syringe pump and connected via a Luer-Lock connector to the 1/8" tubing.

Figure 13: syringe (2500 μl volume) mounted in the syringe pump.

We perform the flow rate determination with fit windows of 1500 s and 10 s (only for the second round of measurements). The deviations of the flow rates are plotted against the position of the syringe plunger (see Figure 14).

We want to determine the deviation of the syringe pump at the syringe plunger positions between 800 μl and 200 μl.

As the characteristic spindle rotation time is 6.24 min for this flow rate, we can average over 9 cycles which represents a volume of 561.6 μl from 800 μl to 238.4 μl. The corresponding average values are added as full circles in Figure 13.

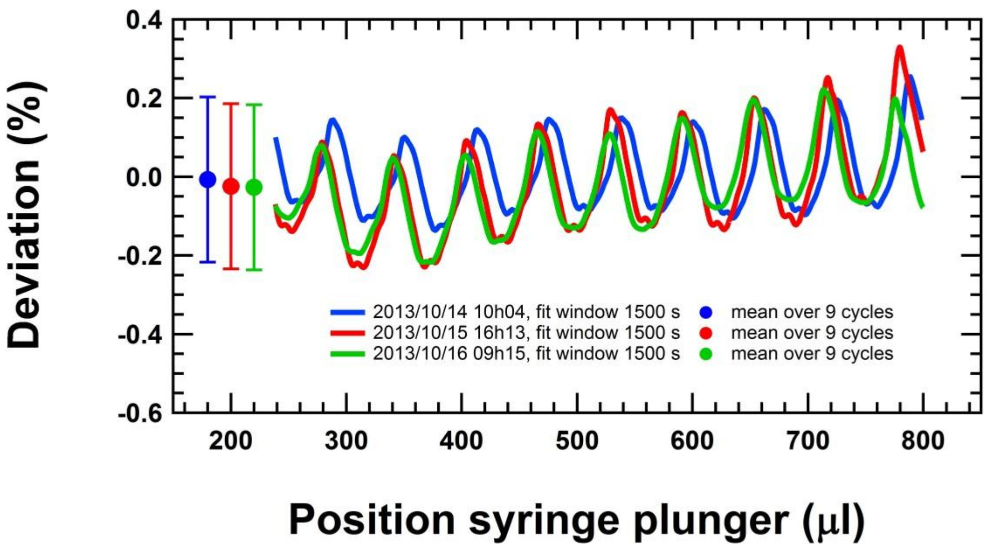


Figure 14: The determined deviations of the flow rates with a fit window of 1500 s are plotted against the position of the syringe plunger. The averages over 9 cycles from 800 μl to 238.4 μl which represents a volume of 561.6 μl are added as full circles.

We performed measurements to establish the reproducibility of the syringe pump with the chosen syringe and got the results number 4 and 5, as can be seen in Figure 15. The full

circles are the averages using a fit window of 1500 s and the open circles are the averages using a fit window of 10 s. It is worthwhile to mention that the first measurement of a new series always slightly differs from the following results. However, we keep this measurement of the second round to establish a reproducibility that reflects the use in general.

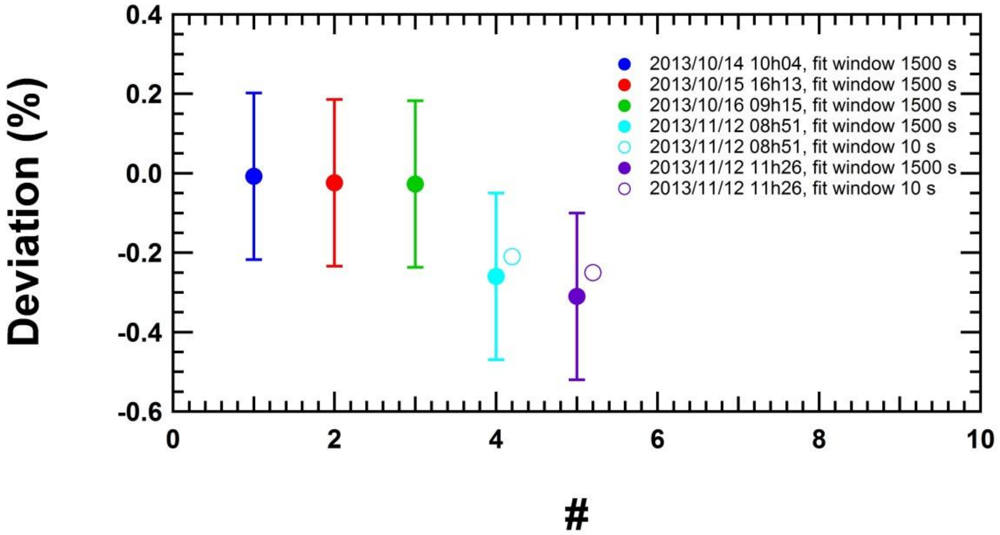


Figure 15: Repeatability and Reproducibility measurements for the flow rate of 10 µl/min using a syringe with a volume of 2500 µl. Measurements 1-3 are part of the first round and the measurements 4-5 are part of the second round. The standard deviation of the measurements 1 - 3 is 0.019 % (repeatability) and of the measurements 1-5 is 0.15 % (reproducibility).

