

Vü Imaging Systems

User Manual



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Disclaimer

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Pop-Bio Ltd

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Safety precautions

Before using the Vü Imaging System read the following safety precautions and this manual. Proper operation of this system and its accessories with help to prevent injuries or damage to the equipment or users

Symbols used throughout this manual



This symbol indicates CAUTION. Not following instructions could lead to injury or damage to equipment or users.



This symbol indicates WARNING Not following instructions could lead to serious injury or death to users.

| | | Power Supply |
|-------------|---|--|
| \triangle | | Use only the power supply provided by Pop-Bio |
| \triangle | 4 | Ensure the mains power to all devices are switched off before connecting or disconnecting cables |
| \triangle | 4 | Ensure mains plug is fully inserted into socket to avoid 'arching' |
| \triangle | | If the device is not being used for long periods unplug it from the mains socket |
| | | Moving the device |
| \triangle | | Always select the 'park' position before moving the device |
| \triangle | | Do not drop the device |
| \triangle | | Place the device on a flat, dry surface |

| \triangle | | Avoid placing the device where it can get wet |
|-------------------------|---|--|
| \triangle | | Place the device away from any heat sources and direct sunlight |
| | | Maintenance and Service |
| \triangle | 4 | Ensure the device is unplugged from the mains before conducting any servicing or maintenance |
| \triangle | | Use only parts supplied by Pop-Bio Imaging |
| \triangle | 4 | Only suitably qualified persons should conduct any maintenance or service operation |
| | | Illumination |
| \triangle | | The Blue LED light present a low risk and in normal operation should not be visible to the user |
| $\overline{\mathbb{A}}$ | A | The UV tube does present a UV hazard however in normal operation should not be visible to the user |
| \triangle | | Avoid attempting to use the system in anyway where Blue of UV light is visible to the user. |

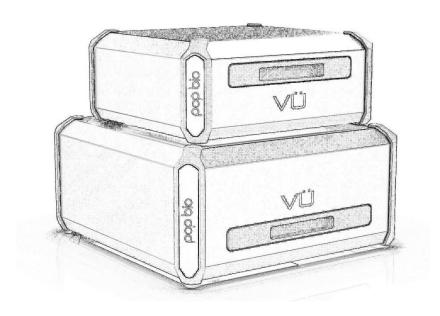
The Vü Imaging system

The Vü imaging system is available in two configurations.

Vü-C is for chemiluminescence applications. The unit has built in RGB LED's which are used for illumination of marker lanes on a blot. No other lighting is available. The Vü-C is used exclusively for chemiluminescent western blot applications

Vü-F is for Fluorescence applications. The unit has built in blue LED's and a 302nm UV tube. Blue light is used for a wide range of 'safe' dye applications. The UV is typically used for illuminating Etbr stained DNA gels. A white light converter is used to created transmitted white light.

This manual covers both systems



Key features of the Vü systems

- Highly sensitive sensor technology able to acquire low light level faint bands.
- A patented acquisition [applied for] system which automatically controls the sensor head unit and processes the acquired image.
- Simple interface to select method of operation and location of saved images.
- Very small foot print taking up minimal bench space
- Easy to install, operate and maintain.

System requirements

| The Vü system can be | operated as follows |
|----------------------|---------------------|
|----------------------|---------------------|

- The Vü system can be connected to a network either by ethernet cable or a WIFI adaptor. There is an ethernet port on the rear of the system and spare WIFI slots for a WIFI adaptor.
- It is also possible to connect the system directly to a computer using the ethernet cable. This is best when a stand-alone installation is required and is probably the most common setup.

The computer should have the following specification:

- PC using Windows 10 or higher
- 200 MB HDD or higher
- 512 MB RAM or higher
- Ethernet connection
- USB for software updates

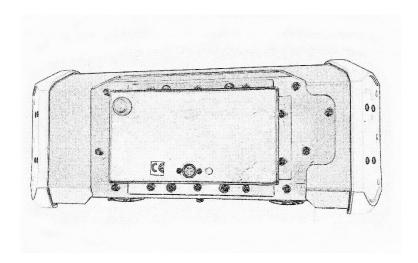
See the separate software installation instructions for connection to a network or stand-alone computer.

Getting to know your system

Front View



Rear View



Setting up the system

Unpacking the system

Carefully remove the unit from its box – remove any packing pieces that are used

Locate the following items included with the system.

Vü-C

- Black plastic blot cover plate
- Power supply
- Appropriate power cable
- Ethernet cable [yellow]
- Plastic tray for blot [10 x 10cm]

Vü-F

- Black tray for blue light applications
- Plastic tray for gels [20 x 20cm]
- Power supply
- Appropriate power cable
- Ethernet cable [blue]
- White light conversion screen
- UV transmitting glass spacer for use with very thin gels

Also supplied with the system[s] is a folder containing software installation instructions, memory stick with Vü software and this instructions manual, Analysis software licence information.

Check for any damage to the unit. Report any damage in the first instance to the shipper and then to Pop-Bio for further instruction or advice. Retain all packing material if the unit needs to be shipped later.

If there are any shortages contact Pop-Bio for instruction or advice.

Installing the unit

Place the unit on a clean, flat, dry surface with the front drawer being easy to reach.

Locate the 4-pin socket on the rear of the unit and plug the lead from the power supply into this.



Using the power cable supplied connect the power supply to a suitable mains outlet [do not switch on the mains socket at this time].

Plug the ethernet cable into the appropriate socket on the rear of the unit.



Plug the other end of the ethernet lead into a stand-alone computer. Depending on the type of computer you have you may need to use an adaptor [not supplied] to connect the ethernet lead to a suitable port.

Switch on your computer first.

Finding the Network name

The initial set up of the system requires that you use a stand-alone computer connected to the system. This is the most common set up. Later you may wish to connect the system to work on a network with each unit [if a Vü-F and a Vü-C are being used] being connected to the facility network using either an ethernet or WIFI connection method. For all connections you will need to use a Network name. This name is assigned to the unit during the manufacturing process. The initial network name given to each system is as follows.

For Vü-F the network name is: vu-fluor-serial number.

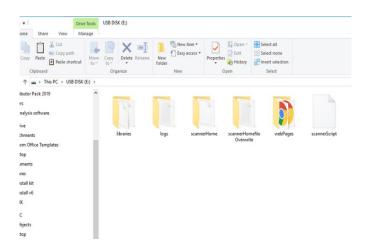
The serial number can be found on the plate on the rear of each unit. Therefore, system serial number 04000001 has been given network name **vu-fluor-04000001**. The next would be vu-fluor- 04000002 and so on.

For Vü-C the network name is vu-chemi-serial number.

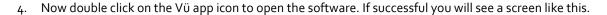
Again, the serial number can be found on the plate on the rear of each unit. Therefore, system serial number o3000001 has been given network name **vu-chemi-o3000001**. The next would be vu-chemi-o3000002 and so on.

