NEO Science

# FluoroBox

## User manual


Ver. 2.4

Nucleic acid Gel Imaging System, FluoroBox User Manual. Ver. 2.4

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# Index

## Overview

FluoroBox is a device which images DNA electrophoresed gel to confirm and analyze the DNA.
The device is optimized to the fluorescent reagents with wavelengths between 450mm and 490mm developed to replace the EtBr. It is consisted of Blue light, a window at the top covered with a camera that can flip open and dark room chamber with doors at each side. Also as it can be used along with UV transilluminator, EtBr images can be obtained.
It's compact size and user-friendliness has been designed for the optimizing convenience of users.
The gel can be observed through the window at the top, and the doors at each side enable gel cutting convenient.
This simple and excellent program enables a user to conveniently secure images and to easily quantify the DNA band.

## Characteristics of FluoroBox

1. Obtaining Image of Agarose Gel with Electrophoresed DNA
The DNA band can be observed by placing the agarose gel with electrophoresed DNA in the chamber. Users can either observe it with the camera or flip open the camera to see it through the window.
For the use of FluoroBox without camera, personal cameras can also be used to obtain image.

2. Image of DNA Staining Fluorescent Reagent to replace EtBr
The blue illuminator can be applied with the reagent which can be excited with wavelengths of 450~490 nm among DNA dye reagents made to replace EtBr. The DNA excited with the blue illuminator can be seen in the eyes through the anti-glare filter and the image of the DNA can be secured through a computer program.

3. Blue illuminator
The blue illuminator is designed to excite the DNA staining reagent. The LED light source with a wavelength of up to 470nm can generate the best effect.

4. Anti-glare Filter

The filter prevents glare and is designed and produced to observe DNA band.

5. Quantitative analysis - Measuring the intensity of fluorescence and drawing the result value

The DNA band can be designated in the form of a square, circle or polygon or automatically. Then, the corresponding area, intensity average value and integrated density value can be measured. The values measured in this manner can be saved as CSV file extension in the form of tables.

6. Simple and Easy Tips for Users

Main features of the program are listed on the side panel for a user to use conveniently. Just a few custom-designated values exist and therefore, users can easily use the program.

## Components


A: FluoroBox

B: White plate

C: Clear plate

D: DC adaptor

E: USB connect cable

F: Program USB memory card

G: User manual

## How to Install and Use

Please install the program in the following manner for use.

1. Connect the DC adaptor to DC jacks at the back of the FluoroBox.

2. Connect the USB cable to the back of FluoroBox and computer.

3. Turn on the computer and activate the NEOimage Program.

4. Push the button to turn on the white light and adjust the focus of the lens by looking at the computer.

5. Turn off the white light and observe the DNA gel by turning on the blue light.

## Driver Connection and Program Installation

1. Driver Connection

1) Insert NEOimage USB memory card into the USB port of the computer.

2) Connect FluoroBox to the computer with the USB cable.

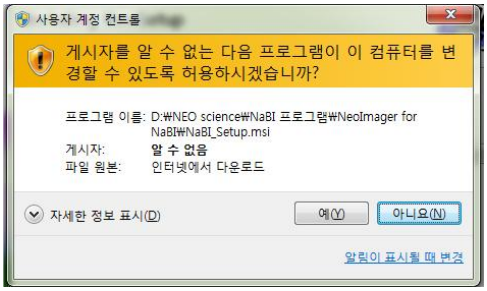
3) Choose the "Mightex Classic USB Driver" as the hardware driver to be installed on the USB card and install the driver.

2. Program Installation

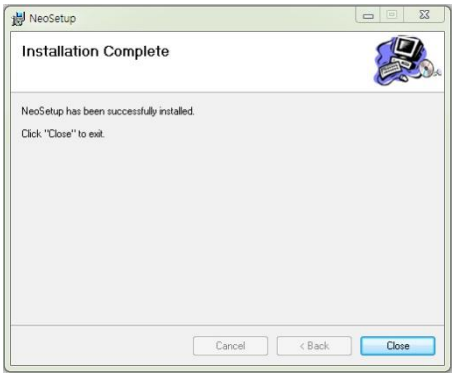
1) Double-click "NEOimage FluoroBox Setup.msi" on the USB card to start installation.

