



Rev 01/22/21

This page intentionally left blank

## Table of Contents

Table of Contents.....	5
1. Introduction .....	7
2. Warranty .....	7
3. Contact Information.....	7
4. Instrument Description .....	8
5. Safety Precautions.....	9
6. Instrument Installation.....	10
6.1 Site Requirements and Operating Environment.....	10
6.2 Unpack and place the instrument where it will be used. ....	10
6.3 Unpack the attachments and accessories. ....	10
6.4 Wipe down components.....	12
6.5 Make Connections .....	12
6.6 Power On .....	14
6.7 Setting Nitrogen Pressures .....	15
6.8 Setting Nitrogen Flow Rate .....	18
6.9 Prime Reservoir and Pump Lines .....	19
7. HMI Navigation .....	24
7.1 Home Screen.....	24
7.2 Methods.....	25
7.3 Diagnostics.....	26
7.4 Records and Updates .....	28
7.5 Training and Maintenance .....	29
7.6 ANKOM Products .....	29
7.7 Who we are.....	29
8. Starting an Assay .....	30
8.1 Confirm Nitrogen Pressure .....	30
8.2 Fill the Solution Reservoir Bottles.....	30
8.3 Assemble Digestion Vessels.....	31
8.4 Load Method.....	32
8.5 Weigh Sample .....	33
8.6 Install Digestion Vessels.....	34
8.7 Add Internal Standard.....	34
8.8 Manually Close Digestion Oven Door .....	35
8.9 Install SPE Columns .....	35
8.10 Install Round Bottom Flasks.....	36

---

8.11	Confirm Solution Reservoirs are Full .....	36
8.12	Start Run .....	36
8.13	Track Progress.....	37
8.14	Abort Run.....	37
8.15	End of Run.....	38
9.	Adjust Method .....	39
10.	Custom Method.....	40
11.	Uploading a New Method .....	41
12.	Faults .....	42
13.	Periodic Maintenance.....	43
13.1	Inspect and clean solution reservoir bottles and filters .....	43
13.2	Inspect and clean the spray nozzles in the digestion oven.....	43
13.3	Regularly clean the instrument surfaces .....	43
14.	Troubleshooting & Replacement Parts .....	44
15.	QC and Calibrations .....	45
15.1	Solution Delivery Calibration .....	45
16.	Appendix A – Reagents.....	48
16.1	Best practices for solution preparation: Vitamin & Cholesterol Analysis.....	48
16.2	Best practices for solution preparation: Fat Analysis .....	48

## 1. Introduction

Congratulations on your purchase of the ANKOM<sup>FLEX</sup> Analyte Extractor. We are confident that this product will effectively serve your needs.

The ANKOM<sup>FLEX</sup> Analyte Extractor is designed to simplify fat-soluble vitamin, cholesterol, and crude and hydrolysis fat determinations. Supporting up to four samples at a time, the system will automatically complete saponification, solid phase extraction (SPE) and evaporation in about two to three hours. It reduces technician variation and increases precision. This manual will provide you with details that will help assure accuracy of your results.

ANKOM Technology designs, manufactures, and markets instruments and support products used by analytical laboratories around the world in the food, feed, bio-energy, agricultural, and environmental industries. ANKOM Technology provides you with products for determining or monitoring fat-soluble vitamins, cholesterol, dietary fiber, detergent fiber, crude fiber, fat, digestibility, microbial fermentation (anaerobic or aerobic) and more.

ANKOM Technology is committed to total customer satisfaction, designing every product based on a thorough assessment of customer needs.

**NOTE:** Please review the entire manual before you begin operating this product.

## 2. Warranty

ANKOM Technology warrants the ANKOM<sup>FLEX</sup> Analyte Extractor against any defects in workmanship or material for one year after the original date of purchase. This warranty does not include damage to the instrument resulting from neglect or misuse. During the warranty period, should any failure result from defects in workmanship or materials, ANKOM Technology will, at its discretion, repair or replace the instrument free of charge. Extended warranties are available upon request.

## 3. Contact Information

We are committed to your total satisfaction and are always available to help you get the most from our products value your comments or suggestions.

For any questions or suggestions regarding your instrument, please contact us at:

Telephone: (315) 986-8090

Fax: (315) 986-8091

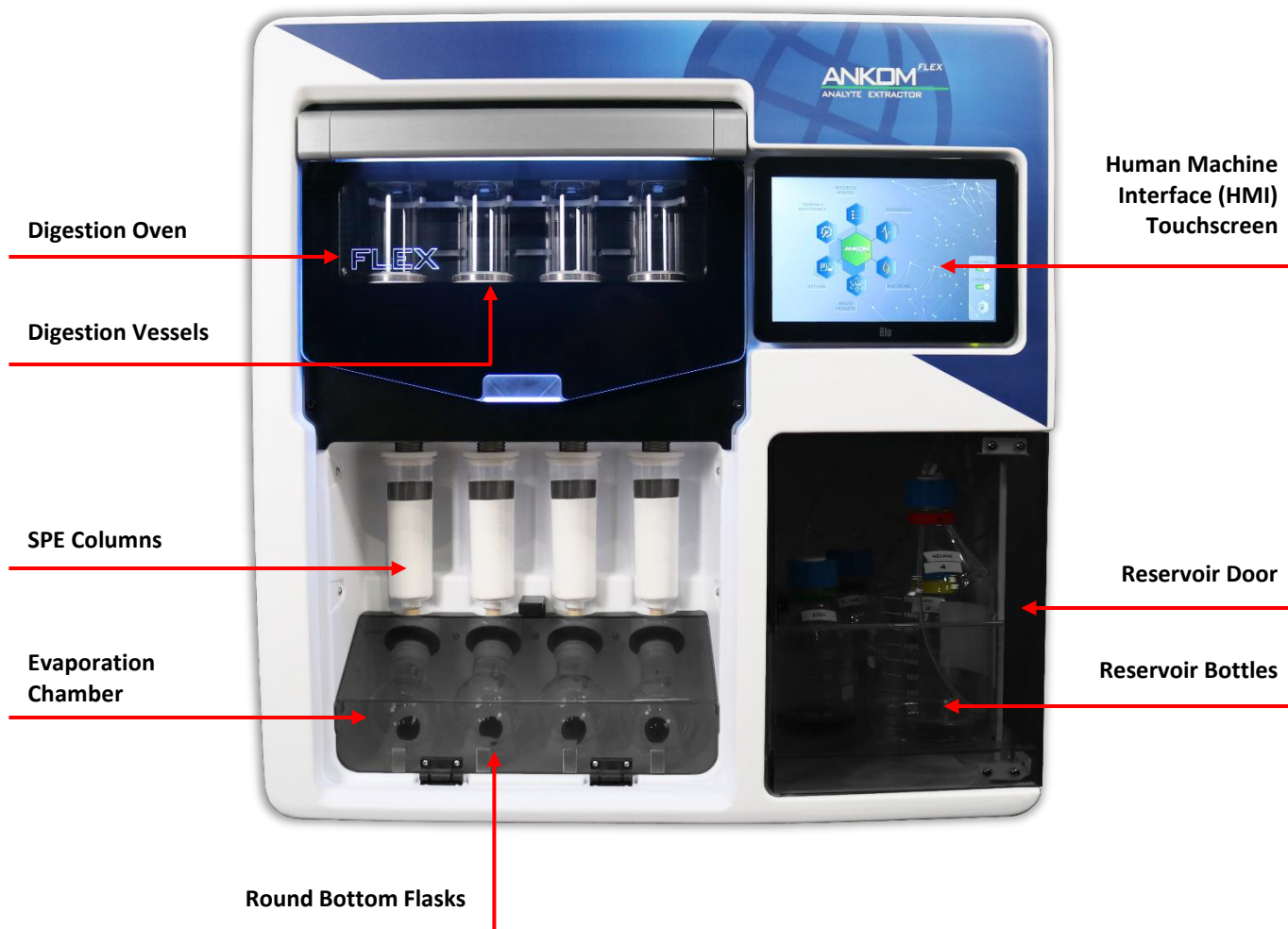
Email: [service@ankom.com](mailto:service@ankom.com)

[www.ankom.com](http://www.ankom.com)


## 4. Instrument Description

The ANKOM<sup>FLEX</sup> Analyte Extractor simplifies fat-soluble vitamin, cholesterol, crude and hydrolysis (total) fat determinations and other possible applications. This fully automated system integrates all the steps involved in these analyses in one instrument. Supporting up to four samples at a time, the system will automatically complete saponification, solid phase extraction and evaporation of the solvent. The system maintains four reservoirs that can be programmed by the user for the solutions required (e.g. ethanol, hexane, KOH, water, etc.).

Supporting standard methods, the FLEX also allows for method development and customization. Digestion vessels have been designed to allow for sample to be weighed directly into the vessels, eliminating transfer error. Filtration takes place without user intervention, further reducing technician labor. A nitrogen atmosphere and positive flow, low temperature evaporation protects sensitive analytes against oxidation and optimizes solvent evaporation. Exhaust lines are ready to be connected to an exhaust system, eliminating under-hood instrument positioning. Once an assay is complete, the isolated analyte is ready for reconstitution and quantitation by HPLC or GC. If the instrument is configured for crude or hydrolysis fat extraction, the isolated fat is ready for gravimetric or fatty acid profile analysis. Sample size can vary up to 10 grams depending on fat and moisture content. Fat-soluble vitamin samples that require larger volumes can be saponified outside of the instrument and aliquots can be introduced for extraction and evaporation.



## 5. Safety Precautions

	<p><b>Hot Surfaces</b> – Do NOT open the Digestion Oven during operation. The interior surfaces can reach 100°C (212°F). <b>Failure to observe this caution may result in burns.</b></p> <p><b>Hazardous Voltages</b> – Do NOT operate the instrument with the back of the Main Electrical Cabinet removed. Hazardous voltages are present during operation. <b>Failure to observe this caution may result in electrical shock or electrocution.</b></p> <p><b>Hazardous Materials</b> – Follow SDS warnings and recommendations for all materials to be used in this instrument, for example: Ethanol, Hexane, and SPE sorbent. Follow both local and federal regulations for vent hood requirements when operating this instrument. <b>Failure to observe this caution may be hazardous to your health.</b></p> <p><b>WARNING:</b> Attempts to override safety features or to use this instrument in a manner not specified by ANKOM Technology voids the warranty and may result in serious injury or even death. This system is designed to meet and/or exceed the applicable standards of CE and CSA.</p>
---	--

**IMPORTANT:** The Power Switch must be in the OFF position before plugging the instrument Power Cord into the power source.

