SepaBean™ machine T

Flash Chromatography System

Software Manual - V1.5.1

Sorbtech
Separate. Purify. Discover.

SANTAI TECHNOLOGIES

Table of Contents

Introduction

SepaBean™ software is an independent research and development assisting software tool. When integrated with SepaBean™ machine, this combination provides an enhancement to the liquid chromatography operations. It not only improves the purification efficiency of the lab, but also promotes development of green chemistry. The software has three functions integrated in it which are online access, offline access, and data sharing. Use of this software improves the separation efficiency significantly and reduces waste of experience resources in chemical experiments using the data sharing feature.

SepaBean™ software is flexible enough to install on most mobile devices, including phones, tablet computers, notebooks, and more. With this ability, the software offers user-friendly remote monitoring of the separation.

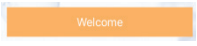
This user manual introduces the steps for the sofware's routine operation in detail. It will be effortless for users to operate the SepaBean™ machine using the SepaBean™ software after reading this manual.

Overview

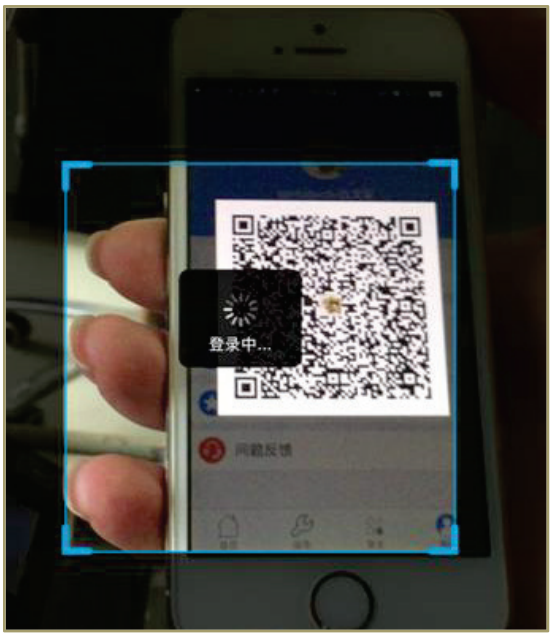
The SepaBean™ software manual covers user registration and login, system configuration, normal phase separation, reversed phase separation, machine maintenance, and history record.

1. User Registration and Login

Click on the SepaBean™ icon  on the iPad screen to enter the login screen. The app will search the device automatically. You can also click on the iPad icon  to search the device. If the local area network (LAN) has many devices, you need to select the device that you want.

Click on  to login.

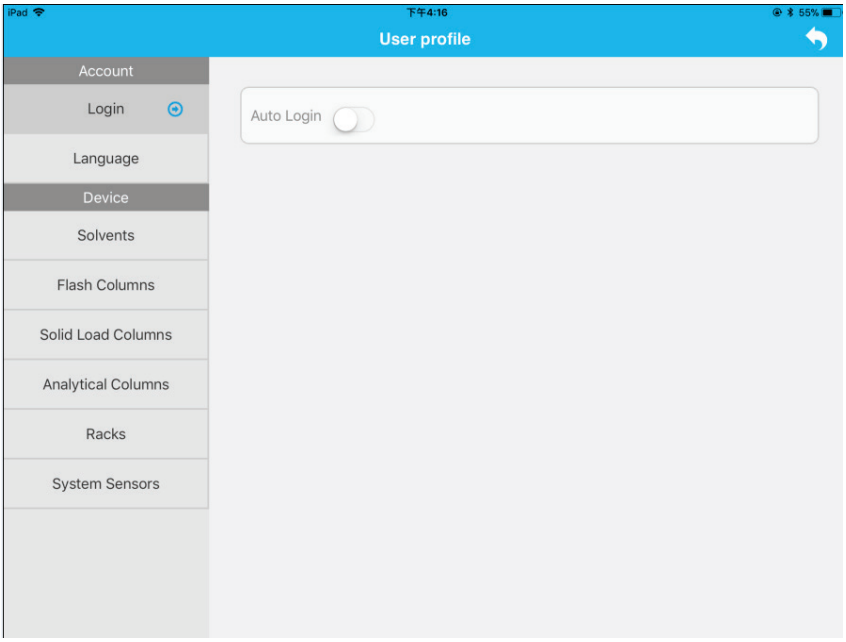
Click on Login to scan the QR code to login.



2. System Configuration

2.1 Account Settings

Click on at the upper right corner to open the system configuration page.



2.2 Login Settings

Click on auto login to switch Auto Login ON or OFF. If switched ON, the auto login enables the user to login directly by skipping the login page. With auto login ON, the software opens the last accessed/ most recent account by default.

2.3 Language

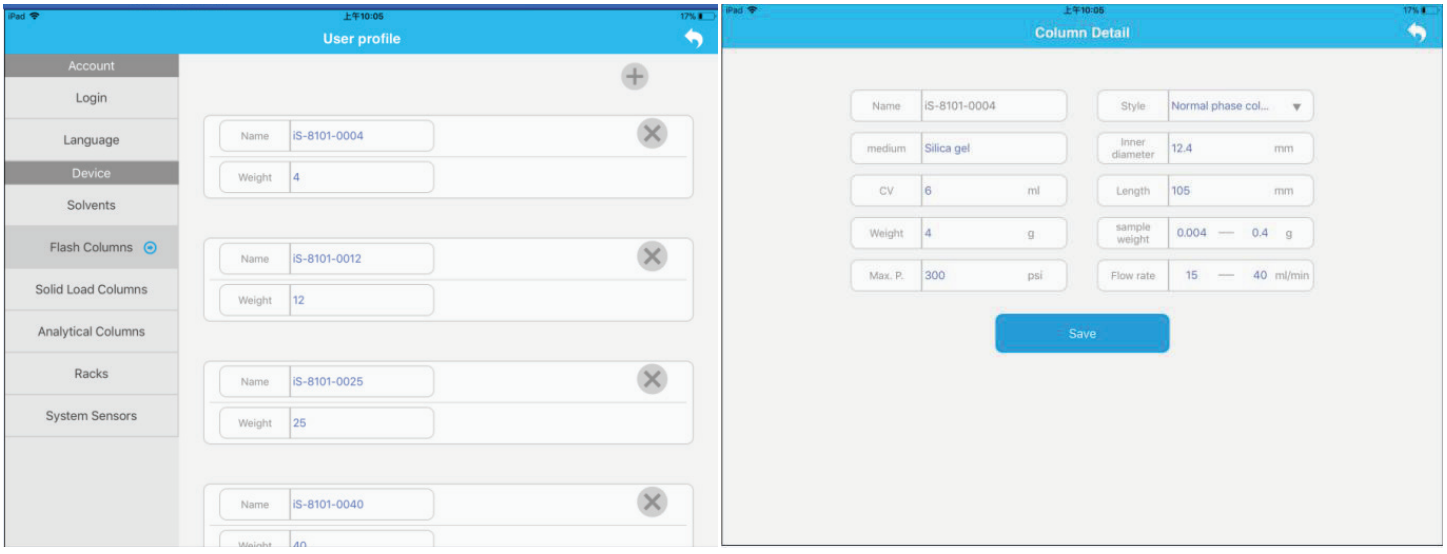
Users select the language by clicking on either “English” or “Chinese.”

2.4 Device

User Management - Displays the multiple user accounts on the screen. Users can logout of the software from this page. Also, users can change the password of the respective account from this page.