2) Follow the instruction and click "Next" to proceed installation.

3) Click "Yes" on the pop up message allowing the publisher to continue installation.




4) When the Installation complete message pops up, installation is completed.




5) Double-click "NEOimage.exe of Desktop and activate the program.

## Quick start

1. Click the NEOimage icon to activate program. (desktop or start menu)

2. Click on the “LIVE” will pop up the live window

3. Turn on the white light.

4. Adjust focus watching the Live window.

5. Turn on the blue light and adjust brightness by setting of the exposure time.

6. Click the “CAPTURE” to take signal picture.

7. Select the required picture and save it under a different name for results backup.

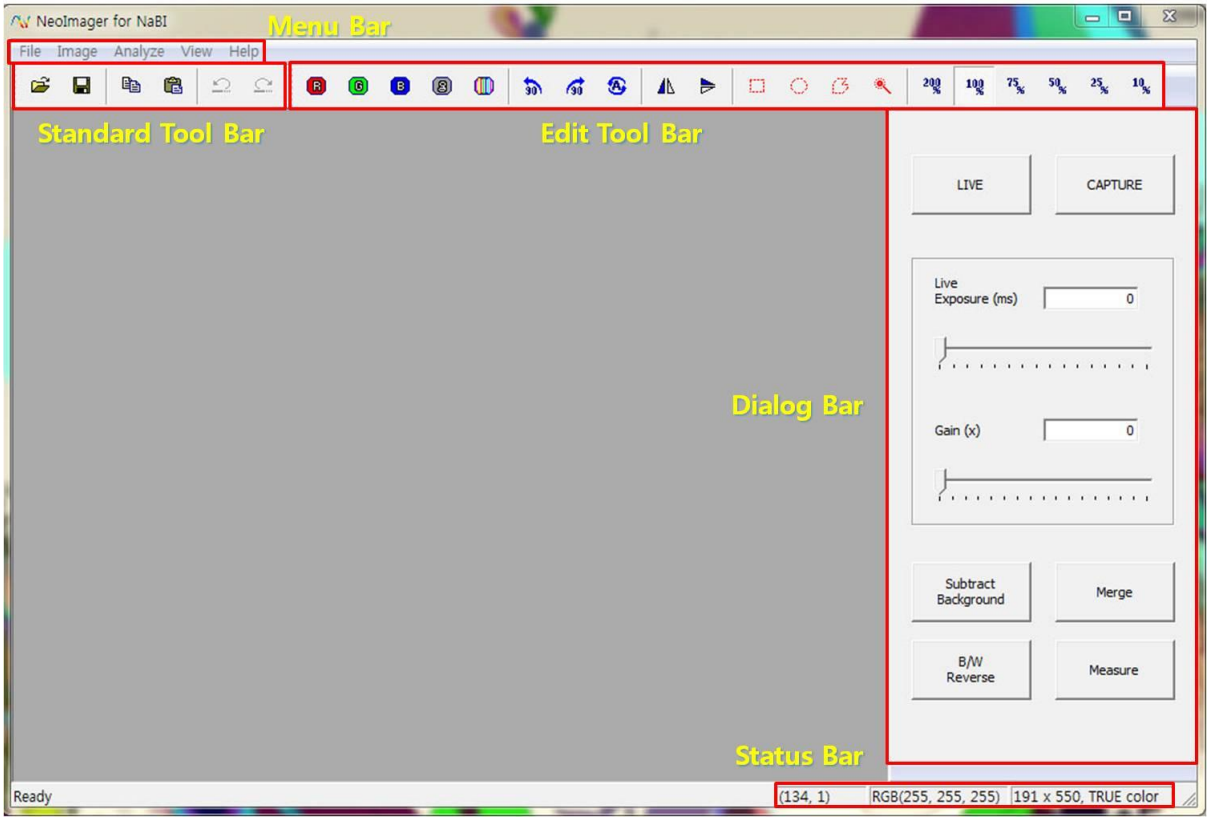
8. Remove the Background using Subtract background.