## 6. Instrument Installation

### 6.1 Site Requirements and Operating Environment

To install and operate the ANKOM<sup>FLEX</sup> Analyte Extractor you will need the following:





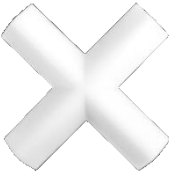

- Adequate Power: 100V–120V ~ 50/60Hz 15A  
220V–240V ~ 50/60Hz 8A
- Must have ability to connect to an exhaust system
- Bench space that can accommodate 77.5 cm (30.5") W x 81.3 cm (32") H x 45.7 cm (18") D and a weight of 54.4 kg (120 lbs.)
- 5.5–6.9 bar (80–100 psi) Nitrogen Supply and connection tubing
- Ambient Temperature Range: 19°–30°C (66°–86°F)

### 6.2 Unpack and place the instrument where it will be used.




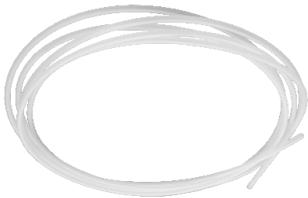
The instrument shipping container consists of three separate cardboard pieces: a top, a bottom, and a sleeve that forms the body of the container. Remove the top and the sleeve of the instrument shipping container. Lift the instrument from the shipping container bottom and place it on a surface that is firm and level. The instrument needs to be near a nitrogen supply and within range to connect exhaust lines to a ventilation hood or exhaust system. The instrument must not be subjected to excessive shock, vibration, dirt, moisture, oil, or other fluids.

### 6.3 Unpack the attachments and accessories.

Within the shipping container is a cardboard box containing attachments, accessories, and a tool kit. Open the box and verify that the following items are present. The tool kit will have a separate packing list included.




<p>Digestion Vessel with port (4 Qty) part # FLEX55</p> 	<p>Round Bottom Flask (4 Qty) part # 9364</p> 	<p>100 ml Plastic Calibration Cup (4 Qty) part # 9483</p> 
<p>Vessel Bottom Assembly (4 Qty) part # FLEX54</p> 	<p>Magnetic Cross Stir-bar (4 Qty) part # 9415</p> 	<p>Black Nitrogen Supply Tubing (1 Qty) part # 8216</p> 



<p>500 ml Solution Reservoir Bottle (3 Qty) part # 5605</p> 	<p>2000 ml Solution Reservoir Bottle (1 Qty) part # 9365</p> 	<p>Waste Bottle Assembly (1 Qty) part # FLEX49</p>  <p>Including 10 ft Exhaust Tubes (2 Qty)</p> 
---	--	---

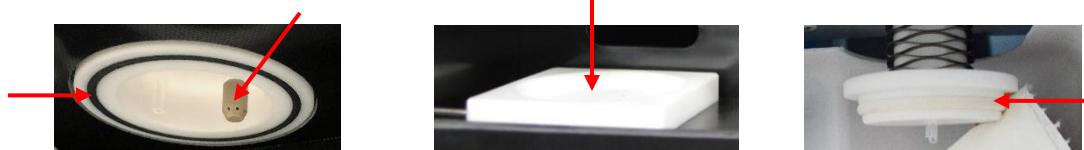
**FLEX Support Items**

The following support items are needed to perform an assay and may be purchased separately.

<p>Prepacked SPE columns part # FLEX-SPE</p> 	<p>Vitamin Filters part # FLEX-VF</p> 
<p>Sorbent Disposal Bags (optional) part # FLEX-SDB</p> 	

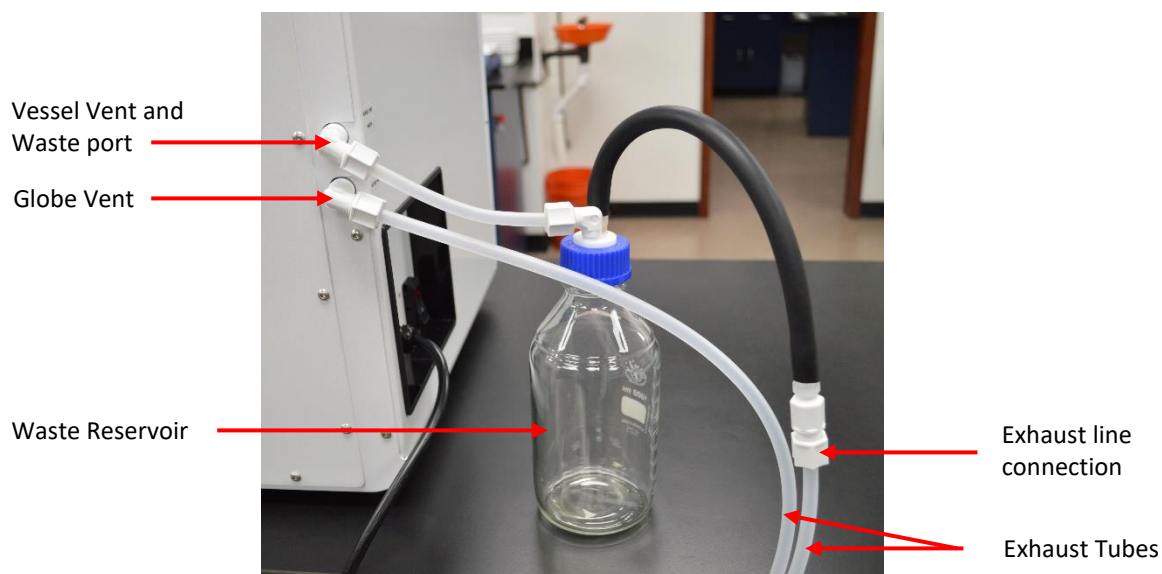
## 6.4 Wipe down components

6.4.1. Wipe the digestion vessel top's spray nozzles and O-rings, vessel bases, and the SPE column top's O-rings with a towel wetted with water. Refer to FLEX Service Procedure FLS011.



## 6.5 Make Connections

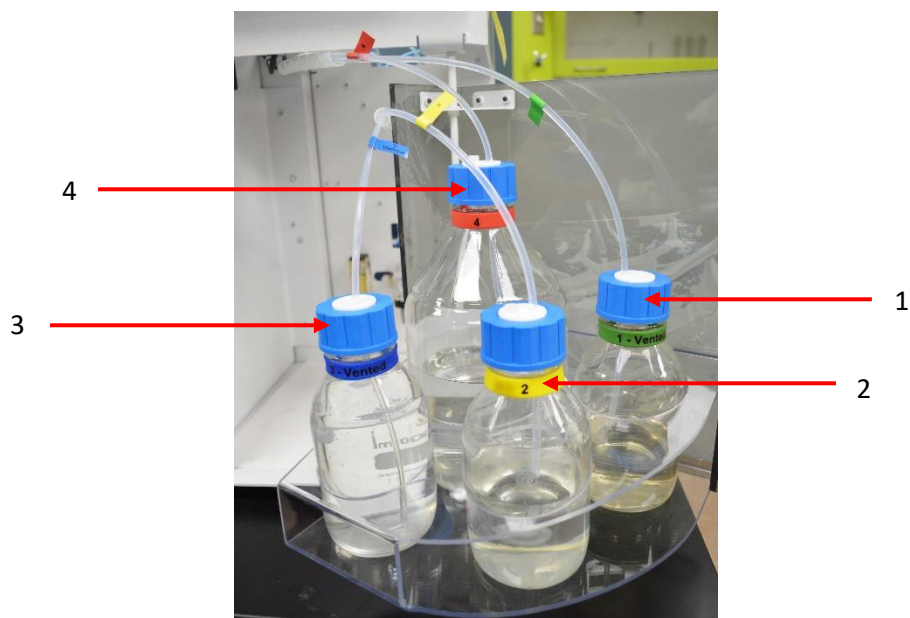
6.5.1. Connect waste assembly - Connect the shorter opaque tube to the vessel vent and waste port on the back of the instrument and the longer opaque tube to the globe vent port. Connect the exhaust line to the waste assembly exhaust line connection. Exhaust lines can be ventilated to an exhaust system. Screw on the cap to the waste reservoir. Refer to FLEX Service Procedure FLS001.



6.5.2. Connect nitrogen supply (N<sub>2</sub> High Pressure) - Attach the black nitrogen supply tubing (part # 8216) to a High Pressure (>50psi) Nitrogen source and then attach to the instrument by pushing the black nitrogen supply tubing into the Nitrogen Supply port on the back of the instrument. Refer to FLEX Service Procedure FLS002.



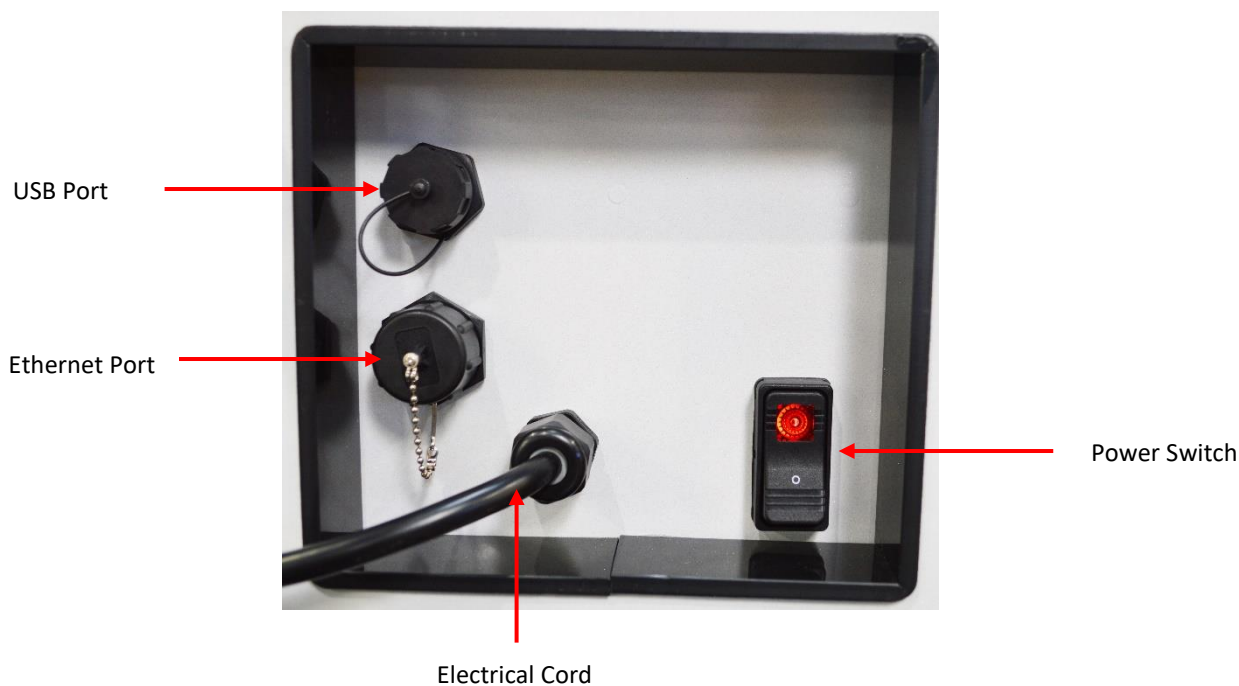
6.5.3. Press the reservoir door to open the door. Install the solution reservoir bottles into the correct numbered positions. Match the colors and numbers of the reservoir bottles to each reservoir cap and tube.