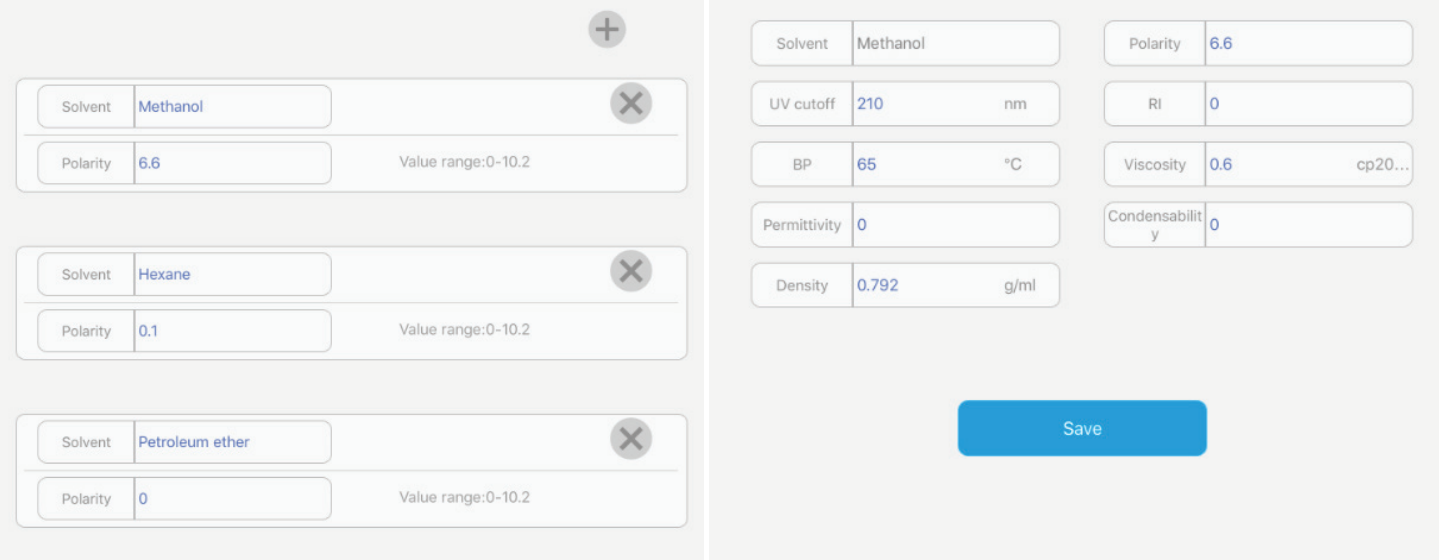
2.4.1 Flash Column

Users can find information about various flash columns on this page by clicking on the column rows. To add a column, click on . Input all the relevant information of that column in the respective fields. Click “save” to proceed. To delete a column, click on after the column detail which you don’t need.



2.4.2 Solvents

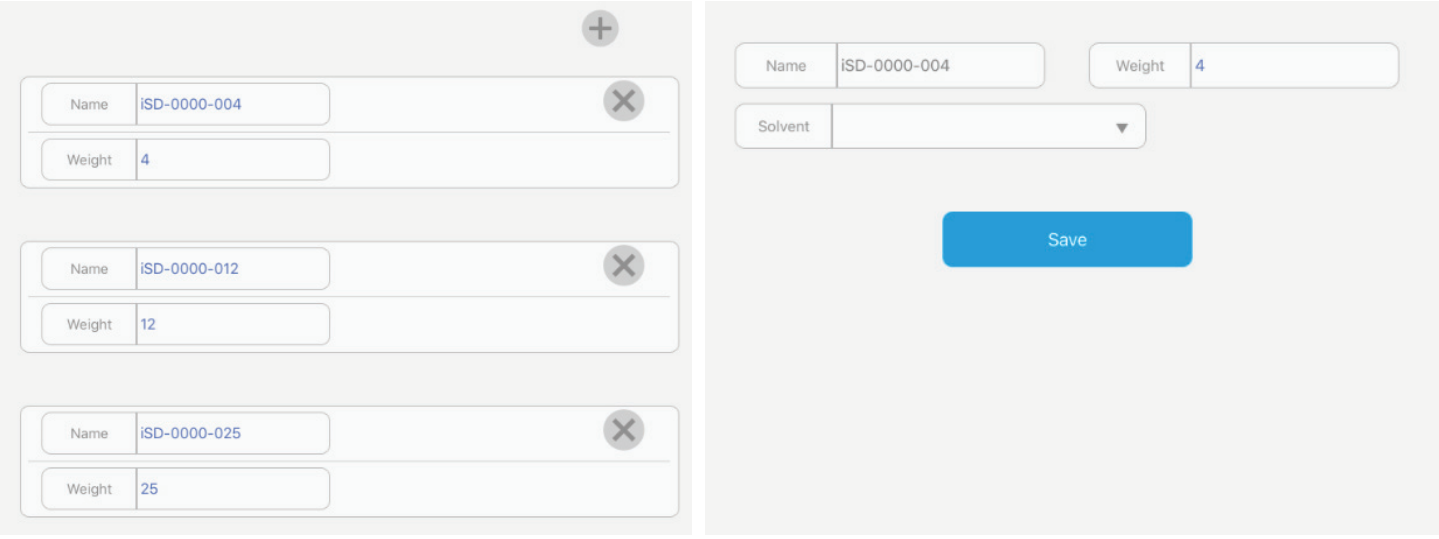
Users can find information about various solvents by clicking on the solvent rows.



To add a solvent, click on  . Input all the relevant information of that solvent in the respective fields. Click “save” to proceed. To delete a solvent that is not needed, click on  to delete the chosen solvent name.

2.4.3 Solid Load Column

The user can find information about various solid load columns by clicking on the respective rows.



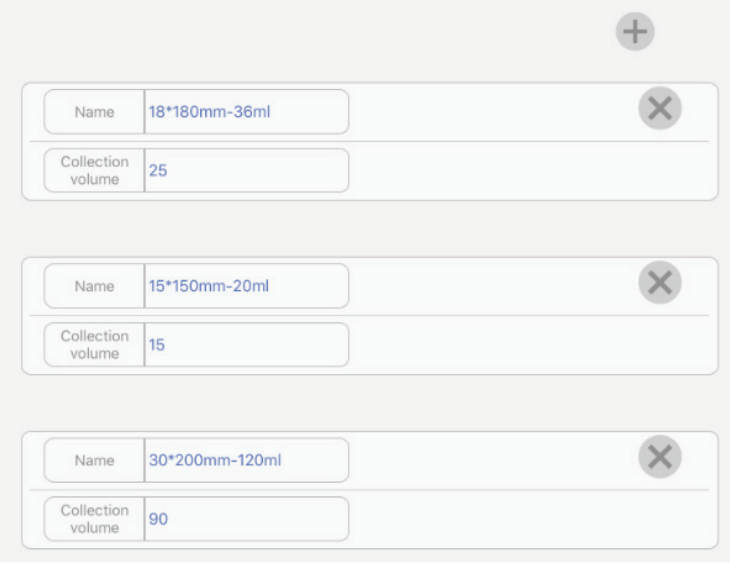
To add a solid load column, click on  . Input all the relevant information of that solid loader in the respective fields. Click “save” to proceed. To delete a solid load column that is not needed, click on  to delete the chosen solid loader name.

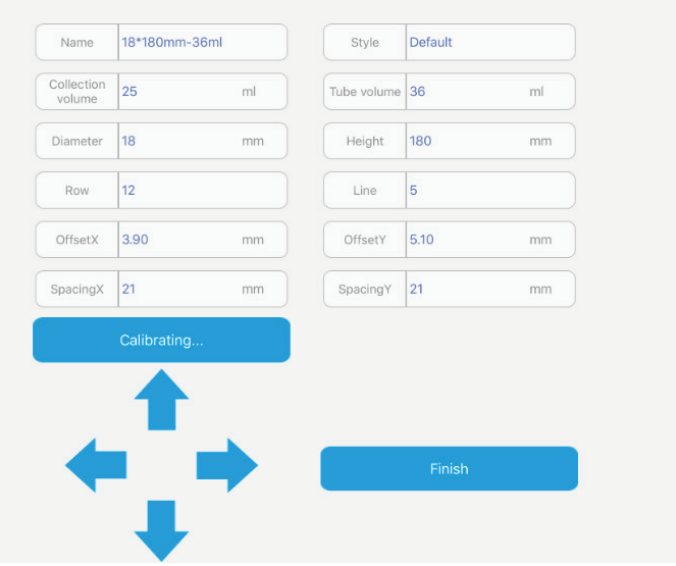
2.4.4 Analytical Column

The user can find information about various analytical columns on this page. Users can add or delete any specific analytical column on this page.

2.4.5 Racks

Users can find information about various racks on this page, and can add or delete any specific rack on this page.



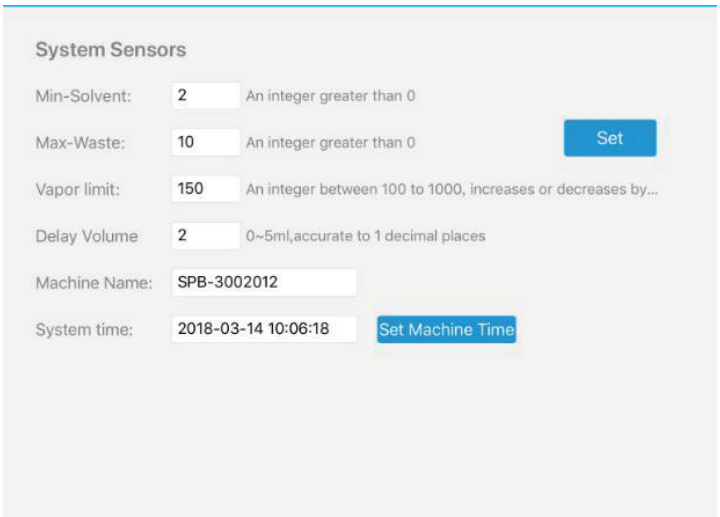


2.4.6 Calibration

By clicking on the specific rack, the user sees the detail of that specific rack. On this page, click on the calibrate button to calibrate the position of the rack with the up, down, left, and right buttons.