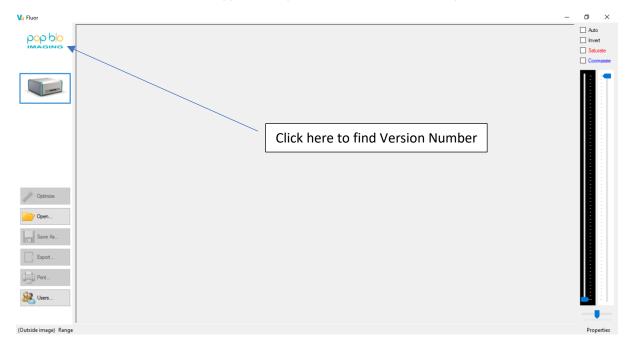
Installing the Vü Software application

- 1. The software can be found on the memory stick supplied with the system or downloaded from the Pop-Bio Imaging website at www.pop-bioimaging.com/download This is a zipped file. Copy the file to the computer.
- 2. Locate the file and now double click the Vü-app icon which will take you to the **Vü app setup** file. It should look like this...



3. Once you double click this setup file the software will be installed on your computer. Follow any onscreen instructions you are asked. When asked if you wish to put a Vü app icon on your desktop it is preferable to say YES





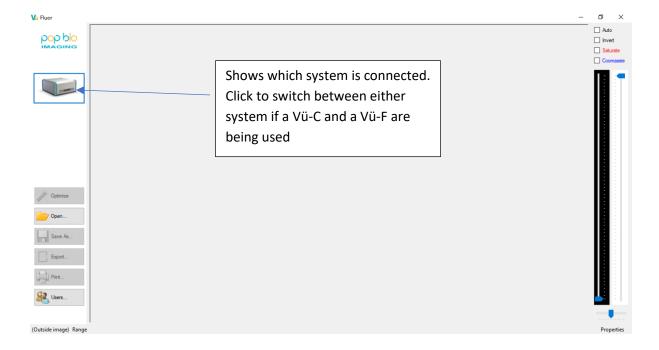
5. To check the version number, you have just loaded on your computer click the Pop-Bio Imaging logo on the top left-hand corner of the screen and see the 'About Vü' option. Click this to see the version number of your software.

Switching on the unit

- Now switch on the power from the mains socket to turn the unit on.
- The LED on the front of the unit will initially glow red while the system does its calibration and setup checks.
- The LED will turn Green when the unit is ready.

Connecting system to computer

The Vü software automatically finds the Vü system if it is connected using a direct link between the computer and system using an ethernet cable. This can sometimes take several minutes depending on the computer. Sometimes you can make the connection faster if you close and re-open the Vu Software. However, once found the computer 'remembers' this name and next time it will find the system immediately. Once connected the Vü interface will show an icon of the Vü system. If you are connecting a Vü-C and a Vü-F then click on the icon to change from one system to the other.



Using the Vü-F Imaging system

Open the Vü software

Select the system you want to use by clicking on the system icon. Select Vü-F [Blue]



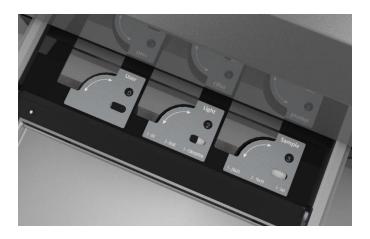
The following describes how to capture a UV image using the simulated gel supplied with the system. We advise all those new to the system to follow this procedure first to learn how the system operates

1. Plug out the drawer from the front of the unit as far as you can. It will reach a 'stop' point.

Take the clear UV plastic tray and place the glass plate spacer on the tray with your simulated gel FACE UP on top of the glass spacer. Make sure the gel is centralised in the centre of the tray. [When using thin gels, the spacer is needed - for all other gels you do not need to use the glass spacer].

For other applications:

- For white light applications use the white light converter [orange screen]. Place the orange screen on the UV clear plastic tray [BRIGHTER ORANGE SIDE FACE DOWN] and the gel on top of the screen.
- For blue light applications use the black tray for blue light [safe dye] illumination.
- 2. Place tray back into the drawer making sure it is located on the 'pins'. Failure to do this could result in the drawer getting jammed.
- 3. Using the thumbwheels on at the back of the drawer select the following:
 - Lighting required [UV, Blue, White conversion] position 1, 2 or 3 Select 1
 - Sample size [20 x 20cm, 10 x 10cm, 5 x 5cm] position 1,2 or 3 Select 2
 - User [numbers o-11]. If you are not using user ID then this should be set to position o. [See more about User ID later] **Select o**



- 4. Push the drawer in all the way.
- 5. The system will start automatically

The system will now begin its process. The time it takes will depend on the application and the default exposure times – more later about exposure times. In this first test you will be using the factory set default settings for the simulated gel.

The image will appear on the computer screen in the Vü software. [Alternatively, it will be sent to another location that has been nominated during the set-up process – see later]. More on how to save and manipulate the gel image is described later.

Open drawer and remove the tray with your gel. Remove your gel.

Ensure the tray is clean and dry for the next user. The tray can be washed to remove any residual fluids when using wet gels.

Now we suggest you try using the system with a gel.

Switching off the Vü

If you wish to switch off the Vü then you need to follow this procedure.

Locate on the rear of the unit the 'park' button



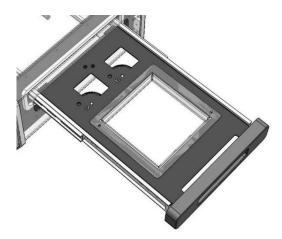
Hold this button in for 2-3 seconds

You may hear the sensor head moving to its park position. Once parked the indicator light glows solid red. In this position the unit can be switched off from the mains and safely moved to another location. Switching the unit back on moves the sensor head out of the park position.

Using the Vü-C Imaging system

Open the Vü software

Select the system you want to use by clicking on the system icon. Select Vü-C [Yellow]



The following describes how to capture a UV image using the simulated blot supplied with the system. The simulated blot requires natural light to activate it. The output will decay slowly to start with and then more rapidly. We suggest you 'activate' the blot each time you wish to use it by placing it close to a window or desk light and then to start the capture process without delay. We advise all those new to the system to follow this procedure first to learn how the system operates

1. Plug out the drawer from the front of the unit as far as you can. It will reach a 'stop' point.

Using the clear plastic tray place your simulated blot anywhere on the tray. Place the blot in the plastic tray – **FACE DOWN**. Now place the black plastic blot cover over the blot. The Vü-C maps from below and hence is looking upwards. Hence the need to have the **blot facing downwards**.

Blots are often imaged still in their saran wrap covering in which case these tend to curl up or add additional thickness. When not in any wrapping blots can also tend to dry out and can curl. Hence, by positioning the blot face down but with a plastic cover on top it keeps the blot perfectly flat and moist. We recommend that saran wrap is always removed.

- 2. Place tray back into the drawer
- 3. Using the thumbwheels at the back of the drawer select the following:
 - Light select whether you are using:
 - 1. Chemi only, Select this option for the simulated blot
 - 2. White light for marker lane only,
 - 3. Coloured light for coloured marker lane only,
 - 4. Chemi+ white light marker lane,
 - 5. Chemi + coloured marker lane.

• User [numbers o-11]. If you are not using user ID the setting should be o. [See more about User ID later]



- 4. Push the drawer in all the way.
- 5. The system will start automatically

The system will now begin its process.