## 6.6 Power On

6.6.1. Plug the electrical cord into an appropriate power source.

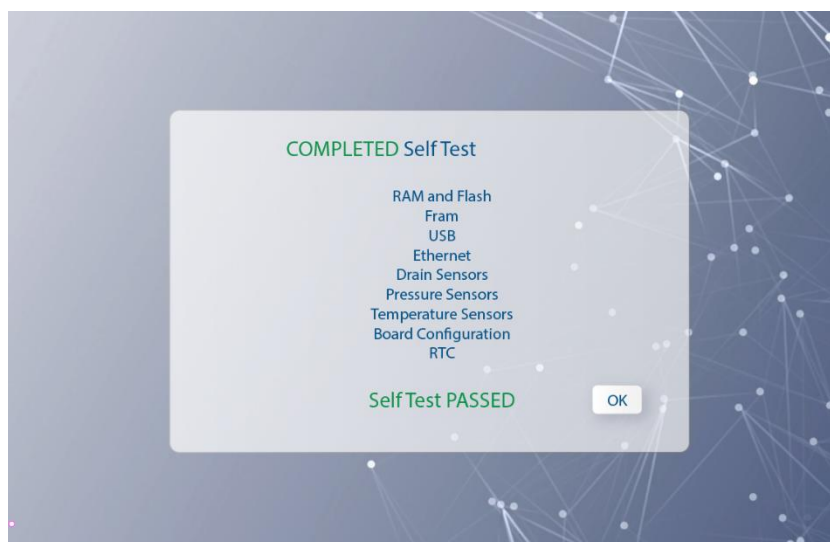
6.6.2. To turn on the FLEX, flip the power switch that is located on the side of the instrument. The red light indicates that it is on.



The FLEX will take about three minutes to power on and run a self-test. The self-test will conclude, and the following screen will be displayed.

**If the pressure sensors do not pass, press "OK" and proceed to the following section to adjust the Nitrogen pressure regulators.**

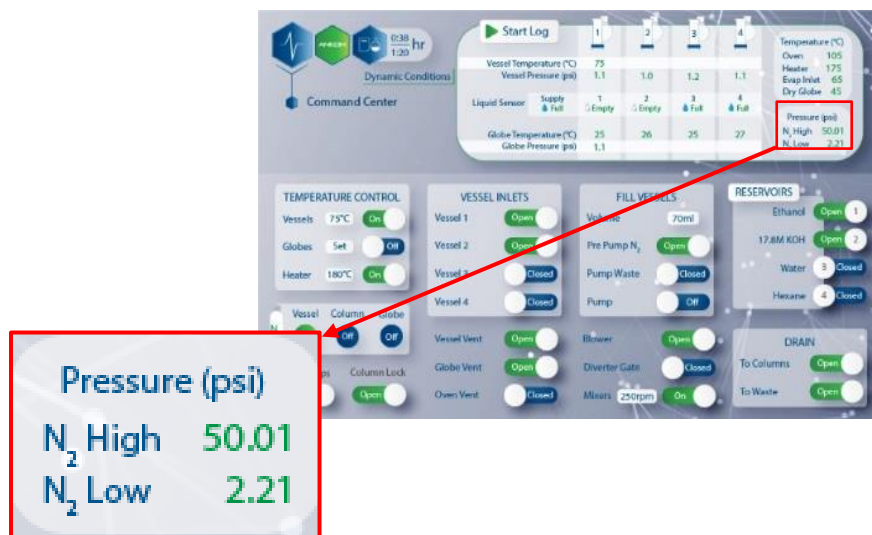
If any other component does not pass the self-test, contact ANKOM.



## 6.7 Setting Nitrogen Pressures

To confirm that the nitrogen levels are set correctly, press the “DIAGNOSTICS” section, and then go into the “COMMAND CENTER.” The N<sub>2</sub> high pressure should be between 50-51 psi and the N<sub>2</sub> low pressure should be between 2.2-2.5 psi.

If the N<sub>2</sub> high pressure does not fall within the acceptable range, follow **Adjusting N<sub>2</sub> High Pressure Regulator** steps. If the N<sub>2</sub> low pressure does not fall within the acceptable range, follow **Adjusting N<sub>2</sub> Low Pressure Regulator** steps.



### 6.7.1. Adjusting N<sub>2</sub> High Pressure Regulator

To increase:

- Pull down the gray dial on the pressure regulator to unlock.
- Rotate dial counterclockwise (to the right) until the pressure increases within the desired range on the HMI touchscreen.
- Push up to lock dial in place.



To decrease:

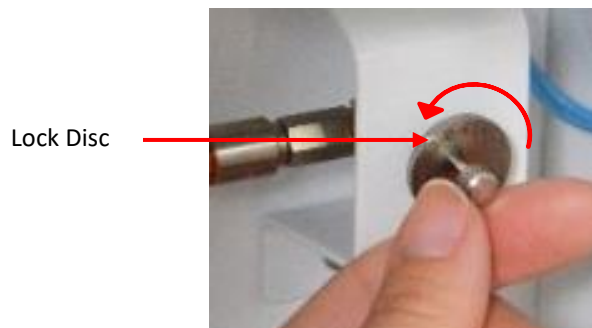
- Pull down the dial on the pressure regulator to unlock.
- Rotate dial clockwise (to the left) until the pressure decreases within the desired range on the HMI touchscreen.
- Push up to lock dial in place.



6.7.2. Adjusting N<sub>2</sub> Low Pressure Regulator

To increase:

- a. Rotate the lock disc counterclockwise (to the left) to unlock.



- b. Rotate the dial clockwise (to the right) until the pressure increases within the desired range on the HMI touchscreen.



- c. In the "Command Center", open the "Pump Waste."
- d. Turn vessel nitrogen "ON."
- e. Wait ~2 seconds.
- f. Turn Vessel N<sub>2</sub> "Off."
- g. Close the "Pump Waste."
- h. Rotate the lock disc clockwise (to the right) to lock.
- i. Confirm that the low pressure is now within range.



If N<sub>2</sub> Low pressure is still below range (2.2-2.5 psi), repeat the steps to increase.

To decrease:

- a. Rotate the lock disc counterclockwise (to the left) to unlock. See lock disc image above.
- b. Rotate the dial one quarter turn counterclockwise (to the left).

**NOTE:** The user will not see a change in pressure on the HMI during this step.



- c. In the "Command Center", open the "Pump Waste."
- d. Turn vessel nitrogen "ON."
- e. Wait ~2 seconds.
- f. Turn Vessel N<sub>2</sub> "Off."
- g. Close the "Pump Waste."
- h. Rotate the lock disc clockwise (to the right) to lock.
- i. Confirm that the low pressure is now within range.



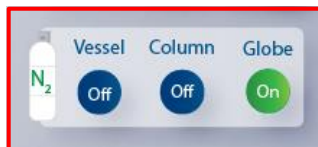
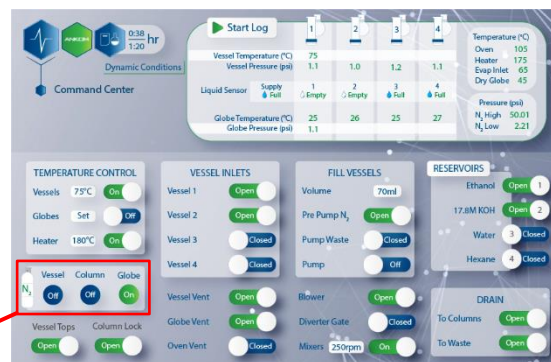
If N<sub>2</sub> Low pressure is still above the range (2.2-2.5 psi), repeat steps to decrease.

## 6.8 Setting Nitrogen Flow Rate

The flow meter should only be adjusted during the initial instrument installation and does not need to be regularly adjusted. Follow the steps below to check and adjust the flow meter.

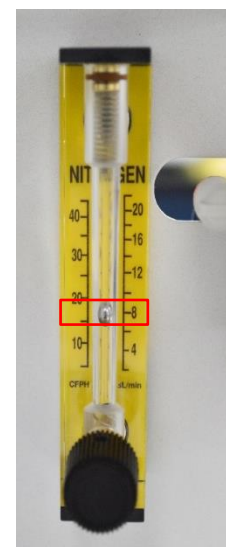
**IMPORTANT:** Make sure round bottom flasks are **NOT** installed in the evaporation chamber when checking the flow meter.

- 6.8.1. From the Home screen, press “DIAGNOSTICS”, then press “COMMAND CENTER.”
- 6.8.2. Confirm that the N<sub>2</sub> High pressure value is within the range of 50-51 psi. If it is outside of this range, refer to “Setting Nitrogen Pressures.”
- 6.8.3. Open the reservoir door on the instrument.
- 6.8.4. Turn N<sub>2</sub> Globe “On.”



- 6.8.5. The float (ball) of the flow meter, located inside of the reservoir area, should be between 7.5-8.5 sL/min.
- 6.8.6. If the float falls outside of this range, adjust the flow meter by turning the control knob counterclockwise (to the left) to increase flow or clockwise (to the right) to decrease flow. This may take several rotations.
- 6.8.7. Once the float has been adjusted to be within the acceptable range, turn Globe N<sub>2</sub> “Off.”

**NOTE:** This step is only necessary outside of installation if the user observes poor performance of evaporation in the round bottom flasks.





## 6.9 Prime Reservoir and Pump Lines

Reservoir and pump lines must be primed **(i) BEFORE the instrument is first used**, and **(ii) when the instrument has been out of use for more than two weeks**. Wear gloves and safety glasses when running the Line Prime method.

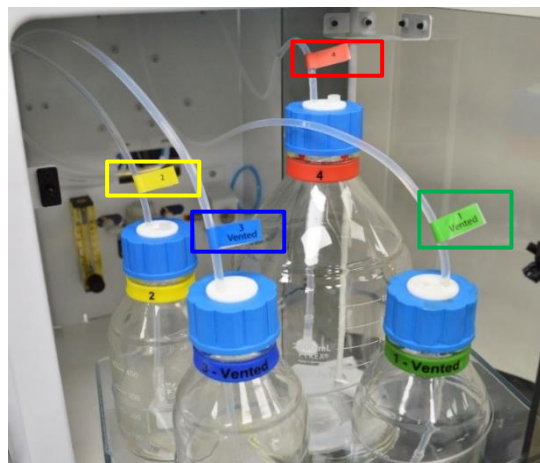
To prime all lines, complete the following steps.

### 6.9.1. Fill the Solution Reservoir Bottles

Fill each solution reservoir with the appropriate reagents. Reagents must match the reagents of the method that the user intends to run subsequently. Refer to Appendix A - Reagents.

You must use the correct reservoir position for each solution as some solvents must not be vented.

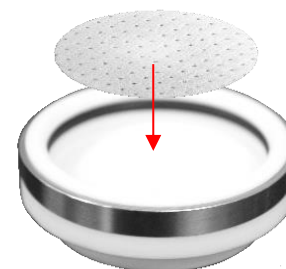
For further details on how to fill the reservoir bottles, refer to FLEX Service Procedure FLS005, found on the ANKOM website.



### 6.9.2. Assemble Digestion Vessels

The following parts are needed when running the line prime method: Digestion vessel (with or without port), filter, and vessel bottom assembly.

6.9.2.1. Select the appropriate filter for the analysis. The filter must match the filter of the method that the user intends to run subsequently. The vitamin filter is used for vitamin and cholesterol methods, and the fat filter is used for a total and crude fat methods. Place the filter (either side up) in the vessel bottom assembly. Make sure that it is pressed all the way down.