9. Select the target signal using the ROI menu in Edit Tool Bar. (Square, circle, polygon and auto)

10. Click the “Measure” to take quantitative result.

11. Click the “Export” to send the quantitative result to csv file.

## NEOimage




1. Dialog Bar

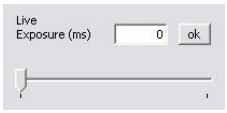
1) LIVE: It shows the status within the chamber in real time. When the white light is on, the general features of the sample can be seen and the fluorescent signal can be observed in real time when the blue light is turned on. The live image is 50% of the real image. When the device is not connected, the live feature does not work.


2) CAPTURE: The image shown on the live screen is captured in the picture form. The newly-created picture is shown in a separate window (50%) on the top left corner of the computer screen and is automatically backed up.

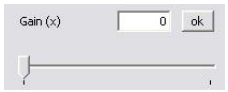

The image is stored in the "C₩FluoroBox data" folder. When a program is shut down without storing a file by accident, the file can be restored in this folder. The obtained file is stored in the "YearMonthDay_HourMinuteSecond.bmp" format indicating the time

when the picture is taken.

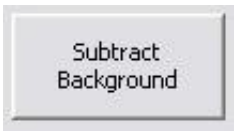
3) Exposure: It adjusts the exposure time when obtaining the image and can be adjusted up to 750ms. Numbers can be entered directly as the exposure time or the length of time can be adjusted by moving the bar with the mouse.

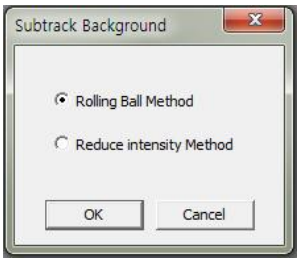



4) Gain: It increases the intensity value on the obtained image and the value can be adjusted up to 16.

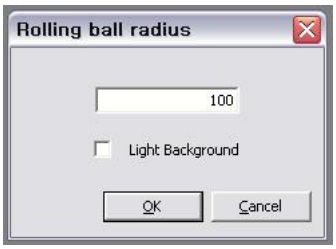



5) Subtract background: It consists of the rolling ball algorithm and the reduce intensity algorithm. When the Subtract Background button is pressed, a window that requires a user to choose one of the activation algorithms pops up.



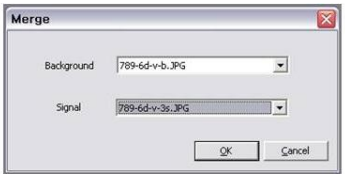

① Rolling ball algorithm: When the feature is activated, a message that says 'Enter the Rolling Ball Radius' pops up. When certain radius is entered and "OK" is clicked, then the background of the picture is removed. As the smaller number of radius is entered, smaller region of the signal will be prominent. However, the intensity gets smaller in case of smaller size of signals and therefore, the number of certain ball size should be entered. When the signal is in black, the Light Background shall be checked and then activated.




② Reduce intensity algorithm: It lowers the intensity of the whole picture by the

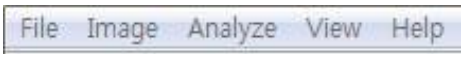
average intensity of the selected background. When this feature is activated, a message requiring a user to choose the area which constitutes the background in the picture pops up. Press OK, and the background of the picture is removed.

6) Merge: It overlaps the ordinary picture and the fluorescent picture of the sample. When the Merge button is activated, a message requiring a user to select the ordinary picture and fluorescent picture opens up. "OK" button is clicked after choosing the corresponding pictures, then the two pictures are merged and a new window is created. If the fluorescent picture has the pixel of less than 20 in the intensity value, the picture will not be merged.