The deviations from the selected flow rate of 10 µl/min are summarized in the table below as well as the measure for repeatability and reproducibility.

Q (µl/min)	Measurement #	Round	Fit window (s)	Deviation (%)	Uncertainty (%)	σ repeat (%)	σ reprod (%)
10	1	First	1500	0.0074	0.21	0.019	0.15
	2	First	1500	-0.024	0.21		
	3	First	1500	-0.027	0.21		
	4	Second	1500	-0.26	0.21		
	4	Second	10	-0.21	To be defined		
	5	Second	1500	-0.31	0.21		
	5	Second	10	-0.25	To be defined		

Table 6: The deviations from the selected flow rate of 10 µl/min presented in Figure 15.

5.7 Flow rate Q ~33 µl/min, 1st set of syringes

Measurement time: 1.5 h

Syringe used: 5000 µl volume, 1st set of syringes

In Figure 16, we can see the syringe mounted in the syringe pump and connected via a Luer-Lock connector to the 1/8” tubing.

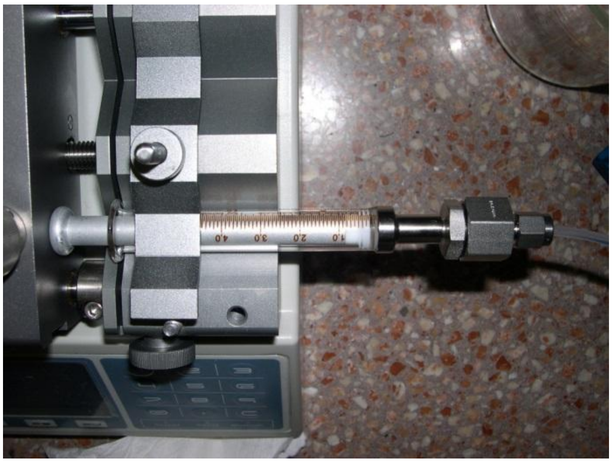


Figure 16: syringe (5000 µl volume) mounted in the syringe pump.

We perform the flow rate determination with fit windows of 1350 s and 10 s (only for the second round of measurements). The deviations of the flow rates are plotted against the position of the syringe plunger (see Figure 17).

We want to determine the deviation of the syringe pump at the syringe plunger positions between 1800 µl and 600 µl.

As the characteristic spindle rotation time is 3.787 min for this flow rate, we can average over 9 cycles which represents a volume of 1124.74 µl from 1800 µl to 675.26 µl. The corresponding average values are added as full circles in Figure 17.

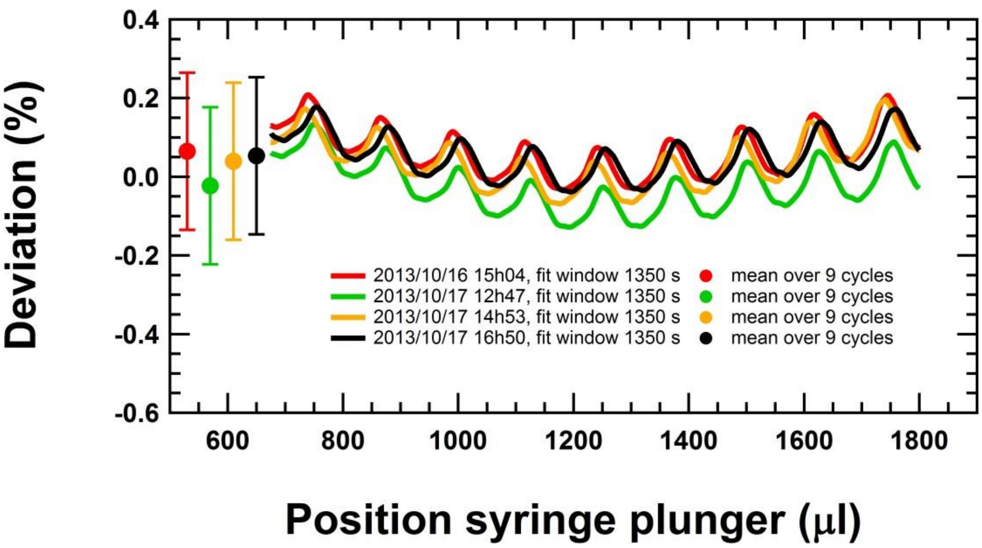


Figure 17: The determined deviations of the flow rates with a fit window of 1350 s are plotted against the position of the syringe plunger. The averages over 9 cycles from 1800 µl to 675.26 µl which represents a volume of 1124.74 µl are added as full circles.