6.9.2.2. Wet the bottom outside edge of the digestion vessel with water. Do not use any other solution, as this will negatively impact the seal.



6.9.2.3. Insert the digestion vessel into the vessel bottom assembly by a slight twist motion.

**IMPORTANT:** Make sure the digestion vessel seats evenly and tightly, with the glass all the way down in the vessel bottom assembly, otherwise the digestion solution will leak.



**6.9.3. Load Method: Line Prime**

From the Home Screen:

6.9.3.1. Press "METHODS."

6.9.3.2. Select the "MISCELLANEOUS" category.

6.9.3.3. Select the "Line Prime" method.

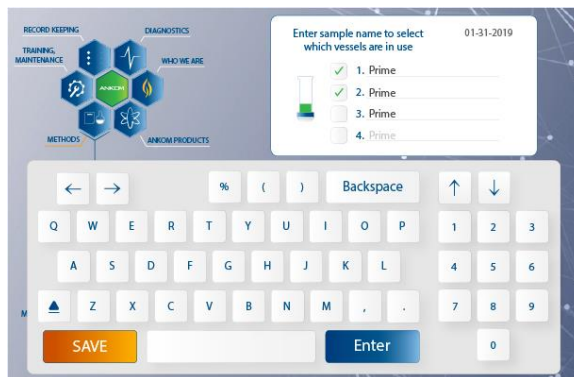
6.9.3.4. The screen will display the steps required to start a run. Press the first step, "1. Enter sample name."



6.9.3.5. Enter "Prime" as the sample name for all four positions.

6.9.3.6. Press "SAVE."

**NOTE:** If "SAVE" is pressed before all sample names were entered go back to the Home Screen and repeat the Load Method steps.



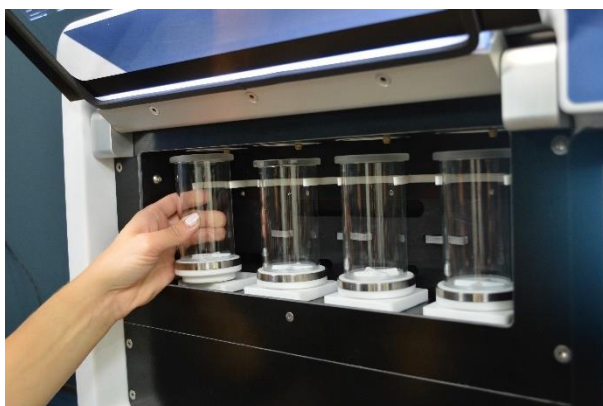
#### 6.9.4. Install Digestion Vessels

6.9.4.1. Open the digestion oven door by pulling it open.

6.9.4.2. Make sure the vessel tops are open. If they are not open, press the "Vessel Top" toggle so that they are in the open position.



6.9.4.3. Install the digestion vessels into the instrument by sliding them into the guide and setting them into the white vessel bases. If you are using digestion vessels with ports, make sure the ports are facing forward.



6.9.4.4. Close the vessel tops by pressing the "Vessel Tops" toggle. You will see the vessel tops descend and seal the digestion vessels.

6.9.4.5. Once all four digestion vessels have been installed, press "YES."

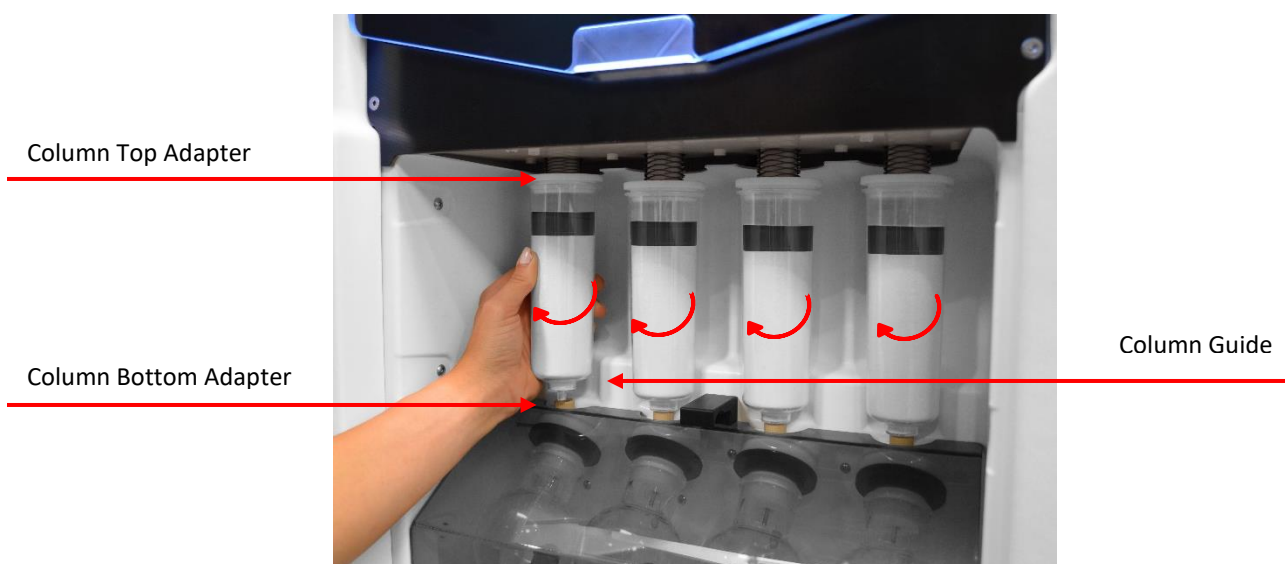
### 6.9.5. Manually Close Digestion Oven Door

### 6.9.6. Install SPE Columns

6.9.6.1. Be sure to remove all shipping packaging from inside the columns.

6.9.6.2. Wipe the inside of the column top with a wetted paper towel (water) to remove sorbent dust before installing each column onto the instrument.

6.9.6.3. Install SPE columns by placing the top of the columns over the column top adapters on the instrument. Push the column upwards and guide the bottom of the column all the way to the back until it hits the guide and set the column in the column bottom adapter on the instrument. **Twist each column a quarter turn clockwise (to the left).**



6.9.6.4. Once all SPE columns are installed, press "YES."

### 6.9.7. Install Round Bottom Flasks

6.9.7.1. Open the evaporation chamber door manually.

6.9.7.2. Turn the large black release ring at each position clockwise (to the right), until it fully tightens.

6.9.7.3. Install glass round bottom flasks over the round bottom flask adapters in the evaporation chamber by pushing each until it seals tightly.



Round Bottom Flask Adapter

Release Ring

6.9.7.4. Close the evaporation chamber door manually.

6.9.7.5. Press "YES," confirming that all four round bottom flasks are installed properly.

### 6.9.8. Confirm Solution Reservoirs are Full

6.9.8.1. Confirm that each solution reservoir is filled with the appropriate chemicals. (Refer to Appendix A - Reagents)

6.9.8.2. Ensure that each reservoir cap is closed tightly.

6.9.8.3. Once all the reservoirs are full, press "YES."

### 6.9.9. Start Line Prime

When set-up steps have been completed, press "START RUN."

The Line Prime method will take 7-10 minutes to complete.



### 6.9.10. End of Line Prime

6.9.10.1. Turn the large black release ring counterclockwise (to the left), to release the round bottom flasks.

6.9.10.2. Remove the round bottom flasks. They will remain clean during the line prime method and can be used for the next assay.

6.9.10.3. Remove and clean digestion vessels for subsequent use. Refer to FLEX Service Procedure FLS011.

**NOTE:** It is very important to thoroughly clean vessel bottom assemblies after use. Use water, and NOT ethanol or acetone, as final rinse solution. The digestion vessels must not be left assembled for extended periods of time (e.g. overnight). If digestion vessels are left assembled for extended periods of time, the O-rings in the vessel bottom assemblies will deform and future integrity could be compromised.

6.9.10.4. Remove the SPE columns. They will remain clean during the line prime method and can be used for future assays.

6.9.10.5. Wipe the digestion vessel tops spray nozzles and O-rings, and the vessel bases with a towel wetted with water.



6.9.10.6. Dispose of waste bottle reservoir materials according to SDS guidelines and internal SOP. This reservoir will contain a mixture of all the solutions that has been used in an assay, which may include organic solvents, strong bases, and acids.

## 7. HMI Navigation

The ANKOM<sup>FLEX</sup> Analyte Extractor comes equipped with revolutionary software that allows the user unlimited options for customization and method creation. The HMI has been created with the user in mind, offering preprogrammed methods, custom method capability, real-time progress tracking, downloadable assay reports, diagnostic center access, and information for troubleshooting and maintenance.

### 7.1 Home Screen

The home screen allows the user to see all the sections available for navigation. The user can enter a section by pressing one of the blue hexagons. To return to the home screen from any screen the user can press the green "ANKOM hexagon". The bar on the right side of the home screen allows the user to open the toggle "Vessel Tops" to gain access to the digestion vessels or to open the toggle "Column Lock" for access to the SPE columns. The "Lock Hexagon" located in the lower right corner of the screen allows the user to safely shut down the instrument.

Sections on the home screen include:

- Methods
- Diagnostics
- Records & Updates
- Training & Maintenance
- ANKOM Products
- Who We Are



## 7.2 Methods

The method section allows the user to select preprogrammed methods or create custom methods.



Faded hexagons represent unavailable methods.

Method availability depends on the FLEX package purchased. Method options include:

### Vitamin A, E, D

This folder contains preprogrammed vitamin methods that the user can run. For example, method “VitAED 75C 45m” is based on a combination of official vitamin methods such as: EN-12823-1, EN-12822, and AOAC 2011.07.

### Total Fat

This folder contains preprogrammed fat methods that the user can run. Methods include Total Fat (Hydrolysis) and Crude Fat options.

### Miscellaneous

This folder contains other preprogrammed methods that the user can run, such as cholesterol and Line Prime method.

### Custom

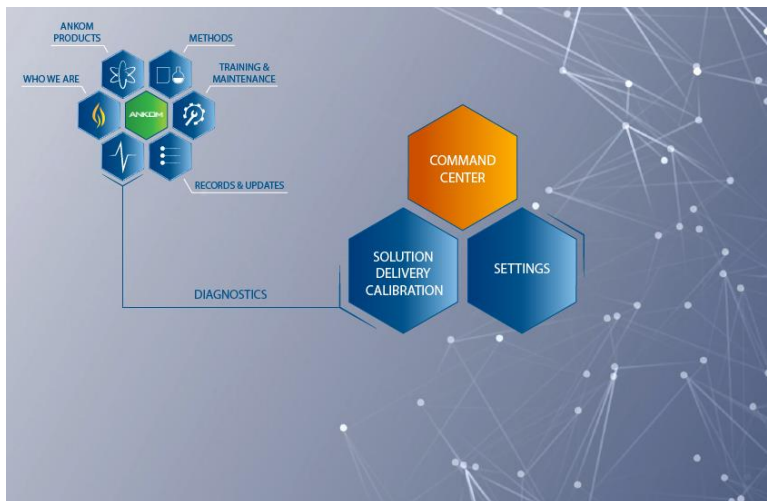
This option gives the user the ability to create custom methods. Custom methods are created by building sequences of modular functions, such as digestion time/temp, solution delivery, mixing, evaporation etc. This section becomes a valuable tool for method development and research.