2.4.7 System Sensors

Users can setup the minimum level of solvent, maximum level of waste, vapor sensor limit, delay volume, machine name, and system time.

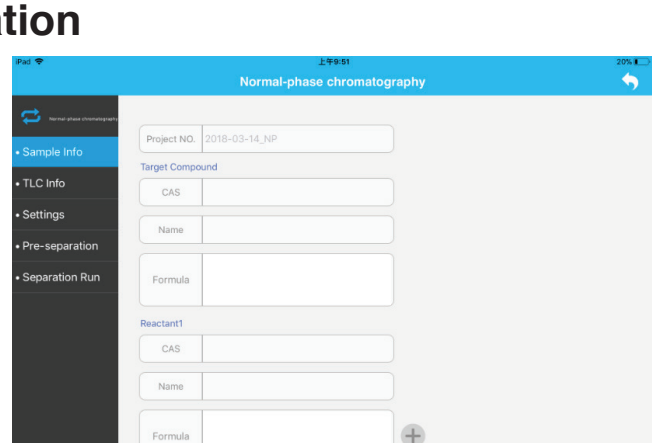


1. **Min-Solvent** - Set the minimum solvent level as per requirement. The default value is 5. The number must be greater than zero. The value must be at least 5 and be divisble by 5.
2. **Max-Waste** - Set the maxium waste level as per requirement. The default value is 5. The number must be greater than zero.
3. **Vapour Limit** - Set the vapour sensor limit. This is used for leak detection. Keep the number low if the system is in good ventilation and keep the number high if the room is shared with other solvent handling instruments. The number must be between 100 and 1000 and divisible by 10.
4. **Delay Volume** - This option is used to synchronize detector signals in case of an external detector.
5. **System Name** - Set the system's reference name.
6. **System Time** - Set the system's date and time.

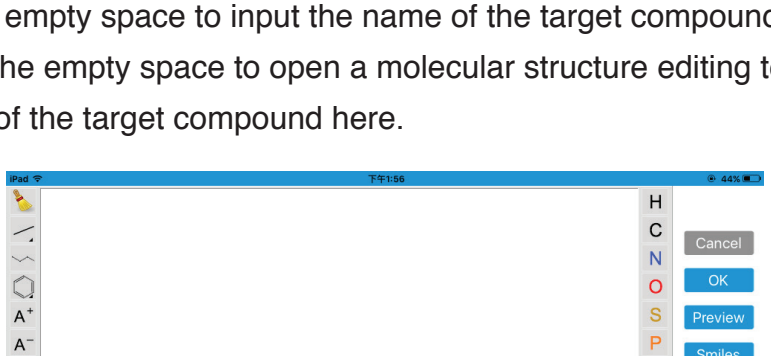
3. User Registration and Login

This module discusses the different method parameters before the start of the run. It has 5 main options: sample information, TLC information, separation settings, pre-separation, and actual start of the separation.

3.1 Sample Information



1. **Project Number** - Users can input a reference number, identification number, or project number of the target compound. The existing project number can be searched by clicking on . A history record of compounds previously run on the system is displayed on the screen for selection.
2. **CAS** - Click on the empty space to input a CAS number. If a project number is searched through the history record and previous data has the CAS number filled in, the CAS field automatically updates.
3. **Name** - Click on the empty space to input the name of the target compound.
4. **Formula** - Click on the empty space to open a molecular structure editing tool. Users can input the molecular structure of the target compound here.

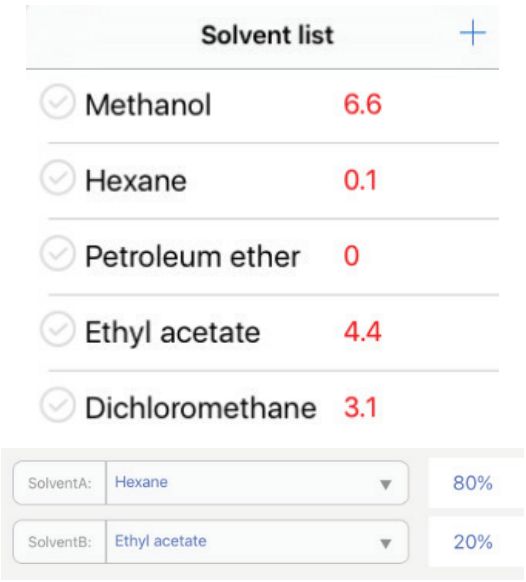


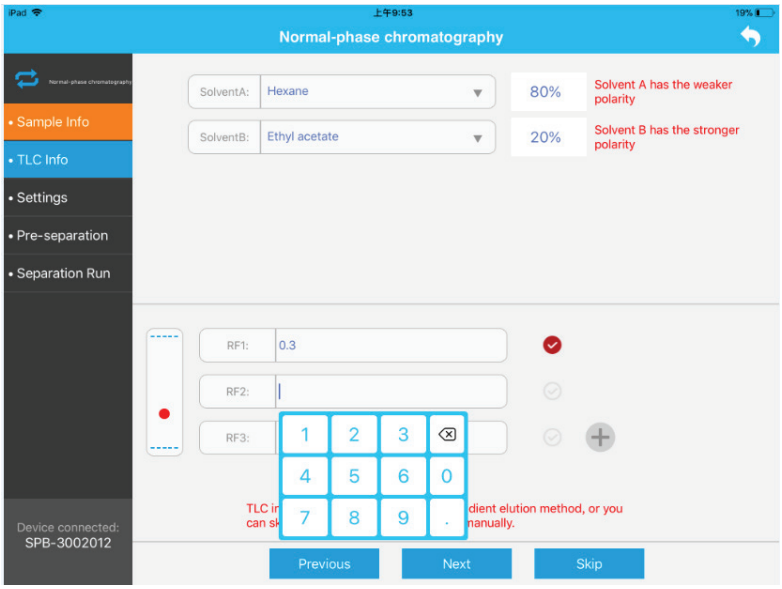
5. **Reactants** - Click on the empty space against the CAS, name, and formula to make the corresponding entries. Users can also add or delete reactants according to needs.

If the user wants to keep the run confidential, the “available for yourself” option can be enabled while keeping the other options disabled. If the user wants to share the run with other users in the company, “available for your company” option can be enabled. Click “next” to go to next screen.

3.2 TLC Information

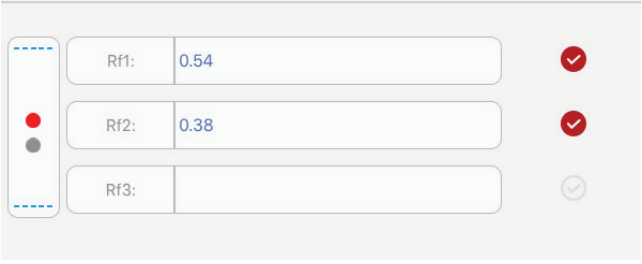
Solvent Selection - Click on the drop-down arrow for solvent A and solvent B to select the solvent from the list. These selected solvents should resemble the solvent system in the TLC run. Click on the % box against each solvent to input the TLC solvent system composition. Once information input is completed, click on “next” to go to the next step.





TLC Rf Values - Input the Rf values of the different spots from the TLC in the software. These Rf values can be of target compound only or, for both target compound and impurities. Maximum four spots on a TLC can be input in this module.

