P-Sperm™ & SQA-Vp
Integrated PIG System

User Guide

Catalog # 5111

Version 1.0

May, 2021

TABLE OF CONTENTS

Section 1: Overview

The SQA-Vp and P-Sperm™ boar management software work together to provide an integrated PIG System (IPS) for boar semen analysis, AI Dosing, extended semen QC and many other functions related to pig reproduction.

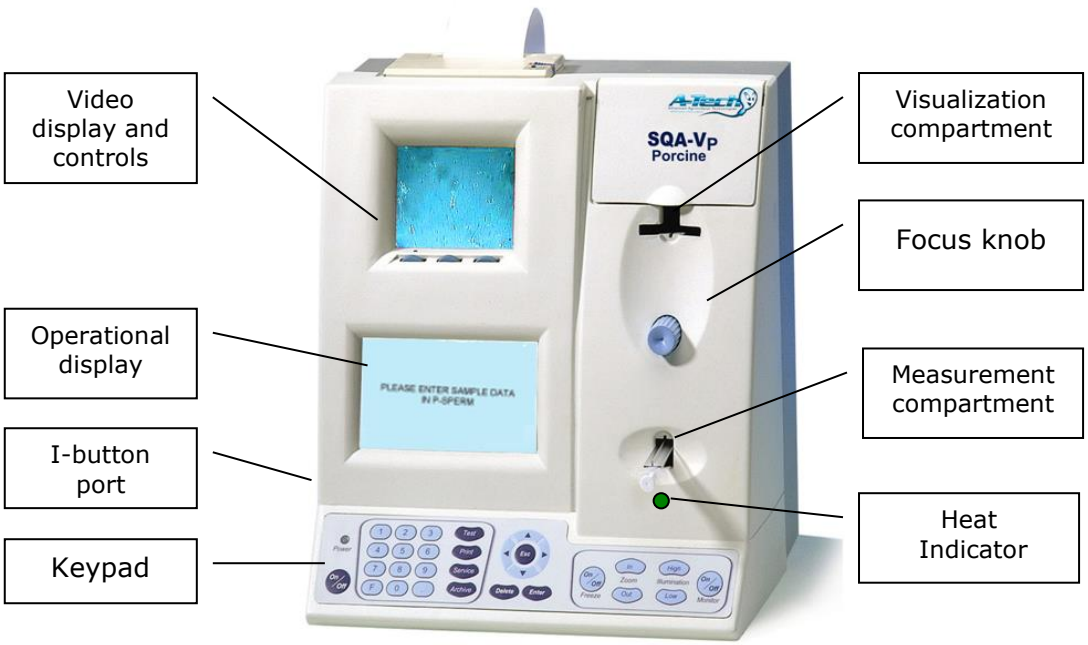
FRESH or **EXTENDED** samples are prepared and aspirated into a multi-use, washable testing capillary. The capillary is inserted into the measurement compartment of the SQA-Vp where state of-the-art technology in electro-optics, computer algorithms and video microscopy produce rapid, accurate and precise analysis of Fresh/Extended boar semen. Samples can be viewed using the on-board video visualization system with magnification capabilities of X300 through X500.

All sample data entry and boar/dosing reports are handled in the PC-based P-Sperm boar management software that operates together with the SQA-Vp as an integrated system. The following functions and features are listed below:

- Rapid data entry utilizing drop down menus that are updated during the testing process or from pre-set tables.
- Fast test results - under a minute to report all semen parameters. Perfect for high throughput industrial boar facilities.
- Easy to use QwikClick feature for counting droplets or assessing detailed morphology defects.
- Automated semen dosing calculation based on single-sire, split or pooled samples.
- Semen dosing reports and data analysis.
- Numeric and graphical Boar information/reports.
- Boar semen samples can be viewed on a large PC screen (video or image) and images can be captured and stored with individual boar records.
- Report options: Fresh Samples, Extended Samples, Daily Production, Morphology (differential), Pooled Batches, Remaining Semen, Individual Boar information.

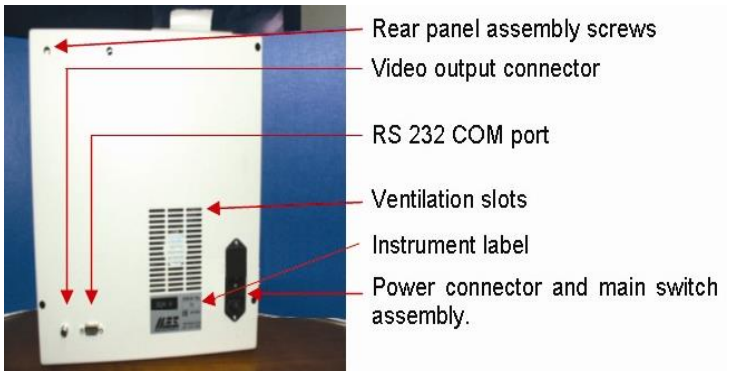
Section 2: SQA-Vp Overview

SQA-Vp



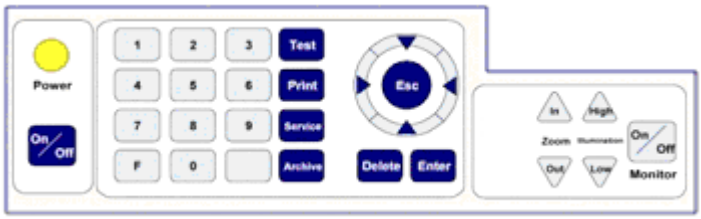
The Rear/Side Panel

The Rear Panel components are as follows:



I-button

Keypad and Navigation

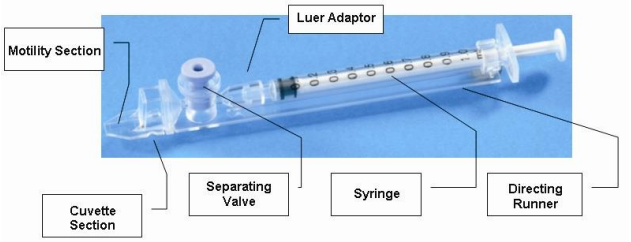


The SQA-Vp front panel Keypad is used for navigation of the SQA-Vp:

- Service Key = Opens the Service Menu.
- ARROW keys = To move within the Service Menu screen.
- ENTER = To select menu options and to move to the next screen.
- ESC = Return to a previous screen or row.
- ZOOM = Set the visualization focus
- Other keys = Calibration/verification (for manufacturer's use only)

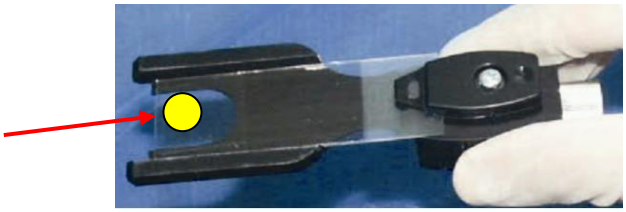
SQA-Vp Testing Capillary

The testing capillary consists of the following parts:



- Plastic, multi-use (animal use only), disposable.
- Can be used in either the SQA-Vp measurement or visualization chamber
- For capillary use/cleaning instructions, refer to the appendix section of this guide.

Slide Adaptor

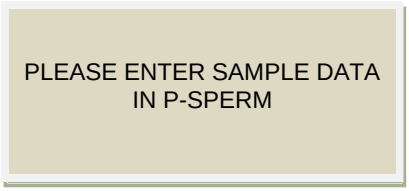


- Use a standard laboratory slide 76 x 25.6 mm and 22 x 22 mm cover-slip.
- Sample should be placed where indicated by the yellow dot.

Section 3: Start-up SQA-Vp

NOTE: Load I-button tests and set system date **PRIOR** to testing (see **SQA-V Service Menu** section for full instructions).

- Turn on the main switch on the rear panel of the SQA-Vp.
- Both the power and heater light on the front panel will illuminate.
- Press the On/Off key on the SQA-Vp keypad.
- The SQA-Vp will perform self-testing and then will display the **#** Tests Remaining
- When ready for testing samples, the SQA-Vp will display the following message:



Section 4: Start-up P-Sperm

WARNING: remember the new password or the system cannot be re-entered.

WARNING: Set-up system defaults prior to using P-Sperm

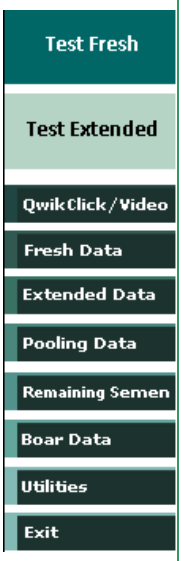
Click on the P-Sperm icon located on the PC desktop to enter the system.

- Enter the **temporary password: fertility** and then click OK.
- A screen for changing the password will appear, enter and confirm a new password.

Section 5: P-Sperm Navigation Overview

Click one of TEN P-Sperm buttons to activate the features described below:

Ten navigation buttons allow easy access to the P-Sperm features.



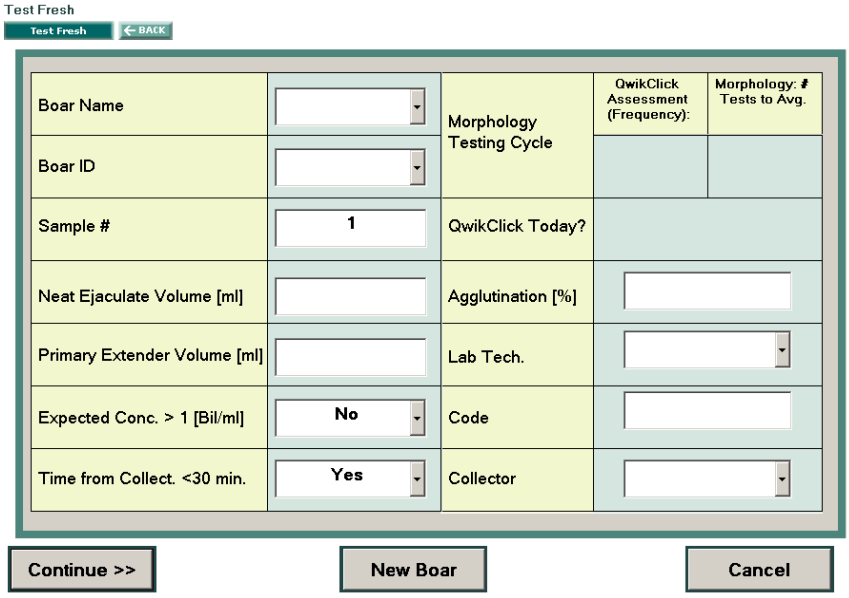
- **TEST FRESH**: Click to begin testing FRESH BOAR SEMEN.
- **TEST EXTENDED**: Click to begin testing EXTENDED BOAR SEMEN.
- **QwikClick/VIDEO**: Click to view samples on the PC and to activate the QwikClick droplet assessment or morphology counter.
- **FRESH DATA:** Click to activate a grid that displays FRESH boar semen information such as test results, dosing settings, graphs, semen images/videos.
- **EXTENDED DATA:** Click to activate a grid that displays information EXTENDED boar semen information such as test results, graphs, semen images/videos.
- **POOLING DATA:** Click to display a grid containing information on Pooled Batches of semen such as batch data, test results, dosing settings/outcomes.
- **REMAINING SEMEN:** Click to display a grid that containing information on semen that remains after pooling or dosing including boar and sample data, extender name/type, test results, etc.
- **BOAR DATA:** Click to activate a grid displaying information on individual boars such as Boar Name and ID, Breed, averages of semen test results, single sire data, split and pooled dosing for a given boar over a selected timeframe and extended semen QC results (linked to the fresh sample used for AI dosing).
- **UTILITIES:** This menu contains Set-up, Controls and SQA-V Self-Test data.
- **EXIT:** Click to close P-Sperm.

Section 6: Sample Testing

- Before testing for the first time or in the event of a change…update the default settings in P-Sperm (Please refer to the Utilities>Set-up section). Enter all boars into the boar table (Utilities>Set-Up>Data Settings>Boar Settings).
- Turn on both the SQA-Vp and P-Sperm to begin testing boar semen.

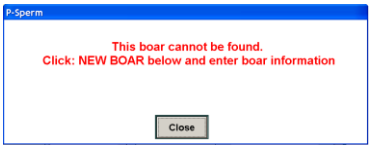
Test Fresh

Test FRESH: Click this button to begin testing FRESH samples. The screen below will be displayed:



ENTER:

- **BOAR NAME** and **BOAR ID** - a drop-down menu will display the complete list of boars that were "loaded" into the boar table. Begin typing and records will be displayed. Click on the desired record and the BOAR NAME and ID will be filled-in. Click the **NEW BOAR** button or go to Utilities>Set-Up>Data Settings>Boar Settings to enter a new boar or set-up groups of boars.
- If **Boar Name** or **ID** is not found, the following message is displayed:



- **Sample #, Neat Ejaculate Volume** and **Primary Extender Volume**.
- **Expected Conc > 1(Bil/ml):** Select YES if the boar produces highly concentrated semen. The default is set to NO.
- **Time from Collection <30 min:** If < 30 minutes, enter YES, otherwise NO.
- **Morphology Testing Cycle** and **QwikClick Today:** Information will be displayed automatically based on the default settings (Refer to Utilities>Set-up).
- **Agglutination:** Can be entered by visual sample observation using the standard slide (not obligatory).
- **Lab Tech., Code** and **Collector**: These are optional fields for each facility.

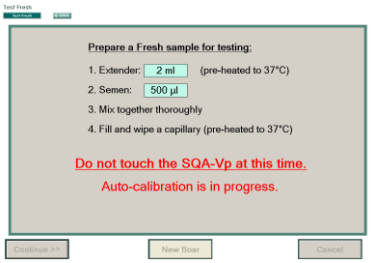
CLICK **CONTINUE>>** to begin testing (SQA-V waiting screen for sample testing should be activated) or **CANCEL** to cancel the test.

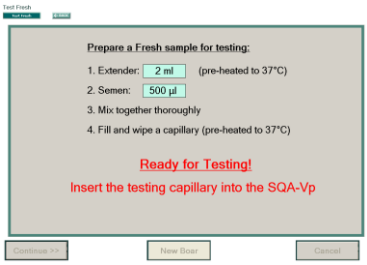
FRESH SAMPLE PREPARATION AND TESTING:

NOTE: When preparing **FRESH** samples for testing the extender volume is always 2 milliliters.

NOTE: **DO NOT pre-heat** the **FRESH** sample! Only pre-heat the extender and the testing capillaries or motility will be impacted.

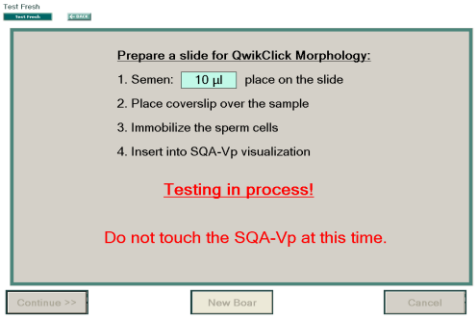
- After clicking the Continue>> button, the sample preparation screens below will be displayed:



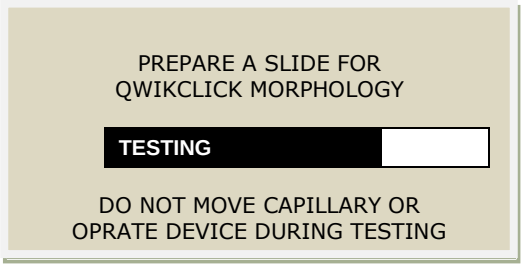


- Prepare a **FRESH** sample based on the instructions.
  - Prepare 2ml aliquots of extender pre-heated to 37^{0}C (98.6^{0}F).
  - Pre-heat the testing capillaries to 37^{0}C (98.6^{0}F).
  - **DO NOT HEAT THE SAMPLE! ONLY THE EXTENDER AND CAPILLARY** or motility will be impacted!
- Follow the screen instructions and do not touch the SQA-Vp or insert a testing capillary until instructed.
- When the SQA-Vp 'beeps', insert the prepared testing capillary into the SQA-Vp when the message: **INSERT CAPILLARY INTO CHAMBER** is displayed.
- Testing will begin automatically and run for 30 seconds.
- If the QwikClick morphology is to be performed, the messages below will be displayed. Prepare the slide while the sample is being tested.

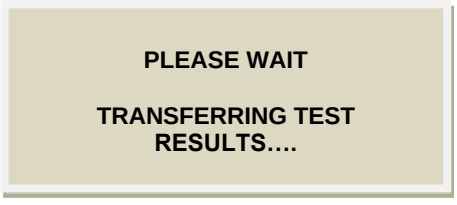
P-Sperm



SQA-Vp



- A "beep" will be heard when the testing is finished and the test results will automatically be transferred to P-Sperm.

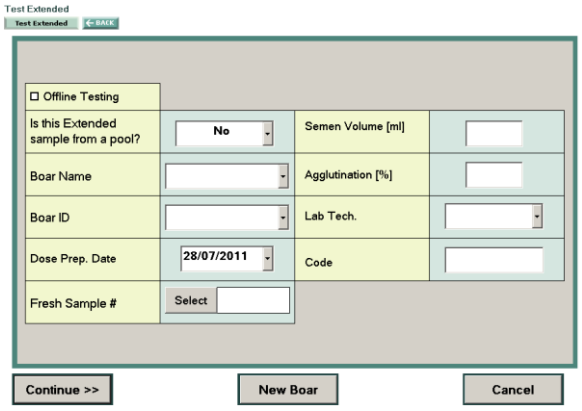


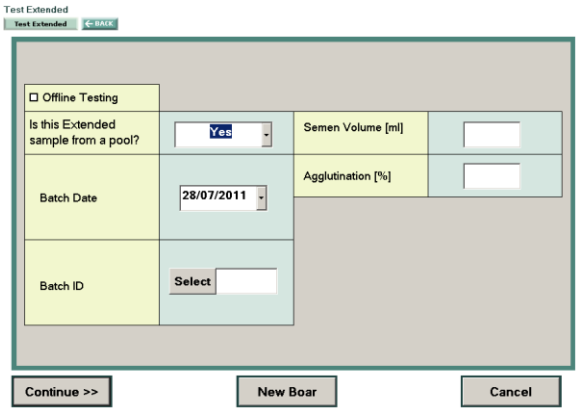
- Then in the SQA-V, Remove Capillary message will be displayed.
- In P-Sperm, the QwikClick Morphology Assessment (QCA) screen will automatically be displayed if the sample requires QCA. If not, the dosing screen will be displayed.
- Once the QCA and/or dosing has been completed, the test results, dosing information and boar/sample data will be saved in the P-Sperm database.
- The Dosing Set-up Report will be displayed automatically (if this function is set) and can be printed out by clicking the Print icon.

Test Extended

Test EXTENDED: Click this button in P-Sperm to begin testing an EXTENDED sample.

One of the two screens below will be displayed depending on whether the Extended sample is from a pool or not (question answered YES or NO – see below screens):

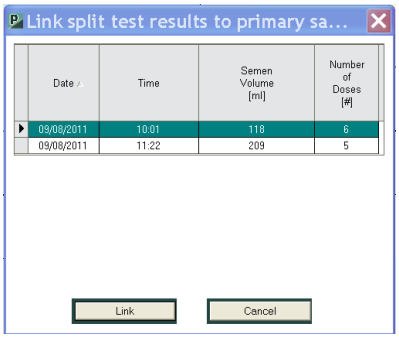




Offline Testing: If this box is checked, whatever is entered into a field will not be saved in the database (Extended records will not be linked to any other data).

<u>Enter the following if "NO" is checked (Is this Extended sample from a pool?)</u>:

- **BOAR NAME** and **BOAR ID** - a drop-down menu will display the complete list of boars that were "loaded" into the boar table. Begin typing and records will be displayed. Click on the desired record and the BOAR NAME and ID will be filled-in.
- **Dose Prep. Date:** Select the date the EXTENDED sample was prepared from the calendar drop-down menu.
- **Fresh Sample #:** Click Select and enter the number of the FRESH sample that was extended for either single-sire or split dosing. If multiple ejaculates were collected from the same boar on the same date, the table like the one displayed to the right will pop-up. Select the appropriate record to be linked to the Extended sample results.



- **Semen Volume**: Enter the volume of AI DOSE.
- **Agglutination**: A visual assessment of % spermatozoa agglutinated.
- **Lab Tech.** and **Code**: These are optional fields for each facility.

<u>Enter the following if "YES" is checked (Is this Extended sample from a pool?)</u>:

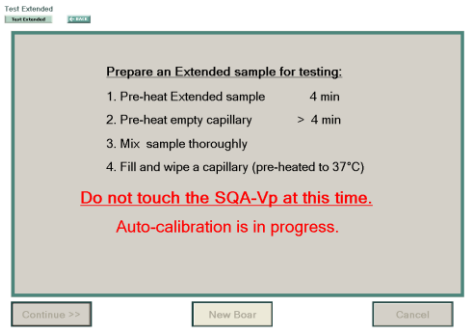
- **Batch Date**: Select the date the pooled batch was prepared from the calendar drop-down menu.
- **Batch ID**: Click Select to input the Batch ID that was extended. Only Batch ID of closed batches will be displayed. If adding AI doses to the batch is not completed, such a batch cannot be tested in Extended mode.
- **Semen Volume**: The volume of AI DOSE.
- **Agglutination**: A visual assessment of % spermatozoa agglutinated.

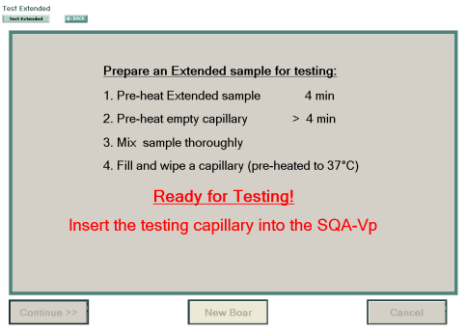
CLICK **CONTINUE>>** to begin testing or **CANCEL** to cancel the test.

NOTE: **PRE-HEAT** the **EXTENDED** semen sample for **ONLY 4 minutes** – do not heat for more or the sample may agglutinate and motility readings will be impacted!

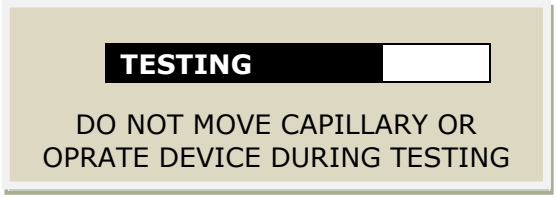
EXTENDED SAMPLE PREPARATION AND TESTING:

- After clicking the **Continue>>** button, the sample preparation screens below will be displayed in sequence:

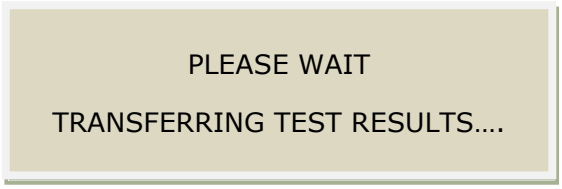


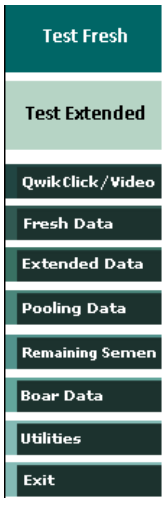


- Prepare an **EXTENDED** sample for testing based on the instructions.
  - Prepare a 2ml SAMPLE pre-heated to 37^0C (98.6^0F) for 4 minutes.
  - Pre-heat the testing capillaries to 37^0C (98.6^0F).
  - **DO NOT HEAT THE SAMPLE beyond 4 minutes** or it may agglutinate!
- Follow the screen instructions and do not touch the system until instructed.
- When the SQA-V 'beeps' and the message: **INSERT CAPILLARY INTO CHAMBER** is displayed, insert the testing capillary into the SQA-Vp.
- Testing will begin automatically and run for 30 seconds.



- A "beep" will be heard when the testing is finished and the test results will automatically be transferred to P-Sperm and saved in the Extended Data grid.





Section 7: QwikClick/Video

Click the **QwikClick/VIDEO** navigation button to activate the:

- **QwikClick** Morphology screen for off-line morphology testing.
- **Real Time Video**: View samples "live" from the SQA-Vp visualization system.
- **Video Settings**: Customize the video set-up.

Note: The QwikClick screen is opened online automatically after the results of FRESH semen sample testing are transferred to P-Sperm.

To pre-set this feature go to: **Utilities>Set-up>System Settings> Administrator > Settings** and **Utilities>Set-up>Data Settings>Boar Settings**.