### Last Run

This option allows the user to select the method that was last executed. This gives easy access to methods that are run on a regular basis.

### 7.3 Diagnostics

The diagnostics section displays the solution delivery calibration, command center, and settings options.



#### Solution Delivery Calibration

This calibration should be done when a new chemical is added or if the user would like to confirm that solution delivery is accurate. Refer to section QC & Calibrations section of the manual.

#### Command Center

This screen contains two sets of information. The top of the screen shows the dynamic conditions of the instrument: real-time temperature and pressures. The lower half of the screen contains toggles that control the functionality of the instrument. These can be toggled for diagnostic purposes.

	1	2	3	4
Vessel Temperature (°C)	75			
Vessel Pressure (psi)	1.1	1.0	1.2	1.1
Liquid Sensor	Supply Full	Empty	Empty	Full
Globe Temperature (°C)	25	26	25	27
Globe Pressure (psi)	1.1			

Component	Temperature	Status
Vessels	75°C	On
Globes	Set	Off
Heater	180°C	On

Component	Status
Volume	70ml
Pre Pump N <sub>2</sub>	Open
Pump Waste	Closed
Pump	Off
Blower	Open
Diverter Gate	Closed
Mixers	250rpm On

Reservoir	Status
Ethanol	Open 1
17.8M KOH	Open 2
Water	3 Closed
Hexane	4 Closed



## Settings

This option contains basic information, such as: serial number, software version, wi-fi networks, system configuration, and factory settings.

### Information

This section displays the instrument serial number and current software version.

### Wi-Fi

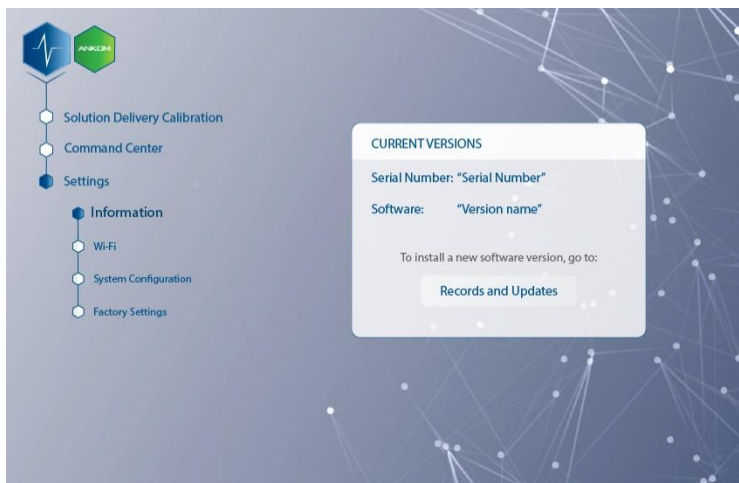
This capability will be functional in future upgrades.

### System Configurations

This section contains settings such as: data logging frequency, date/time, diagnostic functionality, expert mode, language options, and module and fault configurations. To make any system configuration changes, contact ANKOM for the Administrator Password.

### Factory Settings

This section is password protected for ANKOM's use only.



## 7.4 Records and Updates

The records and updates section stores reports and raw data logs and allows the user to install new software updates.



### Records

This section stores various reports and logs. Reports are short summaries that are viewable on the screen and exportable via USB flash drive. Logs are raw data files that can be exported and viewed in excel or another spreadsheet program.

**NOTE:** To export any files from the FLEX, the USB flash drive must contain a folder labeled: ExportFiles

#### Assay Reports & Logs

This section contains reports and logs of all assays done on the instrument.

#### Calibration Reports

This section contains reports of all solution delivery calibrations done on the instrument.

#### Diagnostic Logs

This section contains logs that were generated through the Command Center in the Diagnostics section. Refer to the HMI Navigation – Diagnostics section.

#### Fault Reports

This section contains reports of all faults that occurred during assays.

#### Global Logs

This section contains daily logs of information such as global faults, assay start/stop times, changes in configuration settings, service events, calibration events, when updates were installed, etc.

#### Service Reports

This section contains reports of all services done on the instrument. This section is populated by the user or service technician.

## Updates

This section allows the user to update software and methods. Refer to the "Uploading a New Method" section in the manual.

**NOTE:** After a software update, module and fault configuration settings will be reset to factory default settings. Although these settings have been optimized for all instruments, in rare cases ANKOM will recommend adjusting certain configuration settings that must then be reapplied after a software update. Contact ANKOM for instructions.

## 7.5 Training and Maintenance

This section provides materials and information supporting training and maintenance. Materials can include video tutorials, service procedures, analytical procedures, and SDS documents. These files will be continually updated with each new software version.

If the information needed by the user is not available in this section, refer to the ANKOM website: [www.ankom.com](http://www.ankom.com).



## 7.6 ANKOM Products

This section provides information about ANKOM Technology's products. Instruments include:

- ANKOM<sup>TDF</sup> Dietary Fiber Analyzer
- ANKOM<sup>DELTA</sup> Automated Detergent and Crude Fiber Analyzer
- ANKOM<sup>XT15</sup> Crude Fat Extractor
- Daisy<sup>II</sup> Incubator- In vitro Incubation
- ANKOM<sup>RF</sup> Gas Production System – Gas/Fermentation Measurement
- ANKOM<sup>HCl</sup> Hydrolysis System



## 7.7 Who we are

This section provides a look into ANKOM Technology as a company. Learn more about our American story, our foundation, and our corporate values.



## 8. Starting an Assay

### IMPORTANT:

If the instrument has not been used in two weeks or more, wipe the digestion vessel top's spray nozzles and O-rings, vessel bases, and the SPE column top's O-rings with a towel wetted with water. Refer to FLEX Service Procedure FLS011.

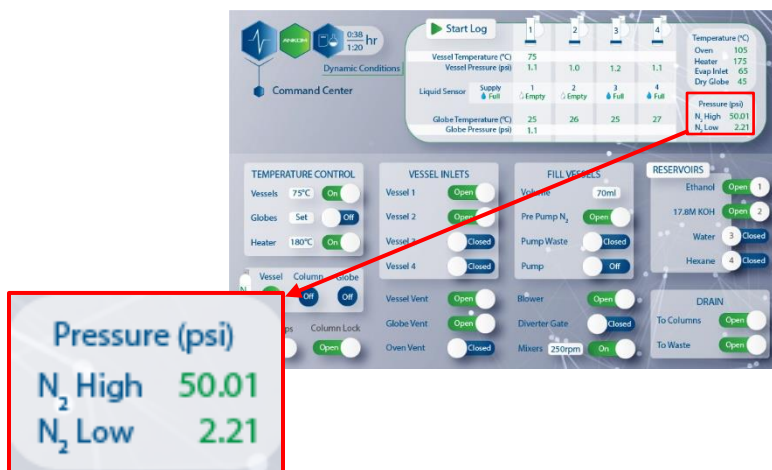
### 8.1 Confirm Nitrogen Pressure

Before starting any assay, make sure that the N<sub>2</sub> High and N<sub>2</sub> Low pressure sensors read within operational range.

From the Home screen, press "DIAGNOSTICS", then press "COMMAND CENTER." Values should be within the following ranges:

N<sub>2</sub> High pressure range: 50-51 psi

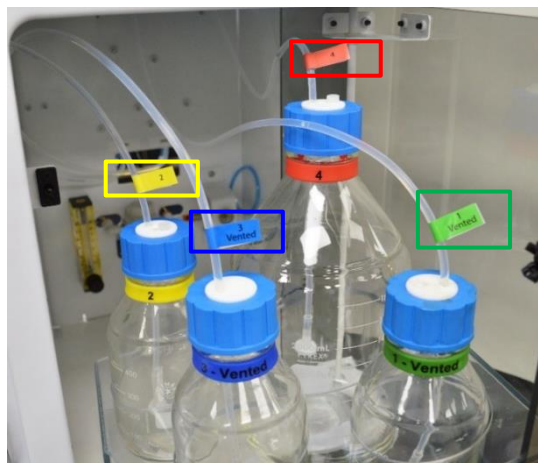
N<sub>2</sub> Low pressure range: 2.2-2.5 psi



**NOTE:** If the pressures do not fall within the specified range, refer to section "Setting Nitrogen Pressures."

### 8.2 Fill the Solution Reservoir Bottles

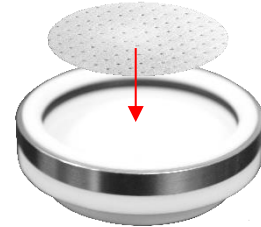
Fill each solution reservoir with the appropriate reagents, refer to Appendix A - Reagents. Be sure to use the correct reservoir position for each solution as certain solutions must be vented or unvented. This is automatically set according to the method the user has selected. For further details on how to fill the reservoir bottles, refer to FLEX Service Procedure FLS005 on the ANKOM website.



### 8.3 Assemble Digestion Vessels

Before user assembles the digestion vessels, make sure that each element is clean and has been wiped down. The following parts make up one digestion vessel: Digestion vessel (with or without port), vessel bottom assembly, filter (specific to fat, or vitamin and cholesterol analysis), and magnetic cross stir-bar. One complete digestion vessel is needed per assay.

- 8.3.1. Select the appropriate filter for the analysis. Place the filter (either side up) in the vessel bottom assembly. Make sure that it is pressed all the way down.



**NOTE:** Make sure the vitamin filter is used for vitamin and cholesterol methods, and the fat filter is used for a total and crude fat methods. Using the incorrect filter will result in plugged lines.

- 8.3.2. Wet the bottom outside edge of the digestion vessel with water. Do not use any other solution, as this will negatively impact the seal.



- 8.3.3. Insert the digestion vessel into the vessel bottom assembly by a slight twist motion.

**IMPORTANT:** Make sure the digestion vessel seats evenly and tightly, with the glass all the way down in the vessel bottom assembly, otherwise the digestion solution will leak.



- 8.3.4. Place the X-shaped magnetic stir bar into the assembled digestion vessel.



### 8.4 Load Method

From the Home Screen:

- 8.4.1. Press "METHODS."
- 8.4.2. Select the desired method category.
- 8.4.3. Select the method of choice. Refer to the ANKOM website for the standard operating procedures of each method.



#### Analysis Options using the ANKOM<sup>FLEX</sup> Analyte Extractor

Method Name	Method Description
<b>VitAED 75C 45m</b>	This method includes ethanolic saponification at 75°C for 45min, extraction through SPE (solvent: n-hexane), and evaporation of the solvent at low temperature. The analyte can then be reconstituted for chromatographic quantitation.
<b>TotFat 3N HCl</b>	This method includes acid hydrolysis of the sample, filtration of the aqueous portion to waste, extraction of the hydrolysate with solvent, filtration with SPE, and evaporation of the solvent. The isolated fat can then be gravimetrically quantified.
<b>CrudeFat</b>	This method includes extraction of the sample with solvent, filtration of the solvent extract through SPE, and evaporation of the solvent. The isolated fat can then be gravimetrically quantified.
<b>Cholesterol</b>	This method includes ethanolic saponification at 75°C for 45min, extraction through SPE (solvent: n-hexane), and evaporation of the solvent at low temperature. The analyte can then be reconstituted for chromatographic quantitation.