We performed measurements to establish the reproducibility of the syringe pump with the chosen syringe and got the results number 5 and 6, as can be seen in Figure 18. The full

circles are the averages using a fit window of 1350 s and the open circles are the averages using a fit window of 10 s. It is worthwhile to mention that the first measurement of a new series always slightly differs from the following results. However, we keep this measurement of the second round to establish a reproducibility that reflects the use in general.

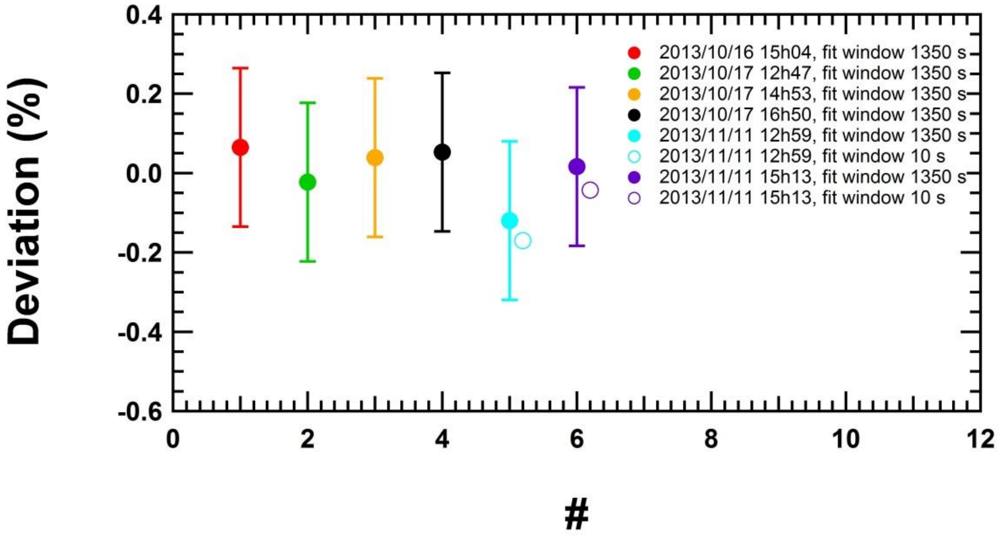


Figure 18: Repeatability and Reproducibility measurements for the flow rate of 33 μl/min using a syringe with a volume of 5000 μl. Measurements 1-4 are part of the first round and the measurements 5-6 are part of the second round. The standard deviation of the measurements 1 - 4 is 0.039 % (repeatability) and of the measurements 1-6 is 0.069 % (reproducibility).

The deviations from the selected flow rate of 33 μl/min are summarized in the table below as well as the measure for repeatability and reproducibility.

Q (μl/min)	Measurement #	Round	Fit window (s)	Deviation (%)	Uncertainty (%)	σ repeat (%)	σ reprod (%)
33	1	First	1350	0.065	0.20	0.039	0.069
	2	First	1350	-0.023	0.20		
	3	First	1350	0.039	0.20		
	4	First	1350	0.053	0.20		
	5	Second	1350	-0.12	0.20		
	5	Second	10	-0.17	To be defined		
	6	Second	1350	0.016	0.20		
	6	Second	10	-0.043	To be defined		

Table 7: The deviations from the selected flow rate of 33 μl/min presented in Figure 18.

5.8 Flow rate Q ~100 µl/min, 1st set of syringes

Measurement time: 0.5 h

Syringe used: 25000 µl volume, 1st set of syringes

In Figure 19, we can see the syringe mounted in the syringe pump and connected via a Luer-Lock connector to the 1/8” tubing.

Figure 19: syringe (25000 µl volume) mounted in the syringe pump.

We perform the flow rate determination with fit windows of 330 s and 10 s (only for the second round of measurements). The deviations of the flow rates are plotted against the position of the syringe plunger (see Figure 20).

We want to determine the deviation of the syringe pump at the syringe plunger positions between 7300 µl and 5300 µl.

As the characteristic spindle rotation time is 6.248 min for this flow rate, we can average over 3 cycles which represents a volume of 1874.4 µl from 7300 µl to 5425.6 µl. The corresponding average values are added as full circles in Figure 19.