QwikClick

QwikClick/Video > QwikClick

Video Settings | Grid | ← BACK

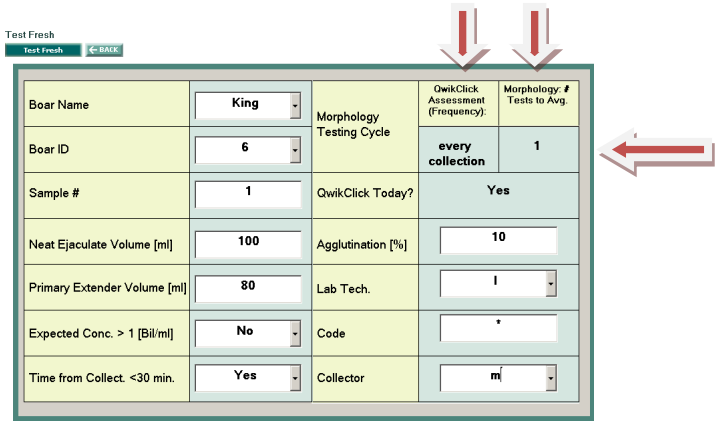
NOTE: Only FRESH samples can be assessed with the on-line QwikClick feature.

This section describes how to use the P-Sperm QwikClick Droplet and QwikClick Morphology Assessment tools.

DEFINITIONS: The QwikClick feature has three settings for assessing porcine morphology if the fully automated option is not preferable:

- **QwikClick Droplet Assessment (QCA)**
- **QwikClick Morphology Counter (on-line)**
- **QwikClick Morphology Counter (off-line)**

- **QwikClick Droplet Assessment (QCA):** Enhance the accuracy of the SQA-Vp automated morphology by assessing % droplets and broken tails (comparable to CASA systems). The easy manual count of % droplets and broken tails are added to the automated results generating a very high level of accuracy. Set up this feature by going to: **Utilities > Set-up > Data Settings > Boar Settings**. Pre-set the frequency for testing boar QwikClick Morphology based on the desired collection cycle (every, every 2nd, 3rd… or 6th collection). This results in good boar morphology management/reporting and can increase testing throughput.
- **The AMI (Average Morphology Index) should be used with the QCA: AMI** is an average of several morphology results from the same boar. The number of morphology test results to average is determined by the operator (minimum of 1 and maximum of 6). Both the frequency and the number of tests to average for computing AMI is indicated in the Test Results table of the QwikClick screen.



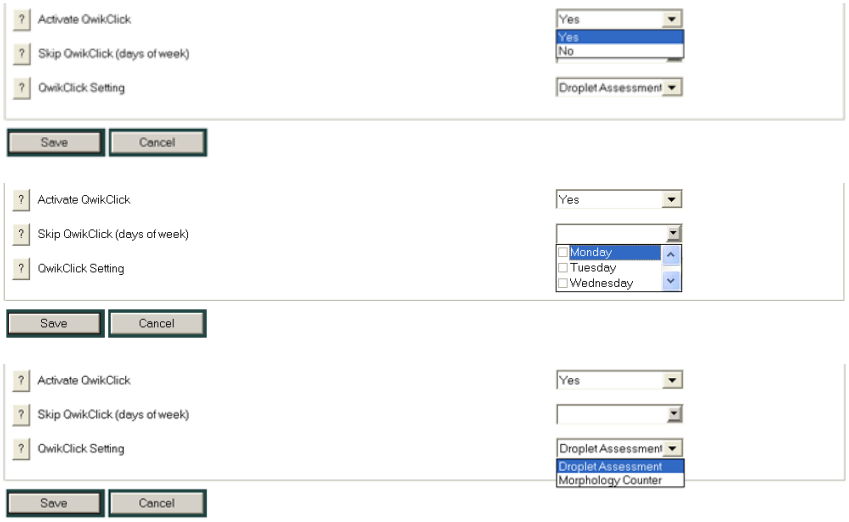
When using the AMI for AI dosing, please note the PASS/FAIL criteria is based on a Morphologically Normal Sperm target or cut-off set by the user. **To ensure quality, boars that have an AMI below the morphology cut-off will automatically be selected for QwikClick morphology testing, regardless of the QCA cycle.**

- **QwikClick Morphology Counter (on-line):** This morphology option should be selected if the user wants to manually assess all morphology defects. The % Normal Morphology resulting from the manual count will be used by the system for AI dosing and the sample will PASS/FAIL based on a pre-set cut-off/target.
- **QwikClick Morphology Counter (off-line):** This feature is for manually assessing morphology defects 'off-line' (without impacting dosing).

How to SET-UP the QwikClick Droplet Assessment (QCA):

- **SYSTEM SETTINGS** (set by administrator): Go to **Utilities > Set-up > System Settings > Administrator > Settings** and select:
  - Activate QwikClick Yes/No
  - Skip QwikClick (days of week)
  - QwikClick Setting: Droplet Assessment / Morphology Counter

QwikClick



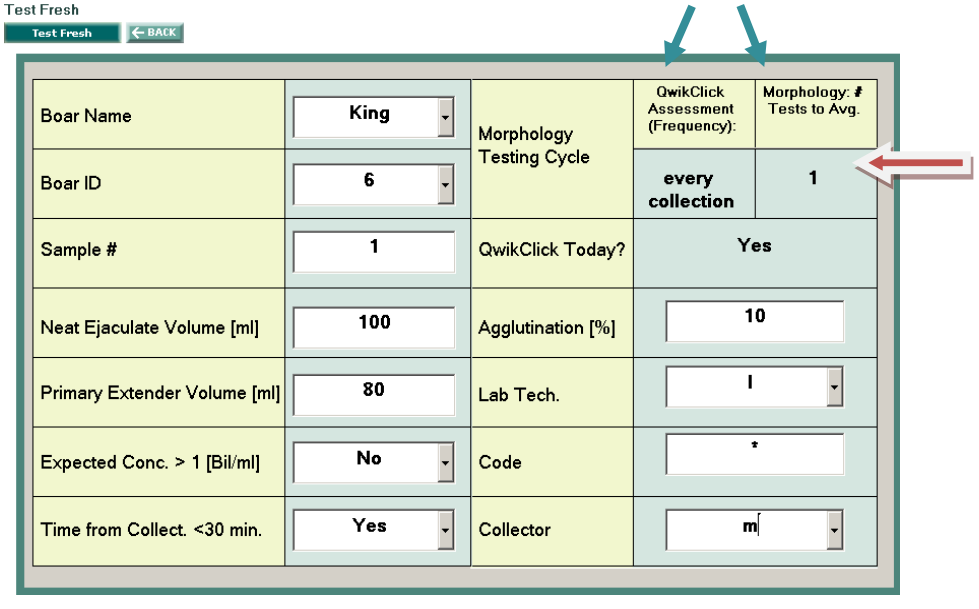
- **BOAR SETTINGS TABLE**: Go to **Utilities>SET-UP>DATA SETTINGS>BOAR SETTINGS** and enter the **QwikClick Assessment (Frequency)** and **Morphology: # Tests to Avg.** Place the cursor over the title cells for an explanation of these settings.

Utilities > Set-up > Data Settings > Boar Settings

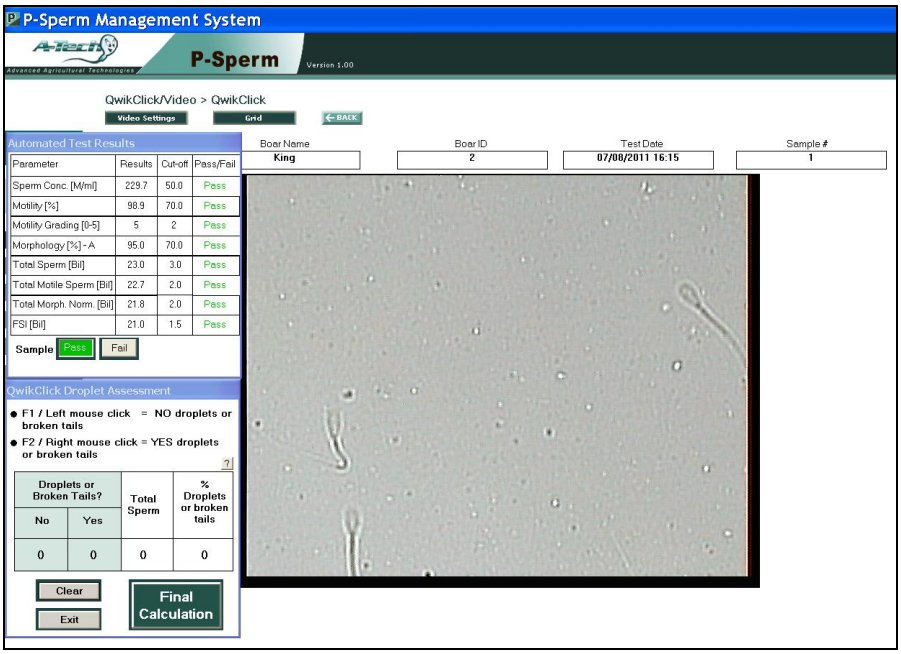
Report Export BACK

Boar Name	Boar ID	Breed	Owner	Location	QwikClick Assessment (Frequency):	Morphology: # Tests to Avg.
					every collection	1
King	2	American Yorkshire	MES	MES	every collection	1
Star	1	American Yorkshire	MES	MES	every collection	1

When a boar is scheduled for QwikClick morphology testing, the **TEST FRESH** data entry screen will automatically provide a YES or NO to the question "**QwikClick Today**?" The QwikClick Assessment (Frequency) and Morphology: # Tests to Avg. will also be shown:



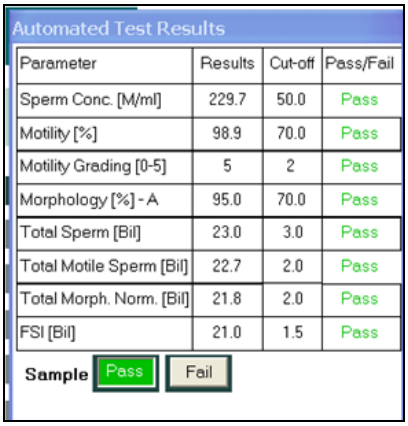
- **If the QwikClick feature has been set-up as a system default** and the boar is scheduled for morphology testing, one of the QwikClick Morphology Counters (depending on default setting) will be displayed when testing is finished. Make a slide and place it into the visualization compartment of the SQA-Vp.
- **If the QwikClick feature is not the system default or the boar is not scheduled for morphology testing,** the QwikClick screen will be skipped and the Dosing Set-up screen will be displayed automatically when a test is completed.
- The QwikClick screen below will automatically be displayed upon completing the SQA-Vp test if the boar is scheduled for QCA or the AMI (average morphology index) is below the cut-off value:



Two floating tables are displayed on the QwikClick screen:

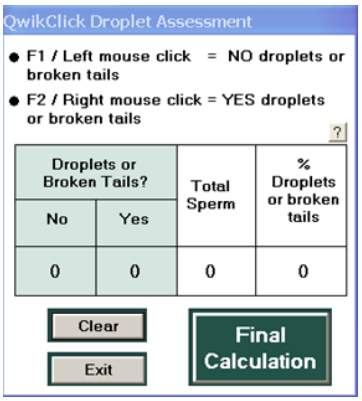
- **Automated Test Results**
- **QwikClick Droplet Assessment** OR **QwikClick Morphology Counter**

The **Automated Test Results** table seen below displays test **Results**, **Cut-off** defaults and **Pass/Fail** results. The sample will PASS if all of the semen parameters have passed the cut-offs. The sample will FAIL if one semen parameter fails.



- Cut-off criteria can be set-up by the ADMINISTRATOR (see the SET-UP section of this User Guide). If not set-up, the manufacturer default cut-offs will be used.
- The P-Sperm can be set-up to automatically or manually PASS or FAIL a sample (see the SET-UP section of this User Guide).

QwikClick Droplet Assessment: Enhance the accuracy of the automated morphology reported by the SQA-Vp by entering % droplets or broken tails to the algorithm.

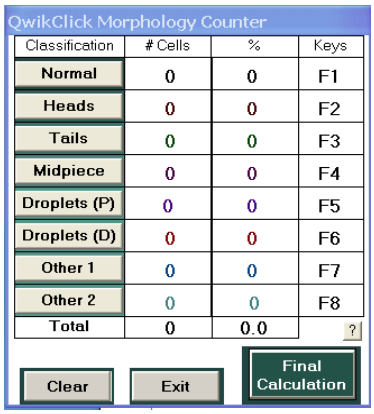


Click ? icon for explanation on how to use QwikClick Droplet Assessment:

- Prepare a slide of non-motile cells (see Appendix section for a protocol) and place it into the SQA-Vp visualization compartment.
- Focus and begin "counting" cells:
  - Use the F1/Left mouse key of a PC to count all sperm cells without droplets (The counting results are displayed online in a table).
- Use the F2/Right mouse key of a PC to count all sperm cells with DROPLETS or BROKEN TAILS (The counting results are displayed online in a table).
- Move to different fields of view by turning the silver knob of the slide holder. When enough cells have been counted, Click: FINAL CALCULATION.
- This completes the count, sends the information to the morphology algorithm and populates the test data and dosing screen with the results.
- Click **CLEAR** to remove the entire count and to start over.

QwikClick Morphology Counter (off-line): Manually assess all morphology defects without impacting dosing or attaching the results to a boar report:

Off-line QwikClick Morphology Counter for full manual morphology assessment.



To activate the **QwikClick Morphology Counter (OFF-LINE):** Click **QwikClick/Video > QwikClick** buttons in P-Sperm. The counter to the right will be displayed.

To use the **QwikClick Morphology Counter** (click ? icon to display instructions).

Note: The function keys that correspond to morphological defects are set by the manufacturer, but can be re-set by administrator in the P-Sperm set-up.

- Prepare a slide of non-motile sperm cells and place it into the SQA-Vp visualization compartment.
- Use assigned function keys (F1, F2, etc.) of a PC keyboard for counting normal cells and morphological abnormalities (one abnormality per spermatozoa).
- Move to another field of view by slightly turning the silver knob of the SQA-Vp slide holder.
- Click **FINAL CALCULATION** to save the results.

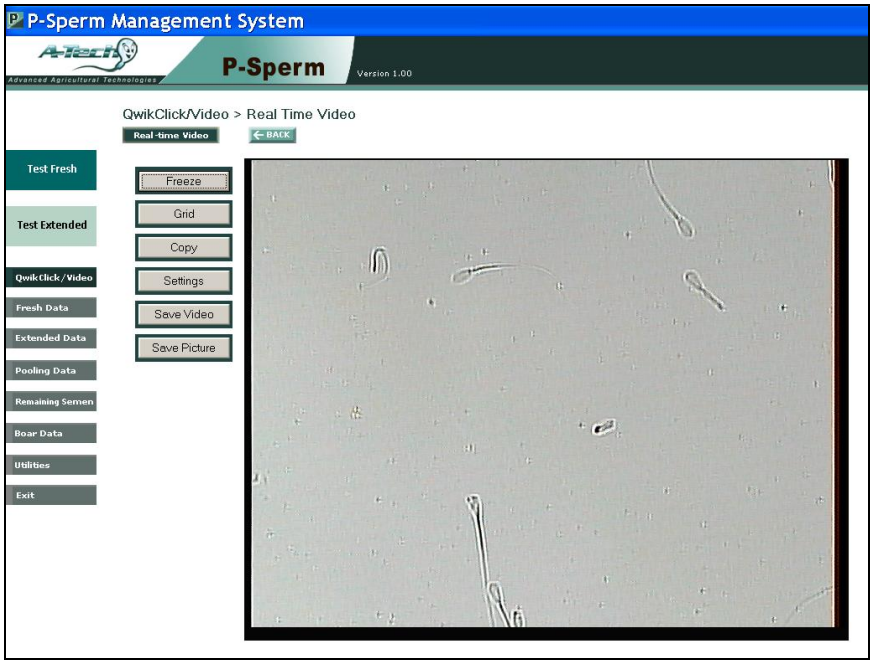
On-line QwikClick Morphology Counter for full manual morphology assessment.

When the **FINAL CALCULATION** button is clicked and a title assigned to a simple test report, the results can be viewed in a report table and/or printed and exported to an Excel file (The results will not be attached to any BOAR record).

QwikClick Morphology Counter On-line: (SEE SET-UP for information how to set this feature): Manually assess all morphology defects and use the % Normal Morphology for AI dosing. This table is used exactly as the OFF-LINE morphology counter above. Differential Morphology results will be seen in the Morphology Report.

Real Time Video

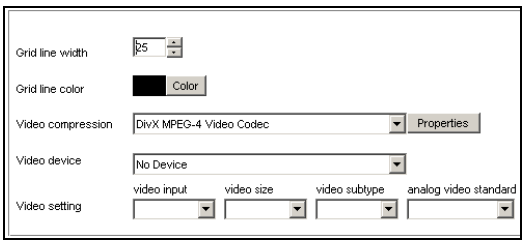
Click **REAL TIME VIDEO** and the following screen will be displayed:



- **FREEZE –** To freeze the screen. Click the REAL TIME button (displayed after image freezing) to un-freeze.
- **GRID –** A grid can be added to the screen to make counting easier.
- **COPY –** Pictures can be copied by clicking the COPY button and then pasting the picture in an external file (WORD, Excel, etc.).
- **SETTINGS –** Video settings can be selected by the user.
- **SAVE VIDEO/PICTURE –** Save video clips (.avi format) and pictures (.bmp format) in a PC file.
- **FULL SCREEN** – Double click on the image to maximize it. Use F1 to freeze/unfreeze and Esc to minimize an image.

Video Settings

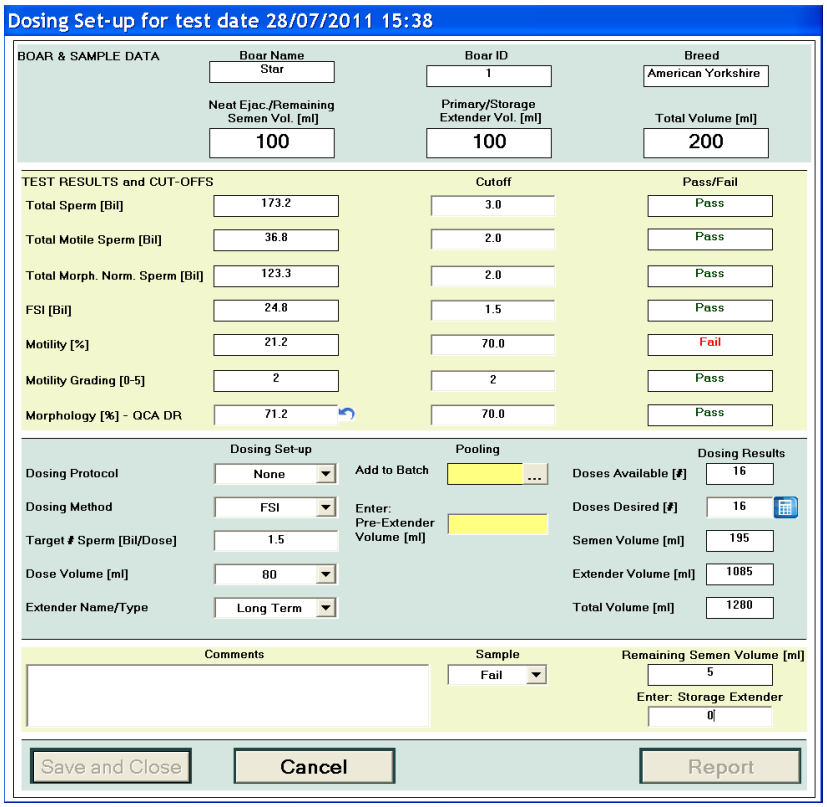
Click the **VIDEO SETTINGS** button to select grid line width, color and video settings. The appropriate video device settings are described in the video device installation procedure supplied with the video device hardware:



AI Dosing

Section 8: AI Dosing

When the FRESH sample testing is completed the Dosing Set-up screen below opens automatically:



NOTE: If manual morphology is to be considered for dosing, enter this into the morphology field by overwriting the displayed value. This will then be considered the morphology default for dosing.

The Dosing Set-up screen has four sections: **(1) Boar & Sample Data, (2) Test Results and Cut-offs; (3) Dosing and Pooling Set-up/Results and (4) Comments/Disposition of Sample.**

Boar & Sample Data: Basic information about the boar and the sample. All of the information in these fields is automatically imported, no data entry is required.

Test Results and Cut-offs: This section displays all the sample testing results, cut-offs (determined by the facility) and an indicator if the sample PASSED/FAILED based on the cut-offs. The sample is considered FAILED if one of the test results falls below the cut-off. The default cut-offs for "Total" parameters is based on the minimum targets for one AI dose.

- FSI – Functional Sperm Index in billions: This parameter is based on Concentration x Motility x Morphology x Correction Factor x (Neat Ejaculate or Remaining Semen Volume)
- The type of morphology assessment is indicated next to the Morphology (%) as follows:

A = Automated; QCA DR = QwkClick Droplet Assessment; QCA MC = QwikClick Morphology Counter; AMI = Average Morphology Index; O = Optional Manual Morphology Entry.

Dosing and Pooling Set-up/Results: Fresh samples can be dosed in a variety of ways using the SQA-Vp technology: **Single-sire, split** or **pooled.** AI dosing can also be performed offline from the **Fresh Data grid** (if the semen sample has not yet been dosed) and from the **Remaining Semen** grids by clicking the Dosing Set-up button.

Single–sire AI Dosing

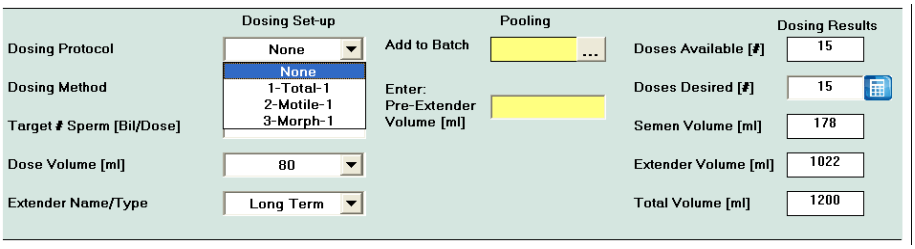
SINGLE-SIRE AI DOSING:

In order to perform Single-sire Dosing, the Dosing Set-up parameters should be defined. The software will store in memory the latest settings. Two options are to dose by **Dosing Protocol** or **Dosing Method**

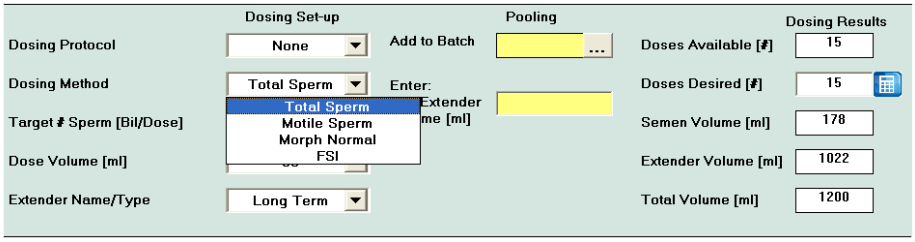
NOTE: The Dosing Set-up parameters can be pre-set by the administrator using protocols (See: Utilities > Set-up > System Settings > Administrator). If protocols are used, the operator only needs to select the DOSING PROTOCOL and the other DOSING SET-UP fields will be displayed automatically and locked.