The Vü-C will very quickly search the 10 \times 10cm area to find the blot and fix this as its scanning area. Blots of all sizes can be detected. The system does this by looking for the white outlines of the blot. This does mean that you **MUST** use a black background when using this auto detect feature and you will need to use a real blot or the white simulated blots. You cannot use the transparent blots on the Vu-C.

The 'auto-find' function takes about 5-10 seconds to complete during which time you will hear the motor being activate.

After the 'auto-find' function the system now does a 'Fast Scan' to evaluate what is on the blot, its position and if the signal is decaying. By default, this is set to 60 seconds. [It can be set to as little as 10 seconds but the normal default is 60 seconds. Some users may have very fast acting blots – possibly their chemi reaction may take less than 30 seconds and hence this setting can be set to suit users' requirements].

An initial image is displayed on the screen showing the size of the blot and some of the bands it has found.

Finally, the system will now make a more detailed scan of the blot. Depending on the information it has gathered during the Fast Scan the system will now select an appropriate exposure time. This scan will take more time. At the end of this scan the image already being displayed will be updated with more signal.

The system has a default called 'Maximum Acquisition Time' which has initially been set for 600 seconds. This means that depending how much time is left after the previous processes have taken place AND if the system considers there is still an opportunity to collect more data it will do a 2nd scan. Again, it will set its own exposure time based on the data.

The system will continue to do more scans up until the Maximum Acquisition Time is up or if the signal has fallen too low to be of any significance to the data already collected. After each scan the image on the display will be updated with more data.

With the simulated bot expect about 2 scans before the process stops. Note that during the 'Maximum Acquisition Time' the user can click the 'Accept' button to stop the process.

Note: if the system has been set up to automatically save images then each image captured during the capture process will be saved [series capture] so that the user can review the images collected. Since each image is an accumulation of the previous ones then the last image should always have the most data.

- 6 Open drawer and remove the tray insert with your blot. Remove your blot.
- 7 Ensure the tray is clean and dry for the next user. The tray can be washed to remove any residual fluid.

Now we suggest you use the system with a 'real' blot.

Switching off the Vü

If you wish to switch off the Vü then you need to follow this procedure.

Locate on the rear of the unit the 'park' button



Hold this button in for 2-3 seconds

You may hear the sensor head moving to its park position. Once parked the indicator light glows solid red. In this position the unit can be switched off from the mains and safely moved to another location. Switching the unit back on moves the sensor head out of the park position.

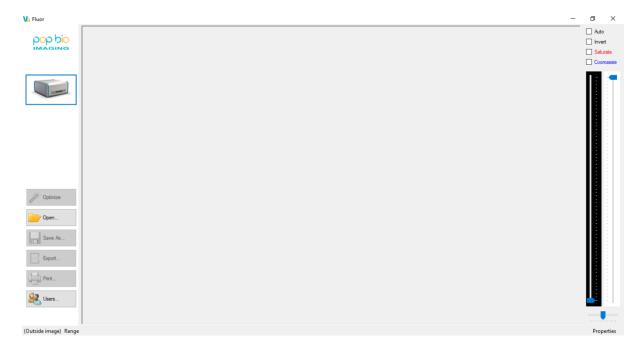
Understanding the Vü software

The Vü systems are built in such a way that the user has the need to control very little. All of the image acquisition and processing is automatic with the intelligent software making the decisions depending on the application/lighting selected by the user. There is no need for focussing, filter selection, sensitivity level selection or exposure time control. This is all programmed into the system for automatic control. Hence the Vü software is very simple and has just a few basic user settings.

Launching Vü software

Once the software is loaded and installed on a computer you can open it by clicking the program icon.

Main Window



Device type



The icon indicates the type of scanner you are connected to. If this is blank, you are not connected to a scanner – check the hostname you are using (see 'Network Settings') and the network connections to and from your computer, and to and from the scanner.

Image display

Zoom

To increase or decrease zoom: roll the mouse-wheel forwards or backwards.

On a laptop, the touchpad may offer different functionality depending on the laptop model and settings:

- the "pinch" gesture: touch the pad with two fingers then open or close fingers to control zoom
- the "scroll" gesture: touch the pad with two fingers held together, then move both fingers together vertically to scroll
- touch the right-hand edge of the pad with one finger and move it vertically along the edge to scroll

Image adjustment

Auto

Click the Auto box to activate the auto-contrast function. By default, this is switched OFF.

When the system captures an image, the software determines the black and white levels that are needed to display a balanced image. Once you have the image you can then use the vertical black and white sliders to enhance it further to the user's personal preference.

It is possible to activate the auto-contrast process by ticking the box ON if so desired.

IMPORTANT TIP

Using the Auto Contrast feature is a very good way of making bands brighter or for making bands more visible which are otherwise not seen on the original captured image. Suppose you capture and image but you think you want to make it brighter or look for 'hidden' bands.

- Turn off the 'Auto' button
- Now zoom into the image so that the faint bands only fill the screen
- Now turn on the 'Auto' button
- Watch as the bands get brighter and brighter.... Automatically....
- Turn off the 'Auto' button again and now zoom out to the normal size
- You should be left with a much brighter image especially in any previously faint area.

Sliders

There are 2 vertical sliders for black and white adjustment. The user can adjust these to an optimum position which suits their own preference.

The horizontal slider below the 2 vertical sliders is the gamma adjustment. Moving this slider will re-distribute the tonal levels of the image making it more acceptable to an individual's perception of what an image should appear like. This is a subjective setting and depends on a person's eyes and how they see black and white. Tooltips on each slider display the black, white and gamma levels as a number.

In all cases where the black, white and gamma levels have been altered it will in no way effect the 16bit TIFF image that can be saved. This is still the image that needs to be used for analysis. Any changes to the image are only saved [or printed] when it is exported as an 8bit TIFF, BMP, JPG or PNG file.

Sat

Click this on to see if the captured image has saturated. Click off to remove this.

Invert

Ticking this **ON** it will reverse the black and white levels of the image. By default, an image captured with the Vü-C or using the white light converter with the Vü-F will result in black bands on a white background – which is the more usual presentation for this sample type.

Coomassie

To capture a Coomassie blue gel you would use the Vu-F and the **W**hite **L**ight **C**onversion screen. Place the Coomassie stained gel on the WLC and select position 3 – Converted - on the thumbwheel Once the image is captured you will see this as a black on white image. If you now tick the Coomassie box to **ON** the image will be displayed in its more usual blue colour. You can save or print this image with its blue colour.

User controls

Open

Vü can open previously scanned images.

Click the **Open...** button to present the Open File dialog, then navigate to find your image file, then click the **Open** button to view that image.

Vü cannot read any image file format other than 16-bit TIFF.

Save As

Vü can export a copy of a scanned image to another file location, perhaps to a USB drive or a network location. Click the **Save As...** button to present the Open Save dialog, then navigate to a destination folder for your image file, then click the **Save** button to save the displayed image to a file.

Vü can only export 16-bit TIFF format.

When saving a chemi+marker image then 3 separate files are saved – see later how to view these images.

This button is disabled when there is no displayed image.