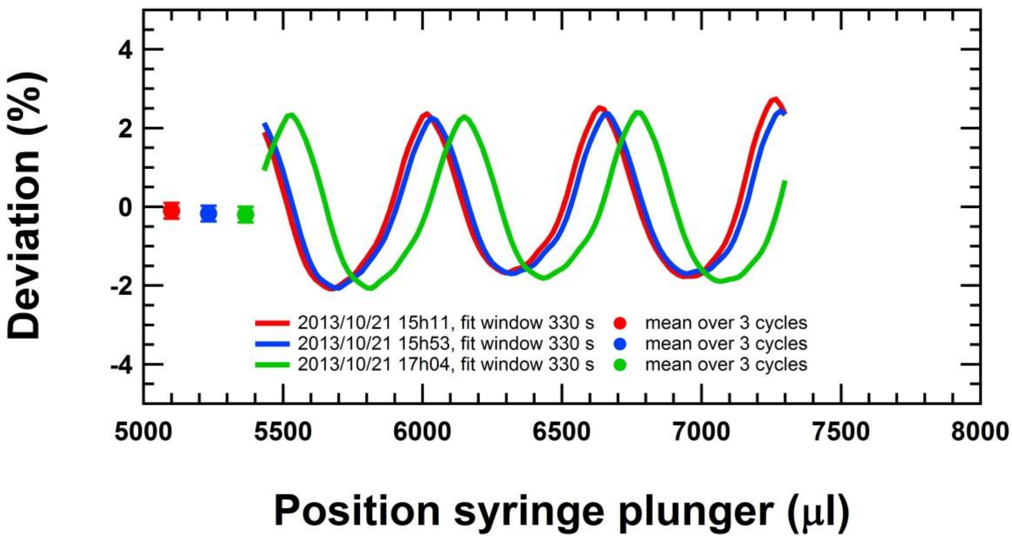


Figure 20: The determined deviations of the flow rates with a fit window of 330 s are plotted against the position of the syringe plunger. The averages over 3 cycles from 7300 µl to 5425.6 µl which represents a volume of 1874.4 µl are added as full circles.

We performed measurements to establish the reproducibility of the syringe pump with the chosen syringe and got the results number 4 and 5, as can be seen in Figure 21. The full

circles are the averages using a fit window of 330 s and the open circles are the averages using a fit window of 10 s. It is worthwhile to mention that the first measurement of a new series always slightly differs from the following results. However, we keep this measurement of the second round to establish a reproducibility that reflects the use in general.

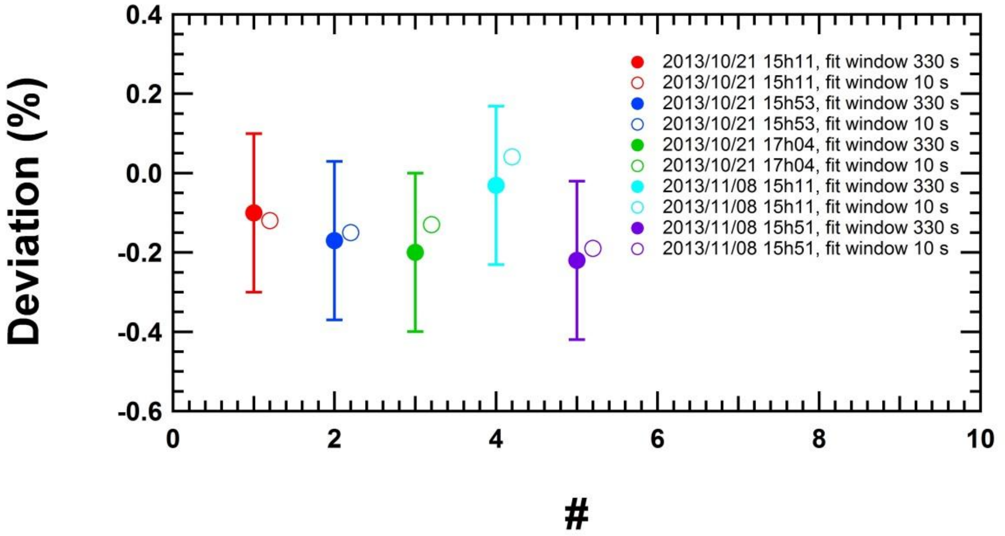


Figure 21: Repeatability and Reproducibility measurements for the flow rate of 100 µl/min using a syringe with a volume of 25000 µl. Measurements 1-3 are part of the first round and the measurements 4-5 are part of the second round. The standard deviation of the measurements 1 - 3 is 0.051 % (repeatability) and of the measurements 1-5 is 0.078 % (reproducibility).

The deviations from the selected flow rate of 100 µl/min are summarized in the table below as well as the measure for repeatability and reproducibility.