**NOTE:** The base methods can be adjusted slightly to change digestion parameters, conserve solvent, etc. Please contact ANKOM for more details.

- 8.4.4. The screen will display the steps required to start a run. Press the first step, "1. Enter sample name."



8.4.5. Enter sample names, starting with line one. The user can enter up to four sample names. Do not skip positions, always start with position one, line one and continue in order.

A vessel position will be run only when a sample name is entered into the corresponding line. If the user wants to run position 1 and 2, then sample names are inputted only in line 1 and 2.



8.4.6. Press "SAVE."

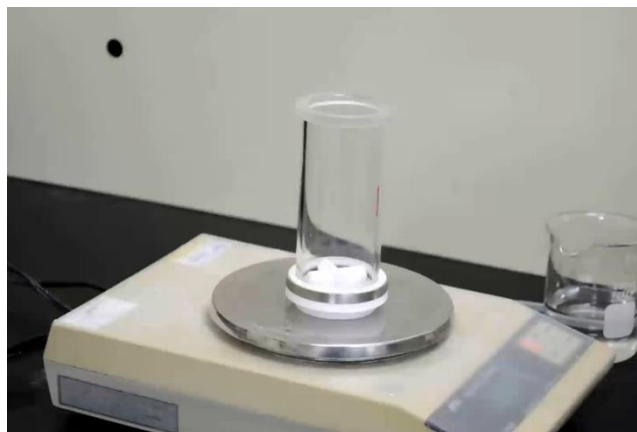
**NOTE:** If "SAVE" is pressed before all sample names were entered go back to the Home Screen and repeat the Load Method steps.

## 8.5 Weigh Sample

8.5.1. Weigh sample into digestion vessels: For more details on sample size per method, refer to the SOPs of the selected methods on the ANKOM website.

If weighing an oil sample, refer to Service Procedure FLS007.

8.5.2. Weigh sample directly into each digestion vessel. It is very important to not exceed the specified sample size. Exceeding the sample size could result in columns overloading.

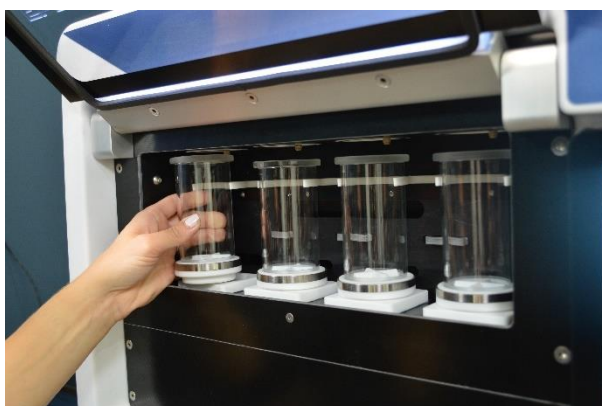


## 8.6 Install Digestion Vessels

- 8.6.1. Open the digestion oven door by pulling it open.
- 8.6.2. Make sure the vessel tops are open. If they are not open, press the "Vessel Top" toggle so that they are in the open position.



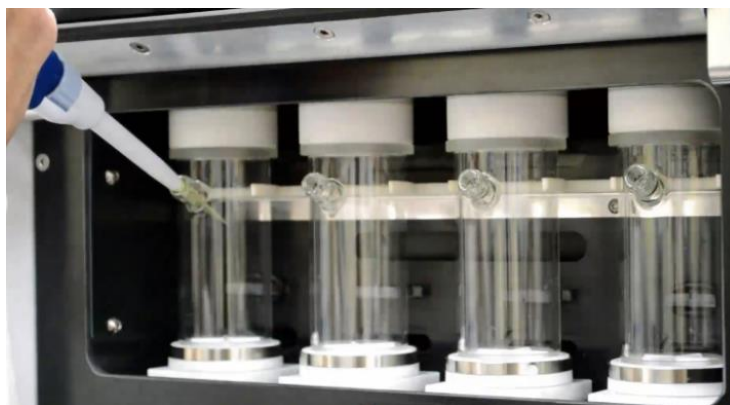
- 8.6.3. Install the digestion vessels into the instrument by sliding them into the guide and setting them into the white vessel bases. If you are using digestion vessels with ports, make sure the ports are facing forward.



- 8.6.4. Close the vessel tops by pressing the "Vessel Tops" toggle. You will see the vessel tops descend and seal the digestion vessels.
- 8.6.5. Once all four digestion vessels have been installed, press "YES."

## 8.7 Add Internal Standard

- 8.7.1. If an internal standard is needed, add the standard into the digestion vessels through the side port of the vessels.
- 8.7.2. Screw the red caps onto the vessel ports. Tighten but do not over tighten.

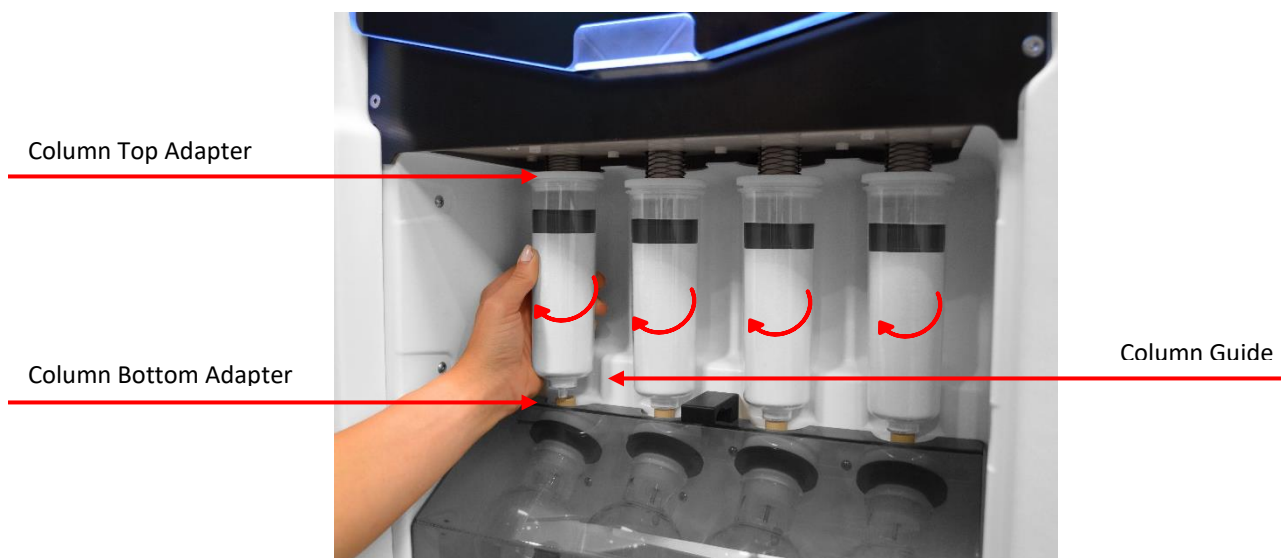




## 8.8 Manually Close Digestion Oven Door

## 8.9 Install SPE Columns

- 8.9.1. Be sure to remove all shipping packaging from inside the columns.
- 8.9.2. Wipe the inside of the column top with a wetted paper towel (water) to remove sorbent dust before installing each column onto the instrument.
- 8.9.3. Install SPE columns by placing the top of the columns over the column top adapters on the instrument. Push the column upwards and guide the bottom of the column all the way to the back until it hits the guide and set the column in the column bottom adapter on the instrument. **Twist each column a quarter turn clockwise (to the left).**



- 8.9.4. Once all SPE columns are installed, press "YES."

## 8.10 Install Round Bottom Flasks

- 8.10.1. Open the evaporation chamber door manually.
- 8.10.2. Turn the large black release ring at each position clockwise (to the right), until it fully tightens.
- 8.10.3. Install glass round bottom flasks over the round bottom flask adapters in the evaporation chamber by pushing each until it seals tightly.
- 8.10.4. Close the evaporation chamber door manually.
- 8.10.5. Press "YES," confirming that all four round bottom flasks are installed properly.



Round Bottom Flask Adapter

Release Ring

## 8.11 Confirm Solution Reservoirs are Full

- 8.11.1. Confirm that each solution reservoir is filled with the appropriate chemicals. (Refer to Appendix A - Reagents)
- 8.11.2. Ensure that each reservoir cap is closed tightly.
- 8.11.3. Once all the reservoirs are full, press "YES."

## 8.12 Start Run

When set-up steps have been completed, press "START RUN."



### 8.13 Track Progress

During an assay, the user can track progress and observe dynamic conditions. Once the run has started, the user can toggle between the following three screens to see details such as: which vessels are being filled, whether draining is complete, which reservoir is engaged, and the current step of the method.

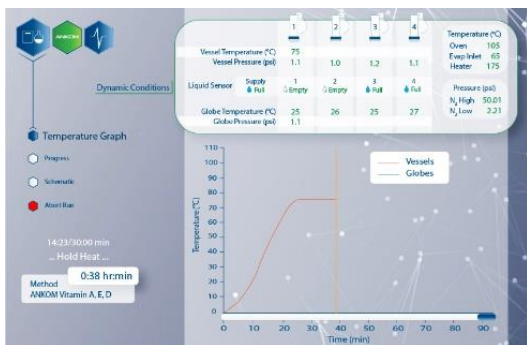
#### Schematic



#### Progress



#### Temperature Graph

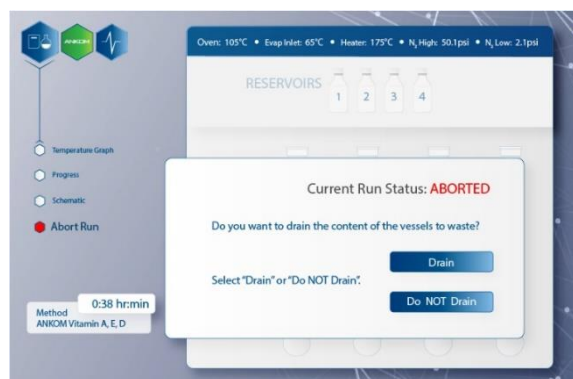


### 8.14 Abort Run

If the user no longer wants to continue with an assay, press: “Abort Run.”

A screen prompt will ask, “Do you want to drain the content of the vessels to waste?”

- If the user does not want to keep the content in the vessels, they will select “Drain.”
- If the user wants to keep the content in the vessels, they will select “Do NOT Drain” –Do not use this option unless CUSTOM mode is enabled or advised by ANKOM.



When a run is aborted, the digestion oven will cool to a safe temperature. Do not remove the digestion vessels if filled with liquid. Be careful when removing the digestion vessels because they may still contain liquid. Digestion vessels have drainage holes that will allow liquids to drain, once removed from the instrument.