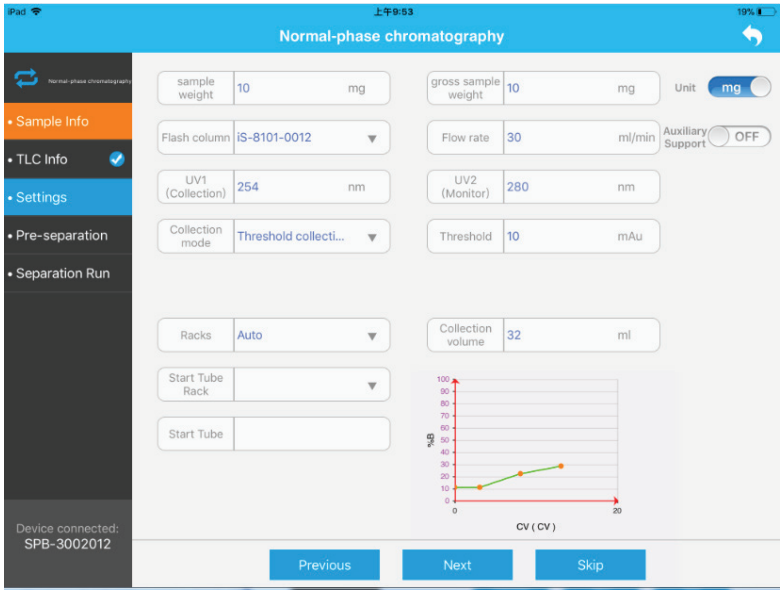
As soon as the Rf values are input, a graphical representation of the TLC can be viewed on the left side showing the position of spots on the TLC according to the Rf values. Once entering Rf values is completed, click on “Next” to go to the next step.



When any of the Rf values are selected, it is displayed as  . If any of the Rf values are selected, then the software will generate a gradient curve automatically. If none of the Rf values are selected, then the gradient curve has to be adjusted manually. Click “next” to go to the next screen.

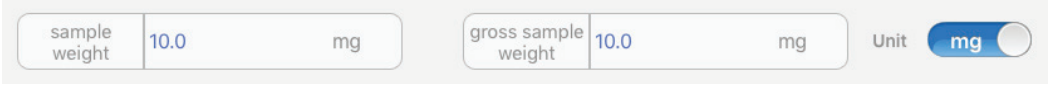
3.3 Separation Settings

This module discusses the various parameters related to the method development for a flash chromatography run.



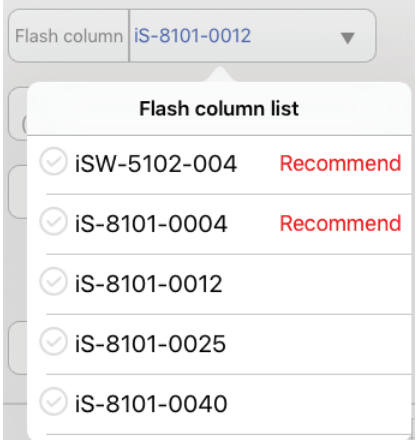
3.3.1 Sample Weight

Sample weight refers to the actual weight of the sample to be loaded in a single separation run. Gross sample weight refers to the total weight of the sample to be separated. If the gross sample weight is more than the sample weight, then the separation run needs to be repeated. This software has a function of automatically repeating the run. The user can toggle the weight unit between mg and g with a toggle switch on the top right corner.



3.3.2 Flash Column Selection

Based on the sample weight, the software automatically recommends the right column. Users can also manually select the appropriate column based on the sample's weight. When the arrow is clicked on, a drop-down list gives a selectivity option.



3.3.3 Flow Rate

The flow rate for the run changes automatically as per the flash column selection. Users can manually override this and select the flow manually. (The flow range is from 5 to 180 mL/min).

3.3.4 Auxiliary Support

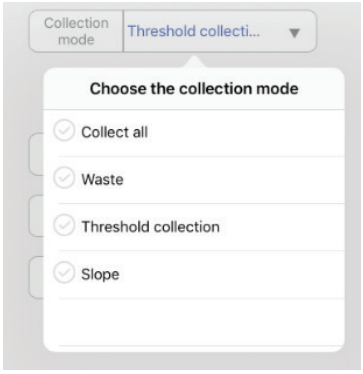
The integrated column holder can accomadate columns up to 330g. If the column is bigger than 330g, it can be installed on the secondary support. In this case, the user needs to switch on the “auxiliary support” by toggling it’s switch.

3.3.5 UV1 and UV2

The wavelength range for the UV detector is from 200 nm to 400 nm, or 200 nm to 800 nm. Collection is based on the detector. Respond according to UV1.

3.3.6 Collection Mode

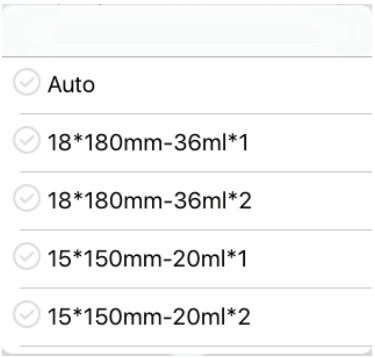
The user can select the way fractions will get collected during the run. The available options are collect all, waste, threshold collection, and slope collection.



- **Collect All** - By selecting this option, the entire run output will be collected regardless if the peaks are eluting or not.
- **Waste** - By selecting this option, the entire run output will be diverted to waste regardless of the peaks eluting or not.
- **Threshold Collection** - When this option is selected, the user needs to input a threshold value for the UV signal in mAu. During the run, when the UV signal exceeds the set threshold value, the fraction collection gets triggered. If the UV signal is below the set threshold, the run output will be diverted to waste. This mode helps the user finish the run with less number of fractions, which in turn saves time in the post-fraction workup. When the slope of the UV chromatogram changes from negative to positive, the fraction collector moves to the next test tube.

3.3.7 Racks

The rack setting has two options. The test tube racks can be set to detect automatically, in which case Santai brand racks must be used with the system. Secondly, the test tube racks can be set to manual by selecting the type of tray at use. With this option, the user can use non-Santai branded test tube racks, which is a great advantage of this system.



3.3.8 Collection Volume

Selection of tray sets the collection volume automatically to a default value. However, the user can change the collection volume to any number below the maximum tube volume. If the collection volume is set above the maximum tube volume, the software provides an error mesage upon pressing “next.”

3.3.9 Start Tube Rack

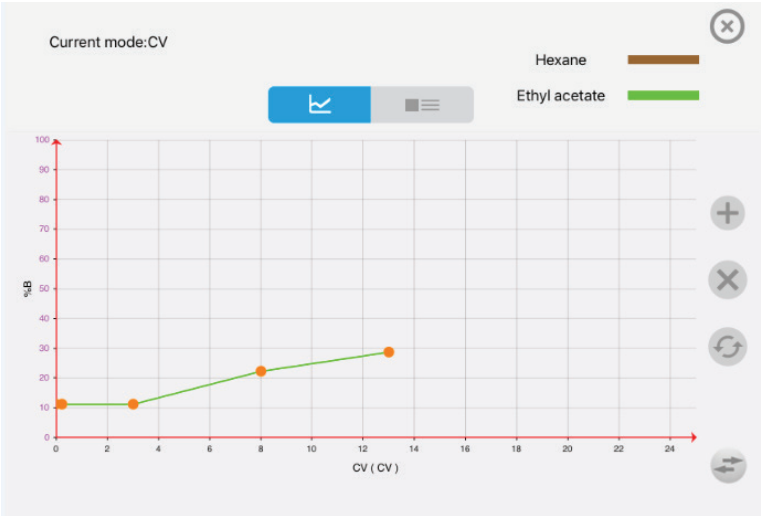
Users can start to collect fractiona from any tube rack. If the user wants to collect from the second tube rack, set the value of the start tube rack to 2.

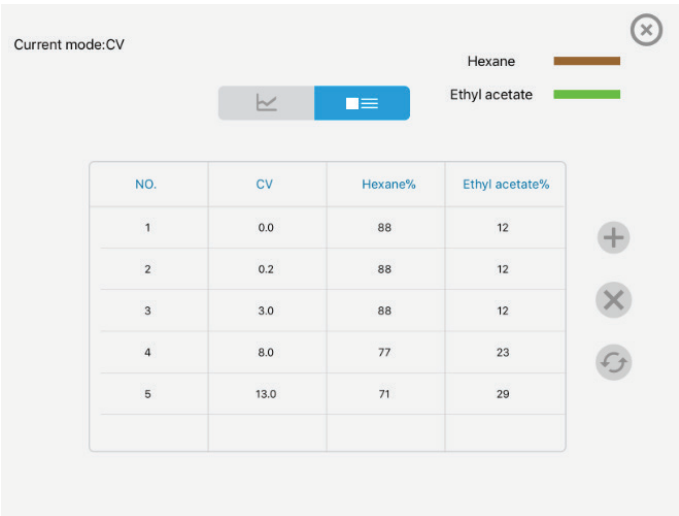
3.3.10 Start Tube

Users can start to collect fractions from any tube. The user only needs to set the value of the tube which will be used as the starting tube.

3.3.11 Gradient Preview

This window shows the gradient profile based on the Rf values set by the user. The user can click on the graph to open an enlarged graph with modification options. There are two options: edit through graph or edit through table. Click  to toggle between the graph and table option.