- **Dosing Protocol**: Set-up protocols (see the Utilities section of this guide) prior to dosing:

- Once a protocol is set-up it can be selected from the drop-down menu in the **Dosing Protocol** field. The **Dosing Method**, **Target # Sperm (Bil/Dose)**, **Dose Volume (ml)** and **Extender Name/Type** fields will then be locked:

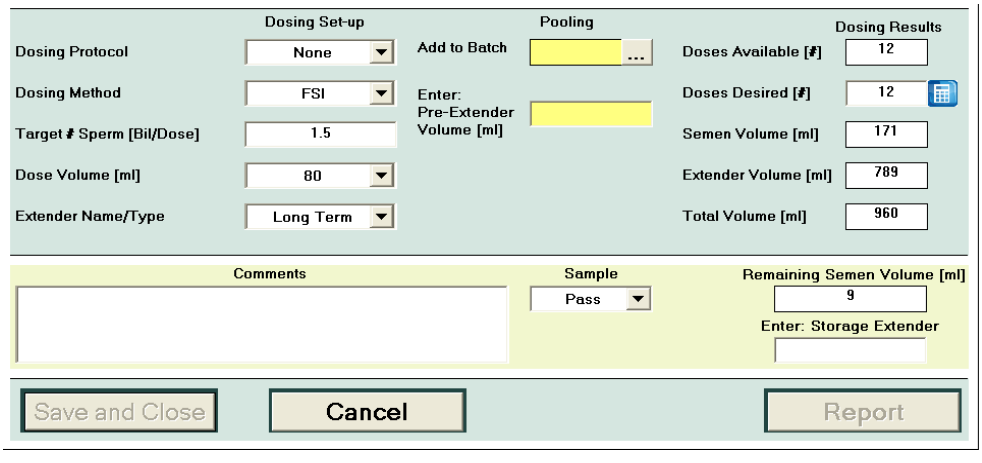


- **Dosing Method**: If 'None' is selected for Dosing Protocol, select from the Dosing Method drop down menu one of four dosing options: **Total Sperm**, **Motile Sperm**, **Morphologically Normal Sperm** or by **Functional Sperm Index (FSI)** and follow the instructions below:



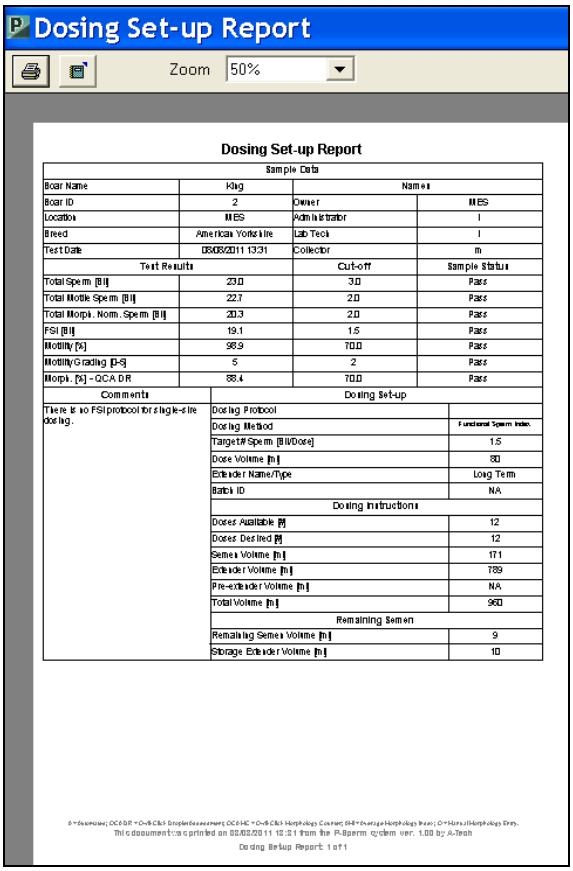
- **After selecting the Dosing Method**, enter the following information:
  - **Target # Sperm (Bil./Dose):** Enter the desired # of total, motile, morphologically normal or functional (FSI) sperm cells/dose.
  - **Dose Volume (ml):** Enter a number or select 80 or 100 ml in the menu box.
  - **Extender Name/Type:** Select from the drop down menu.

- Click  button and the dosing results will be displayed:



- **Doses Available (#):** Total # of doses possible based on the selected dosing settings.
- **Doses Desired (#):** In single-sire dosing, the number of doses desired is equal to the number of doses available.
- **Semen Volume (ml):** The volume of semen to be used for dosing.
- **Extender Volume (ml):** The extender volume to be added to the sample.
- **Total Volume (ml):** The sum of the semen plus the extender volume.
- **Remaining Semen Volume:** The semen leftover (not used) in the dosing process. It can be seen in the Remaining Semen grid (Storage Extender should be added).
- **Sample Pass/Fail:** A PASS will be displayed if all results passed the cut-off criteria. If not, FAIL will be displayed.
- **Comments:** Type comments which can be viewed in the **Dosing Set-up Report** or by clicking on the "**C**" icon in the **Primary Sample Data** grid.

- Click the **REPORT** button to print the **DOSING SET-UP REPORT** (displayed automatically after saving dosing results if this is set by administrator):

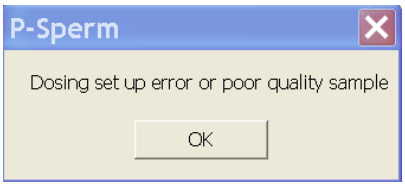


- Click  button to print the report or the  button to export report to a PC WORD file.
- Click **SAVE AND CLOSE** button in the Dosing Set-up screen to save the dosing results or **CANCEL** if the dosing results should not be saved.

Notes:

- **Save and Close** and **Report** buttons are disabled if **SAMPLE FAIL** is displayed or **Storage Extender** volume is not entered. Enter the amount of Storage Extender added to preserve the remaining semen. This is important for split and pooled dosing. A Storage Extender default can be set-up by the administrator (constant volume or a ratio of extender to semen volume).

- If the sample is not large enough or is not high enough quality to make the doses required, the following error message will be displayed:



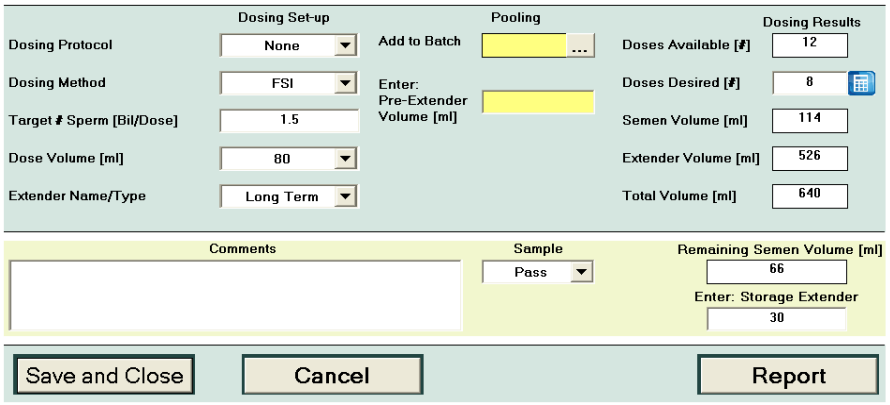
Split AI Dosing

SPLIT DOSING

A single boar ejaculate can be divided into splits and dosed separately. The dosing settings and flow are similar to the single-sire dosing with the following difference:

- Overwrite **Doses Desired (#)** with the number of doses required for a particular split per customer request (This number cannot be more than Doses Available).

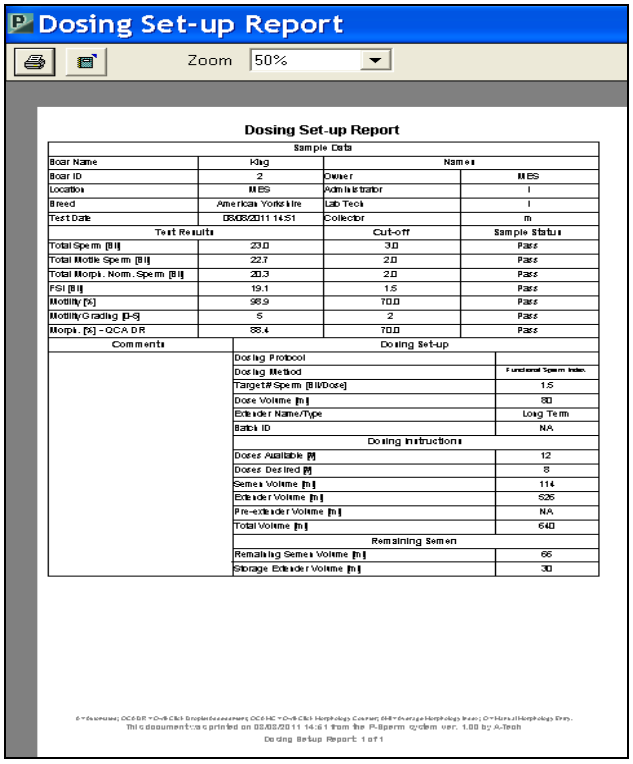
Click  button and the dosing results will be displayed. Enter Storage Extender volume and save results:



- Extend any leftover semen with a Storage Extender to preserve it for the next Split Dosing round. The record is saved in the Remaining Semen grid.
- A report will be displayed automatically if pre-set by the administrator. If not, click the **Report** button before saving to view and print the **Dosing Set-up Report**:

Click on this icon to print a copy of the report.

Click on this icon to export the semen analysis report.



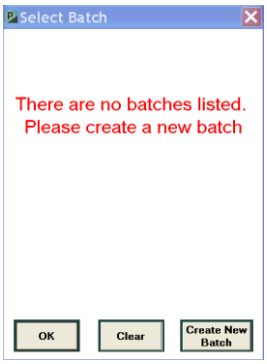
Pooled AI Dosing

POOLED DOSING

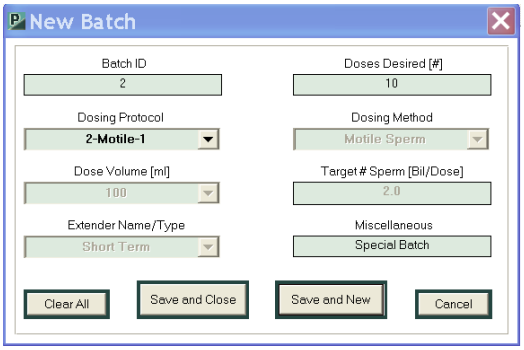
Several boar ejaculates and remaining semen may be pooled if this is part of the acceptable facility protocol. The SQA-Vp will track this process. Pooled Batches should be set-up in advance with dosing targets (see Pooling Data section). The pooled dosing procedure is as follows:

Note: The Create New Batch function can be initiated from the Select Batch or Pooling Data screen.

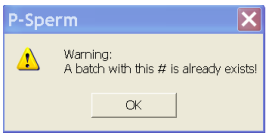
- When the Dosing Set-up screen is opened, click the **Add to Batch** button.
- If **Pooled Batches** are not pre-set, the following message will be displayed:



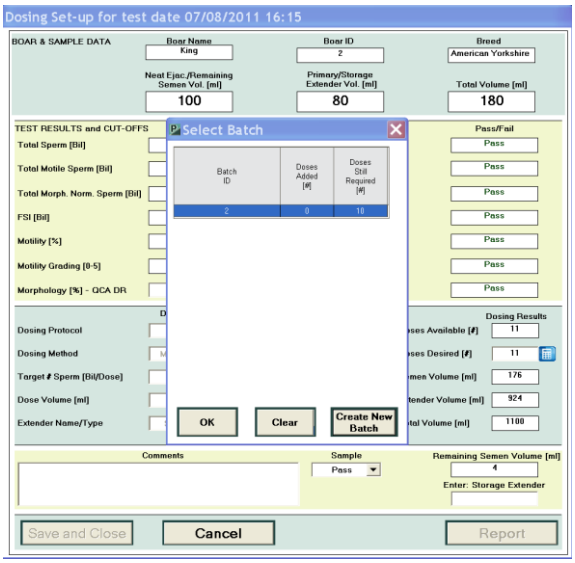
- Click the **Create New Batch** button, and the screen below will pop-up.
- Enter the **Batch ID**, **Dosing Protocol** and **Doses Desired (#)**.



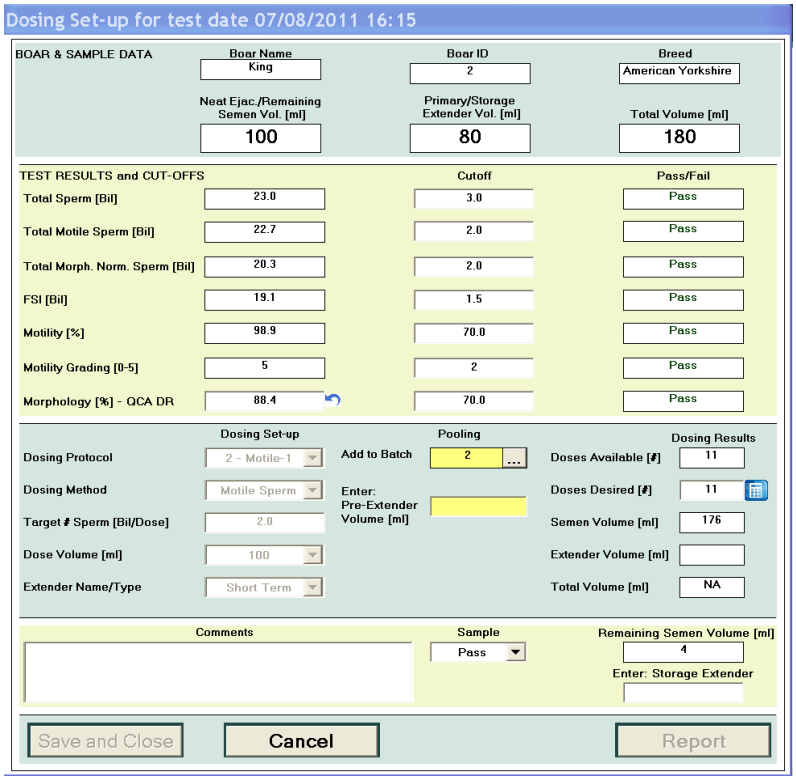
- Data will be displayed automatically based on the Dosing Protocol selected. No changes are permitted. If **Dosing Protocol: None** is selected, manually set all of the parameters.
- The Miscellaneous field is provided for notes or special facility input.
- If the Batch ID exists in the database, the following message is displayed:



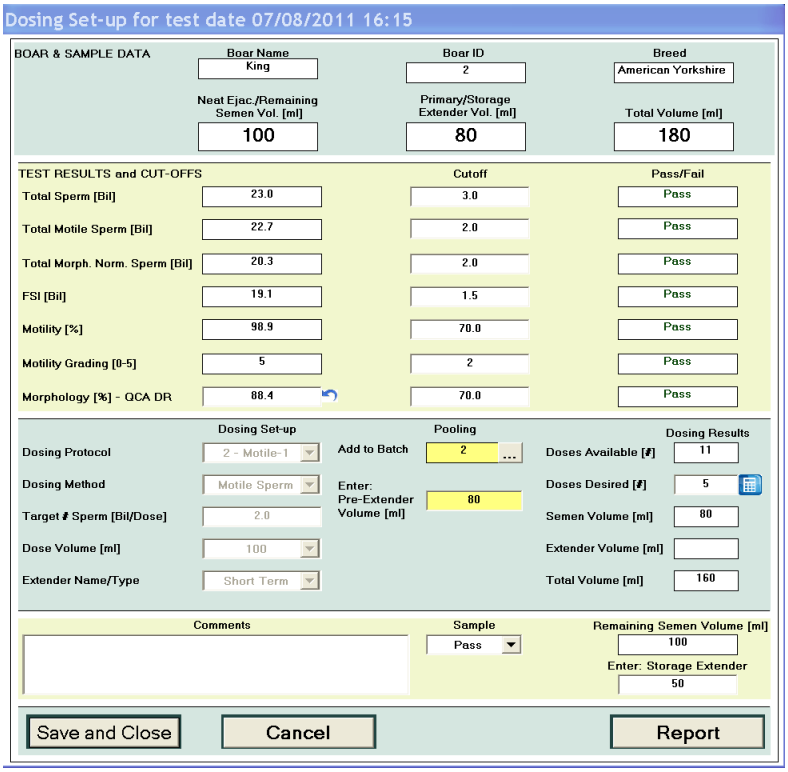
- Press **Save and Close** to save the settings. The batch will appear in the **Select Batch** table:



- The following data is displayed in the **Select Batch** table: Batch ID, Doses Added and Doses Still Required. This information can be compared in order to decide if a given semen sample/ejaculate is suitable for a particular pooled batch.
- Highlight the batch for pooling if several batches are set and press **OK**.
- Press **Clear** to disable selected batch from pooling.
- When a batch is selected for pooled dosing, the Dosing Set-up parameters related to this batch are displayed automatically and are protected from any change. In order not to mix different extenders, the Extender Name/Type should be the same as the Primary Extender used for extending the Fresh semen during collection.
- If the Dosing Set-up screen is opened off-line from the Remaining Semen grid, the system prevents selecting batches with extender types that vary from the extender used in the Remaining Semen.



- In the **Dosing Results** section, Doses Desired (#) can be overwritten based on the Doses Still Required (#) reported in Select Batch table. Enter any number that does not exceed Doses Available (#), otherwise a Dosing Set-up Error message will appear.
- Pooling occurs in steps. Therefore, the initial semen is not fully extended unless Doses Still Required (#) are completely produced and the batch is closed.
- So Extender Volume is not reported at this point. Instead, enter the Pre-Extender Volume added to the semen to preserve it until the final extending process. Usually 1:1 or 1:2 dilution should work.
- When the Pre-Extender Volume is entered, the Total Volume is displayed automatically.
- Storage Extender for Remaining Semen should be entered by the user as well.
- Press **Save and Close** and the dosing results will be saved in the Fresh Data and Pooling Data grids:

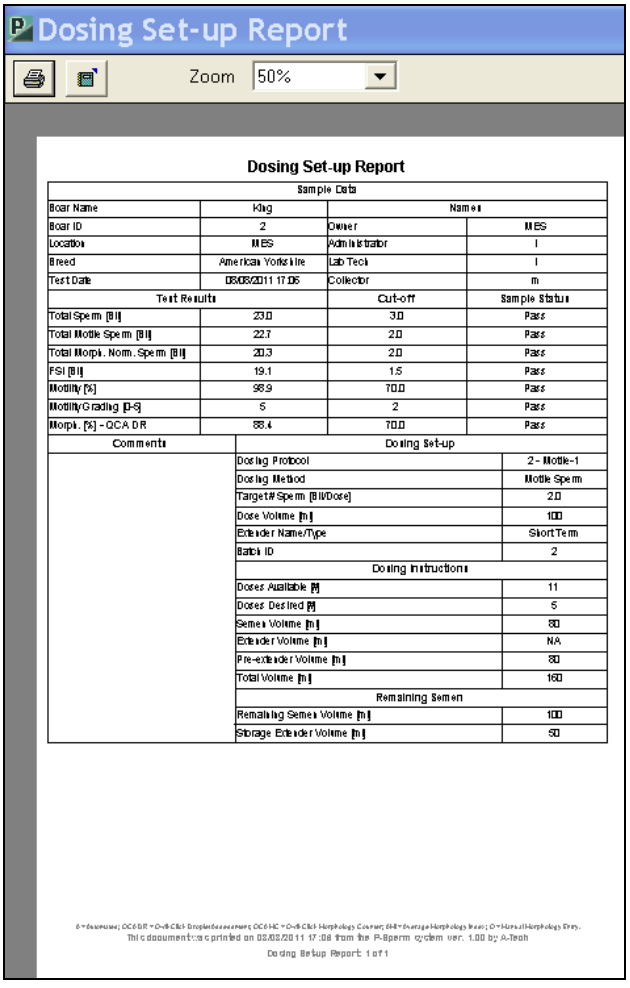


- The report below is displayed automatically if this function is pre-set by the administrator. If not, click the **Report** button before saving to view and print the **Dosing Set-up Report**:

Pooled Dosing Set-up Report

Click on this icon to print a copy of the report.

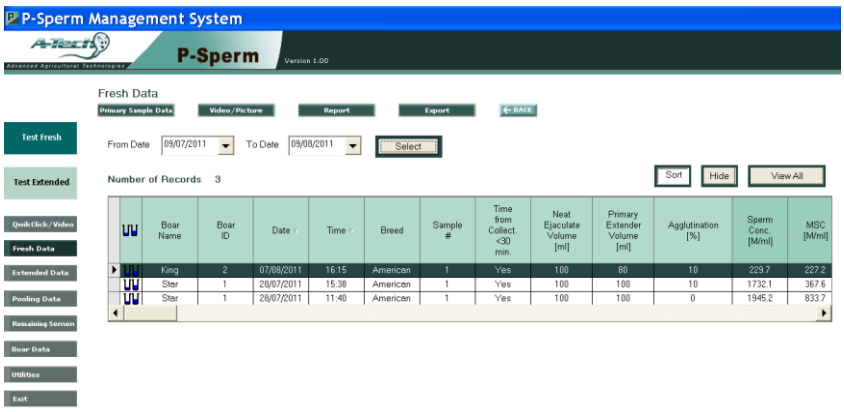
Click on this icon to export the semen analysis report.



Fresh Data Grid

Section 9: Fresh Data Grid

Click **Fresh Data** in the P-Sperm Main Menu to open the grid below:



Click in the grey box of the first column to select one record or drag the arrow to select multiple records. The second column indicates that the sample was dosed. Double click on the icon for details.

 - Single-sire dosing

 - Split dosing

 - Pooled dosing

 - Remaining Semen

 - Wasted Semen

The following information is displayed in the **Fresh Data** table:

Sample/Boar Information:

- **Boar Name** – Entered through the **Boar Settings** screen (Utilities > Set-up > Data Settings > Boar Settings).
- **Boar ID** – A unique boar identification number designated by the user in the Boar Settings table.
- **Date** – The date the sample was tested.
- **Time** – The time the sample was tested.
- **Breed** – The boar breed entered in the Boar Settings table.
- **Sample #** – The sample number designated by the user.
- **Time from Collection < 30 min**. – Indicates Yes or No.
- **Neat Ejaculate Volume** – The ejaculate volume (ml).
- **Primary Extender Volume** – The volume of extender added prior to or during collection of the ejaculate (ml).
- **Agglutination %** – Percentage of sperm cell agglutination noted visually.

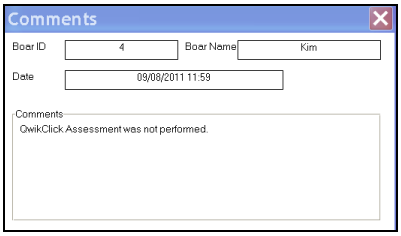
Test Results:

- **Semen parameters**: Sperm Concentration, MSC, Motility, Motility Grading, Morphology, Total Sperm, Total Motile Sperm, Total Morphologically Normal Sperm, FSI – Functional Sperm Index.
- **Sample Information**: Sample Pass/Fail.

Additional Information:

- Lab Tech (name)
- A variety of the following icons indicating different features that are displayed in the other grids as well:

Comments: Click this icon to read the comments in the box below.



Graphs: This icon will appear when there are enough test results to graph. Click on the icon and select the parameter desired to produce a graph.

<> Test results out of clinical range: This icon marks test results are **out of the normal range** that has been determined by the user (please refer to the Utilities > Set-up > Data Settings > Normal Ranges section for details).

! Dosing mismatch: This icon appears when the dosing requirements cannot be implemented. This can happen when the set-up values have been entered incorrectly or the semen sample is low quality.

Picture indicator: This icon appears when a picture is attached to a record. Click to view. Only one picture can be attached to each test.

Video Indicator: This icon indicates a video clip has been attached to the record. Click to view. Only one clip can be attached to each test.

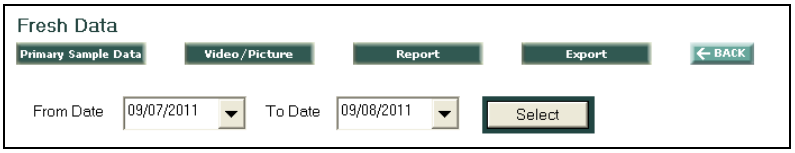
Two buttons appear at the bottom of the **Fresh Data** screen:

- **Dosing Set-up:** Click to open the Dosing Set-up screen. It works ONLY when a sample was not dosed.
- **Delete:** Select a record and click to delete it. This button is enabled only if the sample was not dosed.

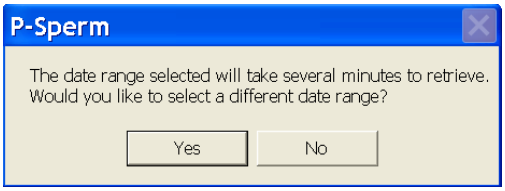
Three buttons are located in the upper right hand corner of the **Fresh Data** screen.

- Click: **SORT** and then click on the column header that contains the information to sort (Date, Boar ID, Sperm Conc., MSC, etc.).
- Click: **HIDE** and then select the columns to hide. The columns will disappear.
- Click**: VIEW ALL** to re-activate ALL of the hidden columns.