## 8.15 End of Run

At the end of an assay, the round bottom flasks that contain the isolated analyte, must be removed for further quantitative analysis.

- 8.15.1. Turn the large black release ring counterclockwise (to the left), to release the round bottom flasks.
- 8.15.2. Remove the round bottom flasks and immediately cover each flask with aluminum foil. Post-processing of the analyte will include a variety of treatments. See relevant standard operating procedures located on the ANKOM website for further instructions.
- 8.15.3. Remove and clean digestion vessels for subsequent use. Refer to FLEX Service Procedure FLS011.

**NOTE:** It is very important to thoroughly clean vessel bottom assemblies after use. Use water, and NOT ethanol or acetone, as final rinse solution. The digestion vessels must not be left assembled for extended periods of time (e.g. overnight). If digestion vessels are left assembled for extended periods of time, the O-rings in the vessel bottom assemblies will deform and future integrity could be compromised.

- 8.15.4. Remove and dispose of the SPE columns according to SDS guidelines and internal SOP.
- 8.15.5. Wipe the digestion vessel top's spray nozzles and O-rings, vessel bases, and the SPE column top's O-rings with a towel wetted with water. Refer to FLEX Service Procedure FLS011.



- 8.15.6. Disposal of waste bottle reservoir materials: This reservoir will contain a mixture of all the solutions that has been used in an assay, which may include organic solvents, strong bases, and acids. Dispose according to SDS guidelines and internal SOP.

If the user would like to turn off the FLEX, go to the home screen, press the "Lock Hexagon" located in the lower right corner and then press "SHUT DOWN." Once the screen is black, the user can turn off the power switch on the left side of the instrument.

**NOTE:** It is recommended to shut down the HMI touchscreen as described above before turning off the power switch. If the instrument needs to be restarted, wait 30 seconds before powering back on.



If after running this instrument the user plans to leave it unused for one week or longer, the user must clear out the fluid lines to ensure proper operation. See "Instrument Out-of-use Procedure" located on [ankom.com](http://ankom.com) for more information.

## 9. Adjust Method

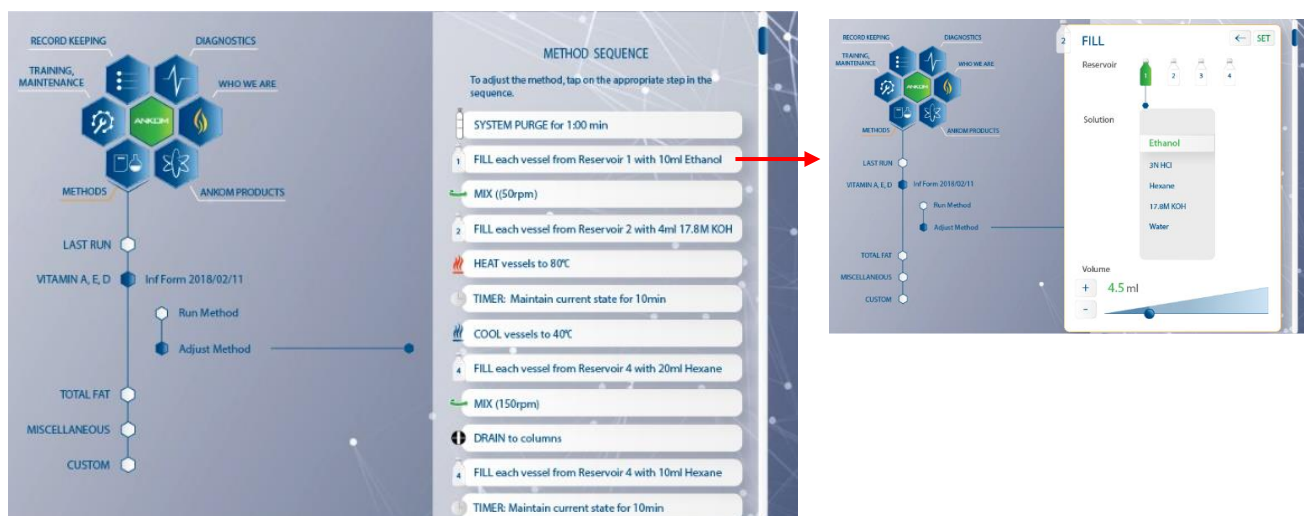
9.1 From the Home Screen, select "METHODS."

9.2 Select the desired method category. Example: "VITAMIN A, E, D"

9.3 Select the method of choice. A list of previously used methods will appear on the right side of the screen. Example: "Vit AED 75C 45m"

9.4 Select "Adjust Method." The right side of the screen will display the method sequence listing each module (step) of the method. Each of the modules (Fill, Heat, Cool, Mix, Drain, etc.) can be selected and individual parameters can be adjusted.

**NOTE:** The method sequence cannot be changed, and modules cannot be removed. The user must upgrade to access the custom section of the software for the ability to create a completely personalized method.



9.5 Select a module to adjust.

- FILL module- Use the slide bar to adjust the volume and select the solution that will go into the vessels.
- MIX module- To adjust the speed of the mixers, use the keypad to specify the mix speed in RPM. Select "SET" to save the parameter or the back arrow to exit without saving changes.
- HEAT Vessels module- To adjust the temperature of a saponification, use the slider bar or the plus and minus buttons. To adjust the duration, tap the current digestion time and use the keypad to specify a new digestion time. Select "SET" to save the parameters or the back arrow to exit without saving changes.
- DRAIN to column module- To adjust drain to column, tap the column or waste icons to specify drain direction. Use the keypad to specify maximum drain time. Select "SET" to save the parameters or the back arrow to exit without saving changes.
- COOL vessels module- To adjust oven cool temperature, use the slider bar or the plus and minus buttons. Select "SET" to save the parameter or the back arrow to exit without saving changes.
- TIMER module- Use the keypad to specify timer. Select "SET" to save the parameters or the back arrow to exit without saving changes.

## 10. Custom Method

This section is only available to users who have purchased an upgradable instrument. For full details on how to create a method refer to the ANKOM website. Custom functionality allows the user to: customize an existing method or build a new method from scratch.

To build a new method, from Home Screen:

10.1 Select "METHODS."

10.2 Select "CUSTOM."

10.3 Select "New Method."

10.4 To build a method the user will drag a module to the right side of the screen under Method Sequence. Each module can be adjusted to the users desired setting.

10.5 Press "SAVE METHOD." The user will type in a method name and select which folder to save the method under.

The user can drag modules to the waste bin to delete them from the method sequence.



The user can adjust each module to desired settings.

Drag to add module

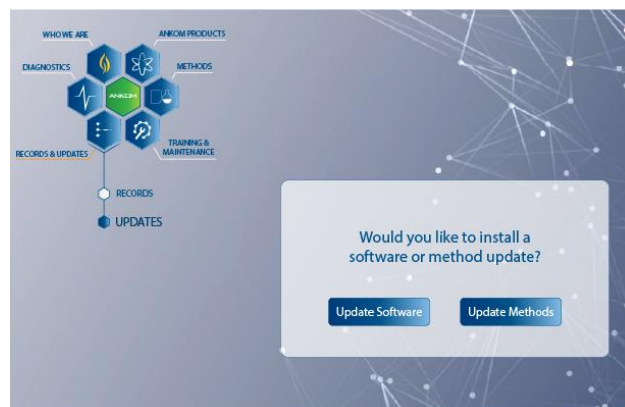
Modules



## 11. Uploading a New Method

This section explains how to upload a new method onto the FLEX instrument.

- 11.1 From the Home Screen, press the "Records & Updates" hexagon.
- 11.2 Press the "Updates" hexagon.
- 11.3 Select "Update Methods."



- 11.4 Insert the USB drive, that contains the new method, into the USB port on the side of the instrument.
- 11.5 The following screen will display to ask you to confirm whether you would like to proceed. If yes, press "Continue."



- 11.6 Once the upload is complete, the screen will prompt you to restart. Press "Restart." The screen will go black and then begin to reboot bringing you back to the home screen. This may take several minutes.
- 11.7 Remove the USB drive from the USB port on the side of the instrument.



## 12. Faults

When the instrument is not operating correctly, a fault message will alert the user. Faults can occur anytime whether an assay is in progress or not. If a fault occurs outside of an assay, an alert will pop-up on the screen. Refer to the "Technical FAQs" section on [ankom.com](http://ankom.com) or call ANKOM Technology.

If a fault is triggered inside of a method, there can be multiple different outcomes:

- The user is alerted, and the method will continue with the assay. No user action required.
- The user is alerted, and user action is required. Follow screen prompts to troubleshoot and press "CONTINUE." If the fault occurs again, refer to the "Technical FAQs" section on [www.ankom.com](http://www.ankom.com) or call ANKOM Technology.
- The user is alerted, and the instrument automatically aborts the run for safety reasons. The user will be asked whether they want to drain the content of the vessels to waste or not. Refer to the "Technical FAQs" section on [www.ankom.com](http://www.ankom.com) or call ANKOM Technology.



## **13. Periodic Maintenance**

### **13.1 Inspect and clean solution reservoir bottles and filters**

On a monthly basis (or more frequently depending on usage), follow the steps below to inspect and clean the solution reservoir bottles and filters.

13.1.1. Inspect the bottles, filters, and internal tubing, for any precipitate, particles, or foreign matter.

13.1.2. Clean filters by running them under fast running water. Replace as needed.

13.1.3. Wash and rinse the bottles out with water and let air dry as needed.

### **13.2 Inspect and clean the spray nozzles in the digestion oven**

On a monthly basis (or more frequently depending on usage), follow the steps below to inspect and clean the spray nozzles.

13.2.1. Periodically observe the spray capability during the addition of solution in the digestion vessels.

13.2.2. Clean spray nozzles by wiping each nozzle with a wetted cloth or paper towel.

### **13.3 Regularly clean the instrument surfaces**

13.3.1. Wipe down the surface of the instrument whenever spills have occurred.

## 14. Troubleshooting & Replacement Parts

The ANKOM Technology website has the most current troubleshooting and replacement parts information. If you have any questions about the operation of your ANKOM<sup>FLEX</sup> Analyte Extractor, or if you need replacement parts, please visit our website at [www.ankom.com](http://www.ankom.com).

## 15. QC and Calibrations

### 15.1 Solution Delivery Calibration

The purpose of the solution delivery calibration is to confirm that the instrument pump is delivering the correct amount of solution. This calibration should be done when a new chemical is added, or if the user would like to confirm that solution delivery is accurate. Wear gloves and safety glasses when running this calibration.

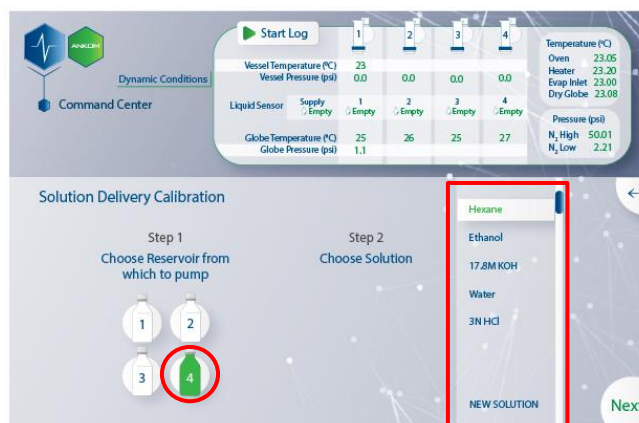
- 15.1.1. Fill each solution reservoir with the appropriate reagents to at least half the bottle volume, refer to Appendix A - Reagents. Be sure to use the correct reservoir position for each solution as certain solutions must be vented (1 and 3) or unvented (2 and 4). Close all reservoir caps when filled. For further details on how to fill the reservoir bottles, refer to FLEX Service Procedure FLS005 on the website.