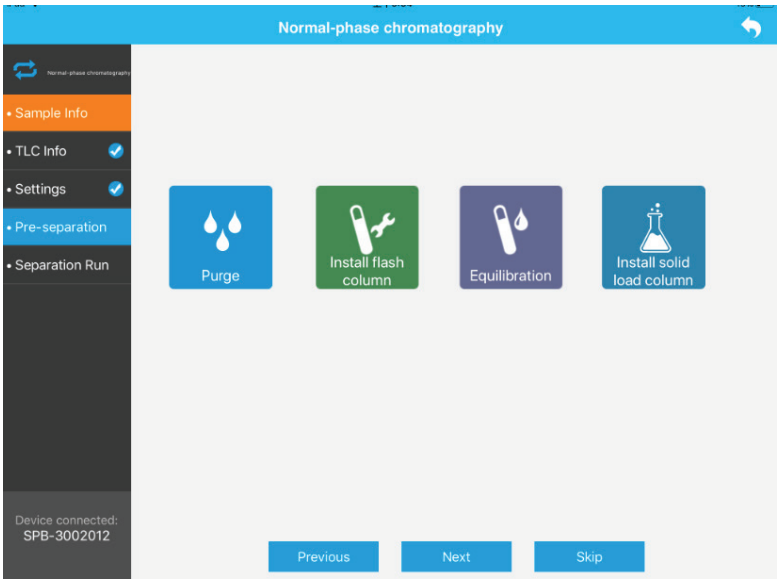




The gradient nodes can be added by clicking  . The slope and duration of the gradient segment can be changed by clicking on the node and then dragging the node to a point of choice. The last created gradient segment can be deleted by clicking on  . By clicking on the bottom right icon, users can toggle the unit of the x-axis between CV and minutes. Click on the top right icon  to exit the screen. Click “next” to go to the next screen.

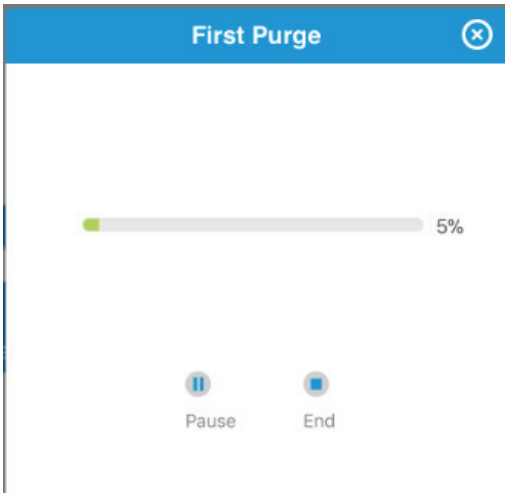
3.4 System Preparation (Pre-Separation)

This module comprises four parts, which is utilized to prepare the system for an actual run, including purge, install flash column, equilibration, and install solid load column. Each of these options can be repeated as needed.



3.4.1 Purge

When clicking on the purge option, a pop-up window opens as below. The user should input the purging time for each line and click on “purge.” The system will purge the strong polar solvent line first followed by the weak polar solvent line. This option ensures that the solvent lines are free from any air gaps and pumping is smooth.

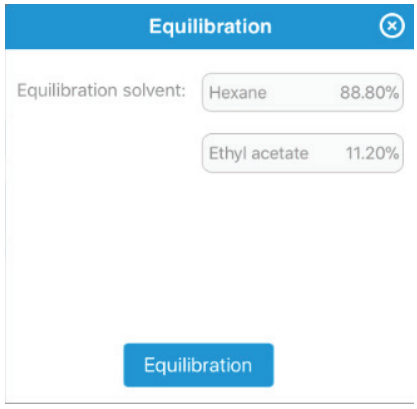


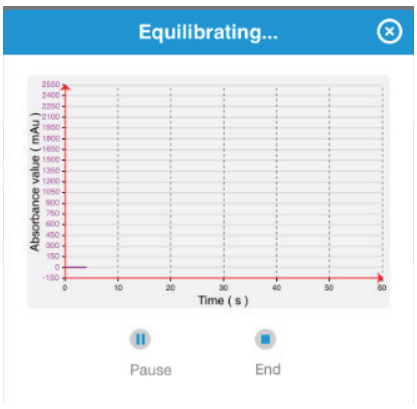
3.4.2 Install Flash Column

When clicking on this icon, the column holder arm moves up autmatically. Users should install the column and then press “finish.” The arm will automatically move down to fix the column in the holder.

3.4.3 Equilibration

When clicking on the purge option, a pop-up window opens. The user can input the starting solvent ratio of the gradient profile, or the system could run equilibration with solvents at the default ratio and the user doesn’t need to do inputs. With this starting ratio of solvents, the column will be equilibrated. Users can manually stop running at any time or wait for the system to automatically stop after running for the default CVs according to the flash column in use.



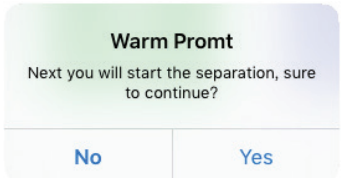


Click on “equilibration” when the ratio is set. It will open a pop-up window with the graph that shows absorbance of equilibration solvent. With this, users can pause or stop the equilibration manually. Otherwise, the system will decide the equilibration time based on the column size and stop automatically after that.

3.4.4 Solid Load Column

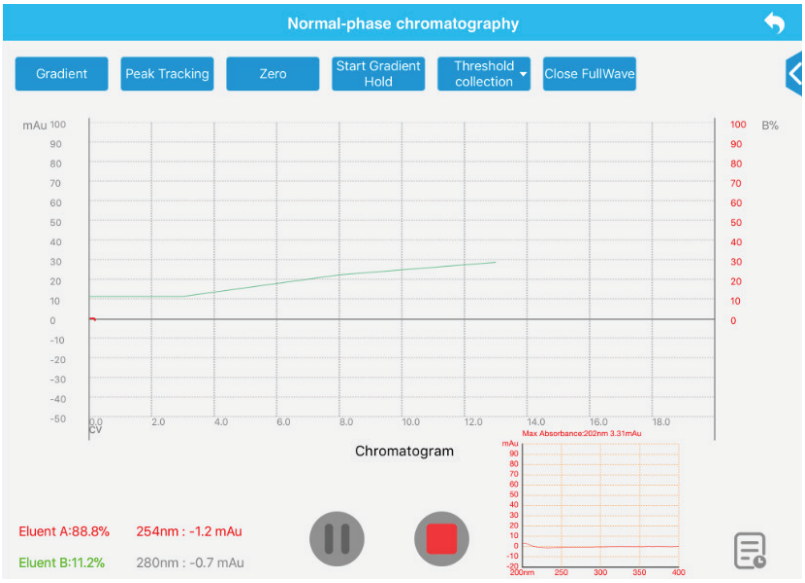
When clicking on this option, the user will see a pop-up window prompting to install the solid load column. At this moment, the user should either install the solid load column, if using a solid sample or inject a liquid sample, if using a liquid sample. The operation is the same for both types of samples. Click on “done” when finished installing the solid load column or injecting the liquid sample.

Click on “next” or “skip” if all the steps are completed. If any step needs to be rechecked or changed, click on “previous.” When clicking on “next” or “skip”, the software prompts user to click “yes” to start the separation.



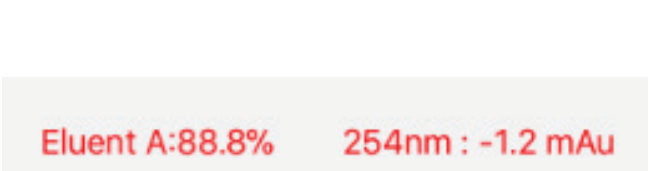
3.5 Running a Separation

This window contains the real time chromatogram with UV absorbance, status bar, control bar, history record access, and access to various parameters related to the separation method.



3.5.1 Status Bar

Click on the arrow icon on the top right corner to access the status bar for access to flow status rate, system pressure, UV1, UV2, collection volume, and UV threshold values. These parameters can be changed during the run by clicking on the respective fields. Also, this bar displays the absorbance value in real time and elapsed time indication.



- **Threshold** - User can change the threshold during the run.
- **Flow Rate** - User can change the flow rate during the run.
- **Pressure** - The system pressure during the run is displayed here.
- **UV1 and UV2** - UV1 and UV2 can be changed during the run. Collection is based on the detector response according to UV1.
- **Collection Volume** - Collection Volume can be changed depending on the shape of the peak.
- **Time** - This is an elapsed time counter.
- **History** - Users can access the history record from this bar during the run.