Export

Vü can export a copy of a modified image to another file location, perhaps to a USB drive or a network location. This means that what you see on the screen is exported so that any image where the levels have been altered or if it has been inverted can be saved.

Click the **Export** button to present the Open Save dialog, then navigate to a destination folder for your image file, then click the **Save** button to save the displayed image to a file.

Vü can export as an 8bit TIF, PJG, BMP or PNG image. Images cannot be re-imported back into Vü software This button is disabled when there is no displayed image

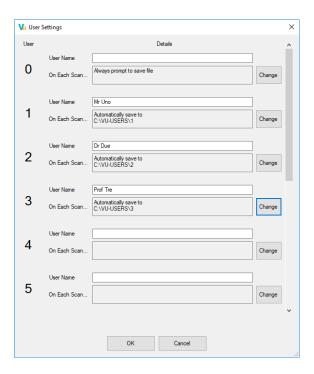
Print

Vü can Print a copy of a modified image to any connected printer. This means that what you see on the screen is exported so that any image where the levels have been altered or if it has been inverted can be printed.

Click the **Print** button to present the Printer dialog, select the printer where you want the image to be printed and then click the **Print** button to send the image to the printer.

Users

On the main window, click the **Users** button to present the User Settings dialog:



User list

A list of users corresponds to numbered positions on the tray thumbwheel.

Change

Click the **Change** button for any user to change automatic behaviour on each scan.

A menu is presented:

Automatically Save All Scans to Folder...

A Browse Folder dialog is presented.

Navigate to a location on your computer or on a shared network drive.

As each subsequent scan is completed for this user, it will be saved automatically as a new file in the same folder.

• Always Prompt to Save Each Scan

As each subsequent scan is completed for this user, the Save File dialog will be presented automatically.

Clear settings for this user

The user name and save action will be cleared for this entry on the list, cancelling any automatic save actions for this user.

User o

The settings for **User o** (number zero) are often used as a "default" or "catch all" if lab users have not yet set up their own save locations. It is recommended that **Always Prompt to Save Each Scan** would be appropriate for this user.

OK

Click the **OK** button to save changes and dismiss this dialog, to return to main window.

Cancel

Click the **Cancel** button to dismiss this dialog without saving changes, to return to main window.

Optimize - Vu-F only

Optimize tries to adjust the exposure conditions to achieve, within the visible region of the image, a peak image intensity that's within the optimal range.

First, choose the region of your image that you want to optimize for. (For example, there may be faint bands within the image that you want to examine specifically. In that case, zoom in until just these bands are visible before you press Optimize. To optimize the whole image, first zoom out so that the whole image is in view.)

You may find that after capturing an image, Optimize is disabled (greyed out).

- If the peak image intensity within the visible region of the image is already within the optimal range, Optimize is disabled.
- If the exposure would need to be increased but the picture was captured with the maximum available exposure, Optimize is disabled.
- If the exposure would need to be decreased but the picture was captured with the minimum available exposure, Optimize is disabled.

Note that, because Optimize works on the area of the image that is visible, the button may enable or disable as you zoom in or out or change the field of view – because the intensities are different within those different regions.

Once you press Optimize and the process starts, the system will try to adjust the exposure conditions and rescan until the peak intensity is within the optimal range.

If the exposure needs to be increased, the whole image will be re-scanned.

If the exposure needs to be decreased, Optimize may choose to re-scan a reduced region containing only the bright parts. Once appropriate exposure settings are determined, it will re-scan the entire (original) region.

Optimize may first increase and then decrease the exposure time (or vice versa).

Additional set-up menu

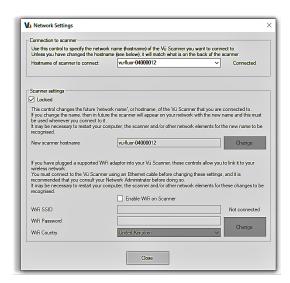
There is an additional setup menu that can be accessed. Click the Pop Bio Imaging logo on the top left-hand corner of the screen.



Here you will find

- Manual scan
- Network setting
- About V

Network settings



Host name

Use this control to specify the network name (hostname) of the Vü Scanner you want to connect to. Unless you have changed the hostname (see below), it will match the serial number of the system.

Vü-F

Serial number format is vu-fluor-o4xxxxxx

The number if found on the rear of the unit

Vü-C

Serial number format is vu-chemi-o3xxxxxx

The number is found on the rear of the unit.

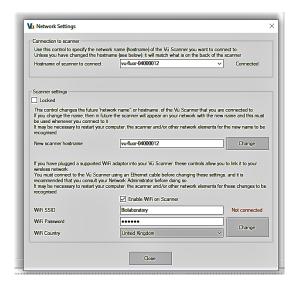
If the scanner cannot be located on your network, Trying to connect appears next to the hostname.

As you enter the name of your $\mbox{\sc V\"{u}}$ scanner, the connection is retested continuously.

When the scanner is located on your network, **Connected** appears next to the hostname.

Unlock scanner settings

Uncheck the Locked checkbox under scanner settings to access the settings for the scanner itself:



Scanner host name

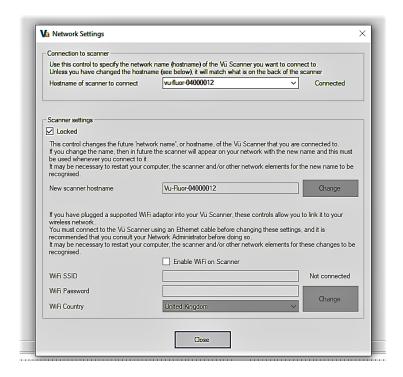
This control changes the future 'network name', or hostname, of the Vü Scanner that you are connected to. If you change the name, then in future the scanner will appear on your network with the new name and this must be used whenever you connect to it.

It may be necessary to restart your computer, the scanner and/or other network elements for the new name to be recognised.

Enter the new name for your scanner then click the **Change** button.

Scanner WIFI

Check the **Enable WIFI on Scanner** checkbox to access the WIFI controls:



If you have plugged a supported WIFI adaptor into your Vü Scanner, these controls allow you to link it to your wireless network.

You must connect to the Vü Scanner using an Ethernet cable before changing these settings, and it is recommended that you consult your Network Administrator before doing so.

It may be necessary to restart your computer, the scanner and/or other network elements for these changes to be recognised.

WIFI SSID, WIFI Password, WIFI Country

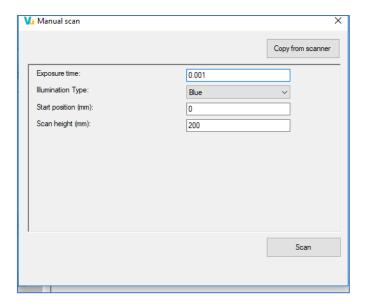
Enter the codes exactly as specified by your Network Administrator, then click the **Change** button.

If you have changed the scanner to another network, after you disconnect the Ethernet cable you will have to configure your computer to also use the same network.

Close

Click the **Close** button to dismiss this dialog and return to the main window.