Q (µl/min)	Measurement #	Round	Fit window (s)	Deviation (%)	Uncertainty (%)	σ repeat (%)	σ reprod (%)
100	1	First	330	-0.10	0.20	0.051	0.078
	2	First	330	-0.17	0.20		
	3	First	330	-0.20	0.20		
	4	Second	330	-0.031	0.20		
	4	Second	10	0.041	To be defined		
	5	Second	330	-0.22	0.20		
	5	Second	10	-0.19	To be defined		

Table 8: The deviations from the selected flow rate of 100 µl/min presented in Figure 21.

For illustration we show in Figure 22 the pulsations measured when we analyse the data with a fit window of 10 s. For comparison, we also show the pulsations obtained when using a fit window of 330 s. As the starting position of the syringe plunger is not well reproducible due to the poor resolution in hand positioning, we set for the blue curve an offset -20 µl and for the green curve an offset -130 µl at the syringe plunger position. We clearly see that the large main pulsations are very well reproducible.

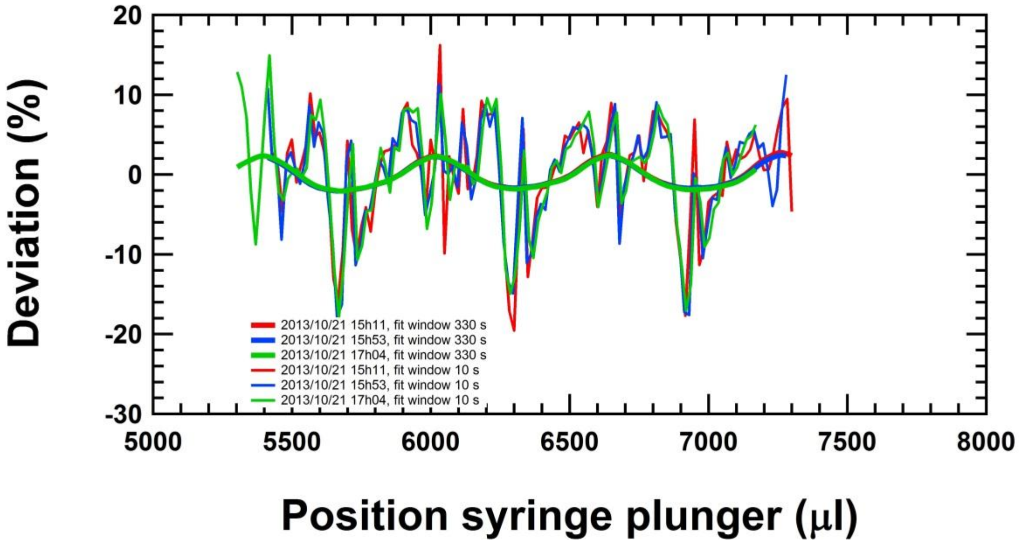


Figure 22: The determined flow rates with a fit window of 330 s and 10 s are plotted against the position of the syringe plunger. (blue) offset -20 µl, (green) offset -130 µl.

5.9 Flow rate Q ~333 µl/min, 1st set of syringes

Measurement time: 0.25 h

Syringe used: 25000 µl volume, 1st set of syringes

In Figure 23, we can see the syringe mounted in the syringe pump and connected via a Luer-Lock connector to the 1/8" tubing.

Figure 23: syringe (25000 µl volume) mounted in the syringe pump.

We perform the flow rate determination with fit windows of 130 s and 10 s. The deviations of the flow rates are plotted against the position of the syringe plunger (see Figure 23).

We want to determine the deviation of the syringe pump at the syringe plunger positions between 7300 µl and 5300 µl.

As the characteristic spindle rotation time is 1.876 min for this flow rate, we can average over 3 cycles which represents a volume of 1874.12 µl from 7300 µl to 5425.88 µl. The corresponding average values are added as full circles in Figure 24.