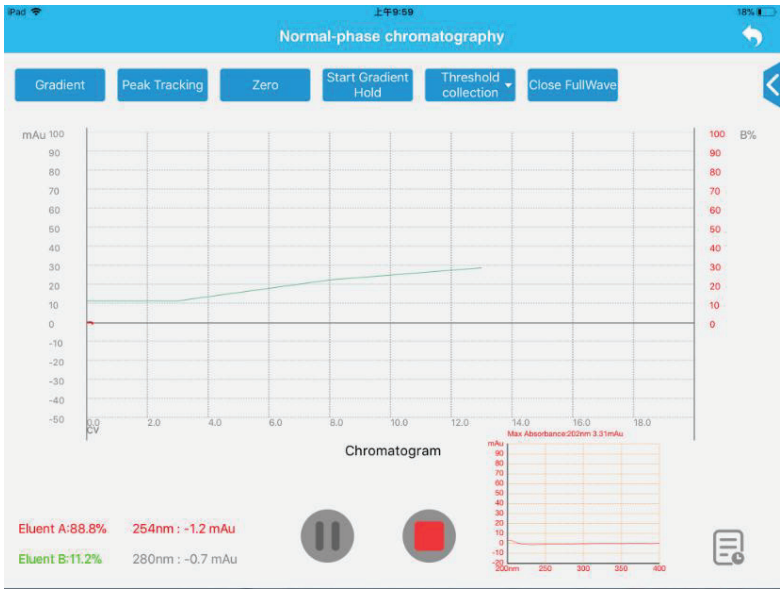
3.5.2 Control Bar

This menu bar provides gradient change, peak tracking, zero, start/stop gradient hold, collect mode, full-wave, next tube, and switching units.

- **Gradient** - User can change the gradient profile from this option. A new gradient segment can be added or an unwanted segment can be deleted.
- **Peak Tracking** - This option enables the user to see the graphical representation of current collecting tube as well as the related tube rack. This allows the user to correlate the peak to the tube rack.
- **Zero** - This allows the user to zero the UV signal. This option can be used when the UV baseline drifts to negative due to the solvent effect.
- **Gradient Hold** - The user can start or stop the gradient hold from this option. User can click on “gradient hold” to hold the gradient % with the value current. To release the gradient hold, click on “gradient hold” again.
- **Collect Options** - Users can switch the collection mode to collect all, waste, or threshold collection.
- **Full-Wave** - Enable or disable showing full-wavelength scanning spectrum.
- **Next Tube** - During separation run, click on next tube. The collector will go to the next tube position. If the collect mode is on waste, user should click on this button, then the collect mode will change to collect all mode.
- **History** - During a separation run, click on  icon. This will enable the user to check with the history records correlating to the current user account.

3.5.3 Chromatogram

The user can see the real time chromatogram with the traces of UV1 and UV2 on this screen.

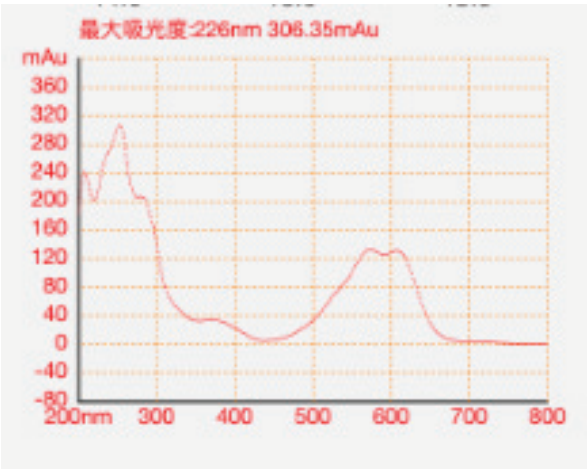


The horizontal x-axis represents either the Column Volume (CV) or Time (T) in minutes. The vertical y-axis represents the absorbance value of the solvent and sample in mAu. The green line indicates the gradient profile by showing the percentage of solvent B. During the run, the software shows the tube number corresponding to the peak on screen. This way, users can track the target compound in the tubes.

- Pause and Stop Buttons - Click on the pause button, the running pauses with the pumping and UV data integration. By clicking on the same button again, the operation resumes.
- Click on the stop button, a pop-up window opens asking the user if the software should stop the separation. Click “OK” to exit or click “cancel” to continue.

3.5.4 Full-wavelength Scanning Spectrum

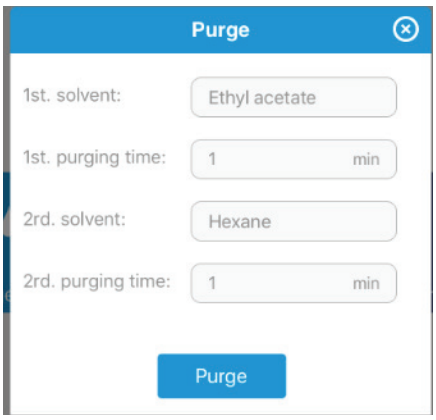
At the bottom right corner of this interface shows the full-wavelength scanning spectrum. The user can utilize the characteristic absorbance of a specific sample to determine whether the sample is eluting out or check with the fraction purity.



3.5.5 Separation End

After the designated time set by the user, the chromatography run will stop automatically. A window will pop-up asking the user to choose stop or continue. If the user clicks on “OK”, the run will stop. If the user clicks on “cancel”, the run will continue for another 1CV.

- When the run stops, the software shows a pop-up window of air purge. If the column and solid load column are to be dried out, the user can input the time and select option “start”. With this mode, air will be blown through the column and solid load column to dry them out. If the user doesn’t want to disturb the equilibrated column, option “skip” can be selected.
- When the air purge ends or when the user presses “skip”, the purge window pops-up asking the user to perform the purging of solvent lines. If the user wants to purge the solvent lines with a strong polar solvent for cleaning, then the time for each solvent line purging should be selected. Click “purge” to start the solvent line purging or click “skip” to skip this step.



4. Reversed Phase Separation

The module of reversed phase separation comprises the following parts: sample information, HPLC module, settings, pre-separation and actual separation, and reserve column.

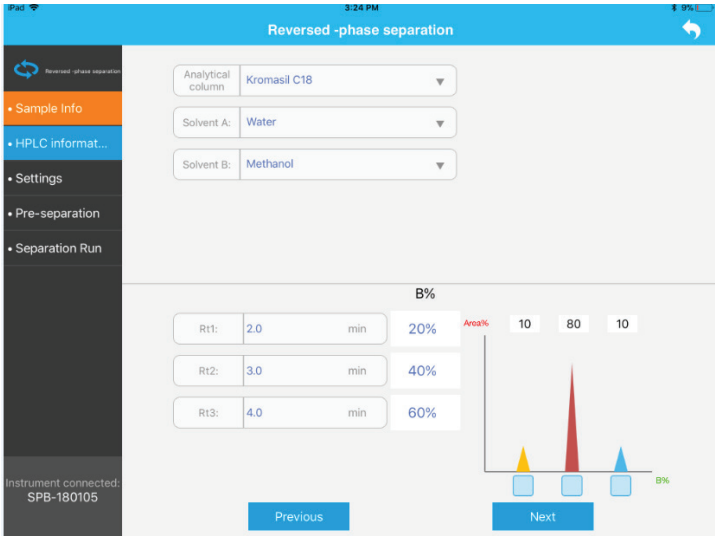
4.1 Sample Information

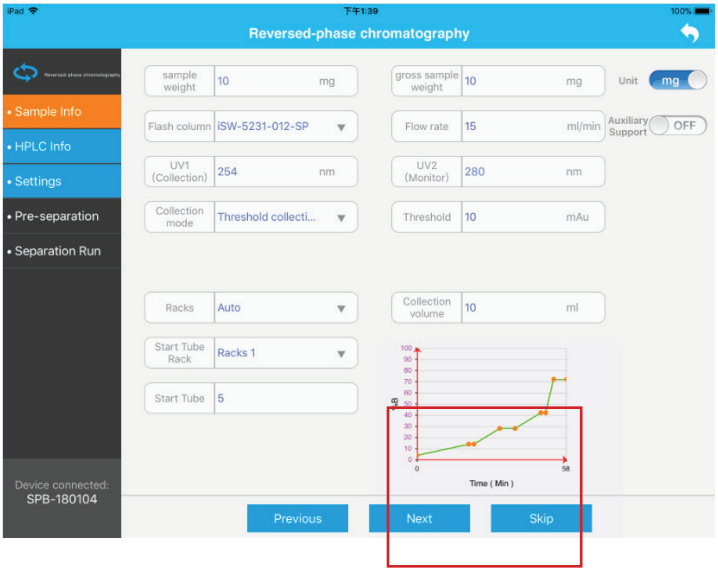
This module is the same as that of normal phase separation. Please refer to section 3.1.