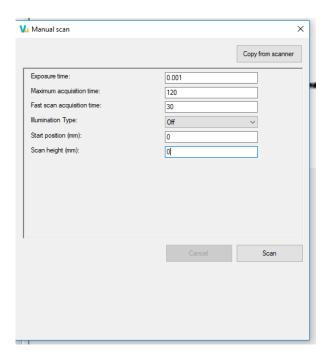
Manual scan



This Manual Scan dialog box shown here is for Vu-F only. Here you can set just the exposure time, illumination type and scan distance. In the example this has been set for an exposure time of 0.001 seconds, using blue light and for a total scan distance of 0 – 200mm.

Once you have made your settings press scan and push the drawer in. The system will then complete one scan using these settings. Unless you repeat this process the next scan will revert to the default settings.

NOTE: Images are NOT automatically saved when using the Manual Scan function. You will need to manually save the images if you wish to keep them.



This Manual Scan dialog box here is for Vu-C only. Here you can set just the exposure time, illumination type maximum exposure time, fast scan exposure time and the scan distance. Leaving the scan height as zero will activate the auto-blot-finding process.

In the example this has been set for an exposure time of 0.001 seconds, using no light, a fast scan of 30 seconds and a maximum acquisition time of 120 seconds.

Once you have made your settings press scan and push the drawer in. The system will then complete one process using these settings. Unless you repeat this process the next scan will revert to the default settings.

NOTE: Images are NOT automatically saved when using the Manual Scan function. You will need to manually save the images if you wish to keep them

About Vü

To check the version number, you have just loaded on your computer click the 'About Vü' option. Click this to see the version number of your software.

Working with gels and blots.

The Vü system is designed for rapid and easy imaging of all gel and blot types. The capture settings are all pre-set and have been determined by Pop Bio Imaging as the typical conditions for capturing a range of image types. Systems leave the factory with pre-set defaults which will enable the user to obtain good images of the simulated blot and gel. These settings are also good for the majority of real gels and blots. However, we recognise that not all gels and blots are the same and that sometimes settings need to be changed or the captured image needs to be displayed in a different way. It is important to always use good quality gels or blots to try and get the best image anyway. Ideally gels and blots should be 'fresh' otherwise the signal output decays with time and the bands become diffused. Capturing images of old or diffused material is never a good idea. Therefore, always use new samples – never try to use something which is even several hours old. This quide will help you get the best from your Vü system.

Image capture with the Vu-F Fluorescence system

The Vü-F systems have been pre-set with a different exposure time for each lighting setting – UV, Blue, converted [when you set the thumbwheel position]. Usually you can place your gel on the correct tray, place it in the drawer and simply push the drawer in to capture the perfect image.

Once you have captured an image you can see its exposure time by clicking 'click for more' which can be found at the bottom right of the screen. The information also contains other details about the image such as date, time, user, size, resolution.

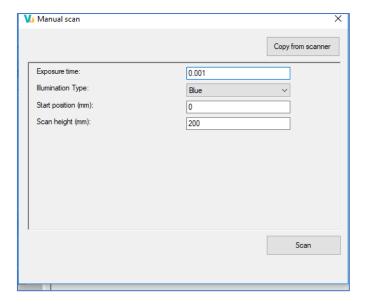
What if I am not happy with the image?

If you think the image is too bright or too faint, then you have some options.

Changing the exposure time

By making the exposure time longer or shorter you can capture more [or less] of the signal coming from the bands. If the bands are too faint then increase the exposure time. If they are too bright then decrease the exposure time.

To make a temporary change to the exposure time click on the Pop-Bio Imaging logo to see the drop-down menu. From this select 'Manual Scan'



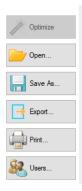
Firstly, select the illumination type you are working with – UV, Blue, Converted.

If the Exposure time box you will see what the default setting is for this illumination type. Now type is a new exposure time you wish to use – either a lower or higher number depending if you want to reduce or increase the image brightness. Now click 'Scan', push the drawer in [if not already in] and the system will do a new scan with the altered exposure time. The system will only use these settings once and will revert to the saved defaults for the next scan.

Changing the exposure time using this settings box does not permanently change the setting for the thumbwheel. It will retain its factory setting the next time you make a capture of a gel or blot. If you want to make a permanent change to the exposure time for a particular thumbwheel setting, then you need to use the 'Configure Thumbwheel' dialog box. See how to permanently change the exposure time later in this manual.

Using the Optimize function

There is another way of changing the exposure time by using the automatic functions of the system. On the left-hand side of the screen you will see an '**Optimize'** button. If you have captured an image and you think the bands are too faint or too bright then rather than changing the exposure setting as described above, you can use the Optimize function instead.



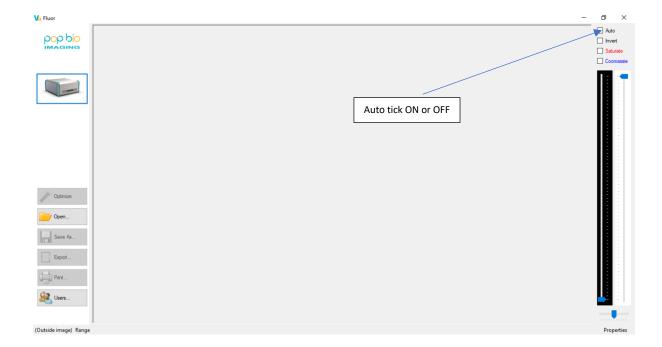
When you click Optimize the software analyses the image you have just captured. It then measures the signal levels. It will then automatically change the exposure setting [either up or down] to what it thinks will give you the best image based on the signal it sees.

Once you have the Optimize image it is a good idea to note the exposure time used [use the 'click for more' information on the bottom right hand side of the screen]. You can either use this exposure time when making 'Manual scans' in future or you may want to use this as a permanent exposure time and therefore configure the thumbwheel with this time as the setting.

Using the image enhancement functions- automatically

The Vü software has some powerful image enhancement functions to help present the best image for the user. This is the best way of producing an enhanced image rather than changing exposure time. This is because typically the Vü system will have already gathered a lot of signal from the gel anyway and all the image really needs is some adjustment to help viewing. The data contained within the image is already fine for analysis but here we are looking at the ways of improving the visual output. Here is how to use the most powerful of these enhancement functions-

If you capture an image with the 'Auto' button ticked **ON** then the image that is captured will be displayed on the screen using the full range of black and white levels – usually 65536 grey levels.



All the grey levels have been used and the image 'stretched' to do this. The result can sometimes seem to reduce the brightness of the bands at the expense of other not so important parts of the image. The simple trick here is to zoom into the image to focus on the areas of the gel or blot which you think are important.

The important thing to notice here is that the faint bands become automatically brighter as you zoom further into the band[s] of interest.

Now un-tick the 'Auto' button and zoom back out to show the full gel again

Now you will have a perfect image. You can now print or export this image exactly as you can see on screen using the buttons on the left-hand side of the interface.

