

QwikCheck *Gold* Equine Sperm Analyzer User Guide

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Section 1: Overview

The QwikCheck™ Gold Equine sperm quality analyzer is used to test and report the quality of FRESH, EXTENDED and COOLED equine semen. The following semen parameters are reported:

Reported Semen Parameters				
Parameter	On Screen	Normal Reference Values	Raw	Extended / Cooled
Sperm Concentration, M/ml	CONC.	100-800*	√	√**
Motility, %	MOTILITY	>50*	√	√**
Progressive Motility, %	PROG. MOT.	>50*	√	√**
Morphology, %	MORPHOLOGY	50*	√	
Velocity, mic/sec	VELOC.		√	√
Total Sperm Number, Bil.	SPERM #		√	
Total Motile Sperm, Bil.	MOTILE SPERM		√	
Total Progressively Motile Sperm, Bil.	PROG. SPERM		√	

*Reference values for equine raw semen: <http://www.vetstream.com/equis/Content/LabTest/lab00123.asp>

**These results are reported in ranges.

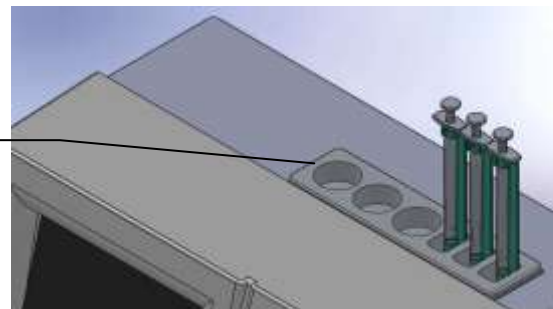
Section 2: System Overview

**QwikCheck™
Gold Equine
Sperm Quality
Analyzer**



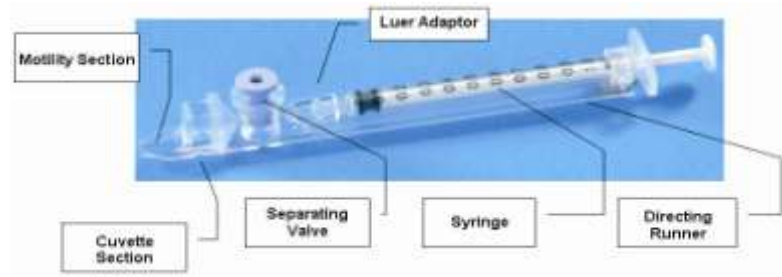
**QwikCheck™
Gold Equine
on-board sample
heating unit**

- On-board sample heating unit will pre-heat:
- 3 testing capillaries
 - 3 – 10ml sample containers





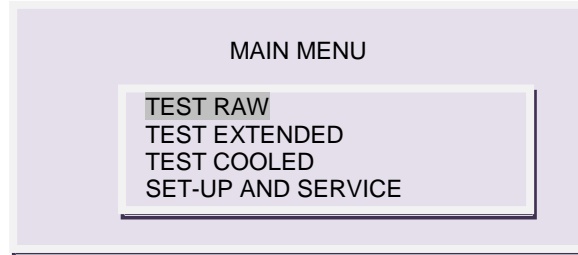
**QwikCheck™
Gold Equine
testing capillary**



- Plastic, multi-use (10X for animal use only), disposable.
- Refer to the appendix section for instructions on how to use/wash the testing capillary.

Section 3: Operating the QwikCheck GOLD Equine

- Turn on the main switch on the rear panel of the QwikCheck GOLD Equine.
- Press the On/Off key on the keypad.
- Wait for the system to complete pre-warming, stabilization and self-testing.
- The number of tests remaining will be reported upon completion of the self-test.
- Press ENTER to view the **MAIN MENU** and select one of the available options:

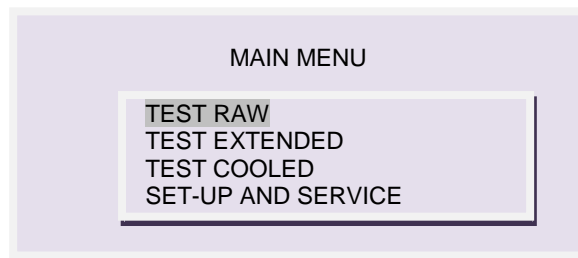


When using the system for the first time please:

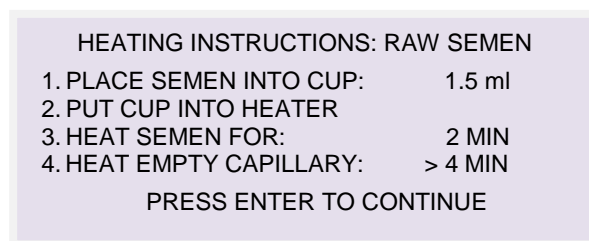
- To load I-Button tests go to: **MAIN MENU > SET-UP AND SERVICE > LOAD I-BUTTON TESTS**. Follow the on-screen instructions.
- **SET-UP AND SERVICE**: Access all system set-up defaults, i-button loading and service screens. For system defaults go to: **MAIN MENU > SET-UP AND SERVICE > DEFAULT SETTINGS (section 5 of this manual)**

Section 4: Sample Testing Running Raw Samples

- Select **TEST RAW** from the MAIN MENU to open the screen displayed below.



- Follow the screen instructions for heating the raw semen and the testing capillary:



NOTE: Load I-button tests and set system defaults **PRIOR** to testing (see Sections 5 and 6 for full instructions)

NOTE: Raw samples need to be preheated for 2 minutes prior to testing.