7) Measure: It designates ROI (Region of interest) on the obtained fluorescent image and secures the number of pixels, intensity value and volume value within the ROI. First, designate the area to be measured as the ROI and click "Measure" to create a new window containing the measured value in the table form. In the table, "ROI" indicates the ID number of the designated ROI. "Area" means the number of pixels within the designated ROI. "Mean of Intensity" is the average value of the measured intensity value from each pixel within the ROI. "Integrated Density" is the value of the "Area" value multiplied with the "Mean of Intensity" value. When the sizes of the fluorescent signals within the ROI are compared, "Integrated Density" values shall be compared. The obtained quantitative value can be stored as the CSV extension by clicking "Export". (It can be opened on the Excel program)


2. Menu Bar




1) File: It opens, closes, saves, saves as, opens Rainbow Standard and closes the obtained picture. Also, it allows a user to open up to nine recent photos without browsing.

2) Image: It undoes, repeats the operation and uses Color, Merge, RGB Split, RGB Merge,

Rotate, Flip and Zoom features.

3) Analyze: It uses Image Calculate, G-ch Analyze, Subtract Background, Set ROI, Measure features.

① GFP analysis: It is a type of Image Calculator and can be used when the signal is GFP. Click it, and a message of "Select Background" pops up. If a user designates a square as the background, the "OK" button on the window is activated. Click "OK", then the result of analysis of the Image Calculation is created in a new window.

② Image Calculation: It removes the background from the fluorescent image. It creates a Long wavelength background (LWB) picture obtained from the background filter of a long wavelength, along with the fluorescent picture. Click the Image Calculation button, and a window requesting a user to select the pictures corresponding to the Signal and Background opens up. Select the corresponding pictures and click "OK", then "Select Background" is generated. When the region corresponding to the background in the signal picture is dragged for selection, the "OK" button is activated in the window. Click the OK button, and the new picture after the operation is created in a new window.

4) View: It selects Standard Tool Bar, Edit Tool Bar, Status Bar, Dialog Bar visible or invisible.

5) Help: It checks the information of NEOimage.

3. Standard Tool Bar

The icons of Open, Save, ROI Copy, ROI Paste, Undo, Repeat, etc. can be easily used.

4. Edit Tool Bar


1) Change Picture Color: The picture can be in red, green, blue and rainbow. The rainbow feature individually shows pixel intensity strength as red, orange, yellow, green, blue and

violet. For a better understanding of the image, the pixel with less than 30 in intensity is not viewed.

2) Rotation: It rotates the picture 90 degrees left or right, or by a custom angle. When the custom angle is a positive number, the picture is rotated to the right and when the custom angle is a negative number, the picture is turned to the left. Also, the picture can be flipped by 180 degrees horizontally or vertically.

3) ROI Designation: The ROI can be designated by selecting the feature of Square, Circle, Polygon or Auto form. The ROI shape is chosen on the selected picture and saves the image by dragging the shape on the picture. The designated ROI can be moved or corrected. However, the designation of the Auto feature cannot be undone.

4) Zoom: The size of the image can be in 10%, 25%, 50%, 75%, 100%, and 200%.

5. Status Bar


It confirms the mouse coordinate, individual RGB intensity, picture size, picture types. The coordinate and the size of the picture are shown in pixels.

## Specifications

| FluoroBox | |
|---|---|
| Hardware | FluoroBox |
| | Camera: 1/2" 1.3M 8bit CMOS |
| | Effective Pixels: 1280 x 1024 |
| | Frame Rate: Up to 24 |
| | Size (W x D x H): 260 x 260 x 390 mm |
| | Interface Connector: Standard USB 2.0 |
| | Light: 470nm LED |
| Software | NEOimage |
| | Image capture |
| | Set target ROI: Manual or automatically |
| | Subtract background |
| | Measure the area, intensity and integrated density |

## Ordering information

| Cat. No. | Product |
|---|---|
| FLB-001B | FluoroBox Blue |
| FLB-001BCM | FluoroBox Blue with CMOS camera |

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