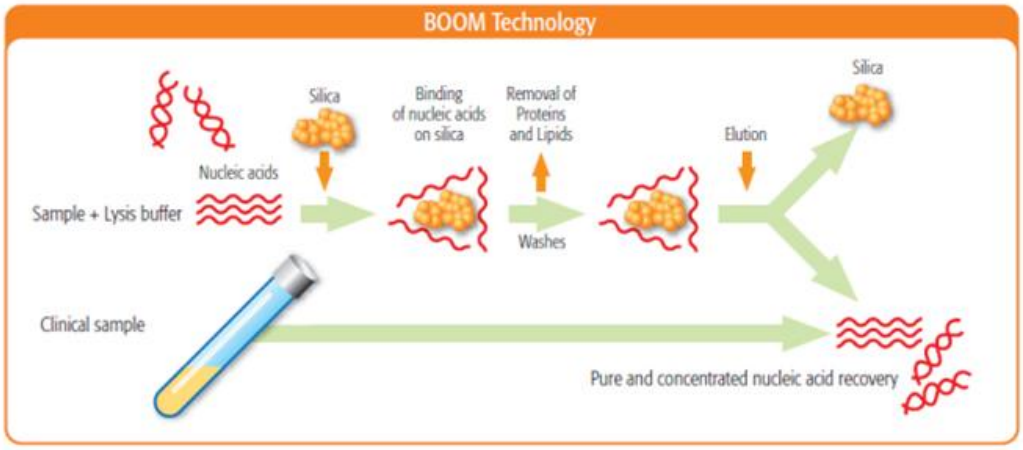
NucliSENS® easyMAG™ Operating Procedure

PURPOSE

This procedure provides instructions for operation of the NucliSENS® easyMAG™ which is an automated extraction system. The extraction chemistry uses the BOOM method for the purification and concentration of total nucleic acid (RNA/DNA) from biological specimens. Viral particles or bacteria in the specimen are lysed in the presence of the chaotrope, guanidinium isothiocyanate releasing the nucleic acids. Magnetic silica binds the DNA and RNA under high salt conditions. After several washes, the concentrated nucleic acid is released into an elution buffer used for amplification.



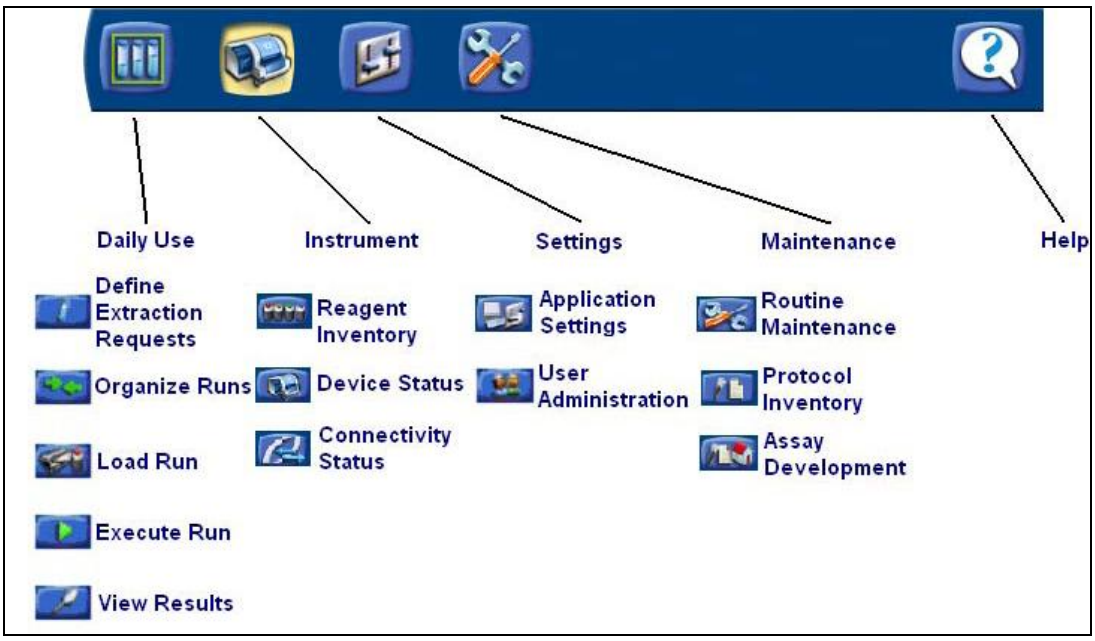
EQUIPMENT REQUIRED

Equipment	Reagents	Supplies
Vortex	5% Extran	Lab coat
Pipets, 20 µl, 100 µl, 200 µl, 1000 µl	70% alcohol	Paper towels/Kimwipe towels
BioHit multichannel pipette	water	Sharps container
BioSafety Cabinet	Sani-Cloth Bleach Wipes (10%)	Micro tubes 1.5 ml, RNase/DNase free
NucliSENS easyMAG	easyMAG Buffer 1	EasyMag disposable vessels & tips
	easyMAG Buffer 2	Nitrile gloves (powder-free)
	easyMAG Buffer 3	Gripper rack, rm 2
	easyMAG Lysis buffer	Gilson pipette tips, 30 µl, 100 µl, 200 µl, 1000 µl
	MagSil	BioHit pipette tips
	Extraction Controls	

