ITSP PAL Intermediate Course for Lab Personnel

ITSP™ SOLUTIONS, INC.

Instructor

Rick Youngblood

Director of Technology

ITSP Solutions, Inc.

Ga Tech Electrical Engineer and Computer Scientist

Lean Six Sigma Green Belt

30 years experience in industrial automation and controls technology including 16 years with Honeywell, Inc.

Certified by CTC Analytics in PAL Service and PAL Programming

5 years experience in ITSP PAL lab automation

ITSP PAL Intermediate Course for Lab Personnel

This course is intended to familiarize lab personnel with the different concepts, processes, and software programs used to run and troubleshoot ITSP methods and PAL performance.

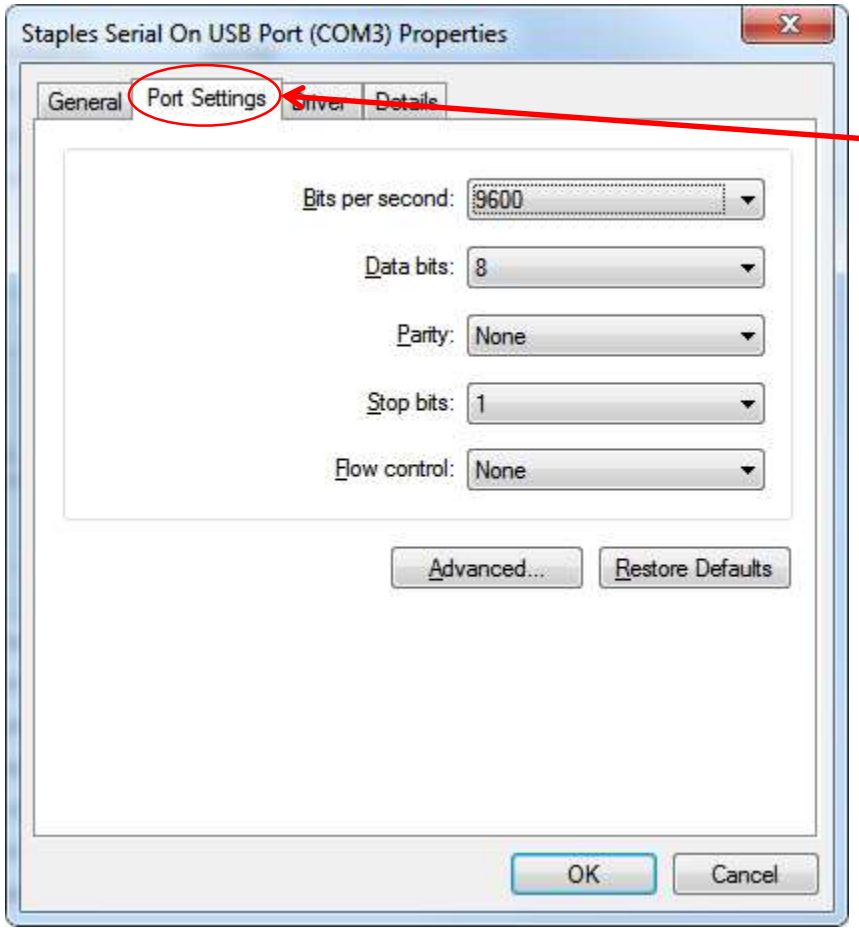
Topics include:

1. Using CTC PAL Loader software to manage PAL firmware from a PC
2. Using CTC PAL Object Manager software to manage PAL objects from a PC
3. Using the local terminal on the PAL to manage objects from the PAL
4. Using CTC PAL Cycle Composer software (simulating supervisory control software, e.g. Agilent MassHunter) to manage methods and sample lists from a PC
5. A discussion of important ITSP concepts

The ITSP PAL Basic Course for Lab Personnel is prerequisite for this course.

PAL Loader Software

PAL Loader software is used to backup, restore, or upgrade PAL firmware.

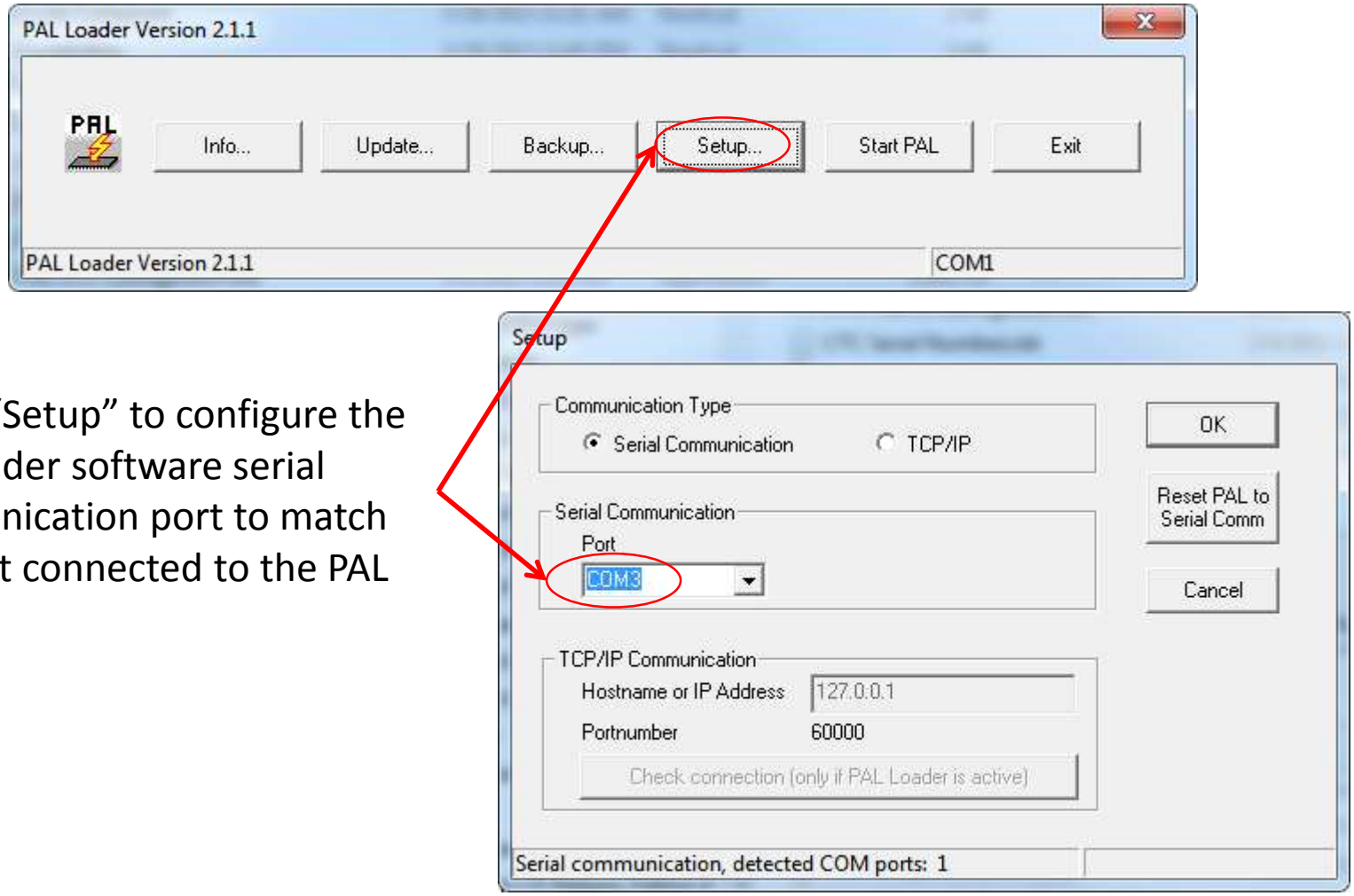


Check the serial port parameters on the COM port of the PC .

Check that no other software is locking the port, e.g. MassHunter.

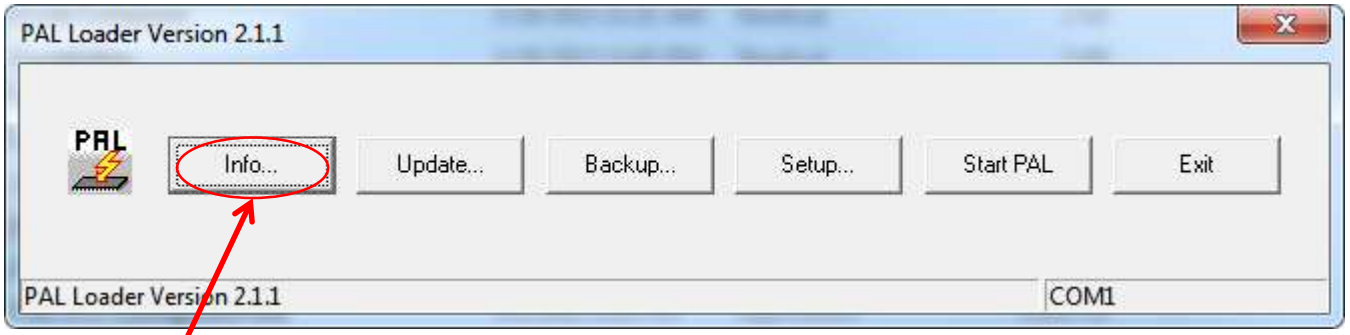
Click on the desktop icon to start the PAL Loader software.