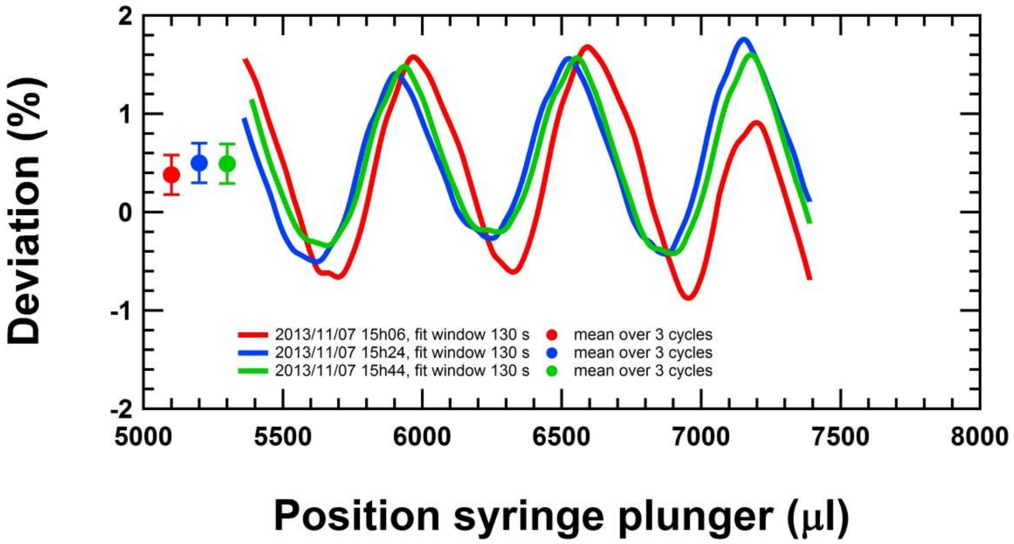


Figure 24: The determined deviations of the flow rates with a fit window of 130 s are plotted against the position of the syringe plunger. The averages over 3 cycles from 7300 µl to 5425.88 µl which represents a volume of 1874.12 µl are added as full circles.

We performed measurements to establish the reproducibility of the syringe pump with the chosen syringe and got the results number 4 and 5, as can be seen in Figure 25. The full circles are the averages using a fit window of 130 s and the open circles are the averages using a fit window of 10 s. It is worthwhile to mention that the first measurement of a new series always slightly differs from the following results. However, we keep this measurement of the second round to establish a reproducibility that reflects the use in general.

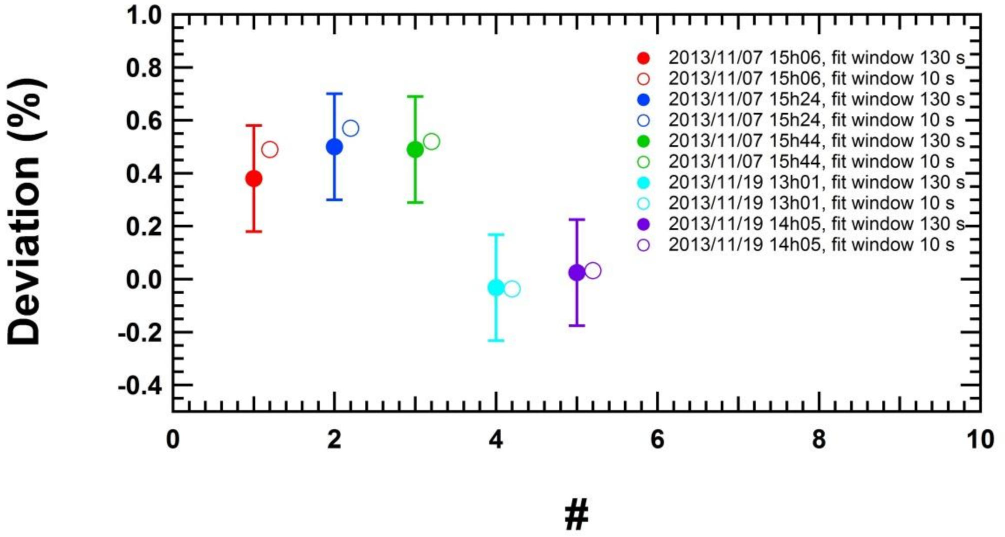


Figure 25: Repeatability and Reproducibility measurements for the flow rate of 333 μl/min using a syringe with a volume of 25000 μl. Measurements 1-3 are part of the first round and the measurements 4-5 are part of the second round. The standard deviation of the measurements 1 - 3 is 0.067 % (repeatability) and of the measurements 1-5 is 0.26 % (reproducibility).

The deviations from the selected flow rate of 333 μl/min are summarized in the table below as well as the measure for repeatability and reproducibility.

Q (μl/min)	Measurement #	Round	Fit window (s)	Deviation (%)	Uncertainty (%)	σ repeat (%)	σ reprod (%)
100	1	First	130	0.38	0.20	0.067	0.26
	2	First	130	0.50	0.20		
	3	First	130	0.49	0.20		
	4	Second	130	-0.032	0.20		
	4	Second	10	-0.037	To be defined		
	5	Second	130	0.025	0.20		
	5	Second	10	0.032	To be defined		

Table 9: The deviations from the selected flow rate of 333 μl/min presented in Figure 25.

5.10 Overview of the results for repeatability and reproducibility

We summarize the results obtained with the 1st set of syringes in one Figure below (Figure 26) where the deviations at the flow rate of 2 µl/min refer to the axis on the left and the other deviations of the rest of the flow rates refer to the axis on the right. Each color code represents repeatability measurements and the 2 different color codes represent reproducibility.