SAFETY CONSIDERATIONS

- Standard precautions; Refer to MB 2.02 Biohazard Containment
- Use of engineering controls: Refer to MB 3.01 Engineering Controls to Prevent Nucleic Acid Contamination
- Caution: NucliSens Lysis buffer and easyMag Wash buffer 1 contain guanidine thiocyanate. Guanidine thiocyanate is harmful by inhalation, in contact with skin and if swallowed.
- Do not perform assay in the presence of vapors from sodium hypochlorite or dust.
- Guanidine thiocyanate in the presence of bleach will form cyanide gas

Figure 1: Guide to easyMAG icons



PROCEDURE A: Follow the activity below for start-up of the easyMAG

easyMAG Start-up

Step	Action
1	Turn power on to the easyMAG and then the computer (**ABC**, Analyzer Before Computer)
2	Once the LED on the instrument turns from orange to green, log into the computer
3	Enter username (bmx) and password (bmx followed by assigned number 1 – 9, ex. bmx3)
4	Wait for the instrument to complete initialization, making sure hour glass is idle
5	**Barcode reagents**. Touch the **Reagent Inventory** icon located on the instrument tool bar located on the top tool bar on the screen 
6	Open the instrument reagent door and barcode the bottle location followed by the bottle identification

PROCEDURE B: Follow the activity below for creating a worklist and run

Create Worklist and Run

Step	Action
1	Using the assay specific worksheet as a layout, organize patient samples and labels to be extracted

Step	Action
a	Number the patients on the worksheet (positions 1 – nn) plus 1 extraction control and 1 negative control
b	Number corresponding patient label according to the worksheet
c	Date one small label
d	Number cap of 1.5 micro-centrifuge tube for each sample to be extracted
e	Matching number on the label to number on the cap, label corresponding micro-centrifuge tube
f	Set tubes in magnetic rack

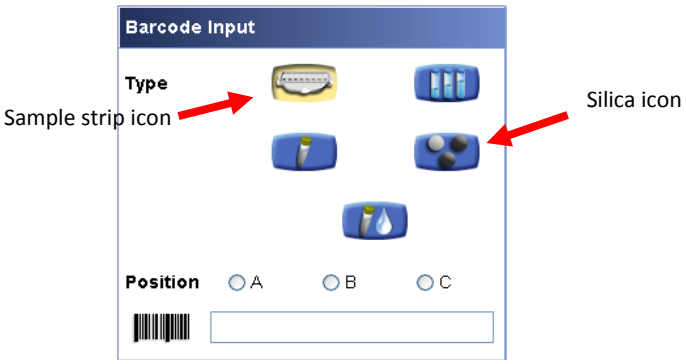
Step	Action
2	Touch the *Daily Use* icon located on the top tool bar on the screen.  This will default to the submenu *Define Extraction Requests* 
3	Enter **Protocol** from drop down box ▪ RVP D 1.01 (RVP) ▪ Generic 2.01
4	Define extraction settings: ▪ Volume (mL): 0.200 ▪ Eluate (µL): default or type in ▪ Matrix: Other or select from drop down menu ▪ Type: Primary ▪ Priority: Normal
5	**Build worklist**: In the Sample ID field, scan patient labels and controls in consecutive order; the carriage return and assignment to worklist will be automatic
6	If sample ID is entered manually, press **Enter** after each manual addition to assign samples to worklist
7	The extraction settings will remain the same for each new entry
8	**Organize Run:** Add samples to worklist by touching **Organize Runs** icon 
9	**Create Run:** Touch the **New Run** icon to create a run name. The date will default with a unique run modifier. 
10	Select workflow options, **On-board Lysis Incubation**  and **On-board Silica Incubation** 
11	Touch OK
12	Sample positions are assigned when the samples are transferred to **Run Layout** using the arrow buttons
13	Transfer samples from the Unassigned List to the Run Layout using the positioning arrows.