PAL Loader – Setup

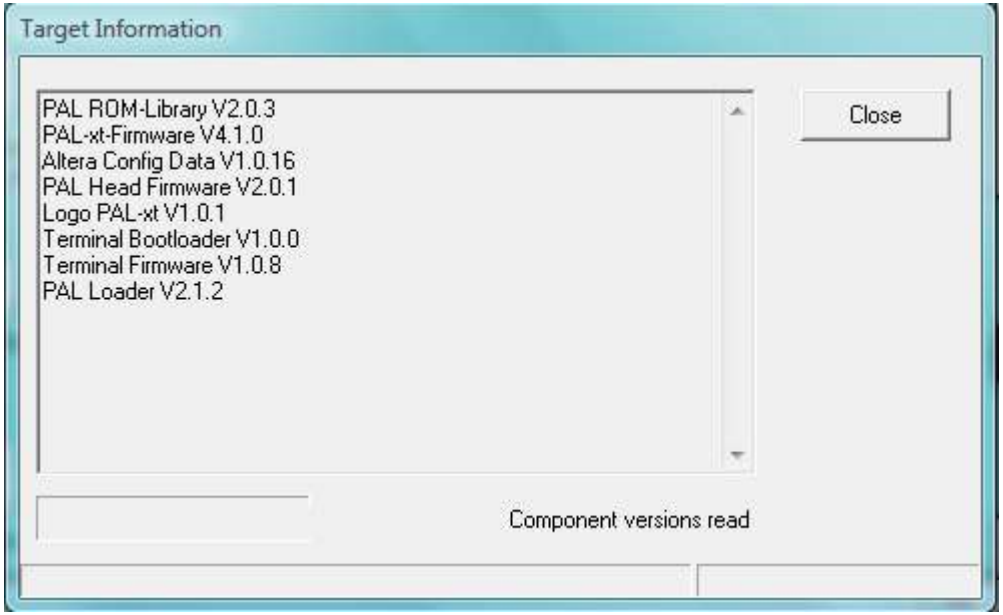


Select “Setup” to configure the PAL Loader software serial communication port to match the port connected to the PAL

PAL Loader – Info



Selecting “Info” reports the current firmware version loaded on the PAL



PAL Loader – Status

There is a red LED light on the back of the PAL X-axis rail that provides information about the current PAL mode and connection/communication status:

- Off: The PAL is in normal operating mode
- Blinking: The PAL is in “Loader” mode
- On: Data transfer is in progress

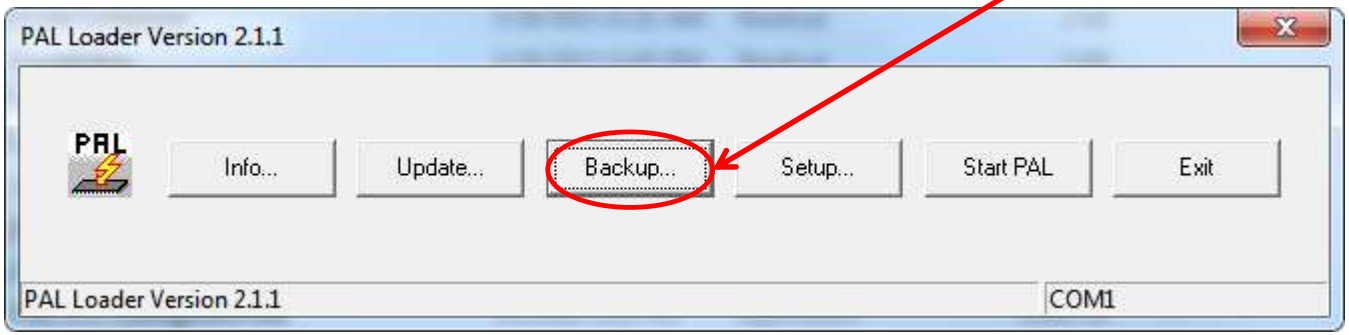
PAL Loader – Mode

If the Loader software is having difficulty establishing a connection to the PAL, after verifying cable connections and port settings, then it is sometimes helpful to force the PAL into Loader Mode. To do this:

1. Turn the PAL power off
2. Turn the power on but when the PAL logo screen appears on the display and 2 beeps are heard, turn the PAL off again (before it completes initialization).
3. Turn the power on again, the PAL will not respond - it is in Loader Mode and will receive commands from the Loader software

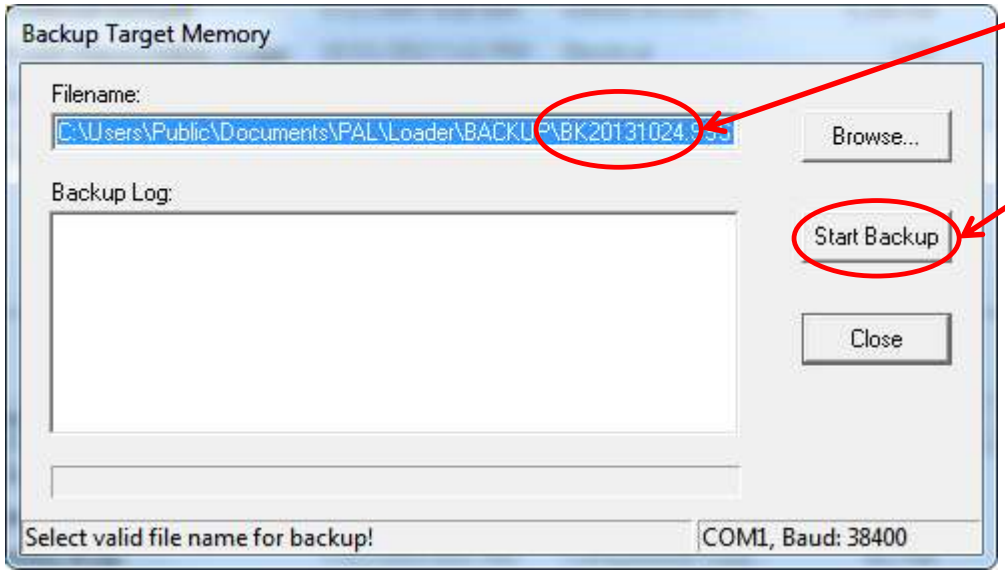
PAL Loader – Backup

Select Backup

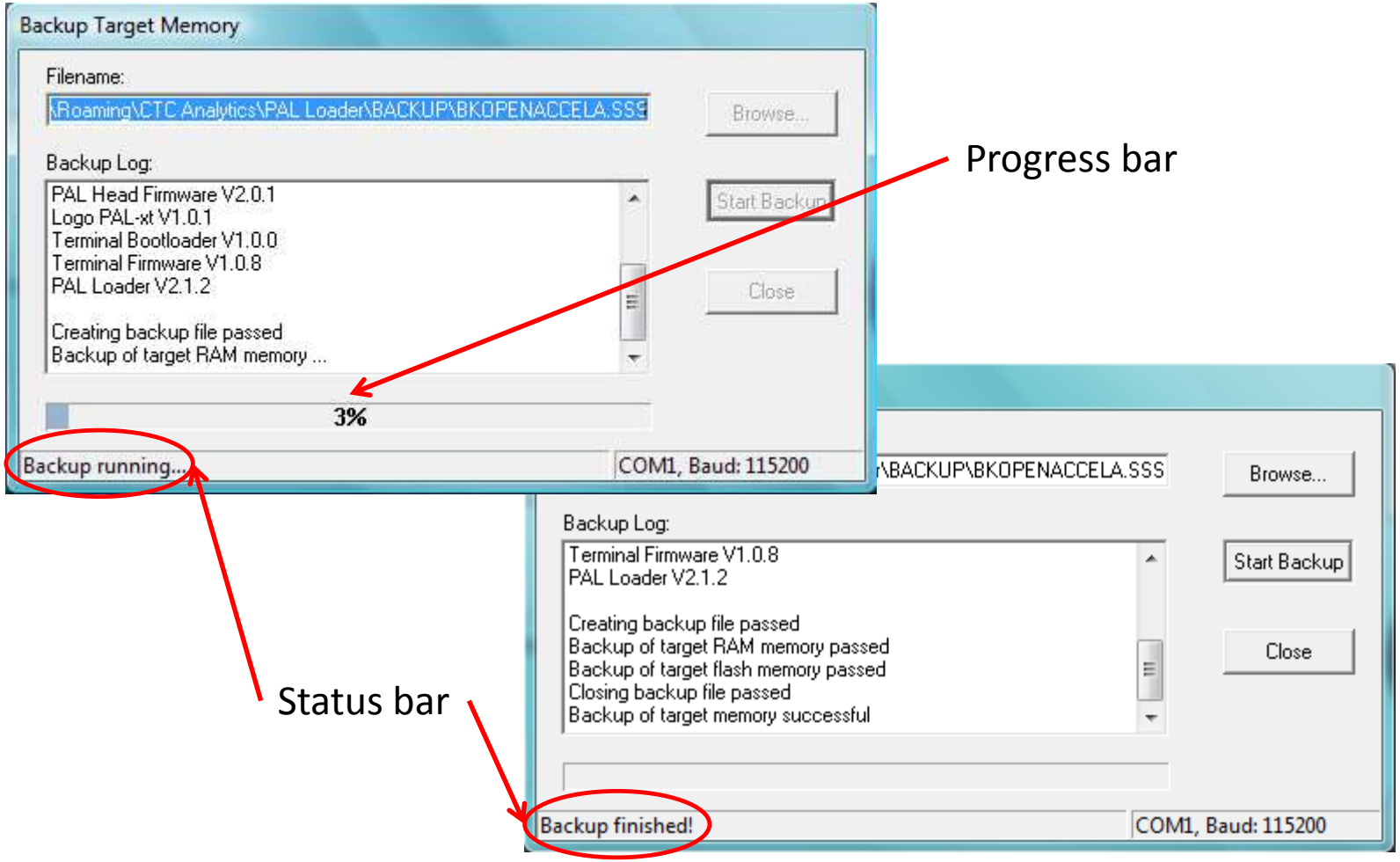


Automatic file naming (can be customized)