Using the image enhancement functions- manually

The default setting is to have 'Auto' switched off. When you capture an image, it is displayed using the black, white and gamma levels which the system decides is best. However, we do not all see the same thing and so you can manually set the levels to what is best for your eyes and the computer display itself. To use these, untick the 'Auto' box if ticked if not already done so. You will find 3 sliders on the right-hand side of the display – their functions are:

- Pixel intensity mapped to black
- Pixel intensity mapped to white
- Gamma

Use your mouse pointer to see the tool tips telling what each slider does

Using a combination of these 3 sliders you can adjust the image to what you want to see. Everybody has a different idea of their perfect image but with these 3 sliders you can make the image suit your own purpose.

There is also an 'Invert' button so that you can reverse the black and white levels.

Again, you can now print or export the image exactly as you can see on screen using the buttons on the left-hand side of the interface.

Image capture with the Vu-C Chemiluminescence system

When capturing a blot image, the system is set up to use its 'auto-chemi' functions to get the best signal from the blot over time.

'Auto-find'

By default, once the capture process is started [blot placed on tray and drawer pushed in] the Vü-C will very quickly search the 10 x 10cm area to find the blot and fix this as its scanning area. Blots of all sizes can be detected. The system does this by looking for the white outlines of the blot. This does mean that you **MUST** use a black background when using this auto detect feature and you will need to use a real blot or the white simulated blots. You cannot use the transparent blots on the Vu-C.

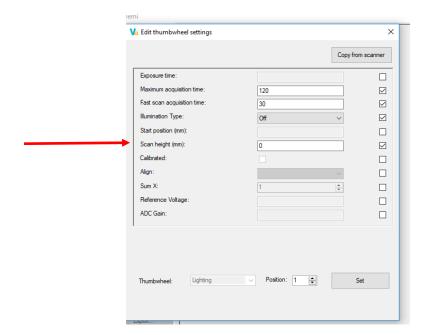
The 'auto-find' function takes about 5-10 seconds to complete during which time you will hear the motor being activate.

The system is by default set-up to use 'auto-find'. You can check this is ON or turn the function off if you wish. You can check this by looking at the 'Configure Thumbwheel' menu. Firstly, make sure that the software is connected to the Vü-C – you will see the Vü-C image on the left-hand side of the interface if it is.

To access the 'Configure Thumbwheel' menu press Ctrl/Shift and click the cursor on the Pop Bio Imaging logo at the same time. Then you will see the options-

- Configure Thumbwheels
- Manual Scan
- Commission

Click 'Configure Thumbwheels to open the following dialog box



Look at the 'Scan height (mm)' box – if there is a Zero [o] in the box this means that the 'auto-find' blot function is selected and you need do nothing more. If on the other hand you want to revert to scanning the whole of the 10 x 10cm area then make sure you type 100 into this box. However, most users will want to keep the automatic 'Scan-for-bolt' function as their default.

'Fast Scan'

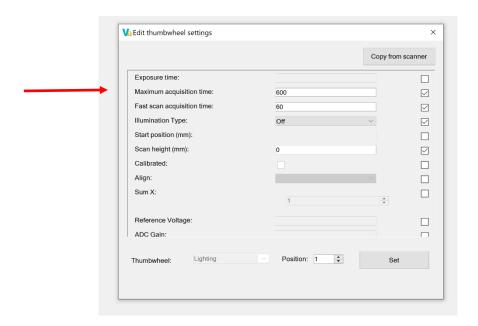
By default, the Vü-C has been set to do a rapid capture in 60 seconds. You can check this setting by going to the 'Configure Thumbwheel' menu as described above. [Note the example shows the 'Fast Scan' set to 30 seconds. This is a typical setting for use with the simulated blot and those rapid blots which decay after only 30 seconds]. The 'Fast Scan' option of just 30 seconds may be too short for some reagents. Maybe the reagent takes longer than 30 seconds to have a strong signal output. Hence, a longer process time would be better. For example, the reagent may have its strongest output at say 2 minutes. For the majority of blots a setting so 60s is typical but you can change it to what you want.

After the quick initial 'auto-find' process, which takes under 5 seconds, the system now does a 'Fast Scan'.

The purpose of this rapid scan is to gather signal from the blot which is then displayed on the screen. The system can then decide from the level of signal and the decay rate if it can gather a stronger image by making a 2nd capture.

'Maximum Acquisition Time'

After the 'Fast Scan' the system now has enough information to do another scan. To ensure there is a time limit on how long the system takes to do this a 'Maximum Acquisition Time' is set by default this is set to 600 seconds. You can check this setting or change it by going to the 'Configure Thumbwheel' menu as described before.



You can change this to a longer time or shorter time depending on the type of reagent in use. However, the recommendation is to leave the setting at 600s.

Explanation of the process

Assuming that the Thumbwheel settings have been left with their factory defaults. – this is the normal automatic settings for the system. The system will then operate in Automatic mode using those pre-set parameters.

- 1. Ensure that the Vü software is connected to the Vü-C
- 2. Place the blot FACE DOWN on the clear tray -anywhere will do
- 3. Place the Black plastic blot cover on top of the blot press to remove any bubbles that may be present.
- 4. Select thumbwheel position 1 for 'chemi'
- 5. Push the drawer in
- 6. A very rapid initial scan takes place to locate the position of the blot- this takes around 5 seconds during which time you may here the motor operating
- 7. Now the system will start it's 'Fast Scan' since the default is set to 60 seconds this will be the length of time it takes to display the 1st image.

During this fast scan the system will be calculating what the signal output is doing and what the decay curve looks like. If after this fast scan the system decides that there is enough signal AND there is little point in continuing because it can see the signal decay is rapid then the system will stop and just display its first image [this would be typical when capturing images from very fast reagents which decay quickly]

However, many commercial Western Blot reagents exhibit much longer decay times -1 hour or longer in some cases although the best signal output is more usually up to 5 -10 minutes. When the Vü-C is set with a 'Maximum Acquisition Time' [default 600s] the system will continue to do repeat captures after the first fast

image up to the maximum acquisition time. You will therefore see a series of progressive image updates being displayed with each one having more signal than the previous one. There could be 2, 3, 4 or more scans depending on the signal. Each time the image displayed is updated

This will continue as follows-

The system will stop after the first image has been captured because it determines this is the best it can get.

Or

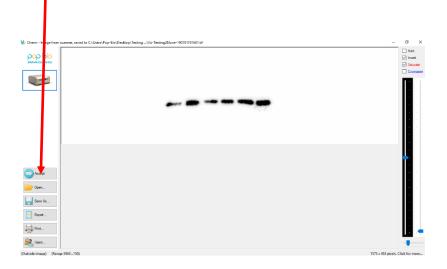
Image captures will continue up to the default time - in this case 600s

Or

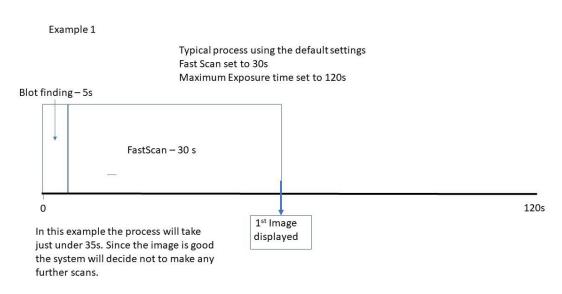
The system detects that because the signal has decayed beyond a certain level it is no longer useful to continue

Or

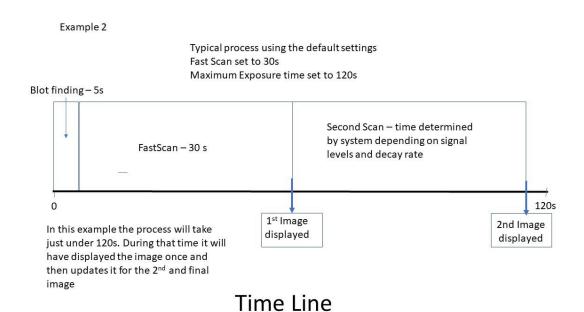
The user is happy with what they are seeing on the screen [maybe 2nd, 3rd or 4th image] and clicks the 'Accept' button. This will stop the process.