4.2 HPLC

This window requires the user to input the HPLC conditions for the sample which will be run on the SepaBean™ machine. It is optional and the user can opt not to input any values in this window. In that case, the user needs to adjust the gradient profile manually. If the HPLC conditions are input, the software will automatically decide the best fitted flash method for the sample. The user needs to input the parameters below.

- **Analytical Column** - From the drop-down list, the user can select the HPLC analytical column used for the sample analysis. (System default is Kromasil C_{18})
- **Eluent A and Eluent B** - The user should input the HPLC solvent conditions in these options. (System default eluent A is water and B is methanol)
- **Target Retention Time** (Selection) - Filling in the reservation time and area according to HPLC, the system will build a separation gradient automatically.



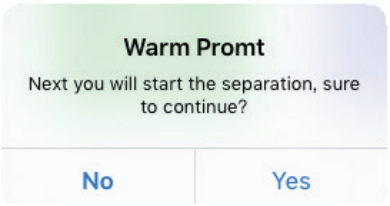


4.3 Separation Settings

This module is the same as that of normal phase separation. Please refer to section 3.3.

4.4 Pre-Separation

This module is the same as that of normal phase separation. Please refer to section 3.4. Click on “next” or “skip” if all the steps are completed. If any step needs to be rechecked or changed, click on “previous”. When clicking on “next” or “skip”, the software prompts if the user wants to start the separation. Click “yes” to start the separation.

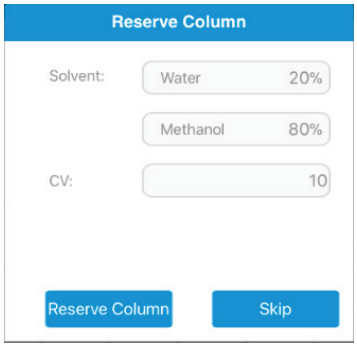


4.5 Chromatogram

This module is the same as that of normal phase separation. Please refer section 3.5.

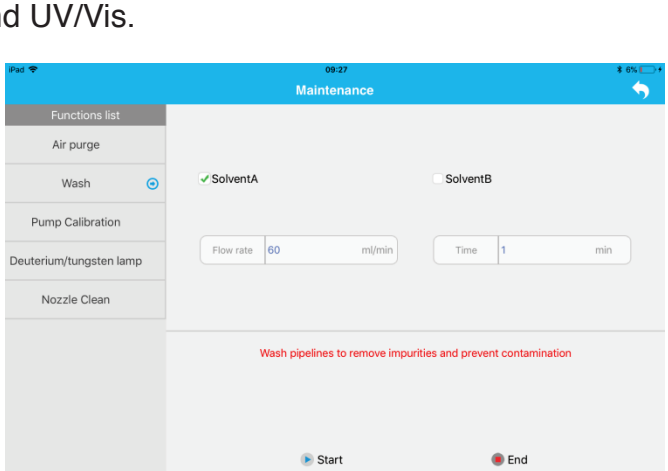
4.6 Reserve Column

When the separation is ended, the interface will pop-up a window to help the user to properly reserve the column. The user should input the gradient and CV, or the system will run with the solvents at the default ratio with no parameters. This process could effectively prevent damage to the flash columns.

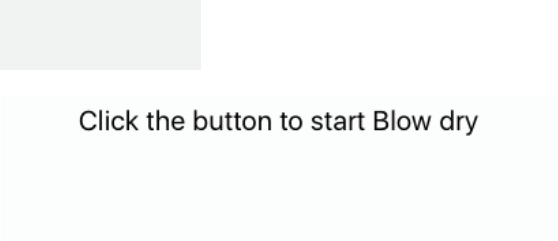


5. Machine Maintenance

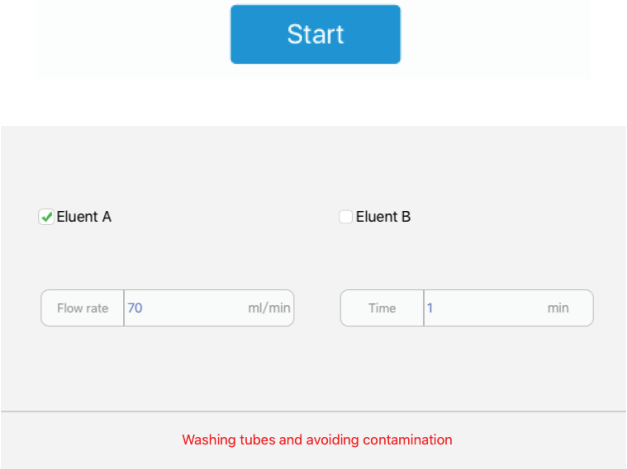
Click on the  icon to open the machine maintenance module. This module includes air purge, purge, pump calibration, and UV/Vis.



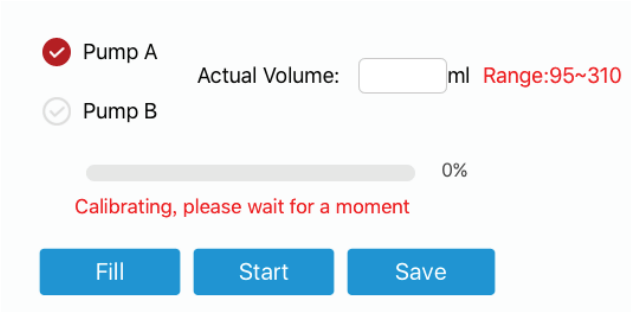
- **Air Purge** - Users can click on “start” to start the airflow through the solid load column and flash columns. This way both the columns will be blown dry manually.



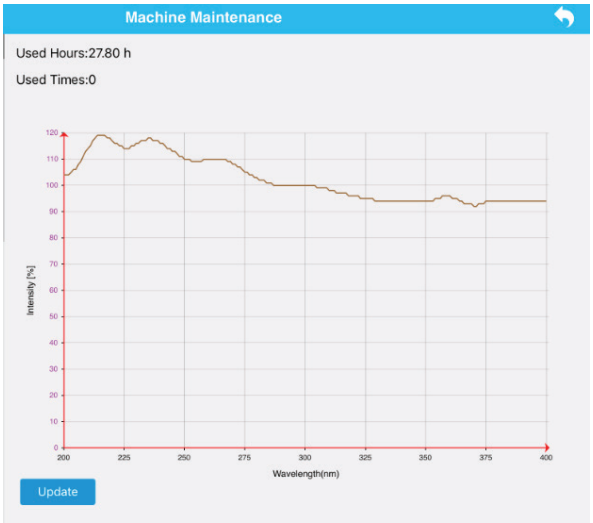
- **Purge** - Users can manually purge the solvent lines with the solvents on this page. Users should select one solvent at a time, input the flow rate and duration time to start the purging. Clicking on “stop” will stop the purging before the duration time ends.



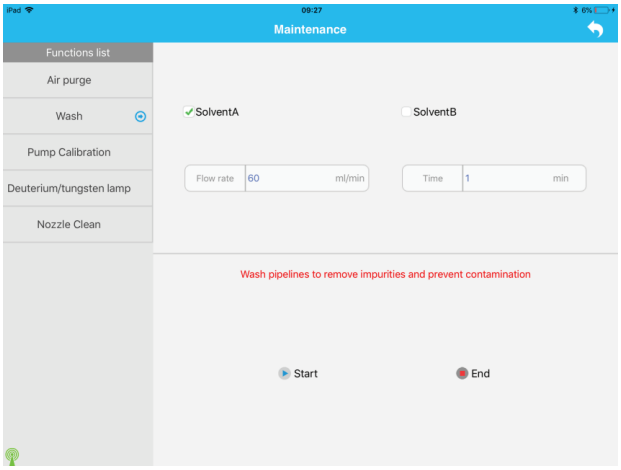
- **Pump Calibration** - To enable this function, users should remove the column and check if the tubings are properly connected. The user should select the pump to be calibrated and click on “fill” to start the pump. A 1000mL measuring cylinder should be kept at the waste tubing. Then, the user should click on “start” and collect the elutions in the measuring cylinder. At the end, the actual volume collected should be input in the blank box provided. The calibration must be saved by clicking on “save”.



- **Deuterium/Tungsten Lamp** - This page shows how many hours and times the light source (deuterium lamp or halogen lamp) has been used. Also, the characteristic peak profile of the light source is shown in the picture to the right for reference.