Start Backup

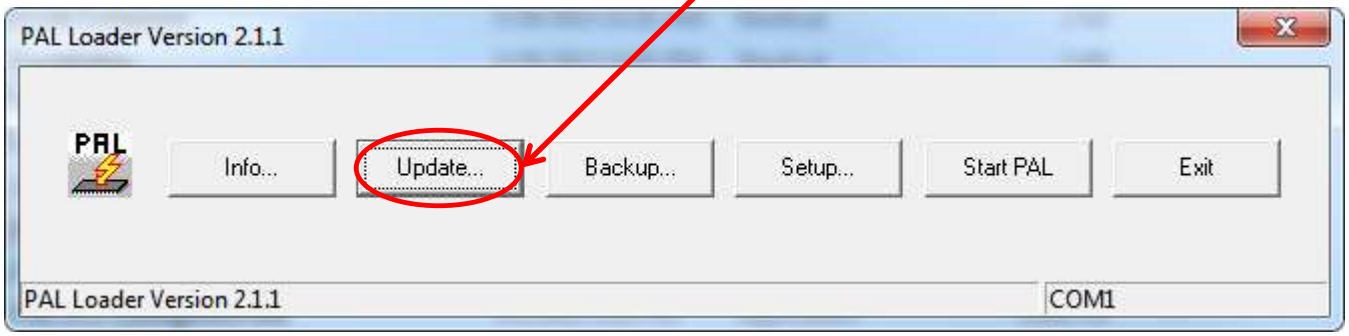


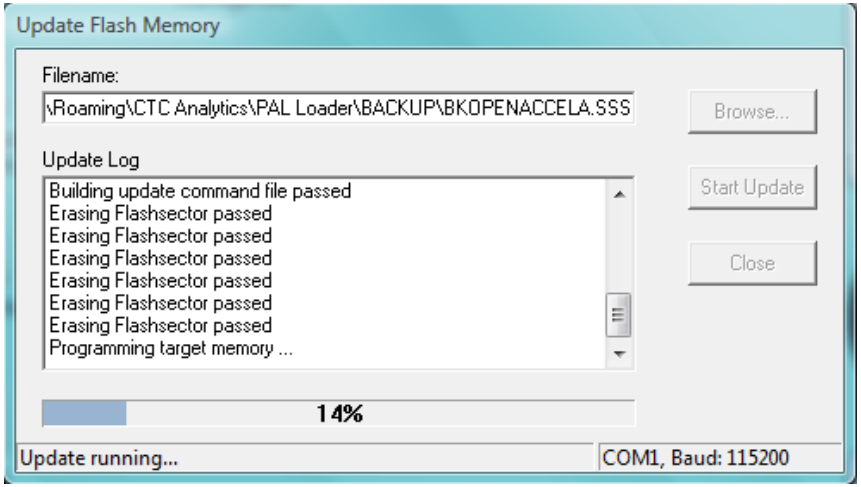
PAL Loader – Backup

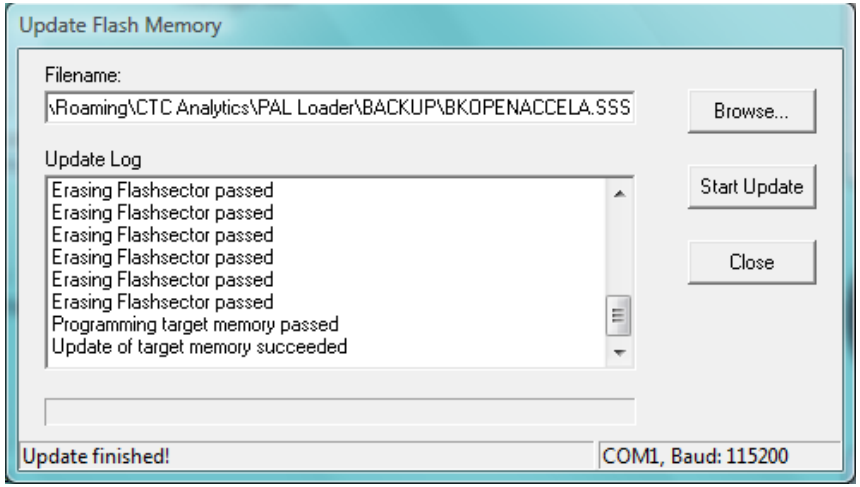


PAL Loader – Update

Select ”Update“ to either upgrade the PAL firmware version level or to reload a firmware backup file (*.sss) to the PAL

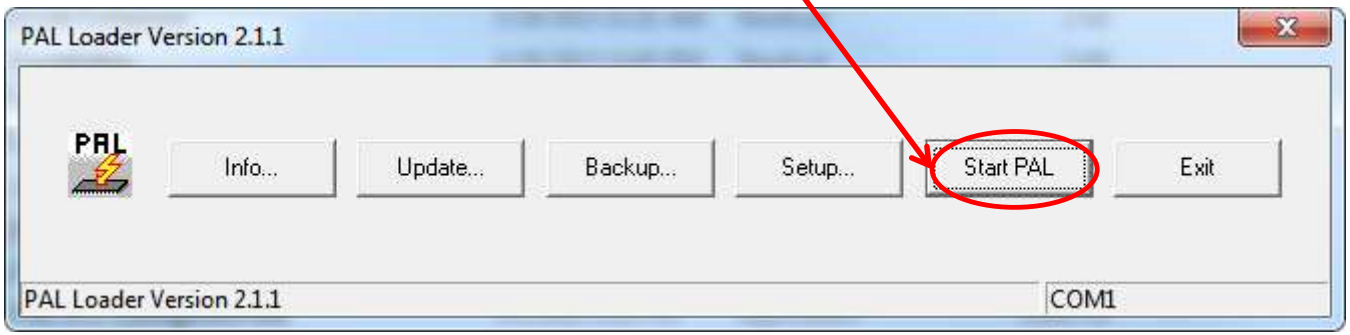






PAL Loader – Start PAL Before Exit

After the update or backup is finished, before exiting the Loader software, always select "Start PAL" to switch the PAL from "Loader" mode to "Running"



PAL Firmware Troubleshooting

Backup File

- It is highly recommended to perform a backup with the PAL Loader software after an installation of, *or any change to*, a PAL.
- If any uncontrolled changes are made or a damage does corrupt the firmware, e.g. an electric surge, the user always has a fast way to recover.
- First, attempt a PAL reboot to see if that fixes the problem.
- If not, then the original complete backup file can be re-loaded using “Update” with the PAL Loader software, and the PAL will be up and running within a few minutes.

Anytime any settings or configuration of a PAL have been changed, a new backup file should be made.

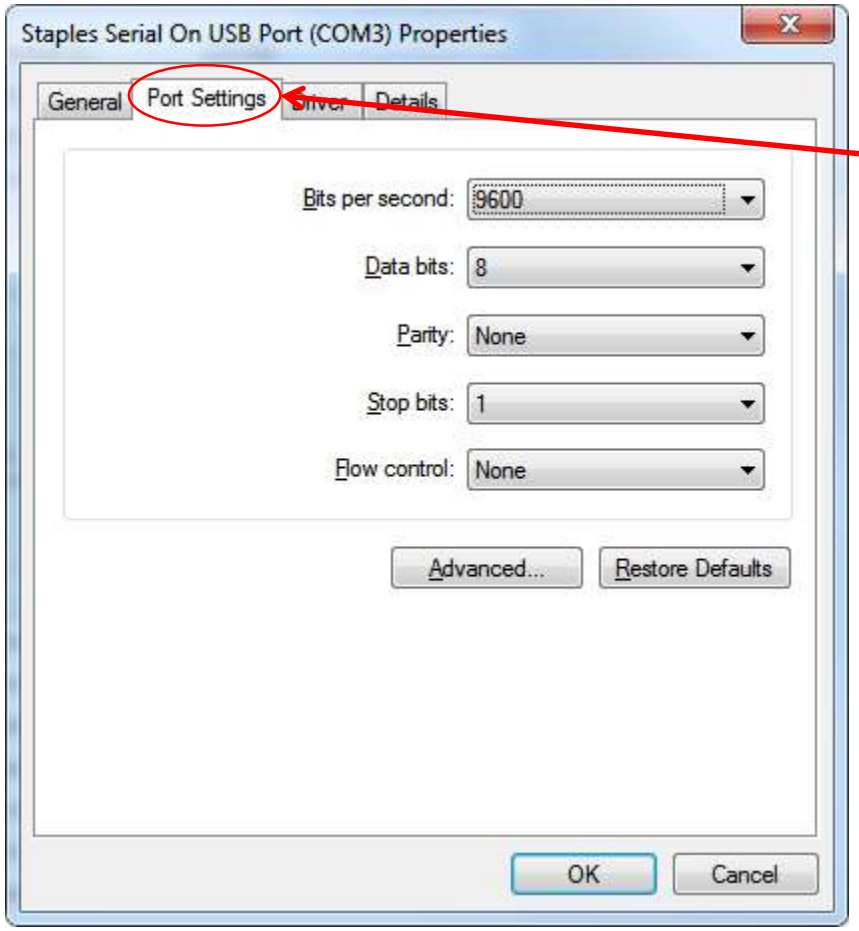
PAL Firmware Troubleshooting

Firmware Errors

- Naturally, any obvious possible source of an error should first be checked and eliminated.
- Firmware errors can occur in various forms, e.g. only some parts may be corrupted and a specific object might create a problem.
- Many errors can be eliminated by simply restoring a backup file and this fix step is very fast and can easily be done by lab personnel.
- Experience has shown that firmware errors related to an object can often be eliminated by simply reloading the backup file.
- Typical cases include:
  - Plunger sensor not detected
  - Head communication error
  - Vial not detected
  - An “STR” (String) error code reported on the message line of the local terminal

PAL Object Manager Software

PAL Object Manager software is used to load new objects to the PAL.