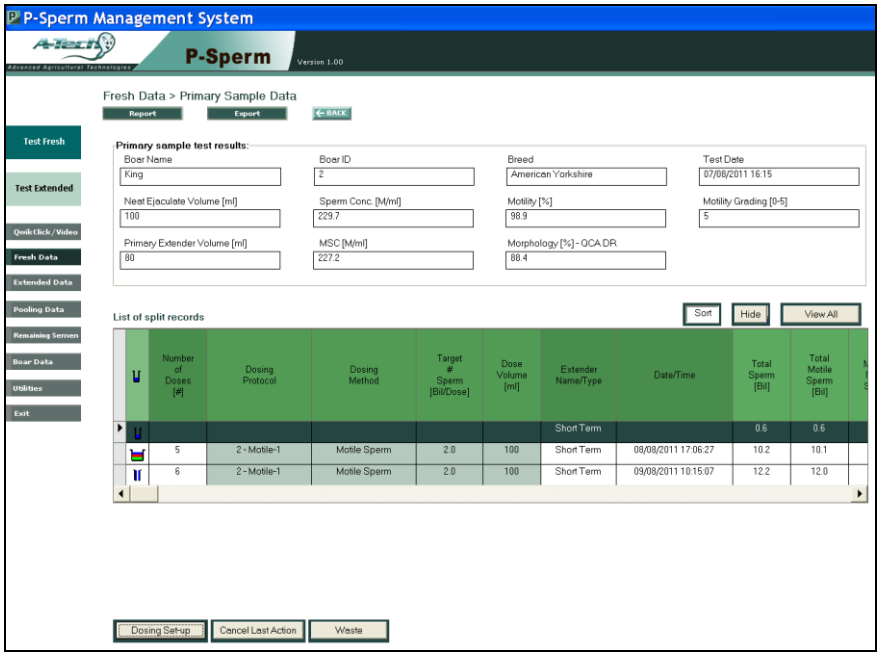
Four buttons and a date range setting are displayed at the top of the **Fresh Data** screen:



The date range is set to display data only in the selected range. The default is one month, however this can be re-set by the user. If the select a dates are too wide a range, the following message is shown:



Click **Primary Sample Data** or double click on the icon of a selected record to display how the Primary Sample was used:



Primary Sample Test Results are displayed in the upper part of the screen. A variety of icons shown in the grid indicate the type of dosing and remaining semen:

 - Single-sire dosing

 - Split dosing

 - Pooled dosing

 - Remaining Semen

 - Wasted Semen

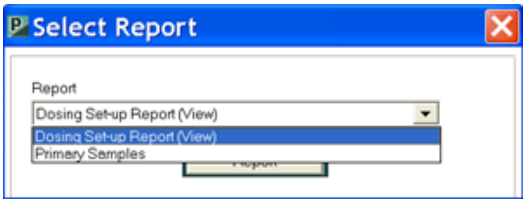
Dosing results, semen parameters with cut-offs and PASS/FAIL results are displayed.

Three buttons are located **at the bottom** of the screen:

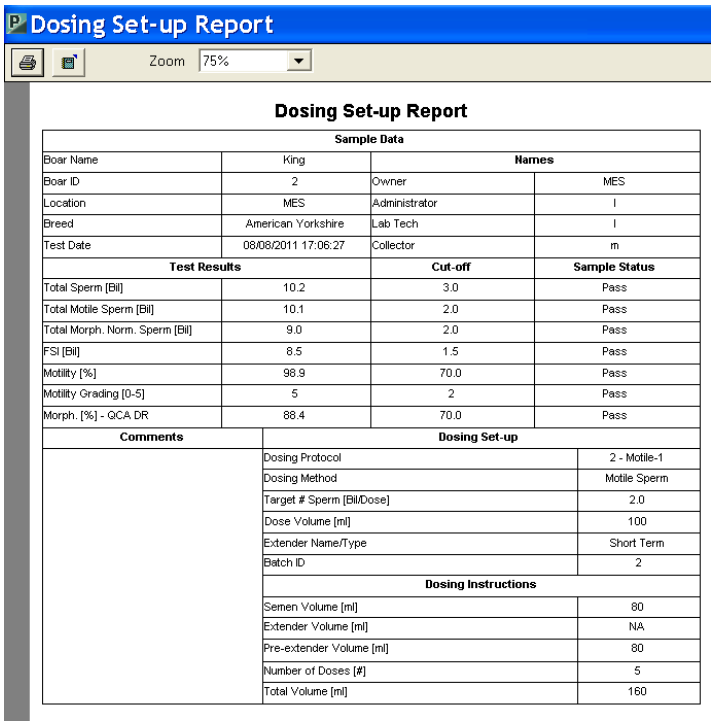
- **Dosing Set-up** – opens a table for dosing Remaining Semen.
- **Cancel Last Action** – cancels the last dosing results.
- **Waste** – to delete any Remaining Semen from the Remaining Semen grid.

Two buttons are located **at the top** of the **Primary Sample Data** screen:

Click **Report** button and **Select Report** box will be opened:



Select **Dosing Set-up Report** to view the report shown below:

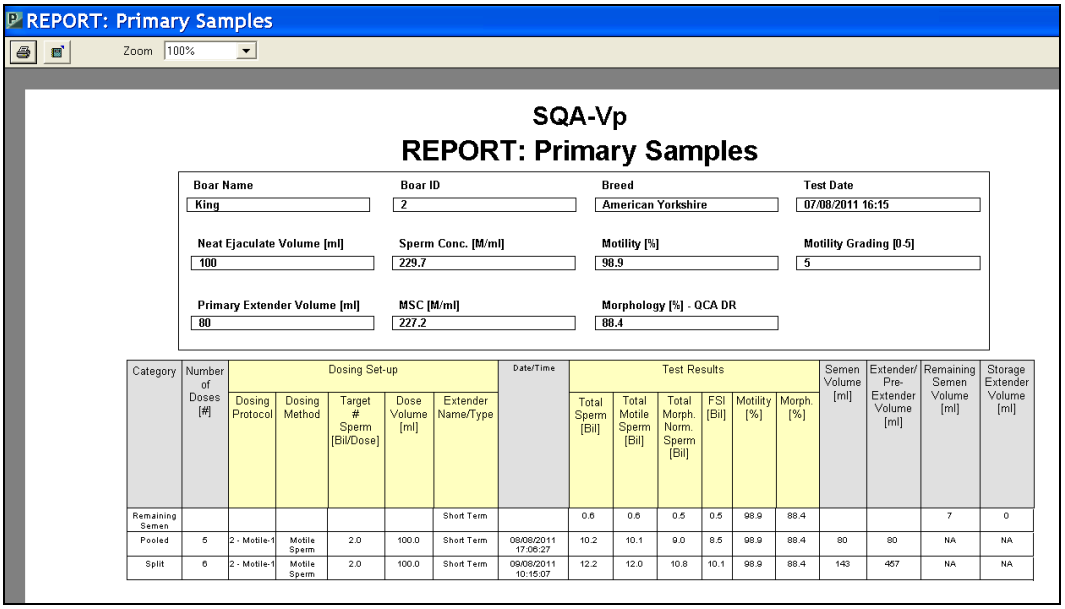


Click on this icon to print a copy of the report.

Click on this icon to export the semen analysis report.

- Click the **Printer** icon to print the report.
- Click the **Export** button to save the data in Excel format (Excel software required).
- Use the **ZOOM** option to minimize/maximize the report view.
- **Exit** the report by clicking the "X" in the upper right hand corner of the screen.

Select **Primary Samples** to view the report shown below:



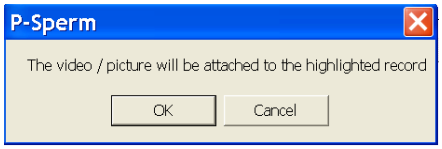
This Report represents the corresponding grid in a printable format.

- Click: the **Export** button in the **Primary Sample Data** table to export data to Excel.

Capture Image: To save a video clip or picture and attach it to a record:

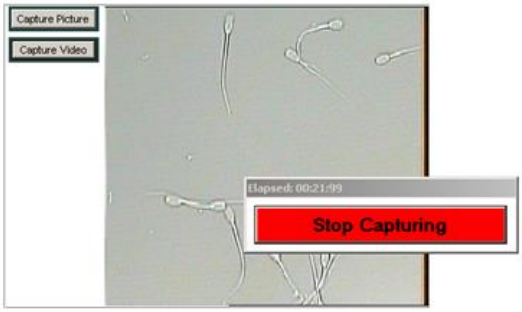
- Open: **Fresh Data**
- Insert a slide with a semen sample into the visualization system of the SQA-Vp.
- Highlight a record in P-Sperm to attach the video/picture.

- Click the **second button** of the **Fresh Data** screen – **Video/Picture.** The following message followed by the video screen will be opened:

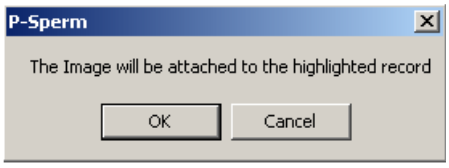


NOTE: The picture will be attached to the latest test, if no record is selected.

- Click: **Capture Picture** or **Capture Video** to save and attach a picture or video clip to the selected record. Click: **Stop Capturing** to end the video capture process.

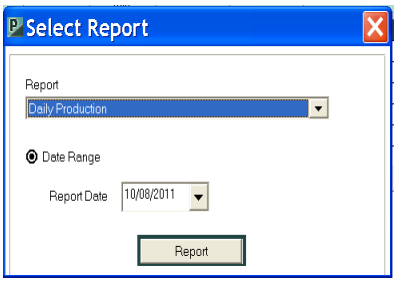


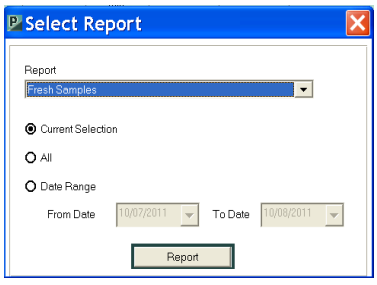
- The message below will be displayed to indicate that the image was saved and an icon will now indicate that a picture or video is attached to the record.

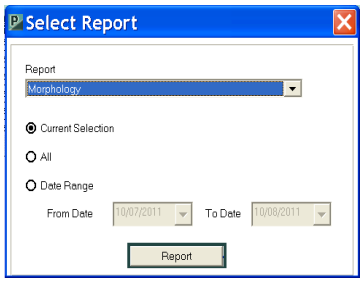


- To **delete** a picture or video simply click on the icon in the **Fresh Data** screen and click on the "Delete" (located in the lower corner of the viewing screen).

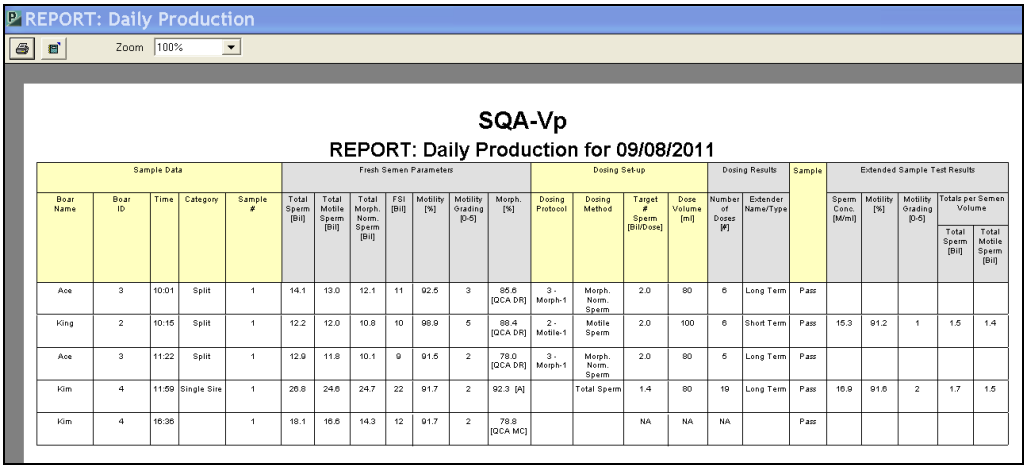
Click the **third button** of the **Fresh Data** screen – **Report.** The following box will display three report options. Select a date range or accept the current system default previously set up, or select ALL:







Select **Daily Production** to view and print the corresponding report combining Boar/Sample Data, Fresh Semen Parameters, Dosing Set-up and results and Extended Sample Test Results:

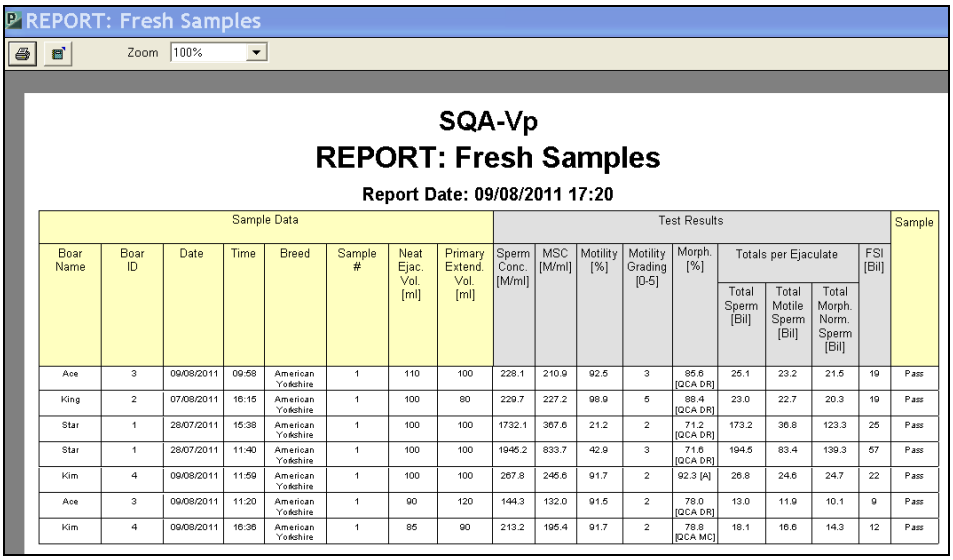


- Use the page bar at the bottom of the report to move between pages.
- Click the **Printer** icon to print the report.
- Click the Export button to save the data in Excel format (software required).
- Use the **ZOOM** option to minimize/maximize the report view.
- Exit the report by clicking the "X" in the upper right hand corner of the screen.

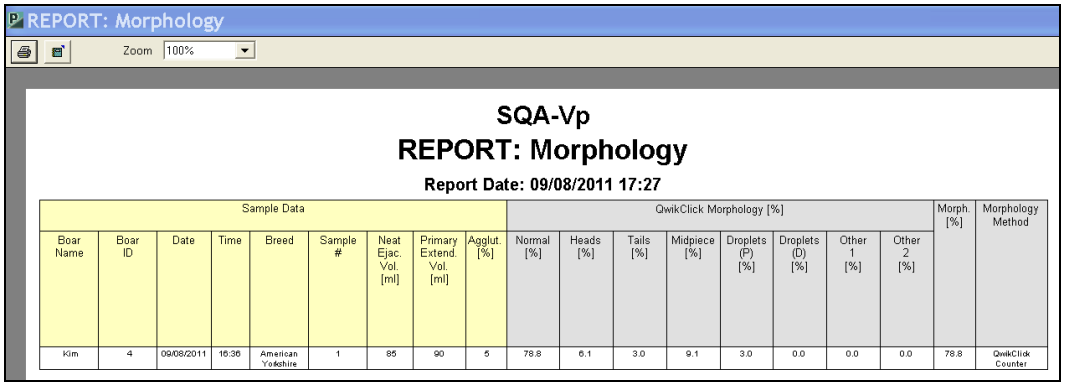
Click on this icon to print a copy of the report.

Select **Fresh Samples** to view and print the following report displaying Sample Data and Test Results for the selected period of time:

Click on this icon to export the semen analysis report.



Select **Morphology** to view and print the Morphology report which displays Sample Data and QwikClick differential morphology results for the selected records:

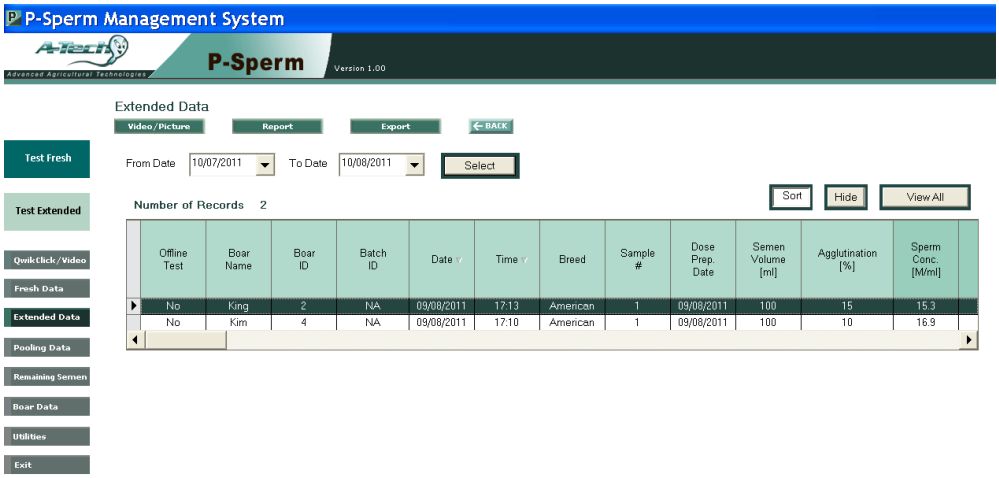


Click the **fourth button** of the **Fresh Data** screen: **Export** and the box below with five report options will be displayed. Use the current (date range) selection, select ALL records in the archive or input a new date range. Click the EXPORT button to send the data to EXCEL (software required)

Extended Data Grid

Section 10: Extended Data

Click **Extended Data** and the screen below will be displayed. This screen will also automatically be displayed after EXTENDED tests are run in the SQA-Vp:



Test Parameters reported for Extended samples:

- Sperm Concentration, M/ml
- MSC, M/ml
- Motility, %
- Motility Grading, (0-5)
- Total Sperm (billions)
- Total Motile Sperm (billions)

Additional Features:

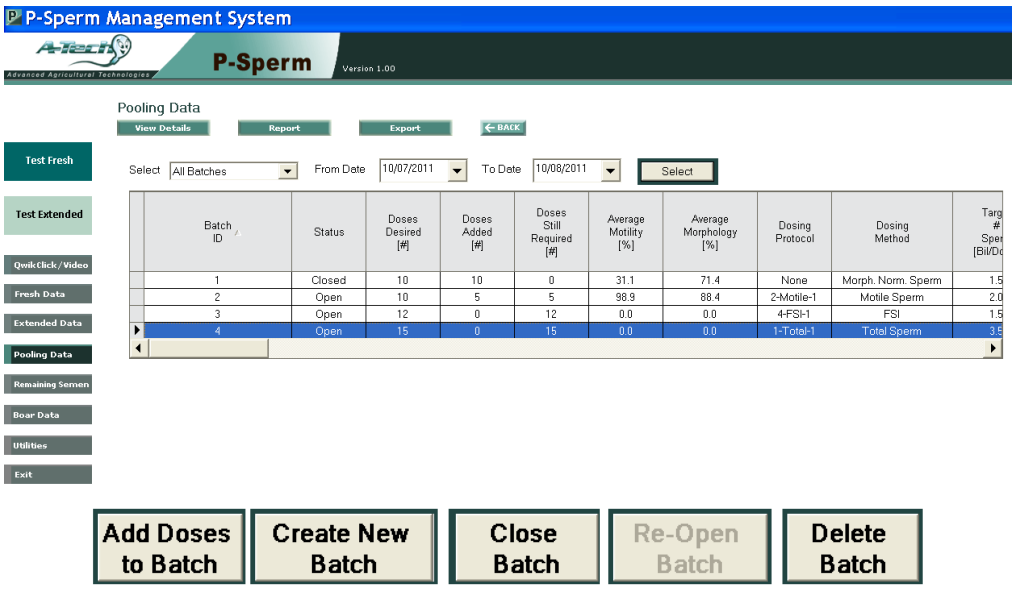
- Sample Pass/Fail
- Name of Lab Tech
- Graphs
- Indicator for "out of normal range"
- Pictures, video clips

The **Extended Data** screen has the same option buttons for displaying data as the previous Fresh Data table:

The same buttons are displayed at the top and bottom of the **Extended Data** table as were previously discussed in the **Fresh Data** table: **Video/Picture, Report & Export and Delete.** The function of these buttons is the same as previously described.

Section 11: Pooling Data

Click **Pooling Data** and the screen below will be displayed.



The following information is displayed in the **Pooling Data** table:

Sample/Boar Information:

- **Batch ID** – A unique batch identification # assigned by the user in the New Batch table.
- **Status** – Open/Closed
- **Doses** – Desired, Added and Still Required.
- **Semen Parameters** – Average Motility and Morphology.
- **Dosing** – Settings and volumes.
- **Date** – The date the batch was created.
- **Time** – The time the batch was created.

There are **five buttons** at the bottom of the **Pooling Data** screen:

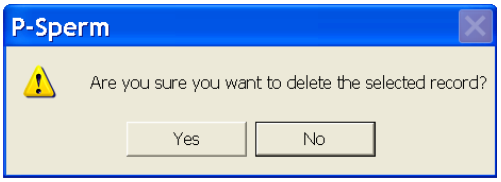
Add Doses to Batch: Click to add doses to an existing OPEN batch(es) of pooled semen.

Create New Batch: Click to set-up a new batch. The batch setting information is saved and shown in the Pooling Data grid and linked screens and reports.

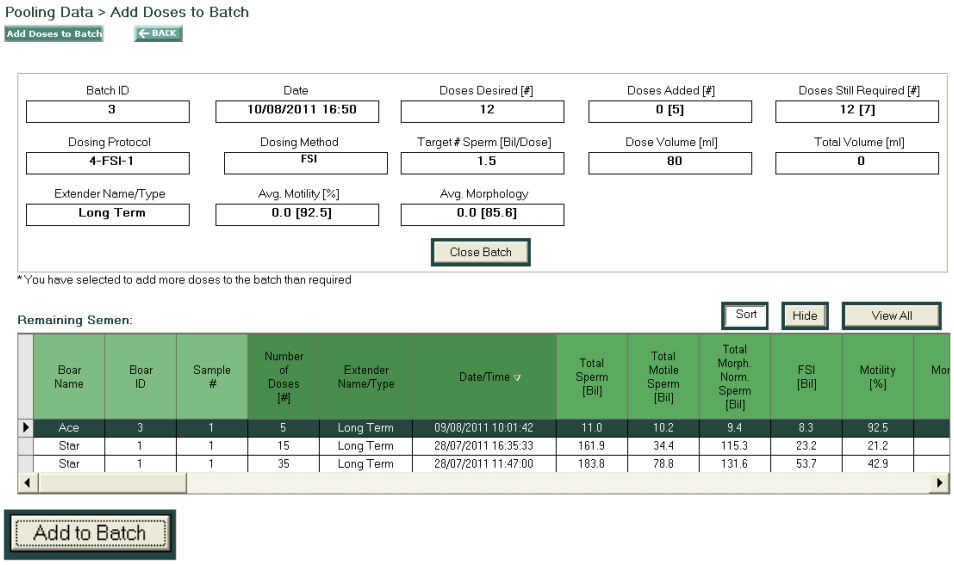
Close Batch: Click to close the batch. When a batch is closed no further doses can be added unless it is re-opened.

Re-open Batch: Click to re-open a previously CLOSED batch to add more doses.

Delete Batch: Click to deleted a batch if 0 doses were added. The message below will be displayed. Click **YES** and the batch will be deleted:

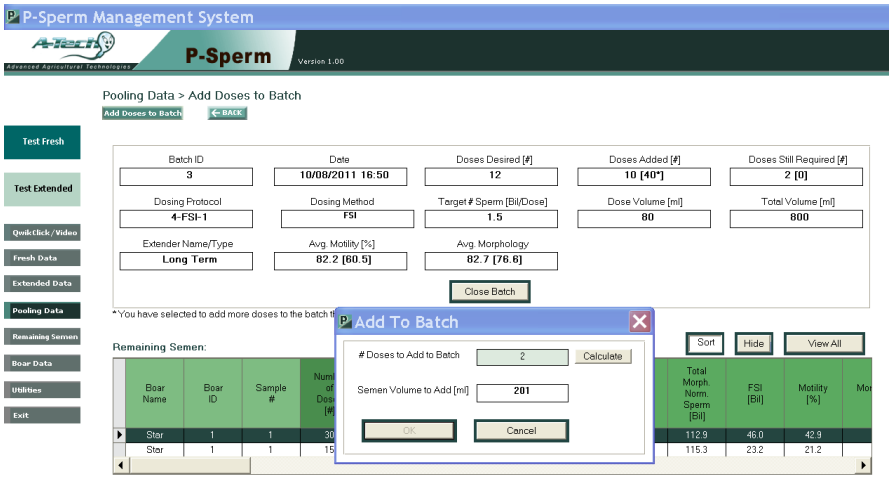


To add doses to an existing batch, click the **Add Doses to Batch** button after selecting the batch from the POOLING DATA grid. The following screen will open:



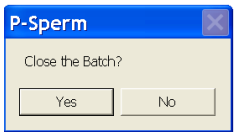
Batch information is displayed in the first section of the screen. The **Remaining Semen** records that contain the SAME Extender Name/Type will be displayed. These can be used to add to the open batch. Select a record by clicking the in left hand column.