- **Nozzle Clean** - Select solvent A or solvent B and fill in the flow rate and run time, then click on the “adjust” button to adjust the nozzle position. Put a beaker or other container under the nozzle and click on the “start” button to clean the nozzle.



6. History

This page enables the user to access to all the past run files in chronological order. The user can browse through the records and choose a specific record to check the chromatogram and other method parameters related to the name. By opening a history record, users can review the experiment parameters, eluting method, the chromatogram, and the full-wavelength scanning spectrum.

It should be noted that the user can tap and slide on the chromatogram, as well as the full-wavelength scanning spectrum. By this operation, the user can check the fraction purity according to the characteristic absorbance peak of the chosen fraction.

Save Key

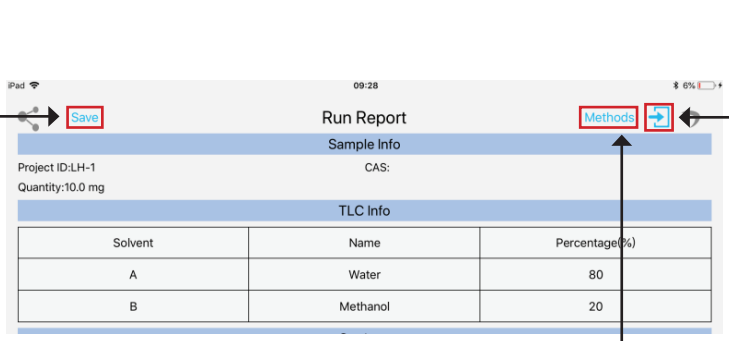
Save this interface as a picture so the user can print the picture

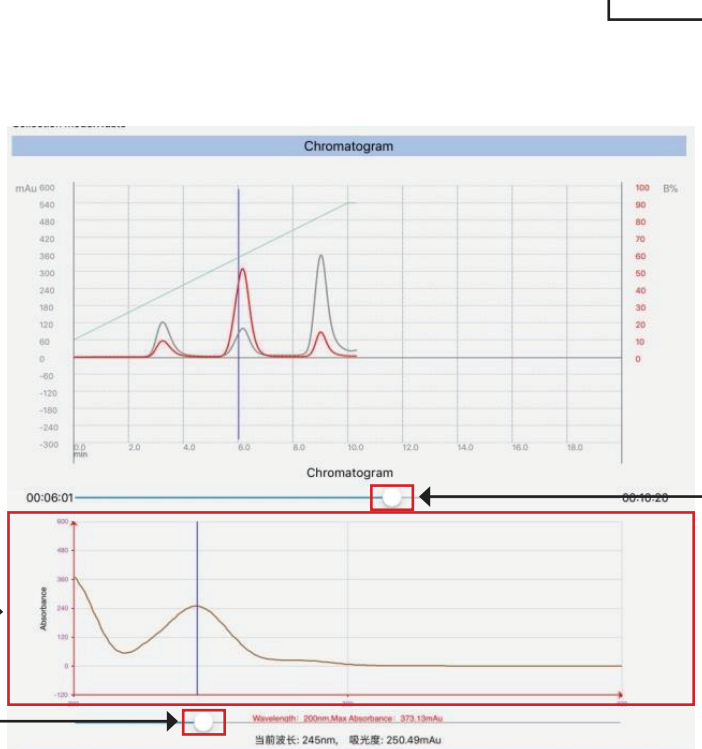
Reference Method Key

Click on this button to enable this method to fill in the separation settings automatically

Save Methods Key

Click on this button to save this method. Then, different devices in the same LAN can view this method





Slide this button to check the wavelength scanning spectrum of the current component

Wavelength Scanning Spectrum

Slide this button to check the maximum wavelength of the wavelength scanning spectrum

7. Appendix

SepaBean™ Machine Login Methods

The user needs to register an account on the ChemBeanGo™ website or app. Only e-mail registration is accepted for use outside of China in the current version.

Website: www.chembeango.com

App Download:

iOS: Search “ChemBeanGo™” in the App Store and download.

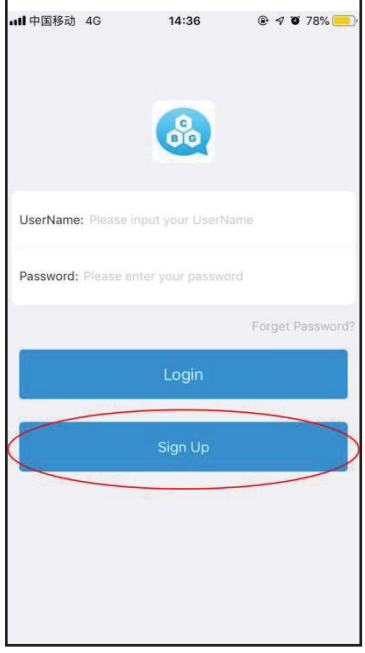
Android: Scan the QR code.

After registration, users can login to the ChemBeanGo™ app with a registered account.

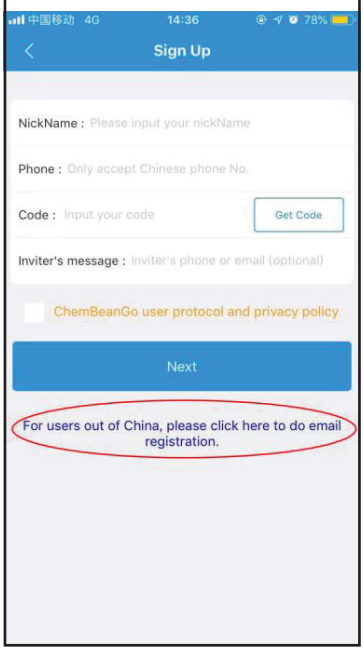
Login Methods

ChemBeanGo Register Instructions

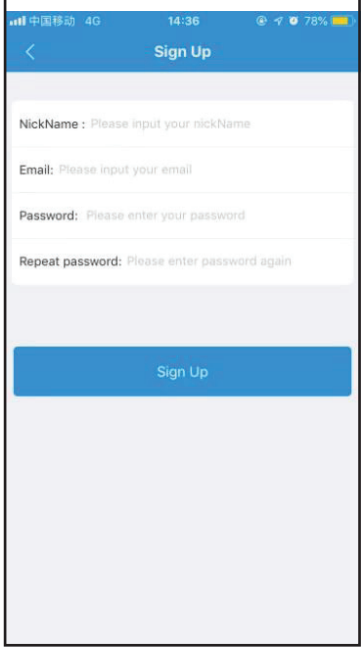
For first time users, you must create a new account. Start “ChemBeanGo” app, click “Sign Up”.



If you are outside of China, please use your email to register. Click the sentence in the screenshot.



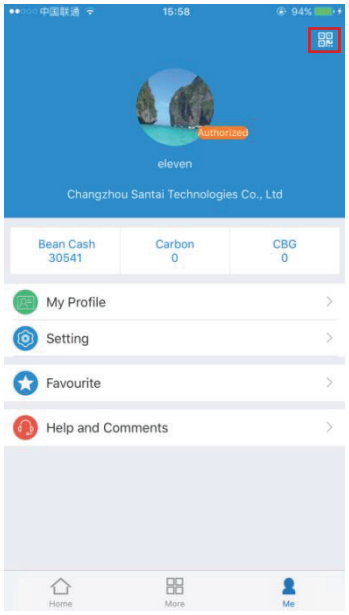
Next, fill in the necessary information, wait for the email verification code, and input it. Click “Sign Up”.



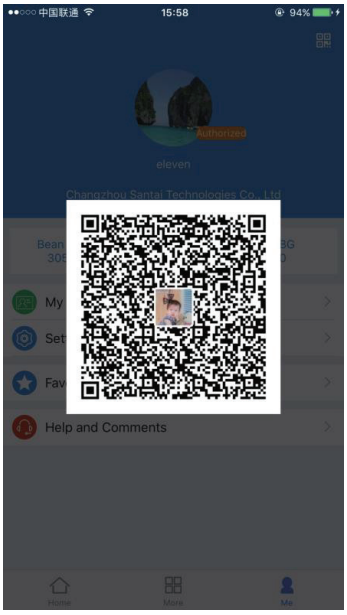
Finally, the registration is complete. Go back to the login page and use the email and password you registered. Then, login and go to the “me” page.

Login by scanning the personal QR code

For all registered users, each of you has a personal QR code on the top right corner of the “me” page.



Click the QR icon and the QR code will be shown.



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Website: www.sorbtech.com Email: info@sorbtech.com

For more information, please call us at 770-936-0326

Sorbtech
Separate. Purify. Discover.