We can make the following statements:

- The repeatability and the reproducibility of the flow rates are very good in general. At the highest flow rate the reproducibility is not as good as expected and the reason for this is not identified.
- The absolute deviations at the lowest flow rate are much larger than all the others. Note, the same syringe was used to perform the measurements at the flow rate of 10 µl/min.

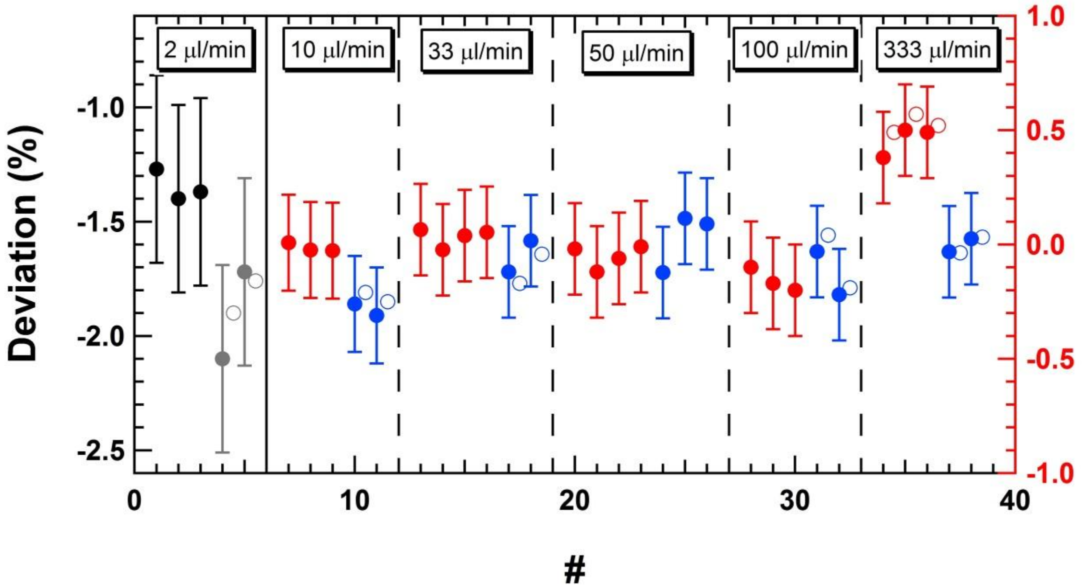


Figure 26: Deviations at the flow rate of 2 µl/min refer to the axis on the left and the other deviations of the rest of the flow rates refer to the axis on the right. Each color code represents repeatability measurements and the 2 different color codes represent reproducibility. (full circles) results for large fit windows (>> 10 s) and averaged over full cycles of the spindle rotation. (empty circles) results for fit windows of 10 s and averaged over full cycles of the spindle rotation.

In Table 10 we have summarized the standard deviations of the measurements at each flow rate expressing statistical values for repeatability and reproducibility.

Q (µl/min)	σ Repeatability (%)	σ Reproducibility (%)

2	0.068	0.340
10	0.019	0.150
33	0.039	0.069
100	0.051	0.078
333	0.067	0.260

Table 10: Standard deviations of the measurements at each flow rate expressing statistical values for repeatability and reproducibility.

5.10.1 Slow oscillations

For the flow rates of 10 μl/min and 33 μl/min, we see in the Figures 14 and 17 a slow oscillation along the plunger position of the syringe. The reason for this is unknown, but it is in agreement with the results found in Figure 7, where we showed that we get different deviations of the flow rate at different plunger positions.

5.10.2 Third round of repeatability measurements

A third round of repeatability measurements will be done by METAS during the inter comparison (June 2014). Therefore more data will be available to confirm the repeatability and reproducibility of this syringe pump NEXUS 3000 in the flow rate range from 2 μl/min to 333 μl/min.

5.11 Results with the 2nd set of syringes (backup)

We summarize in the following table the results obtained with the second set of syringes which is a backup set for the inter-comparison in case that a syringe gets damaged.

Q (µl/min)	number of cycles	Syringe volume (µl)	Fit window (s)	Range of syringe plunger positions to average	Deviation (%)	Uncertainty (%)
2	2	2'500	3600	From 700 µl to 575.12 µl	- 2.03	0.41
					- 1.95	0.41
					- 1.97	0.41
10	4	2'500	1500	From 600.0 µl to 350.4 µl	+ 0.15	0.21
					- 0.16	0.21
					- 0.32	0.21
33	4	5'000	1350	From 1400.0 µl to 900.1 µl	+ 0.29	0.2
					- 0.06	0.2
					+ 0.01	0.2
100	3	25'000	330	From 7300.0 µl to 5425.6 µl	+ 0.09	0.2
					- 0.21	0.2
					- 0.27	0.2
333	3	25'000	130	From 7300.0 µl to 5425.9 µl	+ 0.11	0.2
					+ 0.18	0.2
					+ 0.04	0.2

Table 11: Results obtained with the second set of syringes.