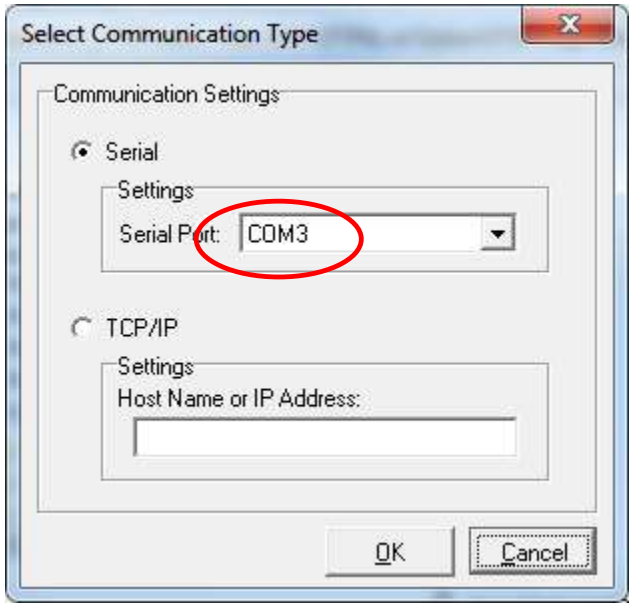
Check the serial port parameters on the COM port of the PC.

Check that no other software is locking the port, e.g. MassHunter.

Click on the desktop icon to start the PAL Object Manager software.

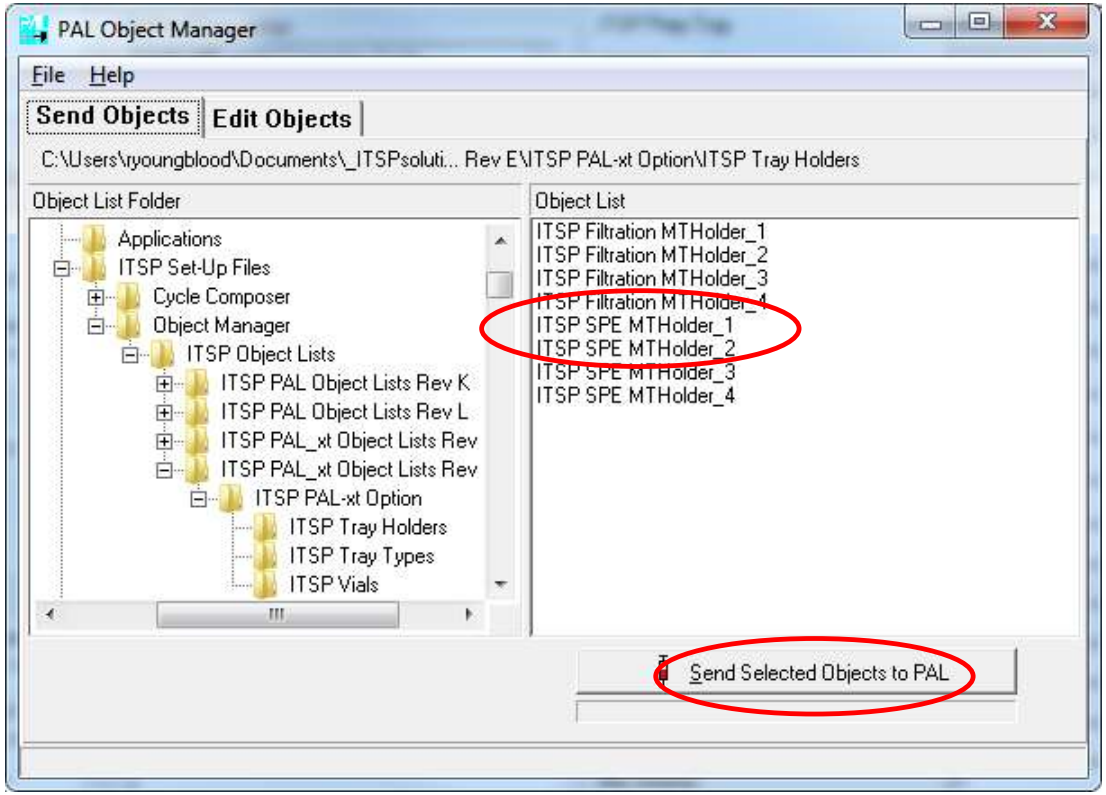
PAL Object Manager – Communication

In the Menu bar, select “File” , “Options”, “Communication” to configure the PAL Object Manager software serial communication port to match the PC serial port connected to the PAL .



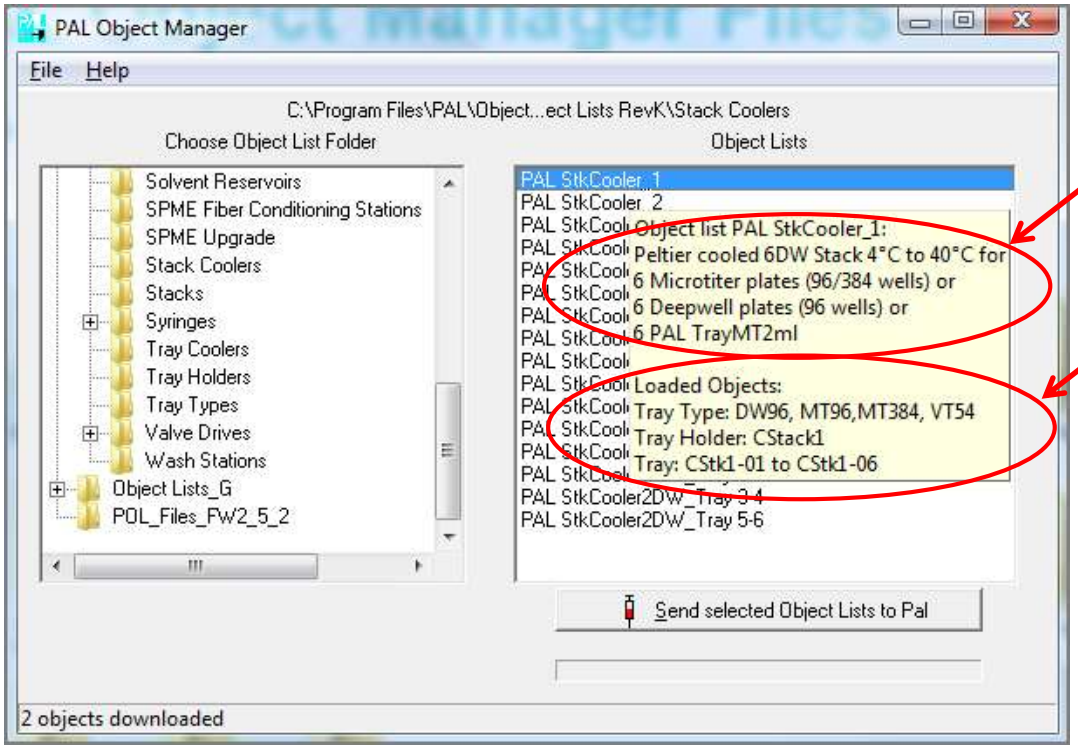
PAL Object Manager – Load Objects

- In the Menu bar, select “File” , “Options”, “Object List Folder” to browse to the appropriate object name within the object list.
- Select the desired object name and click "Send Selected Objects to PAL“.
- When finished, close the PAL Object Manager software.



PAL Object Manager – Object Lists

Additional information is available from the context popup text box when mousing over individual object lists.