Activity	Action
To move all samples	▪ Touch the multi-select icon 
To move individual samples	▪ Highlight sample on the unassigned list ▪ Touch the **Move** icon 
To remove sample from the run layout back to the unassigned list	▪ Highlight sample ▪ Touch the **Remove** icon 
To move samples up/down within a run	▪ Touch the up and down arrows  
To modify the run name or workflow options	▪ Click the **Edit this Run** icon  ▪ Click the save button after changes have been made 
To delete a run	▪ Click the **Delete** button ▪ Samples will be moved back to the unassigned list, not deleted 

14	After samples are assigned a position, touch the Load Run icon 

PROCEDURE C: Follow the activity below to load a run

Load a Run

Step	Action
1	Touch the **Load Run** icon and select the run from the dropdown menu; the last run in the previous screen will automatically be shown 
2	Open the instrument process door and snap aspirator tips into easyMAG; a green square will display to the left of the tips on the screen
3	Place easyMAG extraction strip(s) in carrier rack ▪ Consecutively number each vessel on the strip to correspond to patient samples and controls
4	Add samples to related vessel being careful to avoid air bubbles at the bottom of the well
5	Snap the sample strips into the strip slots ▪ *Caution: Be sure the strips and aspirator tips are seated securely before starting extraction*
6	Scan barcode on the strip and then the position barcode (A, B or C); a green square and ID will display by the strip
7	Touch the **Silica** icon and barcode the silica lot number (located on the silica box cover) 
8	Assign the silica lot number to the sample locations ▪ Highlight the samples on the run list ▪ Touch the **Add icon** 
9	Touch the Silica icon on the top bar to review the assignment 
10	Return to the progress screen; Touch the **Progress View** icon 
11	To print worklist, touch the **Print** icon 
12	Comments can be made by touching the Add Remark icon 

Step	Action
13	**On-Board Lysis** Step / Action a – Add sample to strip vessel b – Insert tips and strips c – Barcode strips d – Touch dispense  e – After 10 min, add internal control (IC) and then 50 µl silica f – Mix with BioHit pipette g – Return and barcode strips h – Close instrument process door i – Touch **Start** icon  **Off-Board Lysis** Step / Action a – Incubate sample in 2 ml lysis buffer b – Transfer sample to strip vessel c – Incubate 10 min d – Add IC and then 50 µl silica e – Mix with BioHit pipette f – Insert tips and strips g – Barcode sample strip h – Close instrument process door i – Touch **Start** icon 

PROCEDURE D: Follow the activity below for unloading samples

Unloading Samples

Step	Action
1	After run is finished, the *hour glass will be idle*; transfer the extracted samples within 30 min to prevent silica contamination
2	Print extraction report from the **View Results** screen 
3	Carefully remove the strips not to disturb the silica button ***Note:*** Check silica buttons; if not present, the extraction must be repeated
4	Transfer the eluates into corresponding 1.5 micro-centrifuge tube in a magnetic rack
5	Take care not to transfer any silica particles, since they may inhibit PCR amplification
6	Allow eluate tubes to sit for 10 min prior to use
7	If the sample is contaminated with silica, retransfer into a clean tube and allow to sit in the magnetic rack
8	If eluates are not going to be used immediately, store under assay specific conditions

PROCEDURE E: Follow the activity below for shutting down the easyMAG

Instrument Shutdown

Step	Action	Related Doc
1	**CBA**, Computer Before Analyzer	
2	Touch the **Logout/Quit/Change Password** icon to power down software 	
3	Wait for the light on the easyMAG to stop flashing red/green and change to a steady orange	

Step	Action	Related Doc
4	Power down the instrument *Caution:* The instrument must be completely powered down before restarting again	
5	Wipe down the instrument with 5% Extran followed by 70% alcohol	MB 3.03 Cleaning and Decontamination
6	Check the drip tray for leakage	
7	Refer to Maintenance Log for daily and weekly maintenance	

MAINTENANCE SCHEDULE: Record on easyMAG Maintenance Log

Step	Maintenance	Frequency
1	Shutdown instrument and PC	Daily
2	Wipe down instrument with 5% extran followed by 70% alcohol	Daily
3	Check drip tray	Daily
4	Check waste bottle	Daily
5	Empty waste bottle	As required
6	Clean o-rings: use cotton tip dipped in nuclease free water	Daily
7	Wipe down vessel carrier with Sani-Cloth Bleach Wipes (10%) followed by water and then 70% alcohol	Daily
8	Maintenance Protocol #1: Clean Dispense needles	Mon – Wed – Fri
9	Check filters	Weekly

REFERENCE

1. NucliSens® easyMag™ 2.0.1 Guide, BioMerieux, 100 Rodolphe Street, Durham, NC 27712
2. NucliSens® easyMag™ User Manual version 1.1, 2005, BioMerieux, 100 Rodolphe Street, Durham, NC 27712

Historical Record

Version	Written/Revised by:	Effective Date:	Summary of Revisions
1	P. Ackerman	07.09.2008	Initial Version
2	P. Ackerman	07.20.2009	Added picture icons
3	P. Ackerman	07.15.2016	Reformatted for CMS upload; added maintenance schedule; changed logo