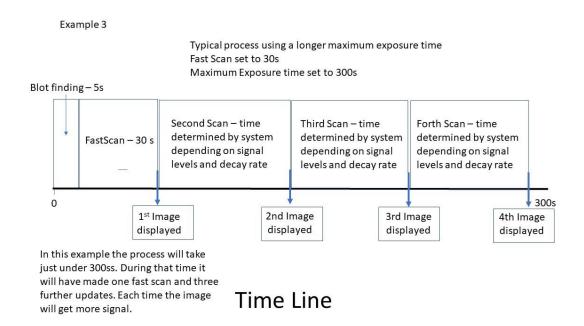
In summary here is what happens – note the different defaults used.



Time Line



VÜ IMAGING MANUAL V5.0



After the system has completed its process images will appear on the computer screen in the Vü software.

If you have set up the system to save images then EVERY image captured will be saved so that you can browse through each image should you wish to use anyone in particular. This is the 'series capture' mode.

When capturing images of the blot and marker lanes you will see the chemiluminescent blot displayed first. Right click the mouse cursor within this image to switch from the main image to marker image or composite image. Each image is saved – Main, Marker and composite.

If you wish to re-open a set of images and view the main/marker and composite again you need to follow the following sequence.

- Open the main blot file
- Press the control key as you open the marker file

You can now view main/marker/composite as before.

Taking care of your Vü

Keep your Vü clean to ensure optimum performance

Cleaning the gel/blot tray

- Remove the tray insert and wash under running water after use. This will avoid long term build-up of dried liquids on the surface of the glass.
- Dry using a non-abrasive cloth or a paper towel.
- Do not use any detergents or solvents to clean the surface.

Maintenance and settings

The Vü has very few parts that require servicing. In the unlikely event of a problem then only suitably qualified service personnel should attempt to remove the cover of the unit. The only routine maintenance that can be undertaken by the user is to change the UV tube in the Vü-F system

Changing the tube

- 1. Ensure the Vü-F has been set to 'park'
- 2. Switch off the unit from the mains supply and remove all cables
- 3. Turn the unit upside down and place it on a firm flat surface. Make sure there is nothing on the surface which could scratch the top of the unit.
- 4. Locate the plate covering the tube holder and remove the screws.
- 5. Inside you will find the UV tube. Simply twist and pull the tube out from its fixing.
- 6. Replace with a new tube and twist it into position in the tube holders.
- 7. Replace the cover plate
- 8. Turn the unit back onto its feet, reconnect and make a test exposure using the UV setting to test that the tube is functioning correctly.

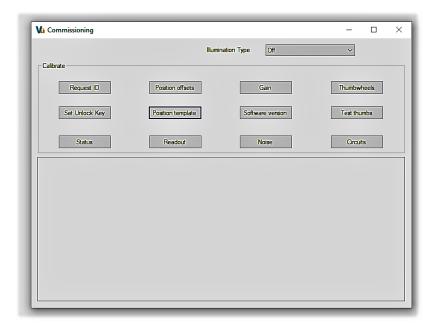
Making adjustments to the operation of the system

To access the settings box where the exposure time and other settings can be changed place the cursor on the system icon [below the Pop-Bio Imaging Logo on the left-hand side of the screen]. Then press control+ shift at the same time.

The following choices will be given: -

- Manual Scan
- Configure Thumbwheels
- Commission

Commission - Service Engineer only



This dialog box is reserved for the use of a trained Service Engineer only. Please see separate service instructions for the use of the options in the box.

Vu Thumbwheel calibration instructions

Calibration

- Start with the drawer open
- Use the Vü app. Open the Commissioning dialog (CTRL + SHIFT + click on the scanner icon)
- Set the thumbwheels to the first combination from the relevant table below (e.g. For a fluor unit set them to 6, 3, 3 left to right so that User is 3, Lighting is 3, Size is 3)
- Press the Thumbwheels button once on the dialog (text says "Waiting for drawer to close")
- Close the drawer, hold it until the text changes (about 4s), then open it again (if the progress bar reaches the left-hand end, the command will terminate. By contrast, you can leave the drawer open for as long as you want)

- For each combination in the list in turn: set the thumbwheels, close the drawer, hold until the text changes, open again. (It doesn't matter if you repeat an item in the list, it doesn't matter if you use a setting that is not in the list, just be sure to use each combination in the list at least once)
- When you reach the final item in the list, close the tray and leave it closed so that the command terminates

Testing

- Start with the drawer open
- On the Commissioning dialog, press Test thumbs
- Repeat for all positions of each thumbwheel: Set the thumbwheels, close the drawer, open, Check the reported values
- Close the tray and leave it closed so that the command terminates

If you see any failures, please save the image that has built up on the image view and send us the file.

Fluor combinations sequence

- 6, 3, 3
- 3, 1, 1
- 6, 2, 2
- 3, 1, 1

Chemi combinations sequence

- 6, 1
- 3, 5
- 3, 4
- 3, 3
- 3, 2
- 3, 1

Vü networking connections and configurations

The following tells you how you can connect to your Vü scanner. It cannot teach you all about computer networks, so you may need to seek further information, advice or guidance from elsewhere. Your IT staff may also wish to be involved in configuring the networking options of your scanner.

Configurations

Vü supports two basic connection configurations:

- A direct connection between a PC and a single Vü scanner through an Ethernet cable. Note that, in this configuration, only these two devices are connected, and that you must use a cable (WIFI is not supported).
- The Vü scanner can be connected to a network, such as the network in your office or laboratory. In this configuration you may have multiple scanners connected to the same network, and multiple computers connected at the same time. In this configuration, the links between each device and the network may be either wired (Ethernet) or wireless (WIFI).

In many ways the configuration and setup of the Vü scanner is similar to the corresponding task for a networked printer or scanner. Such devices are frequently set up using something like the first configuration, and then put to use in a configuration like the second.

IP addresses and DHCP

On TCP/IP networks (the standard computer networking protocol), devices 'find' one another using IP addresses – something like "198.164.1.73". In some cases, certain devices are assigned fixed IP addresses, but in most cases a central service on your network (provided by your router, perhaps, or a computer server) allocates IP addresses to devices that need them so that each device has a unique IP address. This service is called DHCP (Dynamic Host Configuration Protocol), and the Vü scanner requires a DHCP service because it does not have a fixed IP address.