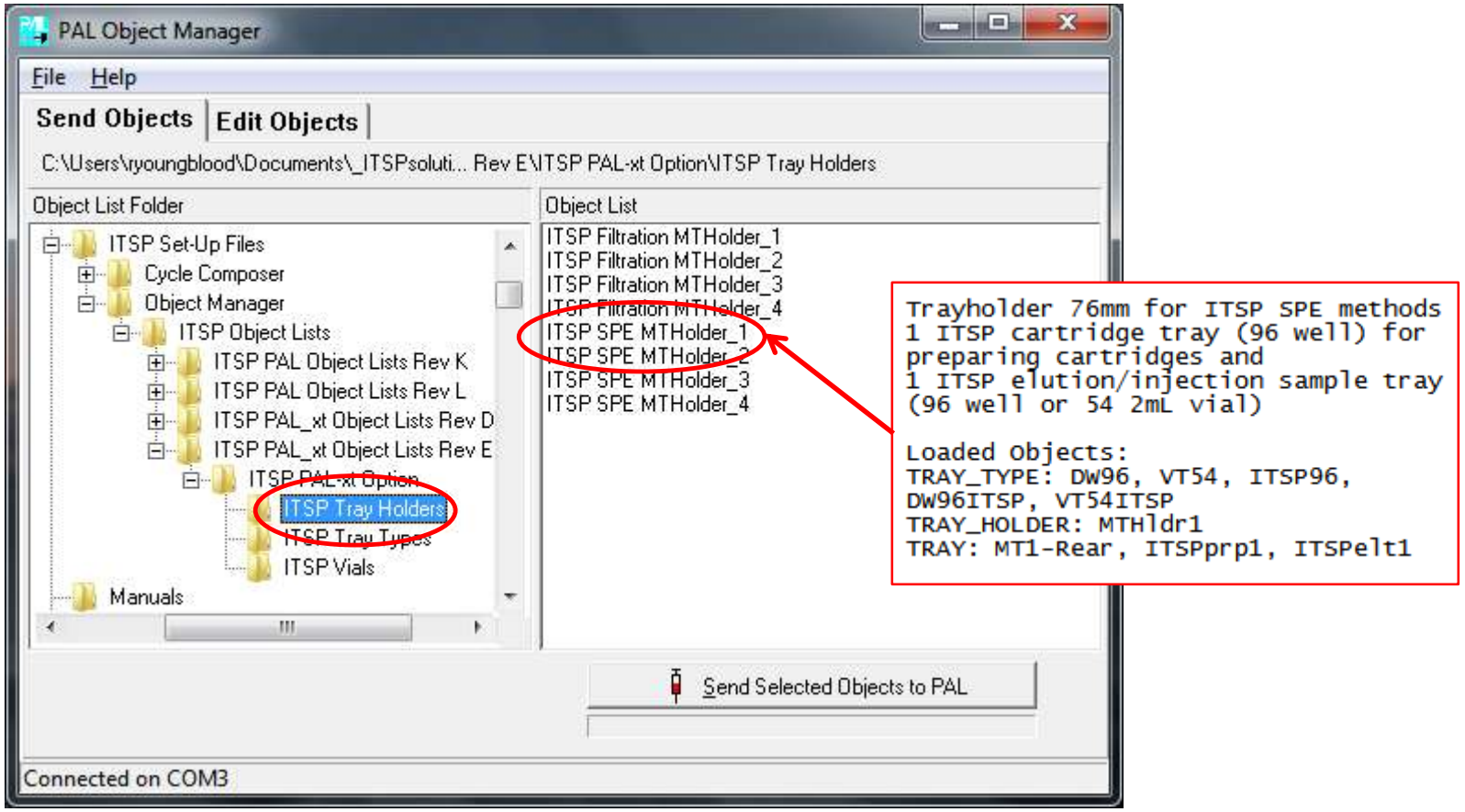


Explanation of the object list

List of loaded objects

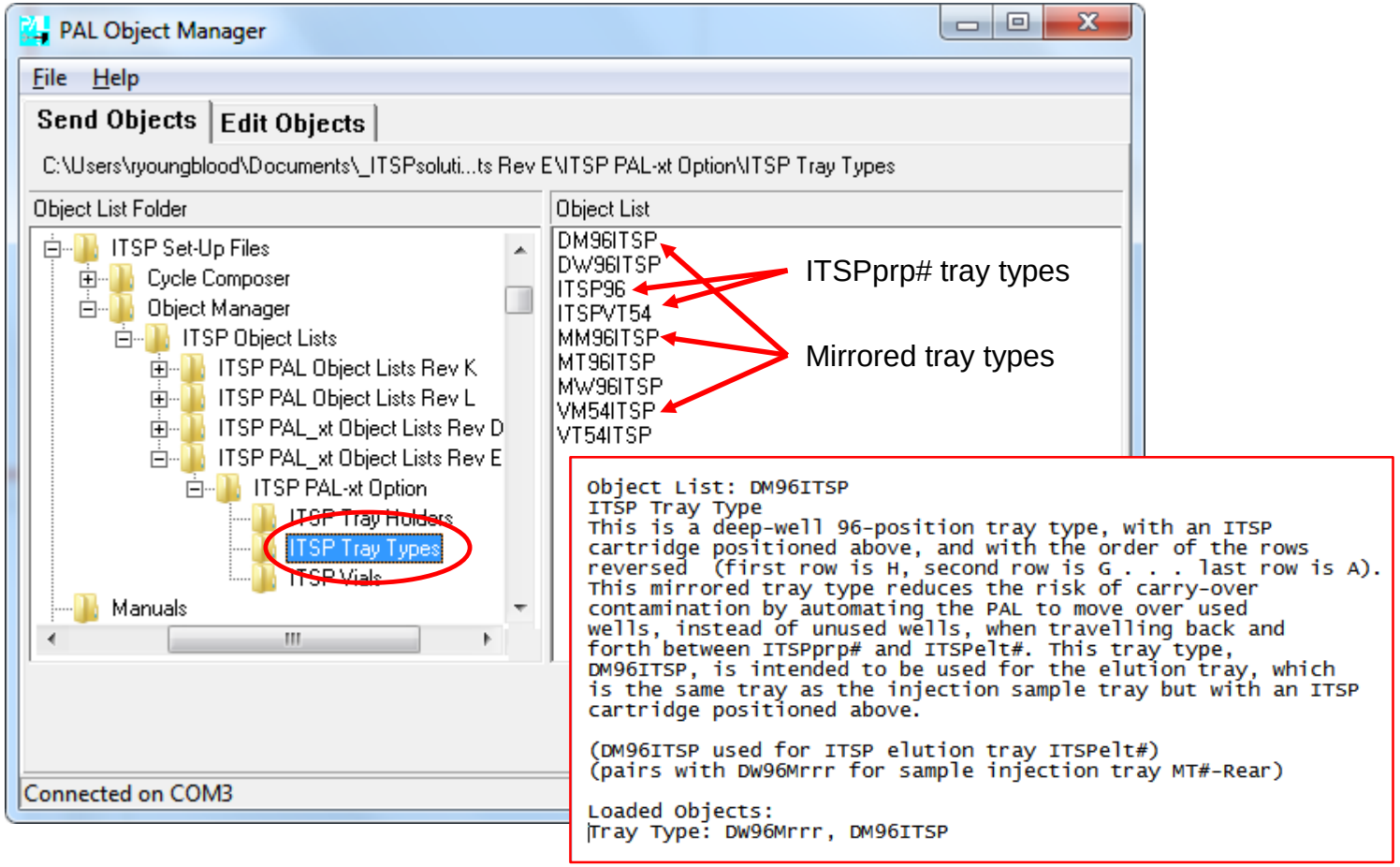
PAL Object Manager – ITSP Tray Holders

Each ITSP tray holder file contains all the tray holder, tray, and tray type objects associated with the ITSP station in that position.



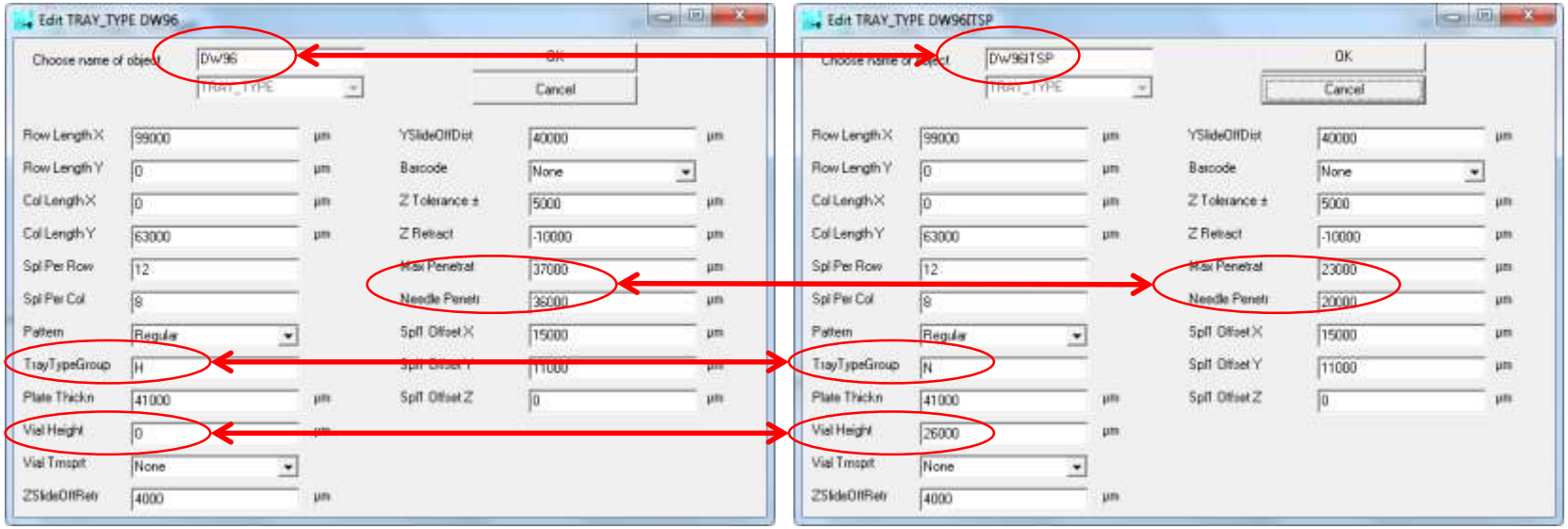
PAL Object Manager – ITSP Tray Types

Each ITSP tray type file contains two matching tray types, one for the elution tray and one for the injection sample tray superimposed on the same physical PAL tray location, e.g. If tray "ITSPelt1" is tray type "**DW96**ITSP", then the corresponding tray "MT1-Rear" will need to be tray type "**DW96**". This is the tray located directly behind the ITSP tray "ITSPprp1".



PAL Object Manager – ITSP Tray Types

- The difference between the two is the presence of the ITSP cartridge at tray “ITSPelt1”.
- Notice the difference in vial height, and needle penetration.
- * The tray type group is also different, to facilitate the use of the PAL local terminal to assign only ITSP tray types to ITSP trays.



PAL Object Manager – Mirrored ITSP Tray Types

If tray “MT1-Rear” is tray type “DW96**Mrrr**”, then tray “ITSPelt1” should be tray type “D**M**96ITSP”. These two tray types compose a coordinated mirrored tray set that is used by ITSP to reduce carryover and to improve chain of custody at the elution plate location.