Click the **Add to Batch** button located in the lower left hand corner to open the **Add to Batch** box seen below:



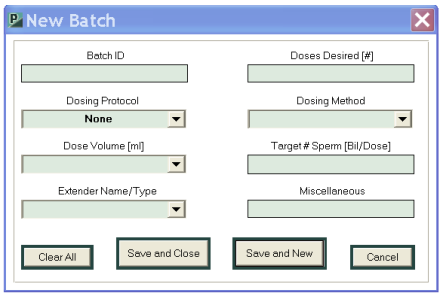
Enter **# Doses to Add to Batch** based on the **Doses Still Required [#]** and press the **Calculate** button. The **Semen Volume to Add [ml]** will be re-calculated as well as the values in brackets for **Doses Added**, **Avg. Motility** and **Avg. Morphology** in the upper section of the screen. These re-calculations demonstrate the impact on the batch of adding the extra dose(es).

Press **OK** to confirm. If all of the **Desired Doses** were added to the batch, the screen below will be displayed: Select **YES** to close the batch or **NO** to keep it open.



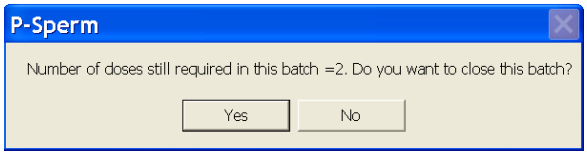
The **three buttons below** are located in the upper right hand corner of the **Remaining Semen** grid of the **Add Doses to Batch** screen. Their function is the same as described previously:

Click: Create New Batch to open the box below:

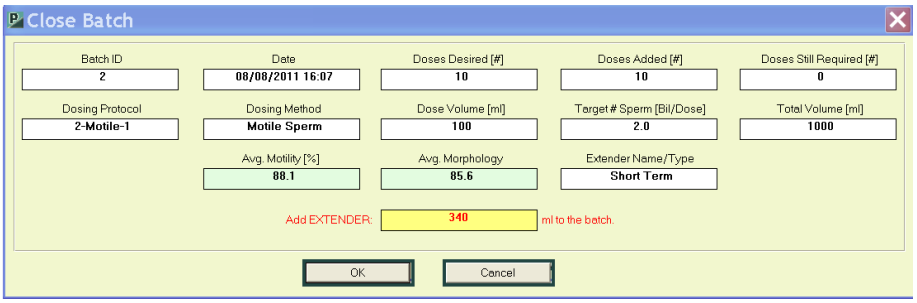


- **Batch ID:** Assign a new number that does not exist in a database.
- **Doses Desired (#):** Enter number of doses desired for this batch.
- **Dosing Protocol:** Select a pre-set protocol from the drop-down menu and Parameters will be displayed based on the Dosing Protocol selected. They cannot be changed in this case.
- If **Dosing Protocol: None** is selected, manually enter the other parameters.
- **Miscellaneous**: This field is not obligatory (provided for some notes).
- If the Batch ID entered already exists a warning message will be displayed.
- Press **Save and Close** to save the batch settings.
- Press **Save and New** to set-up additional batches.

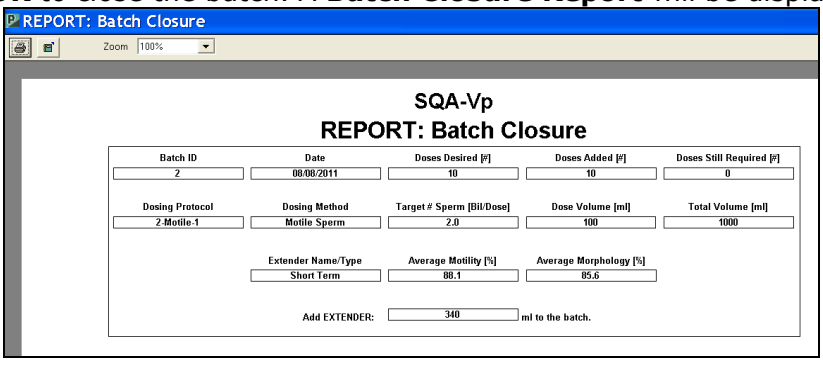
Click the **Close Batch button** and the message below will provide information about the number of doses still required (it will show = 0 if the batch is completed).



Click **YES** and the **Close Batch** box below will appear:



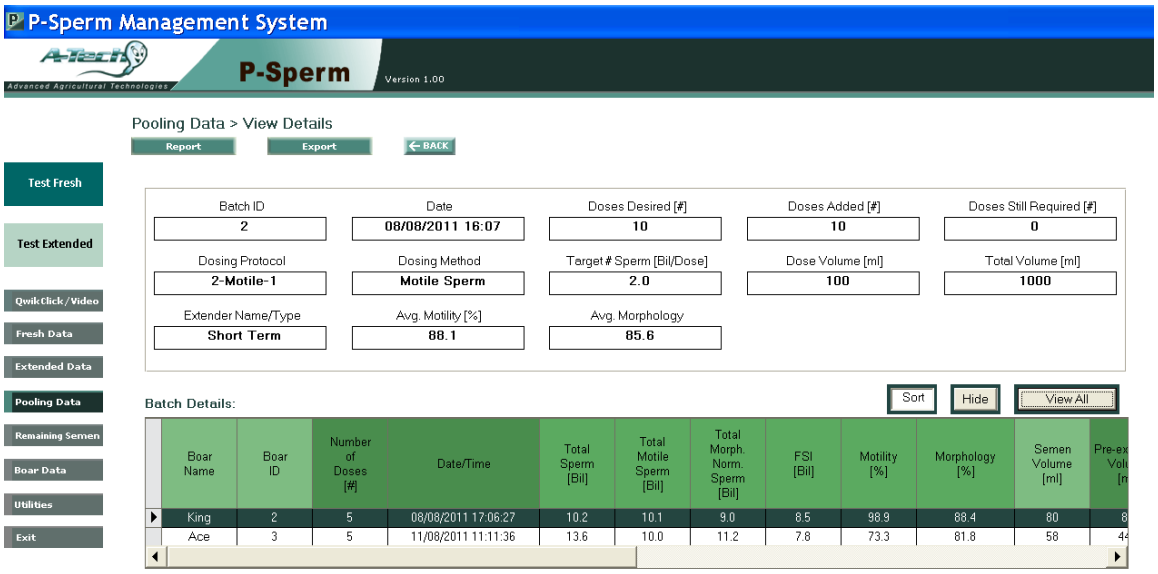
- Batch ID, Date, dosing information, Avg. Motility and Avg. Morphology will be displayed.
- The **Extender** volume to be added to completing the batch is displayed.
- Press **OK** to close the batch. A **Batch Closure Report** will be displayed:



Three buttons and date range table are located at the top of the **Pooling Data** screen:

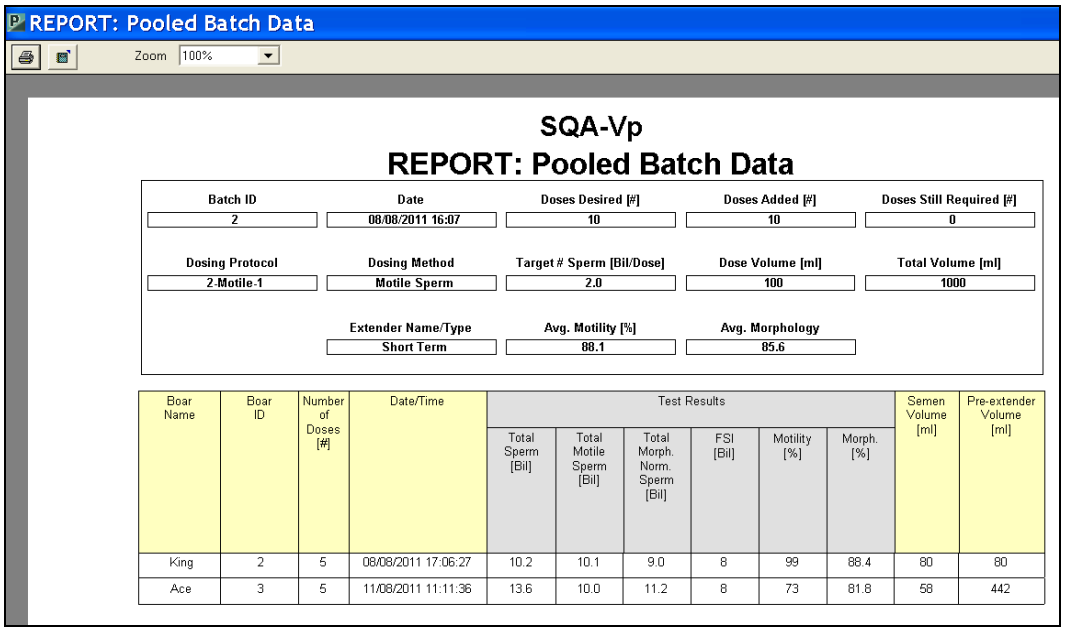
The default date range for displaying data is 1 month, however this can be re-set.

Select a record in the **Pooling Data** grid and click the **first button** – **View Details** or double click the record to view the details of the Pooled Batch.



In the upper section, **Batch** data is displayed. The lower section displays the semen aliquots used in the pool and their associated parameters.

Click the **Report** button to view and print the **Pooled Batch Data** report:



Click on this icon to print a copy of the report.

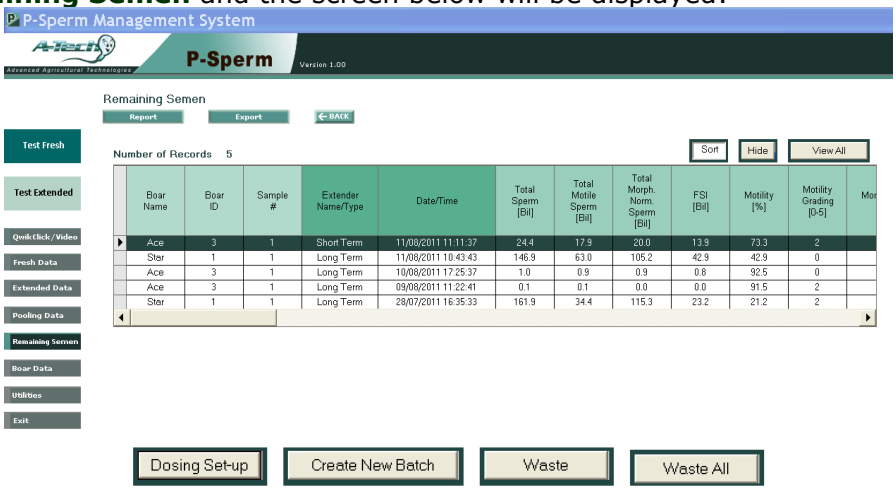
Click on this icon to export the semen analysis report.

Click **Export** button to save the data in an Excel file. The **Report** and **Export** buttons of the **Pooling Data** screen is based on the same principles as in the other screens.

Remaining Semen Grid

Section 12: Remaining Semen

Click **Remaining Semen** and the screen below will be displayed:



Details of the boar, semen parameters and sample information including extender name/type are displayed in the **Remaining Semen** grid.

Report, Export, Sort, Hide and View All buttons function as previously described.

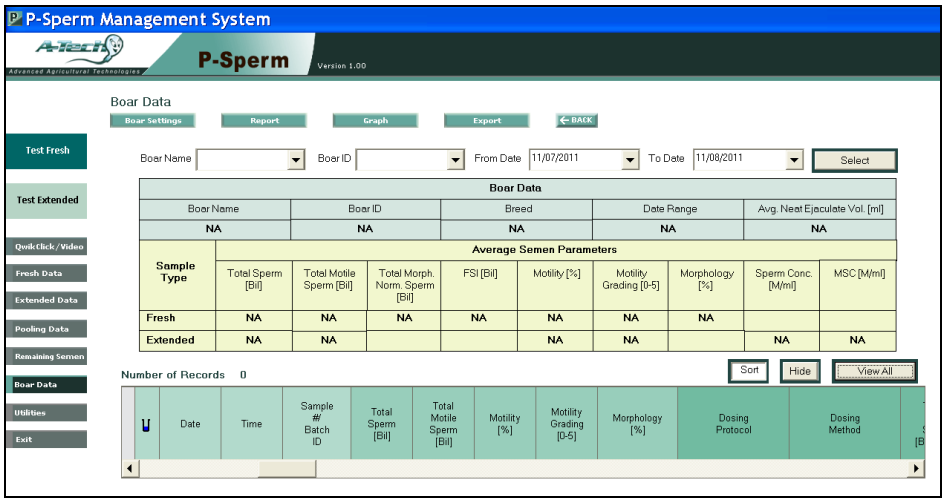
Four buttons at the bottom of the screen are described below:

- **CLICK: Dosing Set-up** to dose the Remaining Semen aliquots (all kinds of dosing, adding to the batches, etc.).
- **CLICK: Create New Batch** to perform the same functions previously described in other screens (**Pooling Data** screen and **Select Batch** box).
- **CLICK: Waste** to delete from the database any Remaining Semen aliquots.
- **CLICK: Waste All** to delete **ALL** the Remaining Semen aliquotes from the grid.

Boar Data

Section 13: Boar Data

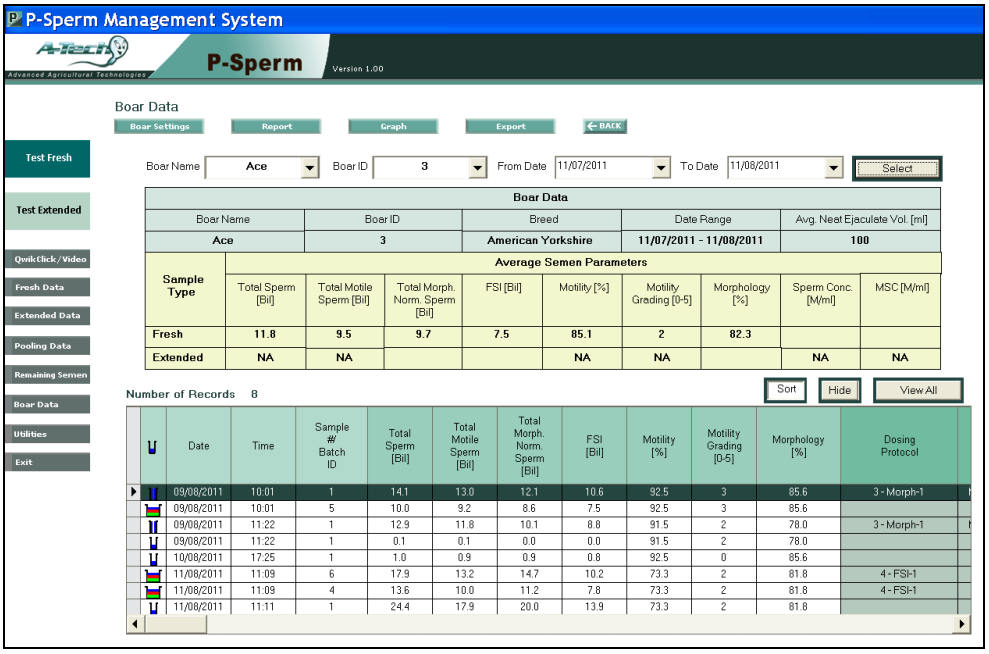
Click **Boar Data** to view and print Boar reports for a specified timeframe. The table below will be displayed:



- Click on the drop-down menu **Boar Name** or **Boar ID** to select the boar.
- Set the timeframe (default is 1 month) and press **Select** to view the report below.

Click on this icon to print a copy of the report.

Click on this icon to export the semen analysis report.

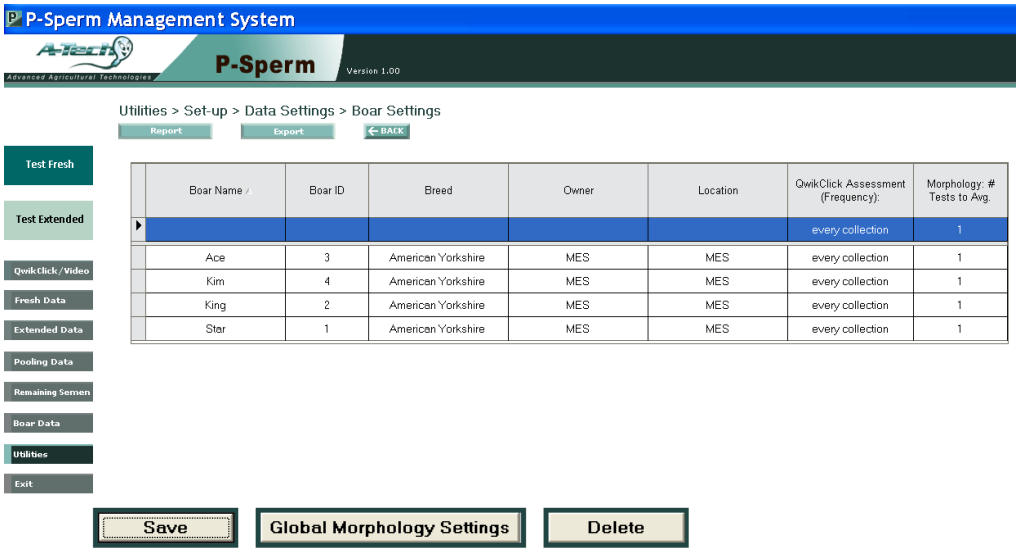


The top section displays the average Neat Ejaculate Volume and Semen Parameters for the boar for the selected timeframe.

The lower part displays complete records of all the boar's semen and dosing history for the timeframe selected.

There are **four buttons** at the top of the **Boar Data** screen.

Boar Settings – Opens the Boar Settings table displayed below:



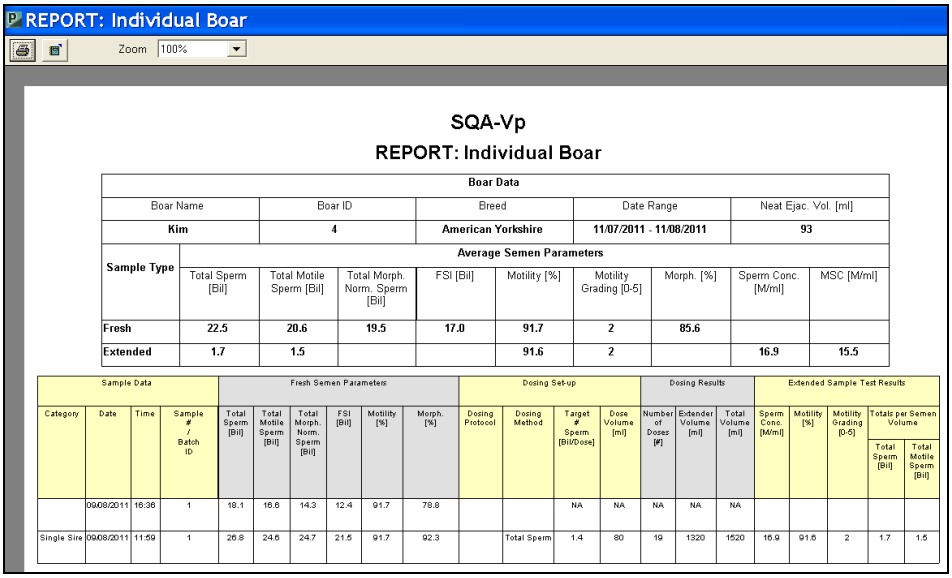
- To add a new boar, click in the top cell and enter the boar information. Automatically updating drop-down menus are provided for some data entry.
- Click the **GLOBAL MORPHOLOGY SETTINGS** button to assign one QCA and AMI for the entire herd. Individual boar settings can be changed by clicking on a particular boar's cells in the table and entering new information.
- **QwikClick Assessment Frequency (QCA):** Enter the frequency for QCA (assessing % droplets in the sample to increase the accuracy of morphology readings). Because morphology testing can be time consuming, in order to increase through-put while effectively monitoring the boar's morphology, select a collection cycle frequency for testing morphology (every, every 2nd, 3rd, 4th, 5th or 6th collection).

- **AMI: Average Morphology Index** is an average of several morphology results from the same boar. The number of morphology test results to include in the average is determined by the operator and can be from 1 to 6 results. The AMI for a given boar will be shown on the QwikClick Assessment or Morphology Counter. **To ensure quality, boars that have an AMI below the morphology cut-off will automatically be selected for morphology testing, regardless of the QCA cycle.**
- **Save or Delete** the settings by clicking the appropriate buttons.
- **Print or Export** (to EXCEL) the **Boar Settings** by clicking the buttons provided.

Click: **Report** in the **Boar Data** screen to open the **Individual Boar Report**:

Click on this icon to print a copy of the report.

Click on this icon to export the semen analysis report.



Boar Data

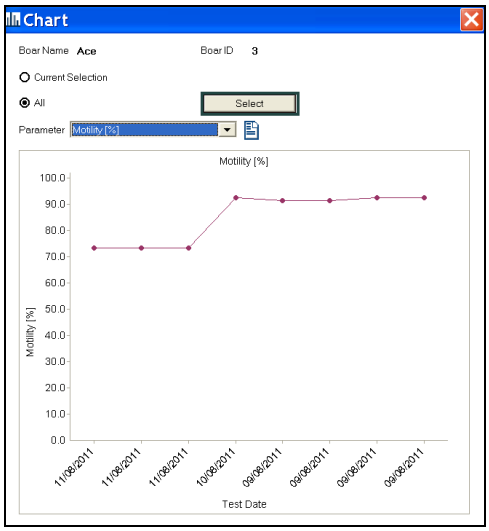
Boar Name	Boar ID	Breed	Date Range	Neat Ejac. Vol. [ml]
Kim	4	American Yorkshire	11/07/2011 - 11/08/2011	93

Average Semen Parameters

Sample Type	Total Sperm [Bil]	Total Motile Sperm [Bil]	Total Morph. Norm. Sperm [Bil]	FSI [Bil]	Motility [%]	Motility Grading [0-5]	Morph. [%]	Sperm Conc. [M/ml]	MSC [M/ml]
Fresh	22.5	20.6	19.5	17.0	91.7	2	85.6		
Extended	1.7	1.5			91.6	2		16.9	15.5

Sample Data				Fresh Semen Parameters						Dosing Set-up				Dosing Results			Extended Sample Test Results				
Category	Date	Time	Sample # / Batch ID	Total Sperm [Bil]	Total Motile Sperm [Bil]	Total Morph. Norm. Sperm [Bil]	FSI [Bil]	Motility [%]	Morph. [%]	Dosing Protocol	Dosing Method	Target # Sperm [Bil/Dose]	Dose Volume [ml]	Number of Doses [#]	Extender Volume [ml]	Total Volume [ml]	Sperm Conc. [M/ml]	Motility [%]	Motility Grading [0-5]	Totals per Semen Volume	
																				Total Sperm [Bil]	Total Motile Sperm [Bil]
	09/08/2011	16:36	1	18.1	16.6	14.3	12.4	91.7	78.8			NA	NA	NA	NA	NA					
Single Sire	09/08/2011	11:59	1	26.8	24.6	24.7	21.5	91.7	92.3		Total Sperm	1.4	80	19	1320	1520	16.9	91.6	2	1.7	1.5

Press the **Graph** button of the **Boar Data** screen to view a graph of a selected boar semen **Parameter** over a specified timeframe.



Clicking  opens the **Boar Data** report with averaged boar parameters and attaches the graph in a printable format.

Utilities

Section 14: Utilities

Utilities contain three sub-menus:

Set-up contains two buttons:

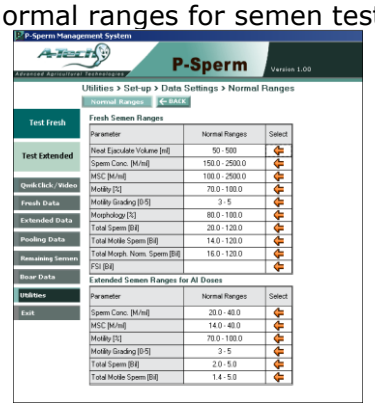
- **Data Settings**
- **System Settings**

Data Settings

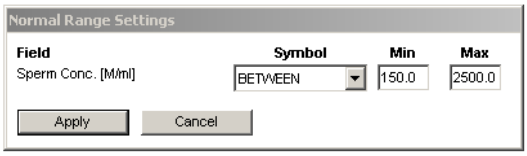
Click **Data Settings** and the following sub-menu will open:

Boar Settings – Please see Boar Data section.