- 15.1.2. From the Home Screen, press “DIAGNOSTICS.”

- 15.1.3. Select “SOLUTION DELIVERY CALIBRATION.” The following screen will be displayed. Follow the instructions on the screen.

- 15.1.4. If calibrating for one new chemical, follow the calibration steps for that specific reservoir only. If calibrating all four reservoirs, follow the specific sequence listed below. Select reservoir 4 to begin.



Sequence:

1. Reservoir 4 - Hexane
2. Reservoir 1 - EtOH
3. Reservoir 3 - H<sub>2</sub>O
4. Reservoir 2 – KOH
5. Reservoir 3 – H<sub>2</sub>O (Run this reservoir again to clear lines of KOH)

- 15.1.5. A list of solutions is provided on the right side of the screen. The user will choose the solution that is in the selected reservoir and then press “Next.”

**NOTE:** The user can add a new solution by pressing “NEW SOLUTION”. The user must enter an accurate boiling point for safety checks. Once this is complete, press “Next”. **The instrument cannot run Diethyl Ether or Acetone.**

- 15.1.6. Label four 100 ml plastic calibration cups, 1-4.

- 15.1.7. Install the plastic calibration cups into the digestion vessel guide, ensuring that the lip of the cup is flush with the guide and that the pour spout is facing towards the user.



Digestion Vessel Guide

- 15.1.8.

**SAFETY ALERT:** Do not press "Start Test" until fully completing the instructions below.

Close the digestion oven door before starting the test. Press "Start Test." The lines will be primed, and solutions delivered. The test will pump 10ml volume to each cup sequentially.

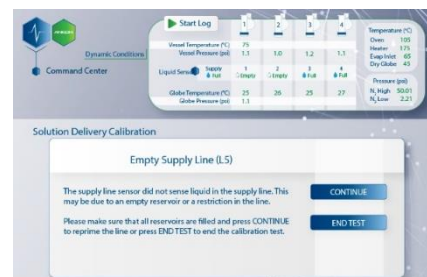


There are two faults that may occur during the solution delivery calibration:

**Empty Supply Line**

This fault is most often caused by an empty reservoir. The user must refill the reservoir and press continue.

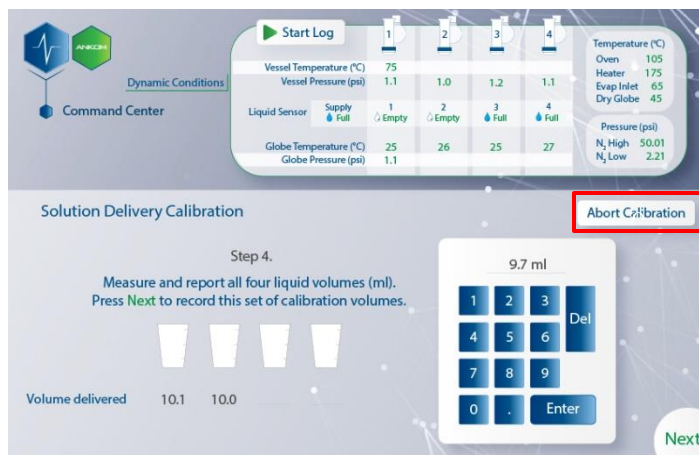
If the message continues to be displayed, it may be caused by a restriction in the line or a leak in the system. Contact ANKOM Technology for assistance.



**Full Supply Line**

This message can occur if the lines are not cleared properly at the end of the solution delivery calibration. Possible causes include left over liquid in the line, full waste container, or mechanical failure. Contact ANKOM Technology.

15.1.9. Open the digestion oven door and remove the plastic cups. Using a graduated cylinder, measure and enter the volumes delivered in each plastic cup to the nearest tenth on the HMI screen. **DO NOT PRESS** "Next" on the HMI screen.



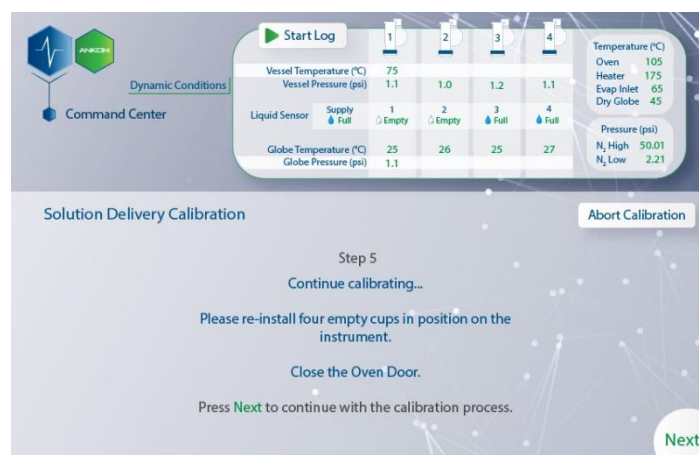
15.1.10. **VERY IMPORTANT:** Are all the volumes between 8.5-11.5ml?

- **If Yes** – Continue calibration by pressing "Next" and follow the prompts on screen.
- **If No** – Abort Calibration and re-try the Solution Delivery Calibration. If volume falls outside this range again (8.5 – 11.5ml), abort calibration and contact ANKOM Technology.

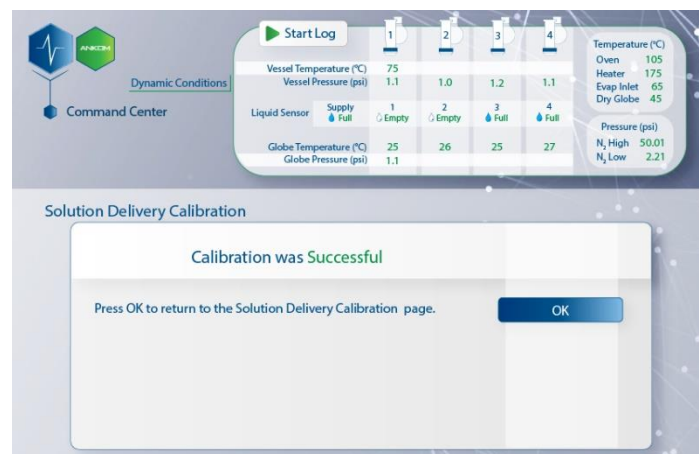
15.1.11. The following screen will be displayed when one calibration series has been completed, but calibration accuracy has not yet been reached.

Continue with the calibration procedure by following the steps on the screen. **The calibration process must be repeated until the delivery volume is within an acceptable accuracy range and the calibration test is passed.**

The system will try multiple times to achieve desired accuracy but will fault if system is unable to calibrate.



15.1.12. This screen will be displayed if the calibration is successful. The pump configuration settings for that solution is updated and will be used during analysis. Press "Ok."



## 16. Appendix A – Reagents

### 16.1 Best practices for solution preparation: Vitamin & Cholesterol Analysis

Where high volume usage makes daily solution preparation less practical, the following guidelines can be used.

Solution	Reservoir	Solution Content	Make up	Shelf Life	Storage Requirements
Ethanol	1. Green	2% (w/v) Pyrogallol (or equivalent) in Ethanol	Weigh 8g ± 0.1g Pyrogallol (or equivalent) into 500ml volumetric flask. Make up to mark with 95% Ethanol	Make up fresh daily.	Actinic glass, Vented Reservoir, Room Temperature (15-25°C)
12.7N KOH	2. Yellow	12.7N KOH	500g KOH + 500ml water	Replenish as needed. Clean bottle every two weeks.	Unvented Reservoir, Room Temperature (15-25°C)
Water	3. Blue	Water	Deionized water	Replenish as needed. Clean bottle every two weeks.	Vented Reservoir, Room Temperature (15-25°C)
Hexane	4. Red	0.05g/L BHT in Hexane	0.05g Butylated hydroxytoluene (BHT) + 1L Hexane	Replenish as needed. Clean bottle every two weeks.	Unvented Reservoir, Room Temperature (15-25°C)

### 16.2 Best practices for solution preparation: Fat Analysis

Where high volume usage makes daily solution preparation less practical, the following guidelines can be used.

Solution	Reservoir	Solution Content	Make up	Shelf Life	Storage Requirements
Ethanol	1. Green	2% (w/v) Pyrogallol (or equivalent) in Ethanol	Weigh 8g ± 0.1g Pyrogallol (or equivalent) into 500ml volumetric flask. Make up to mark with 95% Ethanol	Make up fresh daily.	Actinic glass, Vented Reservoir, Room Temperature (15-25°C)
3N HCl	2. Yellow	3N HCl			Unvented Reservoir, Room Temperature (15-25°C)
Water	3. Blue	Water	Deionized water	Replenish as needed. Clean bottle every two weeks.	Vented Reservoir, Room Temperature (15-25°C)
Hexane	4. Red	0.05g/L BHT in Hexane	0.05g Butylated hydroxytoluene (BHT) + 1L Hexane	Replenish as needed. Clean bottle every two weeks.	Unvented Reservoir, Room Temperature (15-25°C)

# Automation saves time and money

ANKOM Technology is an international company with products that include:

	<p><b>DELTA Automated Fiber Analyzer with Pump System</b></p> <ul style="list-style-type: none"> <li>• Crude Fiber (AOCS Ba 6a-05), ADF, NDF determinations</li> <li>• Automatically adds solutions and rinses</li> <li>• Batch process - up to 24 samples at one time</li> </ul>
	<p><b>XT15 Fat Extractor</b></p> <ul style="list-style-type: none"> <li>• Official Method AOCS Am 5-04</li> <li>• Fully automatic</li> <li>• Solvent recovery at 97% or greater</li> <li>• Batch process - up to 15 samples at one time</li> </ul>
	<p><b>RF Gas Production System</b></p> <ul style="list-style-type: none"> <li>• High sensitivity pressure measurement</li> <li>• Applications include: Biomass-to-Energy analysis (e.g., Ethanol, methane, etc.), Biodegradability, Ruminant Nutrition, Yeast Activity, Beer/Wine Fermentation, Soil respiration, BOD, Human Digestion, etc.</li> <li>• Wireless Computer control and data storage</li> </ul>
	<p><b>Chemicals</b></p> <ul style="list-style-type: none"> <li>• A wide variety of chemicals used for many different lab operations</li> <li>• Pre-mixed solutions available</li> </ul>

Please visit our web site at [www.ankom.com](http://www.ankom.com) for more information.

2052 O'Neil Rd, Macedon NY 14502  
 Telephone: (315) 986-8090  
 Fax: (315) 986-8091  
[www.ankom.com](http://www.ankom.com)