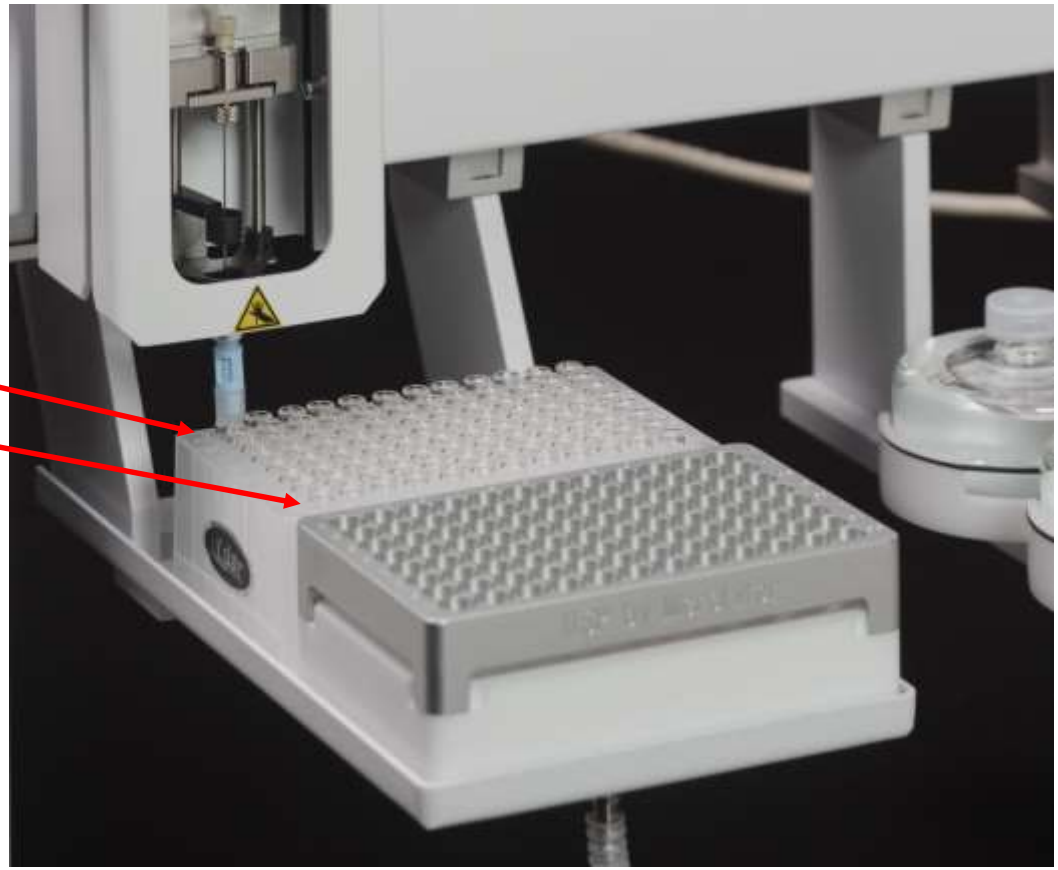
When using mirrored tray types for the elution tray, cartridges in ITSPprp1 row “A” are eluted to ITSPelt1 row “H”, “B” to “G”, etc.

In the elution tray:

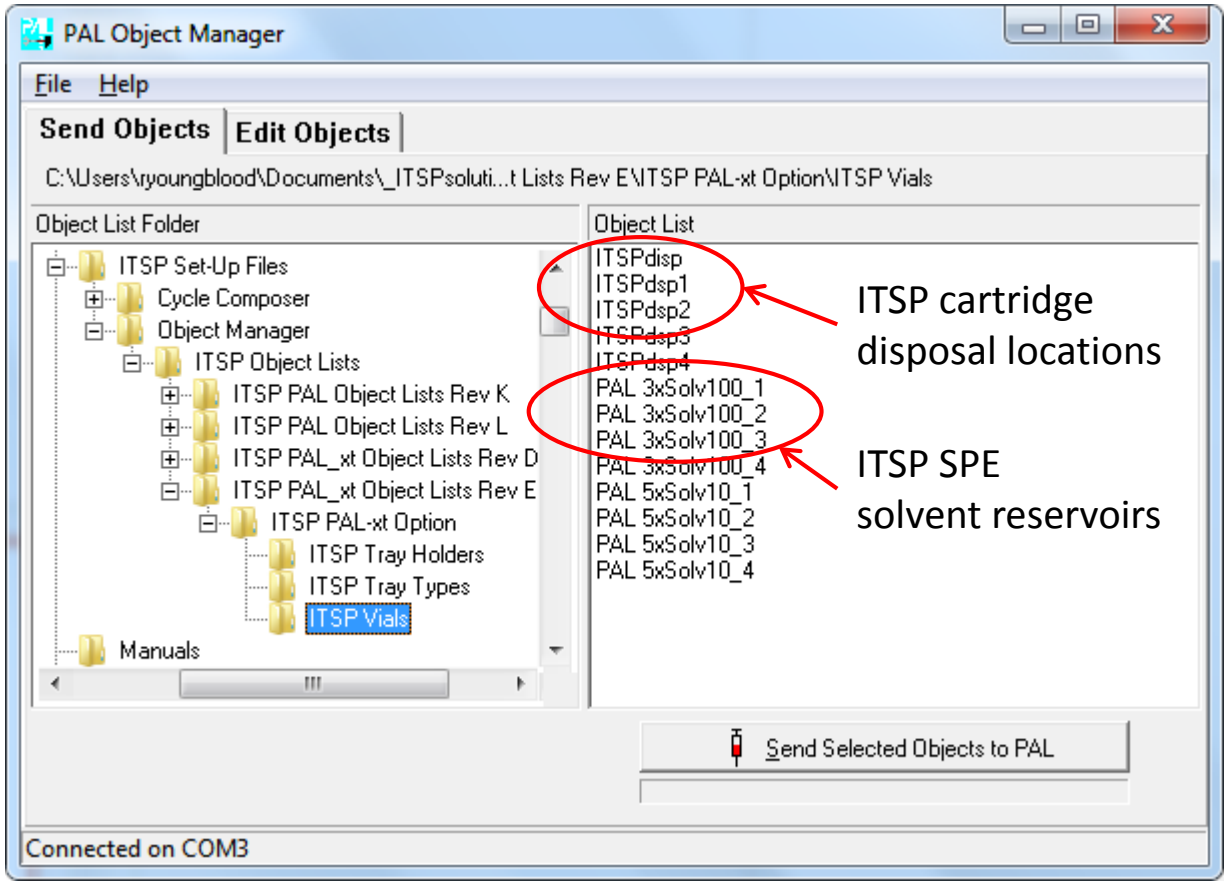
Row “A” is now the *last* row

Row “H” is now the *first* row

Cartridge “A1” elutes to well “H1”, “A2” to “H2” . . . “B1” to “G1”, etc. Therefore, ITSP cartridges are never transported over an unused well.



PAL Object Manager – ITSP Vials



The location to dispose of spent ITSP cartridges is a PAL vial object class.

PAL vials can be used to hold SPE conditioning, washing, and elution solvents for ITSP methods.

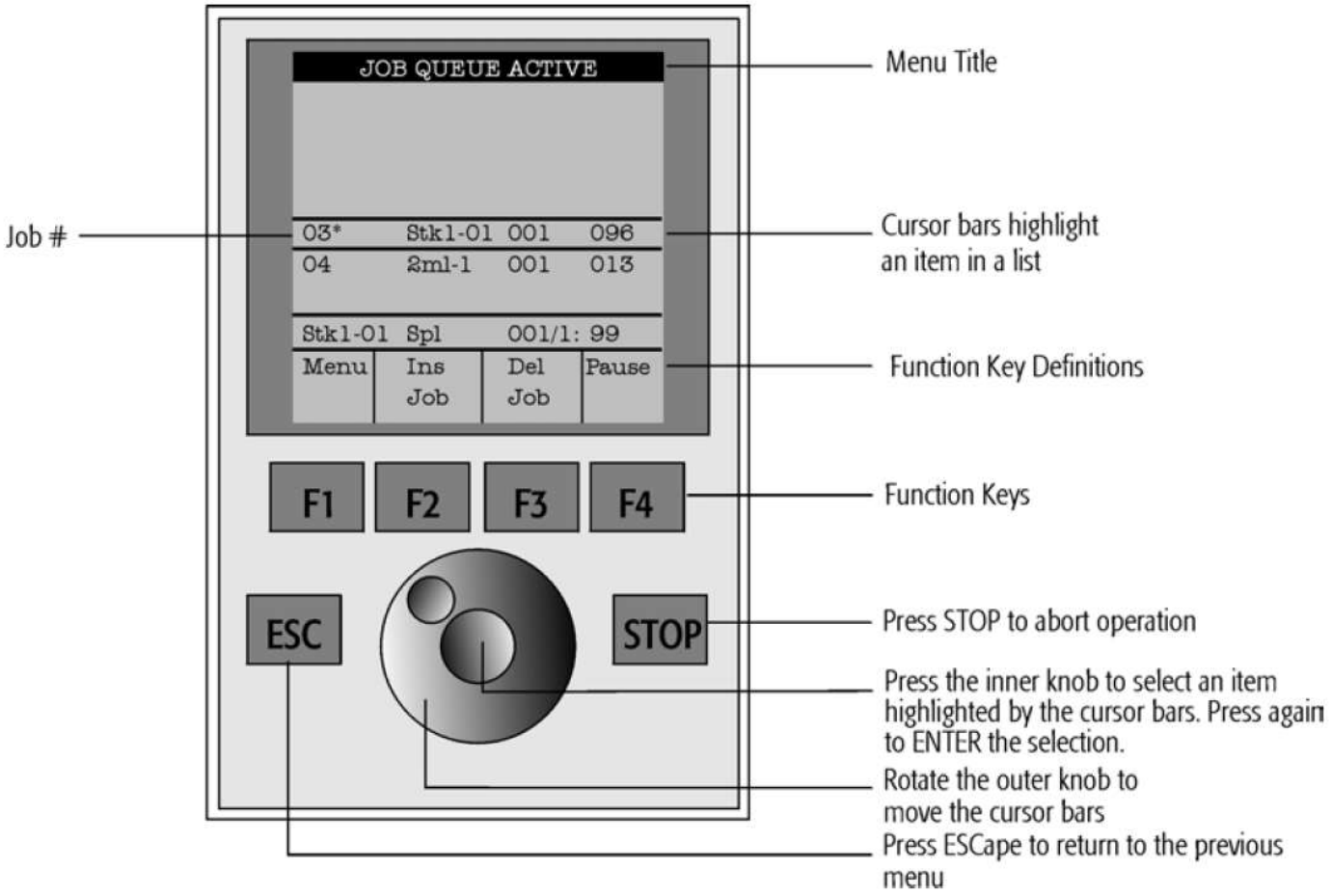
PAL Object Manager

Finally, after loading new objects on the PAL:

- First, teach or verify the position of the new objects using the local terminal.
- Then, make a new backup file of the PAL firmware using the PAL Loader software.