Normal Ranges – Set-up normal ranges for semen test results:



- Click on the ARROW which points to the parameter to be set-up and the **Normal Range Settings** screen will appear:



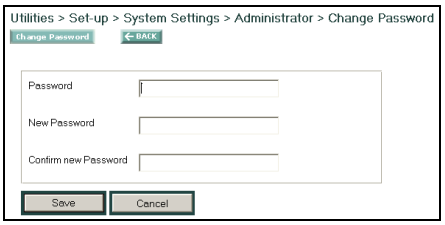
- Enter a symbol (≤, ≥, =, BETWEEN, etc.) and the MIN/MAX values, press **Apply**.

System Settings

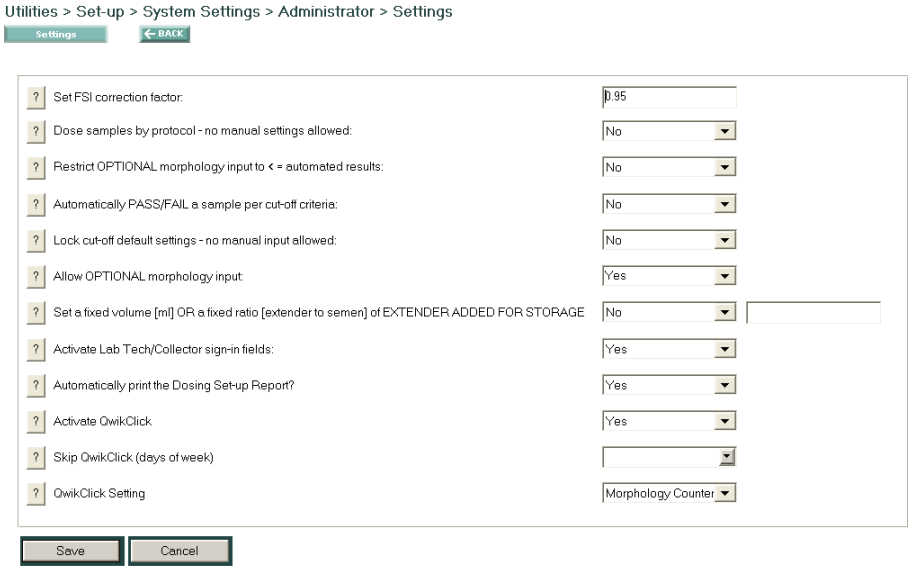
Click **System Settings** to open the five options listed below:

Administrator

This feature is password protected. The default password is **fertility**:



Click **Settings** to display the table below. Click: **?** for information about each setting:



Set FSI correction factor: The FSI (Functional Sperm Index) incorporates concentration, motility and morphology in the formula. FSI can be modified:

- Enter a number less than "1" (0.95/0.85/0.70) and the FSI will be reduced by that factor.
- A number greater than "1"will not be accepted.

Dose samples by protocol – no manual settings allowed: The Administrator can set-up pre-programmed dosing protocols that can be used instead of manually selecting each dosing criteria. After set-up, these protocols are selected from a drop-down menu on the dosing screen. The following options are available for this default setting:

- Select: YES to dose (single or multiple) only by the pre-set Administrator dosing protocols. All manual entry fields will be locked.
- Select: NO to have the option to dose by EITHER a pre-set protocol or by manual selection/entry. All manual entry fields will be un-locked.

Restrict OPTIONAL (manual) morphology to < = automated results: Manual morphology results can be run off-line and entered into a field on the dosing screen. The P-Sperm will then report manual morphology instead of automated. The manual morphology field can be restricted in the following way:

- Select: **YES** and OPTIONAL (manual) morphology results that are greater than the automated results cannot be entered into the manual morphology field. Manual morphology will be reported only if it is less than or equal to the automated results.
- Select: **NO** and the OPTIONAL morphology results and the manual results will be reported instead of the automated results.

Automatically PASS/FAIL a sample per cut-off criteria: After setting up cut-offs for various semen parameters, there is an option to automatically PASS or FAIL a sample based on these cut-offs. If a sample FAILS automatically, the dosing screen will not be visualized. The test results will still be reported. To use this feature:

- Select: **YES** and a sample will automatically PASS or FAIL based on pre-set cut-offs. If the sample FAILS, the QwikClick will be locked and only the EXIT button will be activated. Press EXIT and the test results grid will be displayed instead of the dosing screen.
- Select: **NO** to manually PASS/FAIL a sample and have the option to use the QwikClick Droplet Assessment and view the dosing screen.

Lock cut-off default settings – no manual input allowed:
Default settings for cut-offs can be set by the Administrator. Once these are set, they are reported in the QwikClick and Dosing Screens and PASS/FAIL results are displayed based on these cut-off values. Access to change these fields can be restricted:

- Select: **YES** to LOCK the Administrator defined cut-offs. The fields displayed in the QwikClick and Dosing screens cannot be changed.
- Select: **NO** and the Administrator defined cut-offs can be updated manually in the QwikClick and Dosing screens.

Allow OPTIONAL morphology input:
Morphology results can be run off-line and entered into the OPTIONAL field on the dosing screen. This optional field entry will then be reported on all P-Sperm morphology reports AND will be used to calculate doses (if morphology is part of the dosing protocol) instead of the automated or QwikClick morphology results.

- Select: **YES** to allow the entry of a morphology value in the OPTIONAL morphology field.
- Select: **NO** to block the OPTIONAL field from any input. Only AUTOMATED /QwikClick morphology will be reported.

Set a fixed volume (ml) or a fixed ratio (EXTENDER to SEMEN) of EXTENDED ADDED FOR STORAGE:

In order to preserve the sample for multiple dosing, an EXTENDER needs to be added in Round #1 of the Multiple Dosing cycle.

- Select **YES** to set a defined amount of EXTENDER for the first round (only) of MULTIPLE DOSING.
- Select **NO** to manually enter the EXTENDER volume.

If **YES** is selected, enter EITHER the RATIO of EXTENDER TO SEMEN or a FIXED EXTENDER VOLUME:
RATIO of EXTENDER to SEMEN: Enter a whole number that reflects the ratio of EXTENDER to remaining semen. (Example: Enter "1" to extend a 50cc sample with 50cc of extender; Enter "2" to extend a 50cc sample with 100cc of extender).
FIXED EXTENDER VOLUME: Enter a fixed volume of extender to be added to the sample in the first dosing round.

Activate Lab Tech/Collector sign-in screen:

- Select: **YES** to require a sign-in by the Lab Tech and Collector each time a sample is run and transferred to P-Sperm. The Lab Tech and Collector name will appear on reports.
- Select: **NO** and the sample results will be transferred without this information and will not be tracked in any report.

Automatically print the Dosing Set-up Report?

- Select: **YES** to automatically print the Dosing Set-up Report for both SINGLE and MULTIPLE dosing after the dosing information has been saved.
- Select: **NO** and the report can only be printed by selecting the PRINT button after dosing.

Activate QwikClick

- Select: **YES** to automatically activate the QwikClick morphology counter after running a test.
- Select: **NO** and the dosing screen will automatically be shown after running a test and the automated morphology will be reported.

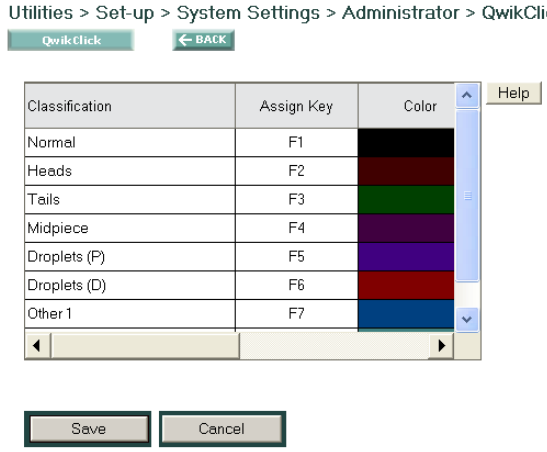
Skip QwikClick (days of week)
Click the drop-dowen menu and check the days of week when QwikClick should be skipped.

QwikClick Setting

QwikClick Morphology Option 1: Droplet Counter (QCA)
Select to automatically activate the QwikClick Droplet Counter when testing a sample. This counter allows the operator to assess the % of droplets seen in the pig semen sample. This droplet factor (%) is then used to re-calculate the automated morphology value for VERY high accuracy that includes this non-motility based defect.

QwikClick Morphology Option 2: Morphology Counter
Select to perform an on-line (links to a pig test report) completely manual morphology differential count. The value for % normal morphology will then be seen in the dosing screen and in all of the reports. The complete manual morphology differential can be seen on a separate morphology report.

QwikClick – to set-up the QwikClick Morphology Assessment table below:



Classify morphology defects:

- Click on any cell in the classification column and type in a description of the morphology defect.
- All cells in the column must be labeled – enter OTHER 1 , 2, etc.
- The NORMAL cell is locked and cannot be changed.

Assign function keys (F1 – F12):

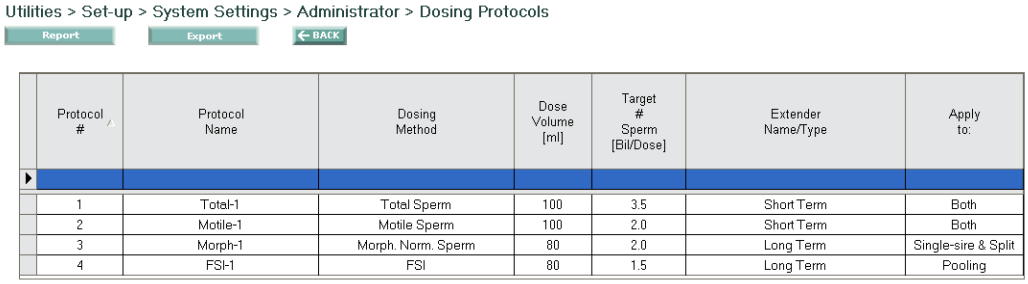
- Click on each cell in the **ASSIGN KEY** column.
- Select a **FUNCTION KEY** (F1 – F12) from the drop down menu in each cell.
- The NORMAL key is locked – it is always F1.

Select COLORS for the FUNCTION keys:

- Click on each cell in the **COLOR** column and select the color that will be assigned to the cell classification.

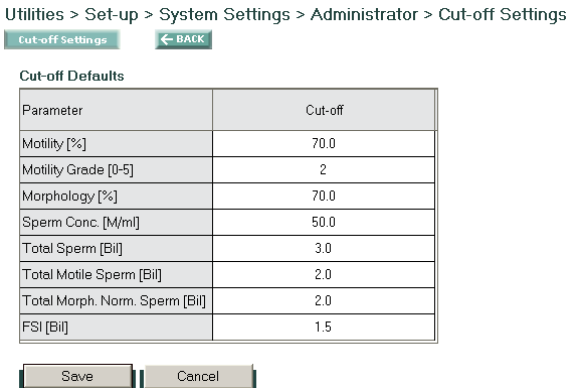
Save settings.

Dosing Protocols can be pre-set. Enter the protocol definitions into the the first line of the table displayed below and click **Save**. The protocol will automatically be available in the DOSING SET-UP Screen(s) selected.



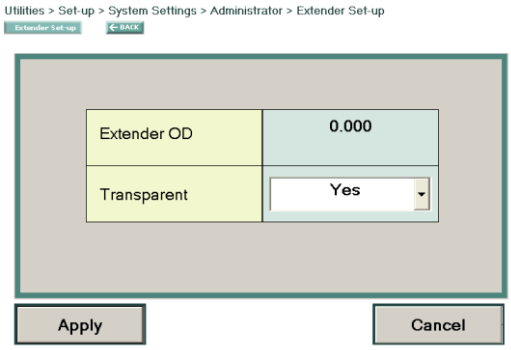
Report and **Export** buttons function the same way as previously described.

Cut-off Settings can be pre-set by entering the cut-off values into the table provided:

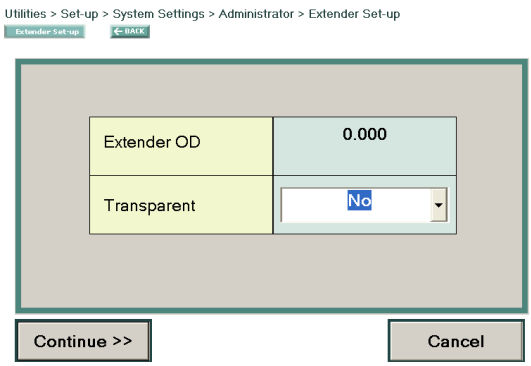


- **Motility Cut-off (%):** Enter the lowest motility acceptable for dosing.
- **Motility Grade Cut-off (0-5):** Enter the lowest acceptable Motility Grade.
- **Morphology Cut-off (%):** Enter the lowest acceptable % normal morphology.
- **Conc. Cut-off (M/ml):** Enter the lowest concentration acceptable for dosing (leave it empty if there is no cut-off).
- **Total parameters & FSI:** Enter the cut-offs that correspond to one AI dose targets (or based on your experience).

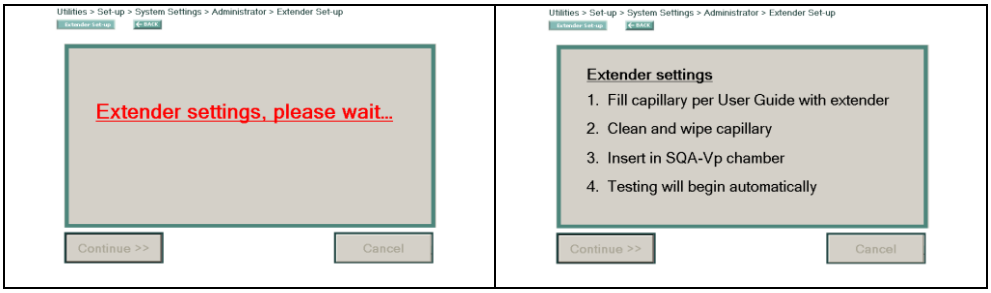
Extender Set-up can be performed from this screen:



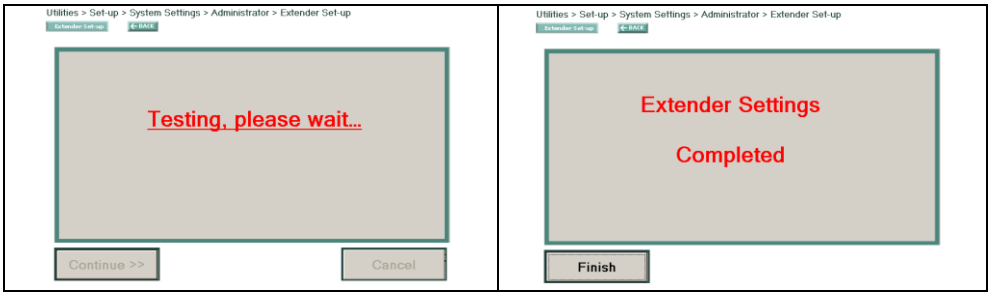
- If **Transparent,** select **YES**, Extender OD will be 0.
- Click **Apply** and results will be saved.
- If **Transparent "No",** click the **Continue** button at the bottom of the screen.



- The following sequence of screens will be displayed:

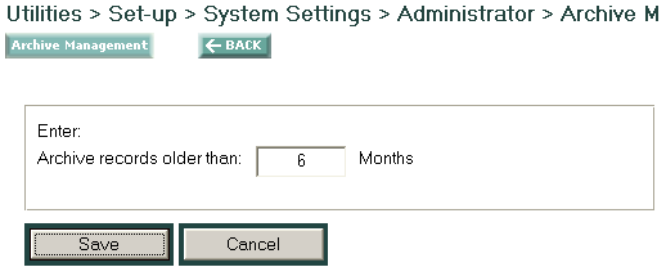


- Follow instructions and the following screens will be seen:



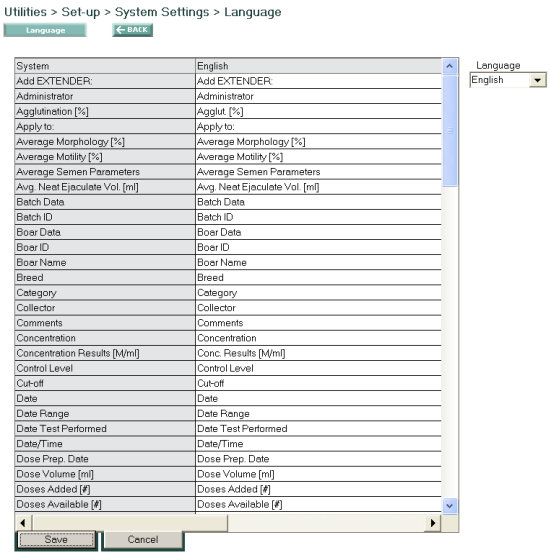
- Click **Finish** and results will be saved.

Archive Management: Click this button and the following screen will be shown:

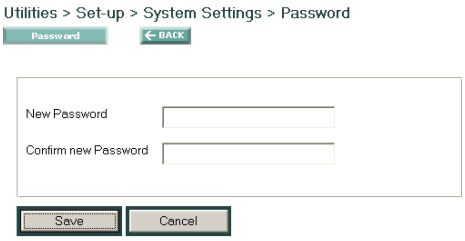


To effectively handle the large database of boar information and permit faster data retrieval, enter **number of months** for archiving records and press **Save**.

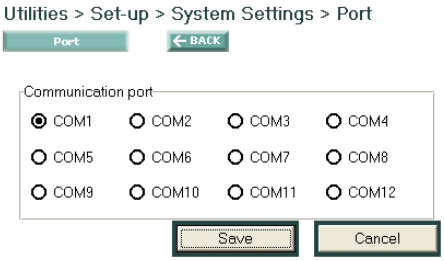
Click: Language to customize the language used in P-Sperm Reports. Choose "Other" from the Language drop-down menu, fill-in the table as desired and click **Save**.



Click **Password** to change to a new password. Enter and confirm the new password in the screen below. Click **Save** to immediately re-set and save the new password.



Port: Click this button to set the communication port for the PC:



Auto Export: This feature requires manufacturer support.

Controls

Controls Testing

QwikCheck™ beads are produced by Medical Electronic Systems: www.mes-global.com)

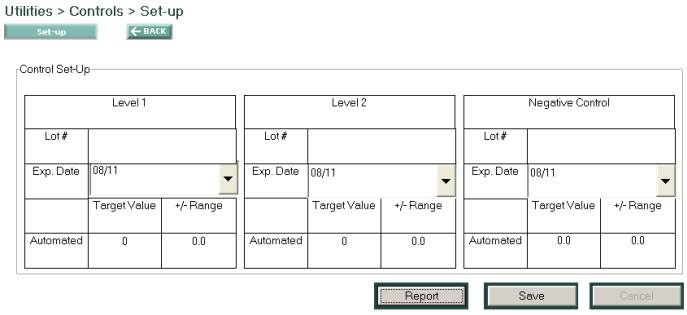
Please note: QwikCheck beads verify that the concentration channel of the SQA-Vp is optimally functioning before a problem impacts the test results.

NOTE:

Remember to set-up Controls before running a new box of QwikCheck Beads.

Click **Utility>Controls** and three options are displayed: **Set-up**, **Test Results** and **Test Controls:**

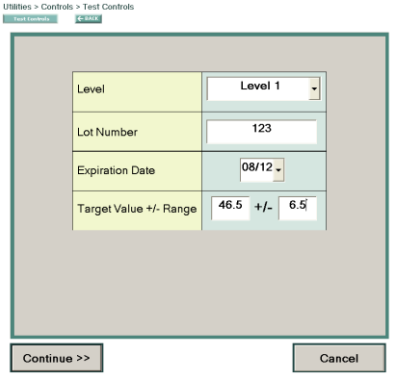
Click **Set-up** and to view the screen below:



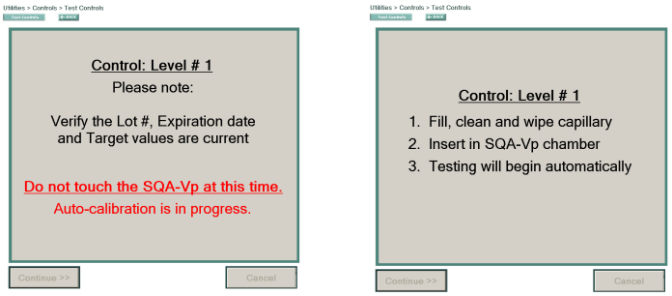
Enter the required information into the table. This information is on the QwikCheck™ beads box. Press **Report** to print out the settings. Press **Save** to apply the settings.

Click **Test Results** to view the control records for a selected timeframe (see below).

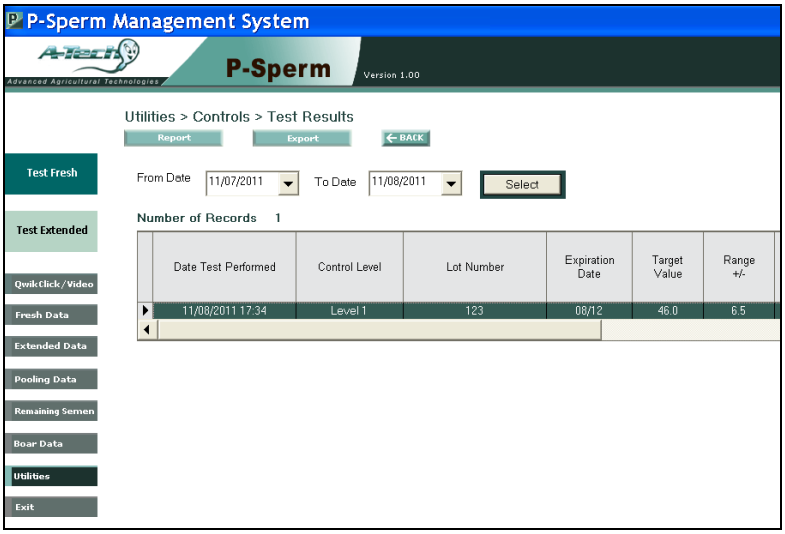
Click the **Test Controls** button and the screen below will be displayed:



- Select the Control **Level**.
- **Lot Number**, **Expiration Date** and **Target Value +/- Range** can be set or over-written in this screen if require.
- Press **Continue** and follow the screen instructions:



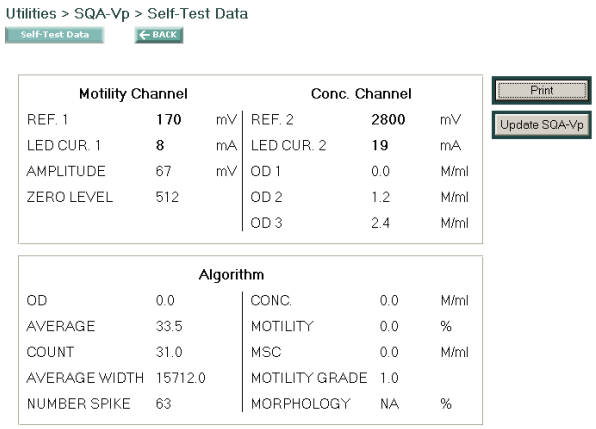
- When testing is completed, the screen below will be displayed:



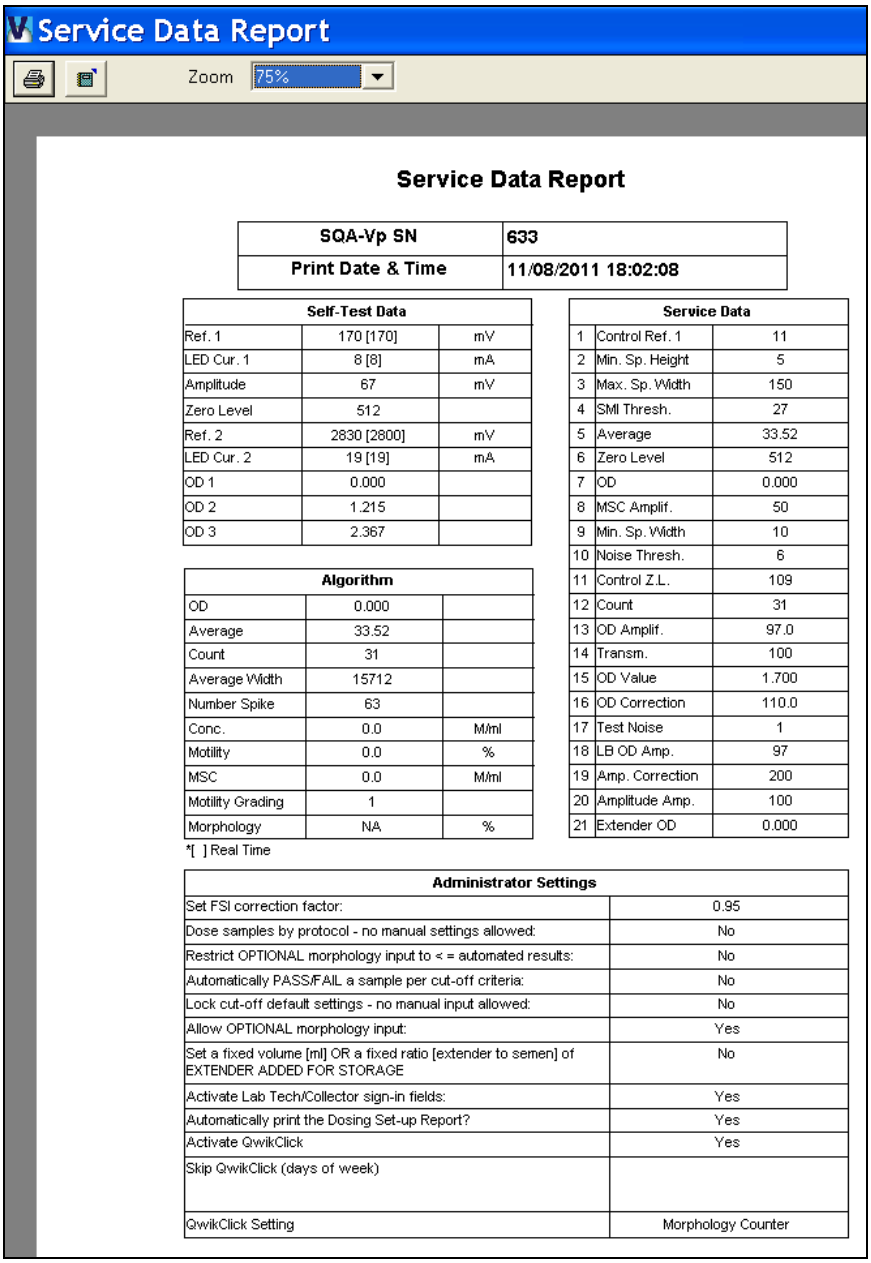
- Click the **Report** or **Export** buttons to print a report or export the control results to an Excel file.