DNS

IP addresses are not very convenient for us humans to use; we prefer names that we can remember more easily. What's more, DHCP means that the IP address assigned to a particular device such as the Vü scanner may change from time to time, so we really want to use a fixed name to refer to it rather than having to use an IP address that may change. The DNS (Domain Name System) allows devices to be referred to by name (in the case of Vü, the name is set to something like "Vü-Chemio3000123" when it is configured). This name is called the scanner's hostname, and while you can change it, it's not advisable in most circumstances.

DNS also requires a central service on your network (your router or other server) to provide a centralised way to map the names to IP addresses. (DNS also enables you to type "www.google.com" instead of requiring you to know the IP address of Google's server.)

You can connect to your scanner using its IP address instead of its hostname – if you know it. (So, if you know your scanner's IP address is "192.168.1.89", you can type that into the 'hostname' field on the Vü app instead of the hostname itself. This can be useful if you change the hostname and then forget it, or if there is some

problem with the DNS service – but you will require some knowledge of networking to find out the IP address assigned to it.)

Direct Connection configuration

When your PC is connected directly to your Vü scanner – and therefore only connected to the scanner – the PC must provide the DHCP and DNS services. A limited DHCP is provided as part of Windows, but it does not provide DNS 'out of the box'. Instead, in this configuration, we use a technology developed by Apple called Bonjour so that we can use our scanner's name instead of the IP address. Bonjour is Apple's implementation of zero-configuration networking, a group of technologies that includes service discovery, IP address assignment, and hostname resolution. Bonjour locates devices such as printers, other computers, and the services that those devices offer on a local network using multicast Domain Name System (mDNS).

It may seem strange that Windows does not provide its own DNS or mDNS system; it is beginning to but it remains somewhat limited. Windows expects to be connected to a 'proper' network and for other devices to be connected to that network rather than a direction connection – and that the network itself will provide the DNS or mDNS service.

Note that Bonjour is only required to be installed on your PC if you are using the Vü scanner in the 'direct connection' mode. You may well find that Bonjour is already installed on your PC; many applications require it.

Windows 10 version 1803

Unfortunately, if you had Bonjour installed and then installed Windows 10 version 1803 as an update, it seems that this 'broke' the Bonjour installation and its mDNS service stopped working. If these circumstances apply to you (the symptoms, for the networking expert, are that you can 'ping' the scanner from the PC using its IP address, but you can't do so using the hostname. Local address), the fix is simply to uninstall Bonjour, then reinstall it. (Running the installation opens the required ports on the firewall).

Sole connection

It may be that, for the direct connection to work, you will need to disable all other network connections on your computer. For example, if your computer is normally connected to your office network by WIFI and you now connect it to the scanner by Ethernet cable, you may need to turn off the WIFI connection to your office network while you work with the scanner. (The solution, of course, is to also connect the scanner to your office network as described below).

.local suffix

In the direct connection mode only, you must add the .local suffix to the hostname of your scanner when you enter it into the Vu app. So instead of "Vu-chemi-03000123" you must use "Vu-chemi03000123.local".

Networked configuration

If you have a 'proper' network in your office or laboratory that provides shared network storage, connection to the internet, shared printers etc. then it's much simpler to just connect the Vü scanner to the network. The network will provide DHCP and DNS, and you can refer to the scanner by name without any difficulty. In this configuration, you also have the option to connect the scanner by either Ethernet cable or WIFI.

An Ethernet connection will always work; it does not require any configuration or setup. Configuring the WIFI does not disable the Ethernet port, so it will continue to work.

WIFI configuration

Setting up the WIFI connection on the Vü scanner is very similar to setting up a Wi-Fi___33 connection on your phone or tablet – the only difference is that you are using an app on your PC to change the settings. Note that this means that you must connect the scanner to the network with an Ethernet cable whenever you change the Wi-Fi___33 settings!

First, plug your supported Wi-Fi___33 adapter into any available USB socket. Next, go to the Network Settings dialog on the Vü app. Unlock the scanner settings and check the box to enable the Wi-Fi___33 on the scanner. Then, provide the required information:

- Wi-Fi___33 SSID: this is the name of your Wi-Fi___33 network. (When you start to connect your phone or tablet to a new Wi-Fi___33 network, a list of the available network names is shown to you; these are the SSIDs of the available networks).
- Wi-Fi___33 Password: you need to provide the password for the Wi-Fi___33 network you choose. WPA is the supported Wi-Fi___33 security protocol.
- Wi-Fi___33 Country: select the country in which you are operating the scanner, because the radio frequency rules vary by country: the transmission power may be limited and some frequency bands may not be allowed.

When you have entered the required information, please press the 'Change' button. When the scanner has connected to the network the status indicator will show "Connected" in green. Once the scanner has connected to the Wi-Fi___33 network you should be able to disconnect the Ethernet connection and still communicate with it; the app will invite you to do so.

(There are some complexities associated with this. Your PC has communicated with the scanner over Ethernet using a particular IP address, which it has been told by the DNS is the correct IP address to use for it. Now, the scanner is connected using a different network - the Wi-Fi___33 network - and probably has a different IP address. So, when the Vü app tries again to communicate with "Vüchemi-o3000123", the PC's networking system may provide the wrong address for it. To avoid this, the app asks the PC's networking system to 'flush the DNS cache' when the Wi-Fi___33 network is first enabled; this means that the PC will 'start from scratch' and establish the correct IP address for the scanner, rather than using the one it had found previously. If you want to force the system to flush the cache again you can open a command prompt and type "ipconfig /flushdns", but this should not be necessary).

Specifications

| | Vü-F | Vü-C |
|----------------------------|---|---|
| Application | Fluorescent gels and blots | Chemiluminescence Western blots |
| Illumination | UV integral 302nm [1 tube] | RGB integral LEDs for marker including coloured markers |
| | Blue light integral LEDs | |
| | White light conversion screen | |
| Sample size (cm) | 20 X 20 | 10 X 10 |
| Sample Drawer | Thumbwheel selection for lighting, ROI and user | Thumbwheel selection for lighting [marker lane] and User |
| Drawer type | Pull out drawer with removable washable gel tray | Pull out drawer with removable washable blot tray |
| Sensor resolution | goMpixel | 51Mpixels |
| Image output resolution | 3550 × 3550 (12.6 Mpixel) | 1530 x 1530 (2.34 Mpixel) |
| Image format | TIFF | TIFF |
| Sensor type | CMOS linear | CMOS linear high sensitivity |
| Process speed | 20-90 seconds - depends on gel and dye used | 2-5 minutes - depends on blot and reagent |
| Sensor lens T-Stop | 2.86 | 1.01 |
| Footprint (mm) | 480D x 426W x 205H | 375D x 353W x 175H |
| Power Supply | 100-240V external | 100-240V external |
| Connectivity | Integral ethernet; optional external Wi- Fi33 | Integral ethernet; optional external Wi- Fi33 |
| Analysis software | 3 copies | 3 copies |
| Warranty | 2 years | 2 years |
| Ordering | 10-1007-04 [UV, Blue LED] | 10-1007-02 |
| Part numbers | 10-1007-05 [UV, Blue/Green LED] | |



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