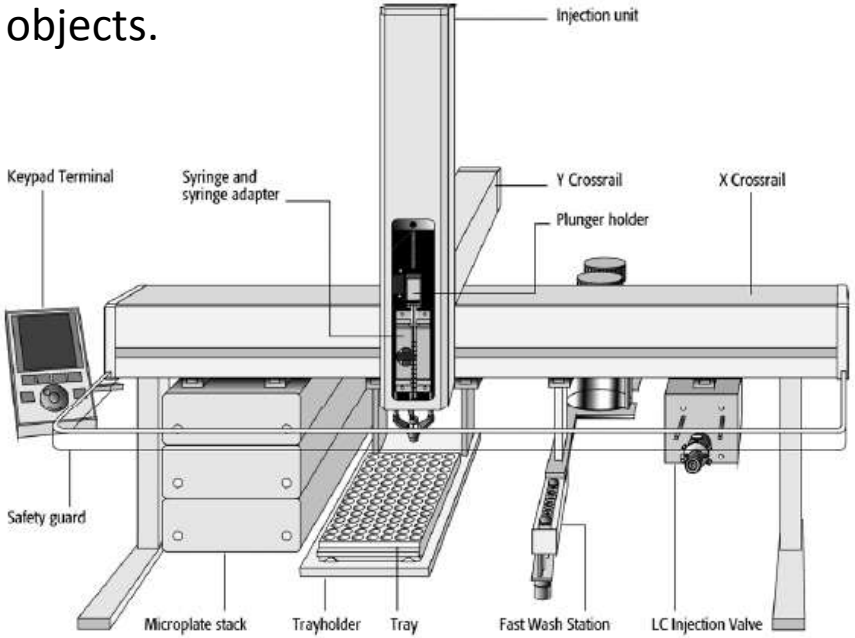
Using the PAL Local Terminal

The PAL Local Terminal is used for various PAL operations, e.g., priming wash stations or changing syringes.



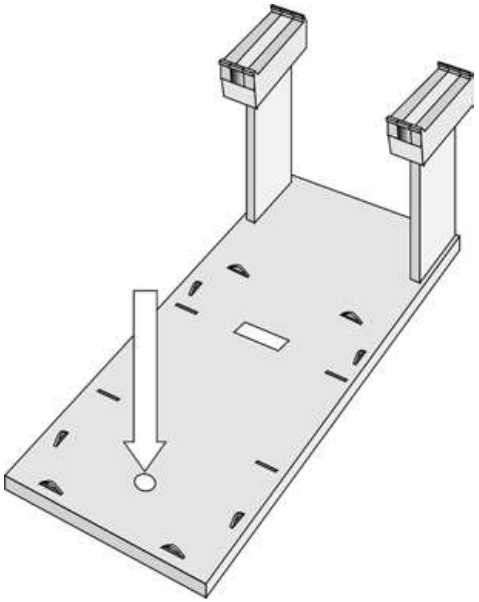
Using the PAL Local Terminal

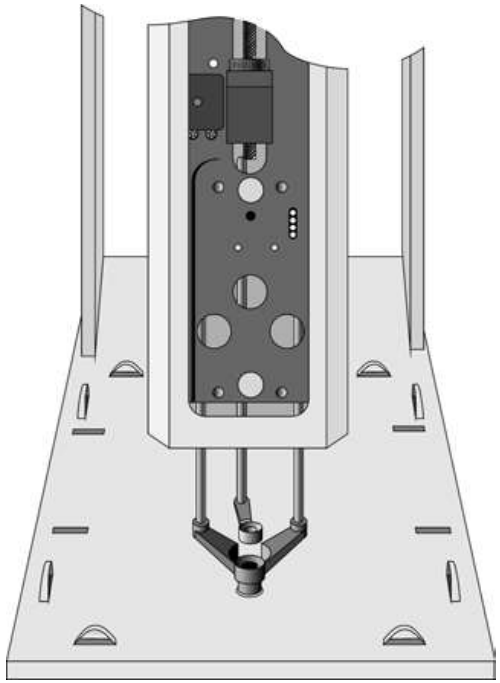
- *Make a firmware backup using Loader before you start making changes to the PAL using the local terminal!*
- *Remove the syringe adapter from the injection unit before performing the following steps! (or else risk «Z»-ing a needle.)*
- Make sure the solvent reservoirs, tray holders, valve, and wash station are properly and securely mounted to the PAL X-axis. The following description is an example of how to teach the reference position for a tray holder. The described procedure is common to all PAL objects.



Tray Holder Reference Position

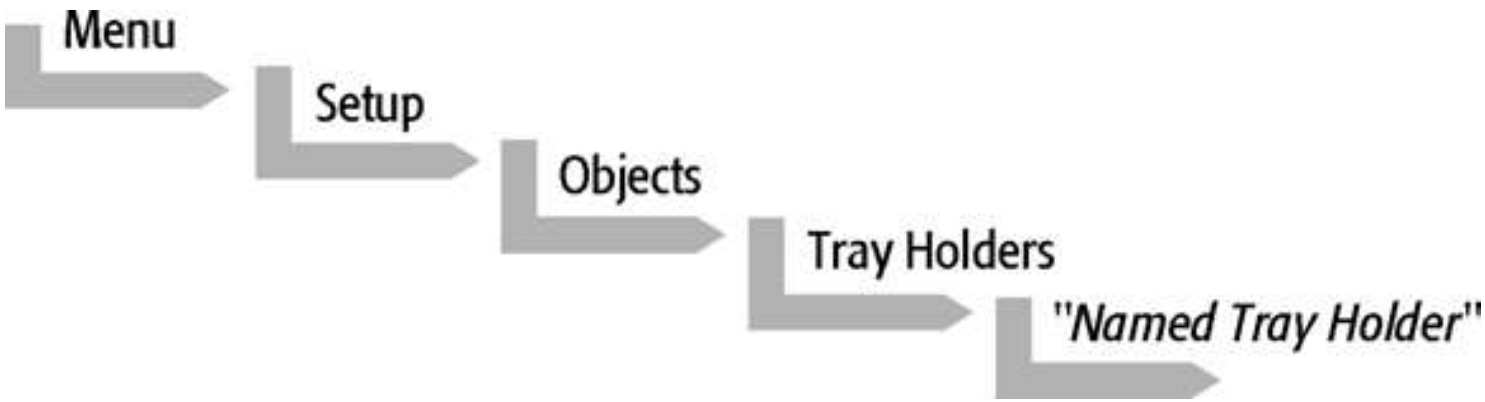
For a tray holder, the reference position is a hole in the base plate. The lower needle guide should be centered in the hole with the bottom of the needle guide flush with the bottom of the base plate.





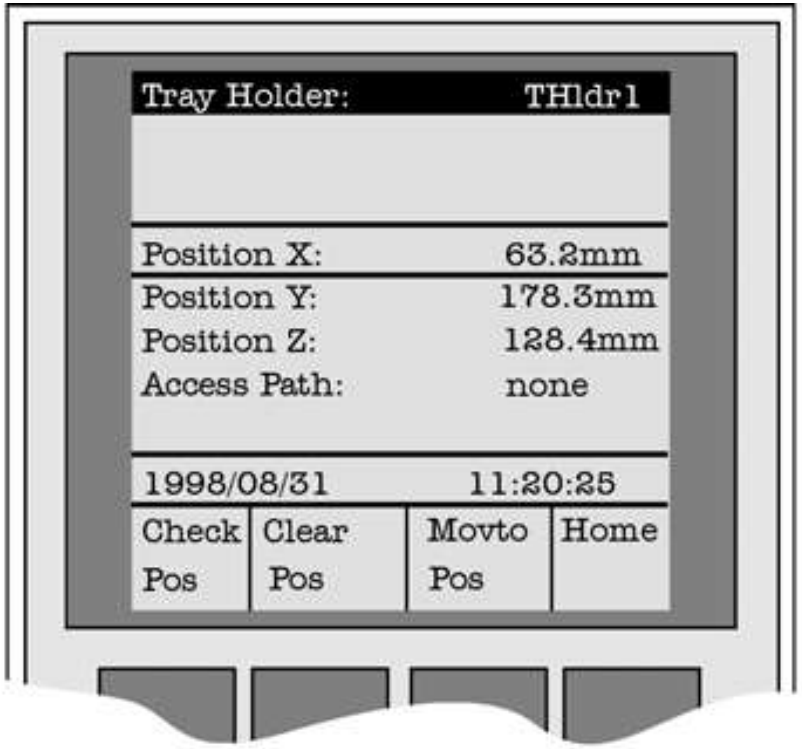
Steps for Teaching an Object

Switch on the PAL power supply. When the "Job Queue" menu is displayed, complete the following sequence (common to all objects): where "Named Tray Holder" represents a predefined tray holder (e.g. THldr1 or MTHldr2).



Steps for Teaching an Object

After selecting "Named Tray Holder" (e.g. THldr1 or MTHldr2), the X/Y/Z- positions for the selected object will be displayed.



Steps for Teaching an Object

- Highlight the item position X with the cursor bars and press “ENTER” (the inner knob). The injection unit will move to the defined X-axis position.
- Rotate the outer knob to adjust the X-axis position to the tray holder reference Position.
- Press the inner knob to “ENTER” the position X value.
- Repeat the steps above for position Y and position Z.
- Press the F3 button "Move to Zero". The injection unit will move to the HOME position.
- Verify the defined X/Y/Z-positions by pressing F1 "Check Pos".

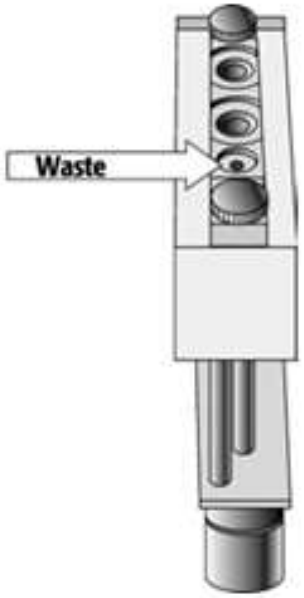
Wash Station & Waste Position

Wash Station (Wash1 or Wash2)

- For a fast wash station the reference positions are two holes above the wash station glass liners. The lower needle guide should be centered in these holes with the bottom of the lower needle guide lightly touching the surface of the wash station assembly.