Utilities > SQA-Vp – Click this button and two options are displayed:

Click **Self-Test Data** and this information will be displayed:



Print the **Service Data Report** which contains additional information:



Service Data Report

Zoom 75%

Service Data Report

SQA-Vp SN	633
Print Date & Time	11/08/2011 18:02:08

Self-Test Data		
Ref. 1	170 [170]	mV
LED Cur. 1	8 [8]	mA
Amplitude	67	mV
Zero Level	512	
Ref. 2	2830 [2800]	mV
LED Cur. 2	19 [19]	mA
OD 1	0.000	
OD 2	1.215	
OD 3	2.367	

Algorithm		
OD	0.000	
Average	33.52	
Count	31	
Average Width	15712	
Number Spike	63	
Conc.	0.0	M/ml
Motility	0.0	%
MSC	0.0	M/ml
Motility Grading	1	
Morphology	NA	%

*[ ] Real Time

Service Data		
1	Control Ref. 1	11
2	Min. Sp. Height	5
3	Max. Sp. Width	150
4	SMI Thresh.	27
5	Average	33.52
6	Zero Level	512
7	OD	0.000
8	MSC Amplif.	50
9	Min. Sp. Width	10
10	Noise Thresh.	6
11	Control Z.L.	109
12	Count	31
13	OD Amplif.	97.0
14	Transm.	100
15	OD Value	1.700
16	OD Correction	110.0
17	Test Noise	1
18	LB OD Amp.	97
19	Amp. Correction	200
20	Amplitude Amp.	100
21	Extender OD	0.000

Administrator Settings	
Set FSI correction factor:	0.95
Dose samples by protocol - no manual settings allowed:	No
Restrict OPTIONAL morphology input to < = automated results:	No
Automatically PASS/FAIL a sample per cut-off criteria:	No
Lock cut-off default settings - no manual input allowed:	No
Allow OPTIONAL morphology input:	Yes
Set a fixed volume [ml] OR a fixed ratio [extender to semen] of EXTENDER ADDED FOR STORAGE	No
Activate Lab Tech/Collector sign-in fields:	Yes
Automatically print the Dosing Set-up Report?	Yes
Activate QwikClick	Yes
Skip QwikClick (days of week)	
QwikClick Setting	Morphology Counter

Verification function is provided for Service Personnel only.

Section 15: Exit

Click **Exit** to close the P-Sperm program. Confirm **YES** or **NO**.

APPENDIX 1:

System Requirements and Installation Instructions

Warning: The frame grabber should be installed prior to using P-Sperm!

System Requirements:

- SQA-Vp with RS232 communication cable and power cable

PC requirements:

Hardware requirements:

- 1Ghz or higher CPU; 256 MB RAM
- AGP Video Display Card with at least 16 MB of RAM memory
- CD-ROM compatible drive
- RS232 communication port (serial) and Two available USB ports

Software requirements:

- Windows XP, 7, 8.1 and 10
- EXCEL (for exporting data)
- At least 40 GB of free hard disk space recommended
- Video resolution ≥ 640x480 (recommended 1024x768)

Video color quality ≥ 16-bit

Installation: Video frame grabber USB device

A FRAME GRABBER must be installed to use the video in P-Sperm. For installation instructions, please to the P-Sperm package insert.

P-Sperm Installation

1. Close any open programs and insert the P-Sperm CD into the PC's CD-ROM. The installation process will begin automatically
2. Click **NEXT** when the screen displays "Initializing Wise Installation Wizard" and the "Welcome" screen appears.
3. Click **NEXT** when the "Choose Destination Location screen is displayed to accept the P-Sperm default folder or click"Browse" to select another folder.
4. Click **NEXT** when "Select Program Manager Group" is displayed. If a message "Digital Signature Not Found" is displayed, click YES.
5. Installation complete: Click OK to restart the PC.

SQA-Vp Connection

- Connect one end of the RS232 communications cable to the PC, the other end to the SQA-Vp.
- Set-up system defaults by following instructions in the SQA-Vp User Guide.

Video Capture Device Settings

- Go to the P-Sperm: QwikClick/Video > Video Settings
- Using the drop-down menus, select the video capture device that has been installed (refer to the Video Capture Installation guide).

APPENDIX 2:
P-Sperm QwikClick Assessment (QCA) and Average Morphology Index (AMI)

QCA Overview:

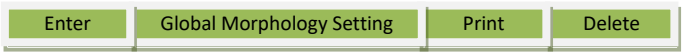
The QwikClick Assessment (QCA) allows the operator to select how often the QwikClick Morphology will be automatically activated for boar morphology testing. Because morphology testing can be time consuming, in order to increase through-put while effectively monitoring the boar's morphology, the operator defines the frequency for testing morphology (every, every 2nd, 3rd, 4th, 5th or 6th collection).

AMI Overview:

The Average Morphology Index (AMI) is an average of several morphology results from the same boar. The number of morphology test results to include in the average is determined by the operator and can be as many as six. AMI is displayed in the QwikClick morphology counter of P-Sperm and can be viewed before a sample is divided into doses. To ensure quality, boars that have an AMI below the morphology cut-off will automatically be selected for QCA morphology testing, regardless of the QCA schedule.

QCA and AMI Set-up in P-Sperm: From the BOAR SET-UP TABLE

BOAR SET-UP TABLE						
Boar ID	Boar Name	Breed	Owner	Location	QwikClick Assessment (Frequency)	Morphology: # Tests to Avg.
					▼	▼
					every collection	2
					every 2nd	3
					every 3rd	4
					every 4th	5
					every 5th	6



QCA (Frequency):

- Select a boar (boars can be sorted by ID, NAME, BREED, etc.). Using the drop-down menu under QCA (Frequency), select which collections should include QCA (every, every 2nd, etc.). Press ENTER to save.
- The frequency of QCA can be set differently for each boar or can be set globally for the entire stud. To set globally, click the Global Morphology Setting and a screen will be activated. Enter the information about QCA (AMI can be set at the same time). Press ENTER to save. If QCA is set globally, individual boars can still be selected and the global setting can be overwritten.

AMI – Morphology # Tests to Avg.:

- Select a boar (boars can be sorted by ID, NAME, BREED, etc.). Using the drop-down menu under Morphology # Tests to Avg., select how many morphology tests to average for the AMI (up to 6). Press ENTER.
- AMI will immediately be displayed on the QwikClick morphology and dosing screens.
- The AMI can be set differently for each boar or can be set globally for the entire stud. To set globally, click the Global Morphology Setting and a screen will be activated. Enter the information about AMI (QCA can be set at the same time). Press ENTER to save.

How to use QCA and AMI in the testing flow:

In the BOAR TABLE of the P-Sperm, each boar is scheduled for both QCA and AMI. When a boar is tested on the SQA-Vp for the first time, irrespective of the operator settings, the QCA screen in P-Sperm will be displayed. Since testing all boars the same day would slow down throughput, an optimal flow should be determined in the P-Sperm right from the beginning of SQA-Vp testing. Assume:

- 250 boars in the AI stud, collected two times/week and MONDAY and TUESDAY are the busiest test days
- The Frequency of QCA is set to "every 5th" (i.e. QwikClick morphology is run every 5th sample of a given boar)
- The AMI is set to "3" (average of 3 tests)

The table below describes how to FLOW the boars for testing QCA/AMI for greatest efficiency and throughput.

# Boars in a stud	Week	Collection	Days of week	# Boars tested	QCA	Notes
250	1	1st	Mo	125	30	MANUAL SELECTION: Perform QCA only on boars whose AUTOMATED morph. < cutoff. Exit QCA screen without droplet counting if AUTOMATED morph. >= cutoff (press Exit button).
			Tu	125	30	
		2nd	We	85	65	MANUAL SELECTION: Perform QCA on all boars not tested during 1st collection. To determine this, look at the Test Results table when the QCA screen is opened. The Morphology will be labeled as "A"(automated), but not AMI (averaged) if the boar was already QCAed at least once. TEST the "A" boars.
			Th	85	65	
			Fr	80	60	
	2	3rd	Mo	125	30	If all the boars have been QCAed during the first two collections, follow the P-Sperm AUTOMATED selection. PLEASE NOTE: If the Frequency of QCA is set to every 5th collection, the QCA screen will not be opened for the 3rd and 4th collections unless the AMI (avg. morph. index) < cutoff.
			Tu	125	30	
		4th	We	85	30	
			Th	85	30	
			Fr	80	30	
	3	5th	Mo	125	30	Follow the P-Sperm automated selection based on the schedule plus samples to be re-tested if AMI < cutoff. The P-Sperm will alert the operator for BOTH types of these QCA tests.
			Tu	125	30	
		6th	We	85	65	
			Th	85	65	
			Fr	80	60	

- 1st Collection: At the beginning in the first two production days, QCA will include ~25% of the boars (**60+** of 250) ejaculates: All samples that have an automated morphology < cutoff (manual selection).
- 2nd Collection: Then in the next three production days, QCA will include ~75% of the boars (**190+** of 250) ejaculates: Those boars not tested during the 1st collection (manual selection).
- Each day when 125 tests are performed, 30 QCA are done.
- Each day when 80-85 tests are performed, 60-65 QCA are done.
- In one week all the boars will have QCA at least one time.

- 3rd and 4th Collection: Follow ONLY the automated QCA schedule: The QwikClick screen will be opened automatically for all QCA scheduled boars PLUS those boars whose AMI [average morphology index] falls below the morphology cutoff set by administrator.
- 5th and 6th Collections: QCA will continue to run automatically (the software will select the QCA scheduled boars PLUS the boars whose AMI falls below the morphology cutoff set by administrator).

APPENDIX 3:
SQA-Vp SERVICE MENU

From SQA-Vp **MAIN MENU** select: **SERVICE MENU** to access one of 5 functions:

SERVICE DATA: Select this option to view three service screens:

- **Service Data screen (Communication screen)**: Establishes communication between the SQA-Vp and the PC (P-Sperm) in order to:
  - Set-up the CONTROLS default settings
  - Report SQA-Vp service information for technical troubleshooting
- **Self-Test Data screen:** Provides system information after Self-Test (Self-Test Data) and after regular testing (Internal Data).
- **Algorithm data screen:** This screen displays algorithm calculations

SERVICE PERSONNEL:

- For technical services personnel only and requires a password to access.

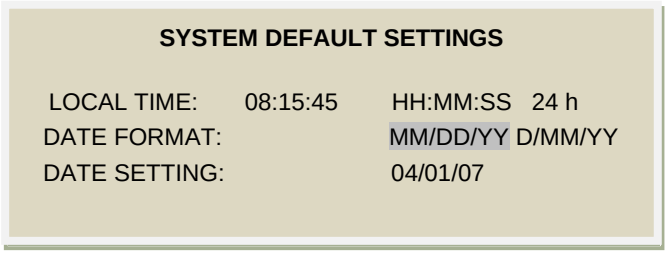
PRINT SELF-TEST DATA AND DEFAULT SETTINGS:

Select to print the SQA-Vp **Self-Test Data** and **Default Settings.** To activate these functions:

- Highlight this option in the **SERVICE MENU** and press **ENTER**.
- Select: **Self-Test Data** or **Default Settings** and press **ENTER.**

SETTINGS Select this option to set the system and sample defaults.

SYSTEM Default Settings:



- **LOCAL TIME:** enter local time
- **DATE FORMAT:** Select the format **DD/MM/YY or MM/DD/YY** using the right/left arrows on the keyboard. Press **ENTER** to confirm.
- **DATE SETTING:** enter current date

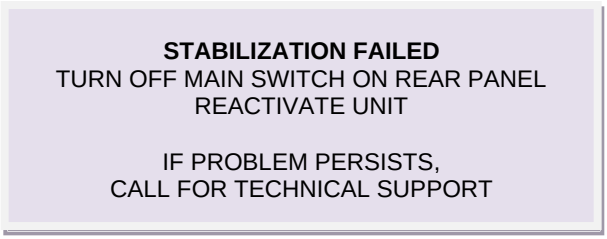
ADD TESTS TO COUNTER: I-button tests

NOTE: If the I-Button is not properly inserted a message: **I-BUTTON NOT PROPERLY ACTIVATED** will be displayed. Remove the button, press ESC and try again.

- Select **SERVICE > SERVICE MENU >ADD TESTS TO COUNTER** and press **ENTER**
- The SQA-Vp screen will instruct the user to: **HOLD NEW I-BUTTON AGAINST PORT AND PRESS ENTER.**
- Make sure the I-Button touches the internal surface and edges of the port. Press the I-button firmly in the port, moving it left and right to make sure it also touches the edges of the port.
- The **#TESTS ADDED** and the cumulative **#TESTS NOW REMAINING** will be displayed on the screen of the SQA-Vp.
- The screen will warn the user if an **EMPTY I-BUTTON** (was) **INSERTED**

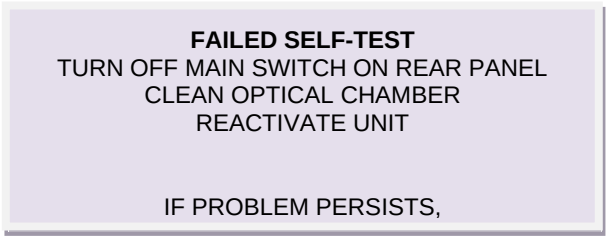
APPENDIX 4:
Troubleshooting SQA-V/P-Sperm Warning Screens

Stabilization Failed:



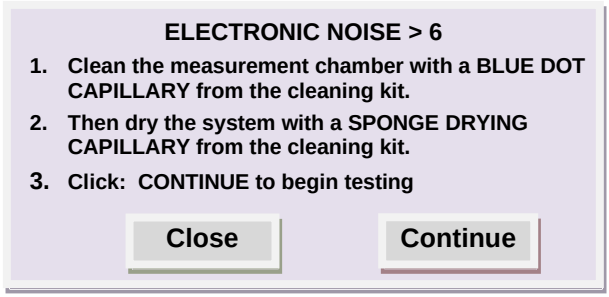
- Ensure there is no testing capillary in the measurement compartment.
- Remove the SQA-Vp from sources of electronic noise (centrifuge, cell phones, etc.).
- Clean measurement compartment (refer to Appendix).
- Reboot the SQA-Vp without a testing capillary in the chamber:
  - Turn system **OFF** then back **ON** at the main switch on the rear panel.
  - Press the front panel **ON/OFF** key to begin Auto-Calibration/Self-Test
  - Call for technical support if failure recurs.

Failed Self-Test:



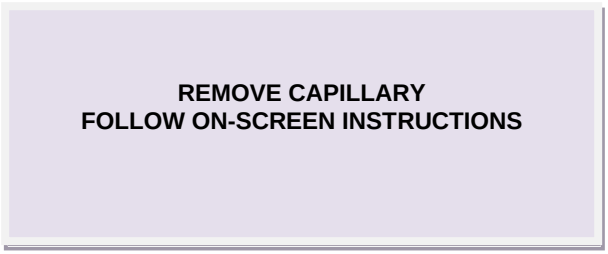
- Ensure there is no testing capillary in the measurement compartment.
- Remove the SQA-Vp from sources of electronic noise (centrifuge, etc.).
- Clean measurement compartment (refer to Appendix).
- Reboot the SQA-Vp without a testing capillary in the chamber:
  - From the rear panel switch, turn the system **OFF** then back **ON.**
  - Press the front panel **ON/OFF** key to begin Auto-Calibration, Stabilization and Self-Test.
- Call for technical support if this message is displayed again. Prepare for technical support by printing a copy of the SQA-Vp Self-Test/Service Data:
  - Press **SERVICE** key. The **SERVICE MENU** will be displayed.
  - Select **PRINT SELF-TEST AND DEFAULT SETTINGS** option.
  - Select **SELF-TEST DATA** and press **ENTER** to print a copy of the self-test/service data.

Electronic Noise:



- The above message will be seen on the computer. Ensure there is no testing capillary in the SQA-Vp measurement compartment.
- Remove SQA-Vp from sources of electronic noise (centrifuge, etc.).
- Clean measurement compartment as instructed on the screen (or in Appendix section).
  - After cleaning, press: **CONTINUE** and re-run the test.
- If this message is displayed again, clean again.
- If this message is displayed a third time, reboot the SQA-Vp:
  - Turn the system **OFF** then back **ON** at the main switch on the rear panel.
  - Press the front panel **ON/OFF** key to begin Auto-Calibration and Stabilization.
  - From MAIN menu: Select **TEST NEW SAMPLE** and re-run.
  - Call technical support if this message is displayed again. Prepare for technical support by printing a copy of the Self-Test/Service parameters:
  - Press **SERVICE** key. The **SERVICE MENU** will be displayed.
  - Select **PRINT SELF-TEST AND DEFAULT SETTINGS** option.
  - Select **SELF-TEST DATA** and press **ENTER** and the service parameters will be printed.

Remove Capillary:



- If the testing capillary has been left in the measurement chamber after testing a sample the message above will be displayed.
- Remove the testing capillary before running a new test.

APPENDIX 5:
Semen Sample Preparation

EQUIPMENT REQUIRED:

- Extender
- Dispenser
- 10 ml Plastic Container
- Pipette
- SQA-Vp capillary
- Heater – set to 40^{0}C (for 37^{0}C sample heating)

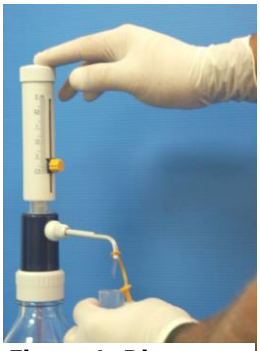


Figure 1: Dispenser

FRESH SEMEN SAMPLES:

- Place 2 ml aliquots of extender into the 10 ml sample containers provided in the test kit (figure 1)
- Pre-heat the extender to 37^{0}C / 98.6^{0}F for **4 minutes** in the SQA-Vp heater.
- Place SQA-Vp testing capillaries in the heater and pre-warm to 37^{0}C / 98.6^{0}F for at least **4 minutes.**
- Do not pre-heat the fresh sample!

Figure 2: Aspirate the required sample volume

SAMPLE PREPARATION

1. Mix the entire semen sample thoroughly.
2. Extract exactly the volume of semen specified by the SQA-Vp onscreen instructions using a pipette (Figure 2).
3. Wipe the tip of the pipette to remove any excess semen.
4. Add the semen from the pipette to the pre-warmed extender in the 10 ml plastic container (Figure 3).
5. Close the plastic container and gently but thoroughly mix the sample for 10-20 seconds (Figure 4).
6. The sample is now ready for testing. Fill a pre-heated SQA-Vp testing capillary with the semen sample.

NOTE: DO NOT PRE-HEAT the **FRESH** semen sample (pre-heat only the extender and testing capillary) or motility and throughput will be impacted!

Figure 3: Add semen to the extender

EXTENDED SEMEN SAMPLES:

1. Mix the extended semen in its original package.
2. Transfer 2 ml of sample into a 10 ml plastic container.
3. Pre-heat the sample to 37^{0}C/98.6^{0}F for **4 minutes**
4. Place SQA-Vp testing capillaries in the heater and pre-warm to 37^{0}C / 98.6^{0}F for at least **4 minutes**
5. Gently and thoroughly mix the sample for 20 to 30 seconds
6. The sample is now ready for testing.
7. Fill a pre-heated SQA-Vp testing capillary with the semen sample and test immediately.

NOTE: Set the SQA-Vp heating device to 40C to obtain 37C heating of the testing capillary and extender.

NOTE: **PRE-HEAT** the **EXTENDED** semen sample for **ONLY 4 minutes** – do not heat for more or the sample may agglutinate and motility readings will be impacted!

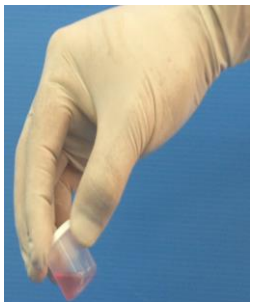
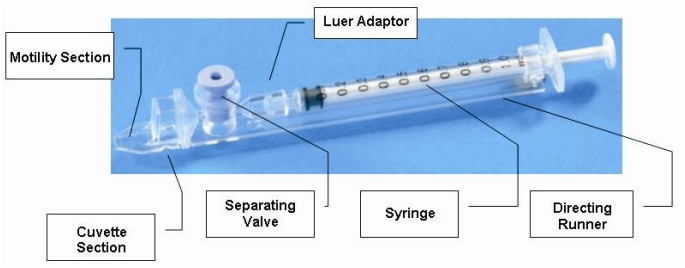


Figure 4: Thoroughly mix the sample.

APPENDIX 6:
Capillary Filling Instructions



1. Push the syringe piston in fully. Place only the thin part of the capillary into the bottom of the diluted sample (Figure 1).
2. Placing two fingers below the piston head pull the piston back slowly while keeping the tip of the capillary well below the sample level and surface bubbles (Figure 1).
3. Continue to aspirate the sample until it appears in syringe – hold in a vertical position and check to see that the sample has completely filled the thin section and the cuvette section and appears in the syringe (Figure 2).
4. Tap on the syringe to make sure there are no air bubbles in the sample.
5. Quickly and thoroughly wipe both the top and bottom of the outer surface of the capillary with a tissue. (Figure 3). Visually confirm that the capillary chambers are still full after wiping - a meniscus will be visible in the thin section of the capillary if sample has been lost due to wiping. If this is seen, push very slightly on the piston to re-fill the thin capillary section.
6. Slowly and carefully push-in the blue valve until it is level with the plastic. The capillary is now ready for testing (Figure 4).
7. Insert the capillary into the SQA-Vp (Figure 5)

Figure 1

Figure 2

Figure 3

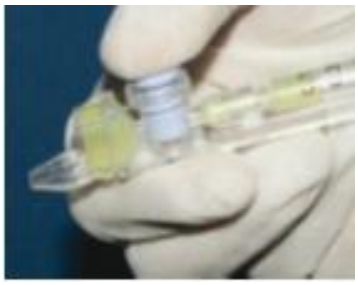


Figure 4

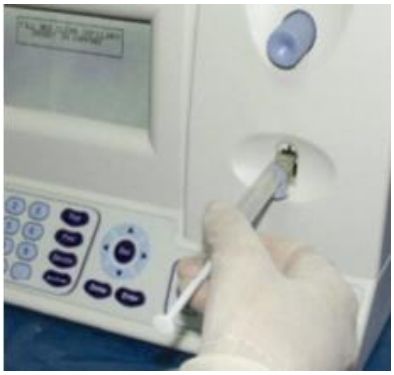


Figure 5

APPENDIX 7:
SQA-Vp Cleaning Instructions

When to clean:

Daily or after every 25 tests
If the system fails **SELF TEST**

Cleaning kit components:

Blue Dot capillaries (fig 1)
Sponge-tipped drying capillaries (fig 2)
Cleaning brush-wooden-handled (fig 4)
Cleaning fluid

CLEANING: STEP 1

1. **TURN OFF** the SQA-Vp.
2. Select a **BLUE DOT** fibrous material capillary.
   - Moisten with **ONE** drop of cleaning fluid, shaking off excess fluid.
   - Insert into the measurement compartment - fibrous material facing DOWN, and move back and forth a few times.
   - Insert a sponge-tipped drying capillary into the measurement compartment and move back and forth a couple of times to dry the chamber. (fig 3)

CLEANING: STEP II

Use the wooden-handled cleaning brush:

1. Insert the brush (bristle-side down) fully into the upper portion of the measurement chamber. (fig 5)
2. Pull the brush out of the chamber while sweeping or "dusting off" the LED (you will feel a step or shelf at the back and top of the chamber – this is the top of the LED). (fig 6)
3. Switch SQA-Vp unit **ON** and observe self-test results. The SQA-Vp should now PASS the self-test. If not, repeat cleaning procedure with the brush.

