

Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

GeneBLAzer[®]/Tango[™]/ CellSensor[®] Setup Guide for beta-lactamase detection on the PerkinElmer EnVision[®] Multilabel Plate Reader

NOTE: The PerkinElmer EnVision[®] Multilabel Plate Reader was tested for compatibility with Invitrogen's GeneBLAzer[®], Tango[™] and CellSensor[®] assays in bottom-read mode. The following document is intended to demonstrate setup of this instrument. For more detailed information and technical support of Invitrogen assays please call 1-800-955-6288, select option "3", then extension 40266. For more detailed information and technical support of PerkinElmer instruments or software, please call 1-800-762-4000 or by e-mail at productinfo@perkinelmer.com.

A. Recommended Optics

PerkinElmer part number	wavelength (nm)	diameter (mm)	
Excitation Filter (2100-5370) MOCA 400/25 Barcode 218	400/25 (or similar)	15	
Emission Filter 1 (2100-5130) Umbelliferone 460/25 Barcode 207	460/25 (or similar)	15	
Emission Filter 2 (2100-5120) FITC 535/25 Barcode 206	535/25 (or similar)	15	
Dichroic Mirror *contact PerkinElmer	(Single or dual detection) Dual detection models: Beta Lactamase D425/490 dual mirror (2100-4230; barcode 661) -or- General bs50/bs50 dual mirror (2100-4050; barcode 651) Single detection models: Beta Lactamase D425 single mirror (2100-4270; barcode 418) -or- General bs50 single mirror (2100-4010; barcode 401)		

NOTE: Before beginning, please ensure that you have installed the excitation and emission filters into the appropriate slots in the Excitation and Emission Filter Slides, respectively. Please note that:



Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

- i) The emission filters must occupy adjacent slots in the Emission Filter Slide.
- ii) The Umbelliferone 460/25 filter should occupy a lower slot number in the Filter Slide than the FITC 535/25 filter (*i.e.* if you have inserted the Umbelliferone 460/25 filter at position 3 in the Emission Filter Slide, then the FITC 535/25 filter must be placed in position 4).

NOTE: The dichroic mirror needs to be manually changed to the bottom mirror position within the machine and selected in the software before use.

B. Instrument Setup

1. Make certain plate reader is turned on, and open up Wallac EnVision[®] Manager software on computer.

🕞 Wallac EnVision	Manager - Instrument mode			Ð
File Edit View Tools	Actions Help			
Back Forward	Dp Optimize Latest TRF op	+ 👌 🍎 📋 ti Lock New Duplicate	Save Delete	
Routine Use Advanced Use	Beta lactamase			6
1. Create Label 2. Create Plate 3. Ruu Wizard 4. Create Protocol Users	Navigation Tree X Image: State of the	General Optimizations Name Excitation Excitation Mirror Excitation filter Emission filter Emission filter 2nd emission filter 2nd emission filter Detector gain 2nd detector gain 2nd detector gain Number of flashes Number of flashes Number of flashes per A/D conversion Reference signal Reference Excitation light (%) Changed	Beta lactamase C Top Bottom General Dual - Bottom 400nm excitation - Ex Slot 4 FITC 535 - Em Slot 6 Umbelliferone 460 - Em Slot 5 5 100 100 100 10 1 295280 4 100 6/11/2004 1:00:00 PM (EnVision) 	<u>۲</u> ۲ ۲
5. Run Assay 6. View Results	Luminescence DELFIA - Time-resolved FI LANCE - Time-resolved Fit Fiters			Taskuuraakasada (Falikida
start	🕫 🕗 🝘 🗾 🔋 💦 🕞 Wallac	: EnVision Mana 👿 Document - Word	dPad	2:03 PM

2. Setup the following label for beta-lactamase.



3. Create a protocol for the assay by selecting New protocol.





4. Name the protocol Beta-lactamase or any suitable description, as shown in this example then click OK.





5. Select Plate type. The EnVision [®] has many pre-set plate definitions such as 384 Costar shown.





6. Define the Operation(s) required for this protocol. Under the Operations tab, select Measurement from the Meas Icon.





7. Select the Label method to be used in this protocol. Select the Beta-lactamase label method created in Step 2.



8. Load plate(s) and optimize the Measurement Height as described in section C. Optimization Setup.



Page 8 of 16

Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

C. Optimization Setup

This section explains how to optimize the Measurement Height on the EnVision[®] using the Assay Start/Optimization Wizard.

Note: In bottom read mode, the Measurement Height is dependent on plate type. Once the Measurement Height has been optimized for a given plate type, only re-optimize the Measurement Height if a different plate type is used.

Note: We do NOT recommend using the Assay Start Wizard to optimize Detector Gain as this typically results in saturation of the PMTs. Instead we recommend manually setting the Detector Gain in the Label (see screenshot on page 2) to a value of either 100 or 150 in both channels



1. Start the Assay Start/Optimization Wizard under the Tools Menu.



GeneBLAzer[®] Compatible Microplate Reader Documentation

Version No.: 01 Jan 10

Page 9 of 16

Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

2. The following window appears. Select Next.





GeneBLAzer[®] Compatible Microplate Reader Documentation

Version No.: 01 Jan 10

Page 10 of 16

Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

3. Select Optimize existing protocol. Click Next.





4. Ensure proper protocol is selected. Click Next.





5. Select Measurement Height and Advanced mode. Click Next.





6. Select Scan well for strongest sample. Click Next.





7. The $EnVision^{\ensuremath{\mathbb{R}}}$ will scan the plate to determine appropriate height setting.

Hallac EnVision Manager 1.11 - Instrument conne File Edit View Tools Actions Help	cted				
Back Forward Up Start Latest	* GeneBLAzer/Tango (BLA)	Edit Run Pause Stop Unload			
Red • Porton Porton Later Routine Use Image: Control Image: Control Image: Control Perform Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control Image: Control <td< td=""><td>* Geneti-Azer/Tango (BLA) Reader Co Processin</td><td>Cancel Cancel C</td><td>View Color scale Color scale Color scale Color scale Color scale Color scale - 1,000 - 800 - 600 - 400</td></td<>	* Geneti-Azer/Tango (BLA) Reader Co Processin	Cancel C	View Color scale Color scale Color scale Color scale Color scale Color scale - 1,000 - 800 - 600 - 400		
Advanced			1		
Press F1 for help Instrument connected EnWsion					
🏄 Start 🛛 🧭 😹 🔹 🎠 GraphPad Prism	anti-green fluorescent pr	📃 Document2 - Microsoft 🛛 🍉 Wallac EnVision Manager 🛛 🦿 Assay Start Wizard	« 🖸 🌠 💐 🔮 3:23 PM		



Setup Guide on the PerkinElmer EnVision[®] Multilabel Plate Reader

01 Jan 10

8. Ensure a bell shaped curve is achieved as shown below (the curve shown is representative and varies by instrument and plate type). Ensure the red vertical line is set under the peak of the curve. If not, slide line with cursor to the appropriate height where the signal is highest. Click Next.





9. Select "Start the assay". Click Finish.