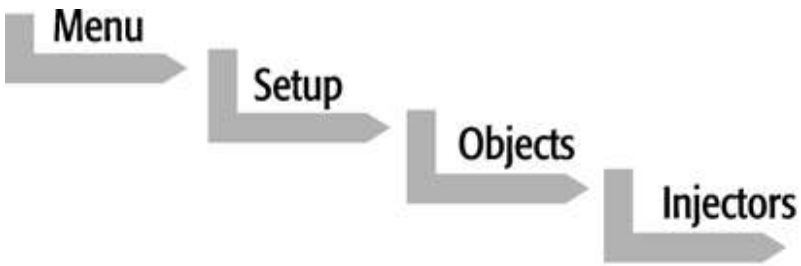
Wash Station & Waste Position

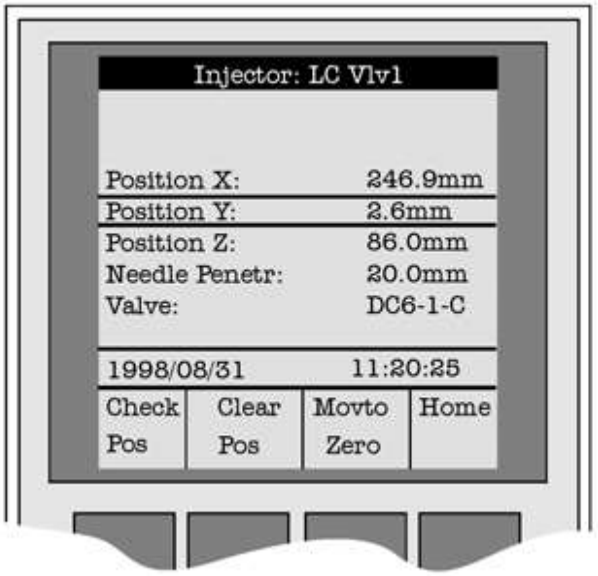
- The Waste position represents an “Injector” within the PAL firmware. It is defined in the object class “Injectors“.
- For the Waste port the reference position is a hole (slightly larger than the needle guide) in front of wash2. The lower needle guide should be centered in this hole with the bottom of the lower needle guide lightly touching the surface of the wash station assembly.



LC Injection Valve

- For an LC valve, the reference position is the valve needle guide fitting mounted on the top valve port.
- The lower needle guide of the injection unit should be centered in the valve needle guide fitting. Adjust the Z-position such that the bottom of the lower needle guide is just touching the surface of the valve needle guide fitting. Then reduce the value by 2.0mm (up).





Training ITSP Trays

The training of trays is not standard with normal PAL usage but it is critical with ITSP to ensure robust PAL performance.

The X-Y target for training an ITSP tray is not the septum in the seal.

Train the X-Y of an ITSP tray to the exact center of the septum at well A1 to allow the needle to penetrate the septum directly above the bore of the cartridge.

The Z Offset of ITSP trays should be trained 0.5mm above the cartridge, because the PAL transports ITSP cartridges with the Lower Needle Guide “blocked”.

Verifying Positions

After all positions have been loaded and trained, then:

- Install a syringe.
- Verify all positions.
- Check the needle penetration for all positions.
  - Menu/Utilities/[Named Tray]/Needle Penetration
- After all positions have been set, perform a backup of the current configuration.

PAL Firmware

Firmware is the base level of control in the PAL autosampler.

- Within the PAL, firmware atoms operate on objects.
- Atoms and objects have parameters.
  - Atom parameters can be:
    - Left undefined by the method developer, or
    - Hard-coded at the macro level of control by the method developer, or
    - Passed 1 level up to the method level of control as a macro (or cycle) variable, or
    - Passed 2 levels up to the sample list level in some cases. (In ITSP methods, those cases are SL.Tray, SL.Index, and SL.Volume.)
  - Object parameters which are left undefined within a macro by the method developer can then still be controlled from the local terminal by lab personnel.

PAL PC File Extensions

- File Extensions
  - **P**AL **ME**thods: *.pme
  - **P**AL **MA**cros: *.pma
  - **P**AL **S**ample **L**ist: *.psl
  - **P**AL **O**bject **L**ist: *.pol (latest_pal.pol and latest_pal.xml, which are the latest PAL configuration, and other object files)
  - PAL firmware backup files: *.sss
  - Cycles: *.cyx
- When e-mailing ITSP Solutions for support with custom ITSP macros or cycles, please include the following attachments:
  - [macro_name].pma (or [cycle_name].cyx for MassHunter cycles)
  - Latest_pal.pol (or latest_pal.xml for MassHunter cycles)
  - Method macro (or MassHunter cycle) parameter settings (e.g., *.pme)

ICC OEM's

Agilent

Thermo SCIENTIFIC

Waters

SHIMADZU

DIONEX

AB SCIEX

VARIAN

BRUKER

DataApex

ITSP SOLUTIONS, INC.

Regular PAL Maintenance

Suggested intervals for other maintenance procedures to ensure uninterrupted operation:

Maintenance Procedure	Interval
Clean the outside of the instrument	Weekly or more often as needed
Replace the syringe plunger and clean the syringe; replace the syringe (needle)	The syringe, and/or plunger, and/or needle need to be replaced on a regular basis depending on the application.
Clean SPE and wash station solvent reservoirs	Weekly or more often as needed
Exchange needle seal in the LC injection port	Monthly or more often as needed
Replace worn bungee cords	Quarterly or more often as needed
Preventative maintenance by certified PAL service technician	Annually or more often as needed

Annual Preventative Maintenance and Repair

- Maintaining the PAL system as described above is considered a daily or weekly PM.
- As a general rule the system should be thoroughly checked and cleaned at minimum once a year. This annual PM should be performed by a certified PAL service technician using a PM Kit from CTC Analytics.

Troubleshooting

The following are suggested steps for troubleshooting problems:

Symptom	Possible Cause	Solution
Clicking sound when injecting to LC valve	Needle penetration for Injector too deep	Re-teach needle penetration
Intermittent error message: “vial not found” error# 115	Z offset value wrong for tray	Re-teach Z offset value for tray
PAL collision error at ITSP object	X, Y, or Z offset value wrong at ITSP object	Re-teach X, Y, or Z offset for ITSP object
PAL transports ITSP cartridge at an angle	X or Y offset value wrong at ITSPprp#	Re-teach X or Y offset for ITSPprp#

Important ITSP Concepts

- ITSP methods are usually run serially on-line from MassHunter eluting to an open well of a 96-well plate.
- ITSP methods are sometimes run in off-line batches from Cycle Composer software.
- When running batches off-line, or when chain-of-custody is paramount, such as in Forensic Toxicology labs, the elution tray should be a sealed 2mL vial 54-position tray using a vial lock:

The cap on the vial must be the MicroLiter #11-0056, available from Wheaton, which has a starburst pre-cut septa.

The MicroLiter Vial Lock #12-0000-VL is available from Wheaton.

This is the only solution approved for eluting ITSP to a closed well or vial.

Certification Test

1. Perform a backup of the PAL.
2. Load a new ITSP tray type object to the PAL
3. Change the ITSP elution tray to the new ITSP tray type.
4. Teach an ITSPprp# tray location.
5. Teach an ITSPelt# tray location.
6. Teach the ITSPdisp vial location.
7. Place all solvent reservoirs and trays in their proper location on the PAL deck and describe to the instructor the purpose of each location.
8. Replace the syringe.
9. Create an ITSP method to run an existing macro utilizing the new tray type and test step-wise.
10. Create an ITSP sample list to run an existing method and test on 2 samples.

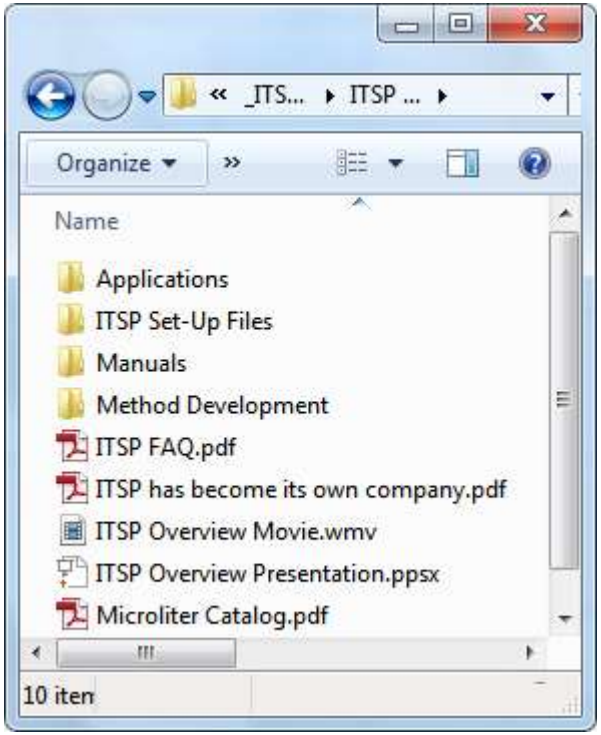
Additional ITSP Resources

The USB memory stick in the ITSP startup kits include:

- ITSP application notes
- ITSP PAL objects
- Example ITSP macros and methods
- Example ITSP automated SPE method development macros
- ITSP setup guide
- ITSP method development guide
- Other resources such as catalogs, videos, and presentations with more information about ITSP.

The USB memory stick in the ITSP PAL training courses include:

- Copies of the course presentations
- Copies of additional PAL reference materials



Instructor Contact Information

Rick Youngblood

Director of Technology

ITSP Solutions, Inc.

706-395-8300

855-395-8300 toll free

Rick.Youngblood@ITSPsolutions.com

www.ITSPsolutions.com

http://www.linkedin.com/in/rickyoungblood/

Mailing Address	Shipping Address
212 Northlake Drive	10 S. Carolina Street
Hartwell, GA 30643	Hartwell, GA 30643

ITSP Pal Intermediate Course for Lab Personnel

ITSP™ SOLUTIONS, INC.