Fig 1

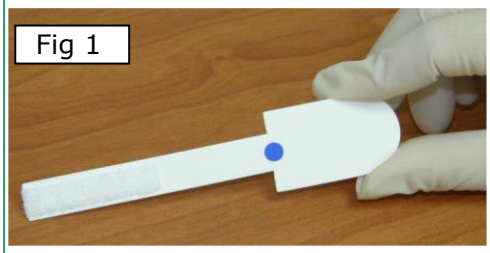


Fig 2

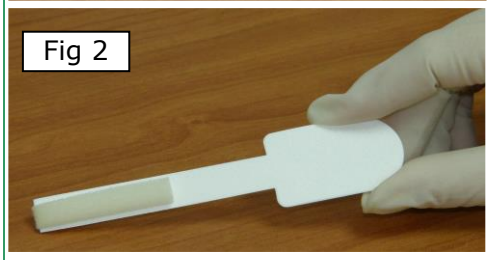


Fig 3

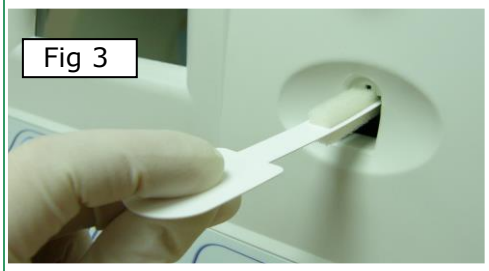


Fig 4

Fig 5

Fig 6

Appendix 8:
Capillary Washing Instructions
(For animal applications ONLY)

Both testing capillaries and 10ml sample collection cups can be washed and re-used up to 5 times by following this EASY procedure:

Testing Capillary

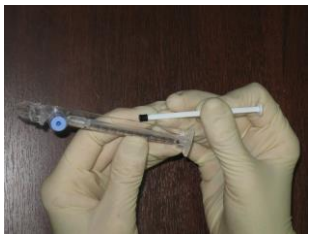
Reposition the blue valve with the jig

Step 1 After running a test:

- Use the white capillary jig to re-position the blue capillary valve
- Expel semen by pumping the plunger a couple of times
- Soak the testing capillary in tap water until ready to wash

Step 2 Set-up: Fill with 1 liter/2 quarts of solution as follows:

- Bowl #1: Tap water (marked "TAP WATER")
- Bowl #2: Distilled water (marked "DISTILLED WATER")
- Bowl #3: Isopropyl Alcohol 70% - 100%



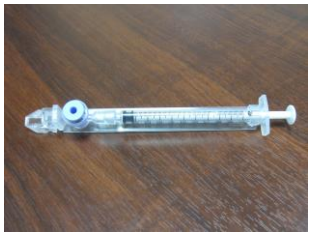
Remove the plunger

Step 3: Expel all liquid from the testing capillary:

- Pump the syringe plunger a couple of times to expel all remaining liquids.

Step 4: Capillary Washing:

- **Bowl #1 Tap Water:** Completely fill each capillary with tap water. Expel the solution into a hazardous waste container. **Repeat 2 times** then go to Bowl 2.
- **Bowl #2 Distilled Water**: Completely fill each capillary with distilled water. Expel the solution into a hazardous waste container. **Repeat 2 times** then go to Bowl 3.
- **Bowl #3 Isopropyl Alcohol**: Completely fill each capillary with isopropyl alcohol and expel the solution into a hazardous waste container. **Repeat 2 times.**
- Remove the plunger from the syringe after isopropyl washing.



Reassemble the capillary

Step 5: Drying the Capillaries:

- Place the capillaries on a flat surface and dry overnight or place in a low heat oven for a few hours until they are completely dry.

Step 6: Final Preparation/Inspection:

- Replace the plunger into the syringe.
- Inspect the capillary for cracks (throw away) or semen (re-wash).
- Note the number of washings by making a dot on the capillary with a water proof marker after each washing cycle.

10 ml sample collection cups

Washing – Please refer to Step 4 Capillary Washing and follow the same process for washing in the solutions in bowls #1; #2 and #3. Turn upside down on absorbent paper to dry overnight.

Appendix 9:
The Visualization System and Sperm Immobilization

The SQA-Vp Visualization System is for analyzing and viewing semen samples. Both the SQA-Vp testing capillary and a standard slide can be read in the visualization system. Pictures and videos of samples viewed in the visualization system can be captured and stored in P-Sperm. The system:

- Operates via control knobs to set focus, brightness, contrast and color, and via the keypad zoom, illumination, and monitor on/off functions.
- Has a magnification range: x300 to x500.

Operating Instructions:

Slide Preparation For QwikClick Morphology Assessment (QCA)

The sperm cells must be immobilized but not overheated or droplets may burst. Follow the instructions below to ensure a quality reading:

- Place slides in the slide heater set to 55°C. Leave slides on the heater a minimum of 4 minutes.
- Place 10 µl of semen in the center but close to the edge of the slide.
- Do not leave on the heater more than 4 seconds.
- Use only a standard slide with a 22mm x 22mm cover-slip.
- Load the prepared slide into the SQA-Vp slide adaptor and insert into the visualization compartment of the SQA-Vp.

Testing Capillary Preparation:

- Insert a testing capillary filled with semen into the visualization compartment of the SQA-Vp.

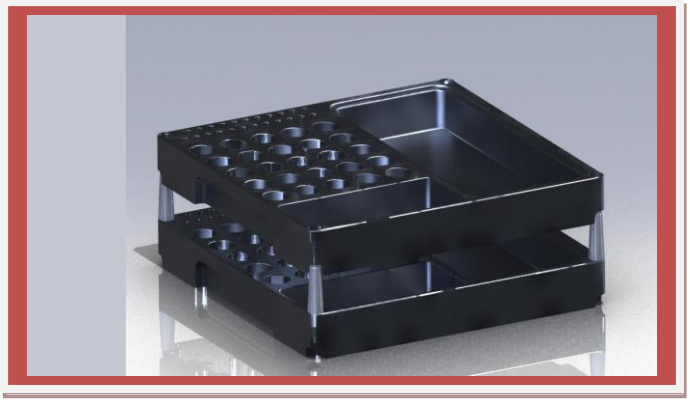
Focusing the Visualization System:

1. The video display illuminates when the SQA-Vp is turned on.
2. To ensure that the visualization system is working properly prior to use:
   a. Press the **HIGH ILLUMINATION** key multiple times to achieve the maximum level setting.
   b. Turn **BRIGHTNESS**, **CONTRAST** and **COLOR** buttons all the way counterclockwise.
   c. Turn **FOCUS** knob fully clockwise.
3. Use **ZOOM IN** for maximum magnification (x500) or **ZOOM OUT** for minimum magnification (x300).
4. Insert the semen sample into the visualization chamber.
5. Turn the **BRIGHTNESS** knob clockwise until the video screen just begins to lighten-up.
6. Turn the **FOCUS** knob counter-clockwise until the image is in focus.
7. Adjust **CONTRAST, COLOR, BRIGHTNESS, FOCUS** and object **ILLUMINATION** controls for optimal image quality.

NOTE: Use caution when turning the focus knob. If resistance is felt it is at the maximum (or minimum) position. Forcing this knob beyond the stopping point may damage the focus system.

Appendix 10: Heating Devices for Sample Testing

For A-TECH Sperm Quality Analyzers with Heating Requirements



Safety:

- Do not pre-fill the SQA testing capillaries with semen prior to heating.
- Wipe any spilled material with a damp cloth after cooling the stages. Do not use chemical cleaning agents.
- Do not stack more than four plates at a time.

Instructions:

- Place 25, **empty** pipette tips in the small holes.
- Place 25 **empty** testing capillaries into the large well.
- Place 25, 10ml plastic cups with caps into the larger holes.
  - FRESH TESTING: Pre-fill each 10ml cup with **2 ml extender**. Close the cup with cap provided. Allow extender to reach 37°C (this takes about 10 minutes on a WARMED stage – this is why 2 stages are provided – one for testing, one for back-up to be rotated so that warmed supplies are always ready for testing)
  - EXTENDED TESTING: Pre-heat the cups – fill with EXTENDED semen just prior to testing. Do not leave the EXTENDED sample in the heater for more than 4 minutes or it will begin to agglutinate.
  - Cap the cups to prevent any evaporation prior to testing.
- Free well – place samples awaiting testing or optional supplies in the free well.

Heating:

- Place un-stacked individual heating stages **WITH SUPPLIES and EXTENDER** on a standard laboratory heating plate set to maintain 37°C for both samples and testing capillaries (this must be calibrated as the heating plate must be set HIGHER to maintain the appropriate sample temperature).
- Allow a one-time initial warm-up of up to 45 minutes for the stages. They will take longer to heat up initially than during the testing period. During the initial heating phase, do not place ANY semen in the wells.
- Once the stages are up to temperature, testing can begin.
- Rotate between the stages - #1 stage for testing; #2 stage for next round so that there is always an operating stage that is warm and back-up stage that is being warmed.

Appendix 11: Glossary of Terms

	Terms	Definition
Menu	SN	Serial Number of the SQA-Vp
	DATE/TIME	The date and time the test was performed
	SAMPLE #	The number assigned to the semen sample
	BOAR ID	The identifying number of the boar being tested
Test Results	CONC.	Total sperm concentration expressed in millions/ml
	MSC	Motile sperm concentration expressed in millions/ml
	MOTILITY %	Percentage of motile spermatozoa: Motile Sperm Concentration divided by Total Sperm Concentration expressed as a %.
	GRADING MOTILITY	Motility Grade score (0-5) is related to progressiveness of motile sperm. Score 0 corresponds to absence of progressive motility whereas score 5 corresponds to the maximum progressive motility.
	MORPHOLOGY	Percentage of morphologically normal spermatozoa.
	SPERM #	The total number of sperm cells per ejaculate volume (Fresh samples) or per semen volume (Extended samples).
	MOT. SPERM	The total number of motile sperm cells per ejaculate volume (Fresh samples) or per semen volume (Extended samples).
Dosing Calculations	EXTENDER VOLUME	The amount (ml) of extender to add to the semen in order to produce the desired number of doses.
	TOTAL VOLUME	Semen volume (neat ejaculate + primary extender volume) + extender volume (ml)
	NUMBER OF DOSES	The total number of doses that will be produced based upon the users set-up parameters
Dosing Set-up	DOSING METHOD	Options to dose by: Total Sperm #; Motile Sperm #; or Morphologically Normal Sperm # in an A.I. dose
	DOSE VOLUME	The desired A.I. dose volume (ml): 80/100/other
	TARGET # SPERM	The number of spermatozoa according to the dosing method desired in an A.I. dose (Billion/Dose)
	MOTILITY CUTOFF	The lowest acceptable level of Motility
	MOTILITY GRADE CUTOFF	The lowest acceptable level of Motility Grade

Appendix 12: SQA-Vp System Specifications

Dimensions: 40 x 30 x 15 cm

Weight: 4 kg

AC power supply: 100 to 250 VAC, 50/60 Hz, 10 VA

Measurement Compartment

- **Sources of radiant energy** - two 880 nm LEDs for motility and spectrophotometry channels
- **Detector system** – 2 photo detectors - Motility and Optical Density

Visualization Compartment

- Green LED illumination system
- CCD, 350 TV lines
- Objective lens: Standard, x20
- Signal Output: PAL standard
- Zoom system for smooth magnification transition from x300 to x500
- Focus regulator

Display(s)

- Operational backlight LCD (16 lines x 40 characters)
- Video backlight LCD (8 x 10 cm)

Printer

- Built-in, Dot Matrix
- Non-thermostatic narrow paper with 20 characters per line (Citizen)
- Ribbon cassette (Citizen)

Keypad

- **Operational keys:** ON/OFF, TEST, PRINT, SERVICE, ARCHIVE, DELETE, ENTER, four cursor buttons, ESC, numeric buttons (0-9)
- **Video control keys**: ON/OFF, ZOOM IN/OUT, ILLUMINATION HIGH/LOW, and MONITOR ON/OFF

Front Panel

- Built-in printer
- Visualization compartment
- LCD video display and controls
- Focus knob
- LCD operational display
- Measurement compartment
- Multi-button keypad

Rear/Side Panel

- Power connector with fuse-holder (fuse 250V, 1A)
- Video connector
- RS232 cable outlet
- I-Button port (side panel)

Specimen Testing Supplies

- **Measurement capillary:** Disposable, multi-use plastic, positive displacement testing capillary (purchase from manufacturer).
- **Standard lab slide:** 76 x 25.6 mm, 22 x 22 mm cover-slip.
- **I-Button:** Required to run tests (purchase from manufacturer)

Archive Capacity

- 500 test records in each (Test Results and Control) archive

Operating System

- **Control:** Keypad
- **Analysis Time:** 45 seconds for a Normal Test
- **Software:** Resides on flash memory and drives all man-machine interface functions, runs algorithms for test measurements and operational screens. System can be upgraded from a PC CD-ROM.
- **Sample Testing Temperature:** 37^0C (98.6^0F).
- **Motility channel input signal:** Analog, up to 5V.
- **Spectrophotometer channel input signal:** Modulated (1 kHz) analog, up to 5V.

Quality Control

- **Internal:** Electronic Self-Test and Auto-Calibration.
- **External:** QwikCheck-beads control media.

PC Compatibility

Minimum requirements for P-Sperm™ software

- **PC:** 1 GHz processor, Pentium 3
- **RAM:** 256 MB
- **AGP-video display card** with at least 16 MB of RAM memory
- **Video color**: At least 16 bit (65,535)
- **CD ROM drive**
- **40 GB free hard disk space** for image capturing and storage
- **Video resolution:** Minimum 640 x 480

Operating system compatibility: Windows 2000, Windows XP, DirectX, DivX

- **Ports:** One serial; two USB ports
- **Monitor:** 15" color

Additional Software (supplied with system)

- **P-Sperm software:** Real time visualization interface between PC and SQA-Vp visualization system, data transfer, video/picture capture and archive.

Operational Temperature and Humidity

- System is operational at 15-38°C.
- *NOTE*: SQA-Vp operates in a wide range of ambient temperatures however the system is calibrated to measure semen samples at 37°C (98.6°F).
- System is fully operational at up to 80% humidity and 31°C.

Maintenance Schedule

- Cleaning daily and after every 50 tests (refer to User Guide – "Cleaning Instructions").

Manufacturer Recommendations

- Operate the SQA-Vp away from devices that may cause electronic noise (cell phones) or other devices causing vibrations such as centrifuges.
- Turn system **OFF** at the rear-panel when not in use for extended period of time.
- Semen is considered a biologically hazardous material and is subject to individual laboratory protocols for handling such materials.

Factory Default Settings:

Date format: DD/MM/YY

Time/Date: Manufacturer's local time/date

Sample Type: FRESH

Automatically print: YES

Display test results: YES

Automatically send to PC: YES

Extender transparent: YES

Appendix 13: Product Performance Data

Abbreviations:
CONC: Sperm Concentration
CV: Coefficient of Variation
M/ml: Million per milliliter

Performance Data Summary:
The performance of the SQA-Vp system for boar semen analysis is summarized in the text, tables and graphs below. Sperm concentration measurements are expressed as 10^6 sperm cells per milliliter (M/ml). Motility is expressed as a percent (%). Unless otherwise noted all testing was performed using fresh and extended boar semen samples.

Calibration:
Each SQA-Vp is biologically calibrated against two reference systems at Medical Electronic System's laboratory using boar semen.

Dynamic Range:

Sample Type	Conc. M/ml	Motility %	Grading Motility	Morphology %
Fresh	0-1500	0-100	0-5	0-100
Extended	0-500	0-100	0-5	-

Precision and accuracy established against a known target (Latex beads)

Background: The precision and accuracy of the SQA-V were compared to a known target value using commercially available latex beads of two concentrations. Latex beads are used commercially to validate automated sperm counting systems. The beads were run on the SQA-V in the same manner semen samples are run on the system.

Limitations of method:
Latex beads cannot:
- Measure sperm motility or morphology
- Correct for inaccurate chamber depths or technician errors

Methodology:
A total of 320 latex bead samples were tested on ten SQA-V systems. The precision of the SQA-V is demonstrated in Table 1. SQA-V concentration readings were compared to the established target values +/- the acceptable range for the latex beads (Fig. 1 & 2).

Accu-beads® published ranges (*Hemacytometer*):
- Vial #1: 47 +/- 7.0 M/ml
- Vial #2: 24 +/- 3.4 M/ml

Table 1: Precision

SQA-V	Latex-beads	CV %
Intra-device Variability	**High 47± 7.0 M/ml**	**≤ 0.01**
	Low 24 ± 3.4 M/ml	**≤ 0.01**
Inter-device Variability	**High 47± 7.0 M/ml**	**≤ 2.00**
	Low 24 ± 3.4 M/ml	**≤ 2.50**

Fig. 1. Accuracy: High Level Control

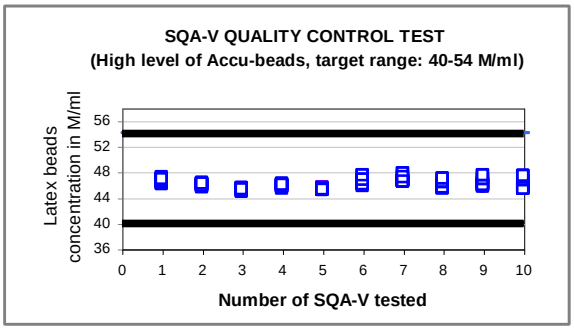
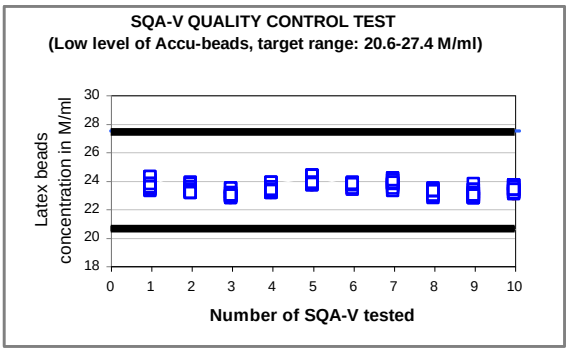


Fig. 2. Accuracy: Low Level Control



Conclusions:

The **CONTROL** mode software of the SQA-Vp (pig) device is exactly the same as the SQA-V (human) system. Both systems also have the same hardware platform. Therefore, the accuracy and precision results obtained on the CONTROL mode of the SQA-V will be the same as that of the CONTROL mode of the SQA-Vp.

Sensitivity, specificity, precision and correlation to manual methods established in MES laboratories and field clinical trials using boar semen samples

Clinical claims:

Sensitivity

- Concentration: 90%
- Motility: 75%
- Grading Motility: 60%
- Morphology: 60%

Specificity

- Concentration: 90%
- Motility: 75%
- Grading Motility: 80%
- Morphology: 80%

Precision (CVs)

- Conc.: 3%
- Motility: 5%
- Morphology: 5%

Correlation to Manual Method:

- Concentration: 0.9
- Motility: 0.8
- Morphology: 0.7

Table 2: Sensitivity/Specificity

SQA-Vp vs. Microscope	Sensitivity, %	Specificity, %
Sperm Concentration	**91.7**	**95.2**
Motility	**80.0**	**79.0**
Grading Motility	**66.7**	**89.1**
Morphology	**66.7**	**84.0**

Table 3: Precision: SQA-Vp intra-device variability (CV, %)

Semen Variable	CV, %
Sperm Concentration M/ml	**2.1**
Motility, %	**4.0**
Morphology, %	**3.6**

Notes:

- Sensitivity and specificity **claims** are lower than actual values noted (Table 2).
- Precision CV **claims** are higher (lower precision) than actual values noted (Table 3).
- Correlation to Manual Method **claims** are less than actual correlations noted (Table 4).

Method comparison:

SQA-Vp was compared to the microscope based on WHO'99 manual guidelines. The SQA-Vp automated readings of the sperm concentration, motility and morphology were compared to microscopic results. A Makler chamber was used according to manufacturer's instructions for manual sperm concentration measurements. A standard slide and P-Sperm software were used to assess manual motility. The stained slides were used for the manual morphology examination. The protocols were based on WHO'99 manual and MES guidelines. The alpha-site clinical trials were conducted at the Lahav farm. A total of 58 fresh and extended semen samples were analyzed.

Table 4: Correlation to manual method

Parameters	Correlation coefficients
Sperm Concentration, M/ml	**0.99**
Motility, %	**0.83**
Morphology, %	**0.71**

Analytical Specificity:

- To achieve analytical specificity a specific wave length of light which is maximally absorbed by sperm cells and minimally absorbed by other cells and seminal plasma is used.
- Low noise and high electronic resolution hardware components and compensation circuits ensure that analytical specificity is optimized.

Limitations of method:

Samples were assessed in duplicate on the automated SQA-Vp system and manually using a microscope. Statistical counting errors and intra-operator variability (subjectivity) may have affected the results of the study.

Performance parameters:

- Sensitivity and specificity were calculated using ROC analysis formulas. The cutoffs normally used for the sperm concentration, motility and morphology were used for calculation of sensitivity and specificity (Table 2).
- Precision of the SQA-Vp device was estimated by calculation of the intra-device coefficients of variation (CV) of the duplicate measurements (Table 3). CV is calculated according to the formula:

  CV = SD / MEAN x 100

  The lower CV, the higher precision of the method.
- Correlation to manual method was established by calculation of correlation coefficients (Table 4, Fig. 3-5).

Fig. 3: Method comparison: Regression plot of SQA-Vp Sperm Concentration in boar semen vs. manual results

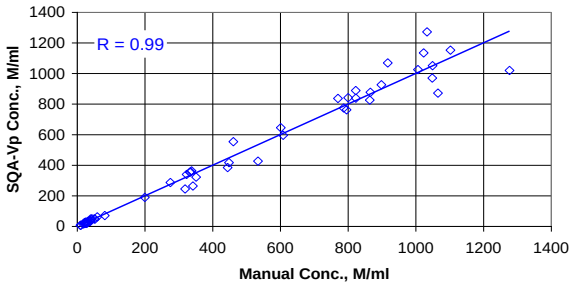


Fig. 4: Method comparison: Regression plot of SQA-Vp Motility in boar semen vs. manual results

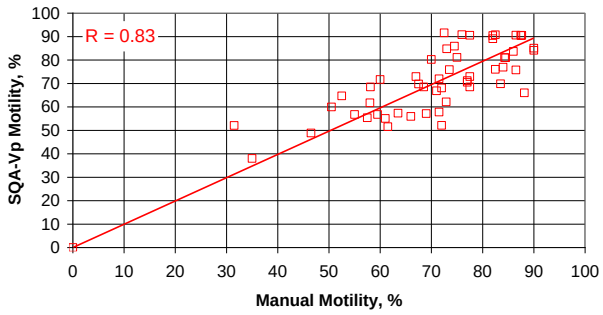
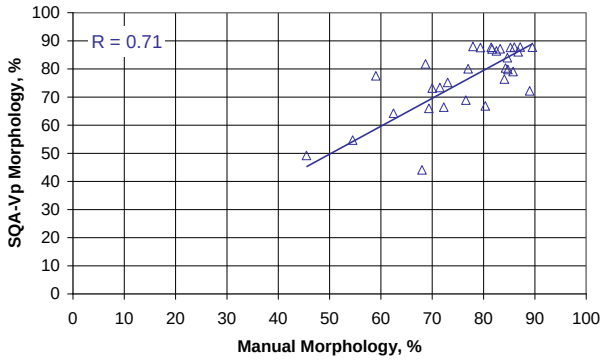


Fig. 5: Method comparison: Regression plot of SQA-Vp Morphology in fresh boar semen vs. manual results



Conclusions:

- The sensitivity, specificity and correlation of the SQA-Vp to the manual method are very high. Therefore the instrument can replace the manual method for assessing, dosing and extending pig semen.
- The SQA-Vp is precise and reports accurate results with low coefficients of variation for the semen variables assessed (<6%).