The results are shown in Figure 27, where all the results from the first and the second set of syringes are summarized in the same Figure.

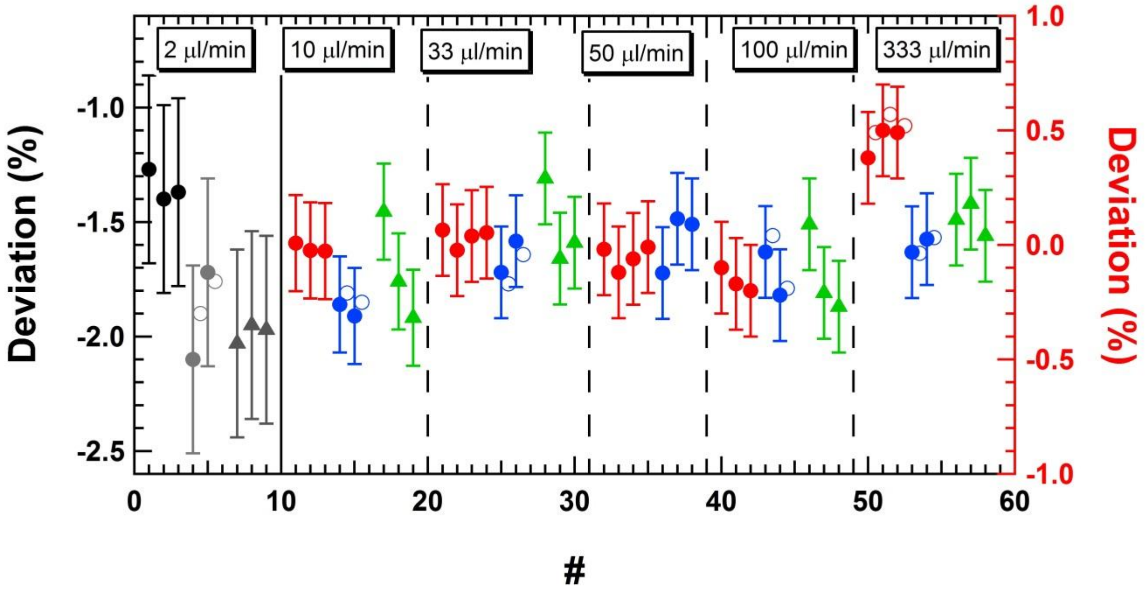


Figure 27: Deviations at the flow rate of 2 µl/min refer to the axis on the left and the other deviations of the rest of the flow rates refer to the axis on the right. The circles represent the results from the first set of syringes and the triangles represent the results from the second set of syringes. Each color code represents repeatability measurements and the 2 different color codes represent reproducibility. (full circles) results for large fit windows (>> 10 s) and averaged over full cycles of the spindle rotation. (empty circles) results for fit windows of 10 s and averaged over full cycles of the spindle rotation.

6 Conclusion

Regarding the reproducibility results for the different flow rates we can conclude that the syringe pump NEXUS 3000 is suited for the inter comparison (task 1.4.2) using a flow generator.

It is worth to mention that the pulsations measured are not negligible and that the measurement uncertainty applied is determined for steady flow and not for pulsating flow. However, we can assume that averaging over full cycles of the spindle rotation of the syringe pump will averaging out any noise effect due to the oversized fit window or the stochastic noise from the fit using a short fit window of 10 s. This will be more investigated when defining the measurement uncertainty for pulsating flow.

Additionally, we will test commercially available drug delivery devices with plastic syringes (WP 3) which are used in clinical treatment and do not have better specifications in accuracy and reproducibility. It is not necessary to perform the inter comparison with a high accurate flow generator as this would be exaggerated compared to the technical assessments of the drug delivery devices. The results will already give an indication of strong deviations in the measurements of the different participating laboratories.

The inter comparison by means of a flow meter calibration will be significant for the validation of the claimed uncertainties, not the inter comparison by means of the flow generator calibration.

We propose to perform the measurements at the following syringe plunger positions and number of full cycles:

Q (μl/min)	number of cycles	Syringe volume (μl)	Start position of syringe plunger (μl)	Volume to be dispensed (μl)	Relevant syringe plunger positions	Measurement time (min)
2	2	2’500	800	300	From 700 μl to 575 μl	150
10	4	2’500	800	600	From 600 μl to 350 μl	60
33	4	5’000	2000	1500	From 1400 μl to 900 μl	45
100	3	25’000	8000	3000	From 7300 μl to 5400 μl	30
333	3	25’000	8000	3000	From 7300 μl to 5400 μl	10

Table 12: Measurement proposition: number of full cycles and syringe plunger positions.