- Press ENTER to view the screen below & start the 2 minute timer for semen warming:

ENTER SAMPLE DATA:

DATE: 01/04/12	TIME: 08 :15
STALLION ID:	123
SAMPLE #:	1
SEMEN VOLUME:	60.0 ml
RICH FRACTION COLLECTED?	NO / YES

- Enter the following STALLION information using the keypad:
 - **Stallion ID:** Up to 8 digits
 - **Sample #:** Up to 10 digits
 - **Semen Volume:** (Gel free): Up to 3 integers and 1 decimal point.
 - **Rich Fraction Collected?** Select YES if only the rich sperm fraction of the ejaculate is collected (i.e. via an Artificial Vagina). No is the automatic default.
- The screen below will open automatically:

WAIT FOR BEEP TO BEGIN TESTING

TIME REMAINING: 01:20

AUTOCALIBRATION – DO NOT TOUCH UNIT

- When the sample pre-heating controlled by the timer is over, the sample preparation screen will open.
- Follow the instructions on the screen.
- Wait for a “beep” and the instruction: **INSERT CAPILLARY INTO CHAMBER** as seen below will be displayed:

SAMPLE PREPARATION:

1. MIX HEATED SEMEN
2. FILL AND WIPE HEATED CAPILLARY
3. INSERT CAPILLARY INTO CHAMBER

- Insert the testing capillary into the QwikCheck Gold. Testing will begin automatically:

PLEASE WAIT
HEATING CAPILLARY

TESTING

DO NOT MOVE CAPILLARY OR
OPERATE DEVICE DURING TESTING

- After ~50 seconds, a “beep” will indicate that testing is finished and the results will be displayed:

RAW SEMEN - STALLION ID: XXXXXXXXXXXX

CONC. 216.8 M/ml	MORPH. 65.6 %
MOTILITY 58.8 %	VELOC. 93 mic/sec
PROG. MOT. 49.8 %	
TOTALS (SEMEN VOL: 60.0 ml)	
SPERM 13.01 Bil	PROG. 6.48 Bil
MOTILE 7.65 Bil	

DOSING? **DEFAULT** / MANUAL / NONE

**AI DOSING**

NOTE:
This screen
is skipped if
the
DOSING:
DEFAULT is
selected.

- At the bottom of the screen above, select the following **DOSING** options:
 - **NONE:** Select NONE to go directly to the main menu without dosing- the results will be printed if the default is set to auto print or press PRINT .
 - **DEFAULT:** Select DEFAULT to dose the sample according to the manufacturer's default settings as seen in the screen below. If these setting are acceptable, press **ENTER**; if they are not, press **ESC** to go to the **MANUAL** dosing screen:

```

      DEFAULT DOSING SETTINGS
      DOSING BY:                PROG. MOTILE
      MINIMUM # SPERM PER DOSE <M>:    1000
      MAXIMUM DOSE VOLUME <ml>:       80
      FINAL CONCENTRATION TARGET: 25 - 50 M/ml

      PRESS ENTER TO CONTINUE
      PRESS ESC TO GO TO MANUAL DOSING
    
```

- **MANUAL:** Select this option to enter customized dosing settings.

```

      SET-UP: MANUAL DOSING
      DOSING BY:  TOTAL/MOTILE/PROG. MOTILE
      MINIMUM # SPERM PER DOSE <M>:    1000
      MAXIMUM DOSE VOLUME <ml>:       50
      FINAL CONCENTRATION TARGET:
      MINIMUM                          25 M/ml
      MAXIMUM                          50 M/ml
    
```

- **For MANUAL dosing:** Enter the desired dosing settings into the screen shown above. Press ENTER and the AI dosing instructions will be automatically generated (the dosing settings will remain in the memory of the system until manually changed):
 - **DOSING BY:** Select the type of dosing preferred – Total # cells, Motile # cells or Progressively Motile # of sperm cells.
 - **MINIMUM # SPERM PER DOSE (M):** Enter the desired # of sperm cells per dose in millions. PLEASE NOTE: The number entered should correspond to the type of dosing (i.e. If dosing by Total # cells the number should be higher than if dosing by # Prog. Motile cells as there are LESS Prog. Motile Cells in an ejaculate).
 - **MAXIMUM DOSE VOLUME (ml):** Enter the maximum volume desired for an AI dose (in milliliters).
 - **TARGET FINAL CONCENTRATION (M/ml):** Enter the acceptable range (minimum and maximum) for Final Concentration of the AI dose (in M/ml).
- Press: ENTER to view dosing Instructions for the ENTIRE ejaculate or a single AI dose:

```

      AI DOSING INSTRUCTIONS:
      # DOSES:                6      1
      SEMEN VOLUME (ml):     60    10.0
      EXTENDER VOLUME (ml): 324    54.0
      TOTAL VOLUME (ml):    384    64.0
      # TOTAL (MOTILE/PR. MOTILE) SPERM PER DOSE (M): 1044
      FINAL CONCENTRATION (M/ml): 33.8
    
```

- If Raw semen Concentration is < 50 M/ml, dosing is skipped per the message below:

```

      THE SPERM CONCENTRATION
      IS TOO LOW [<50M/ml]
      CENTRIFUGATION REQUIRED FOR DOSING

      PRESS ESC: REFER TO TOTAL SPERM
      RESULTS FOR MANUAL DOSING

      PRESS ENTER FOR THE MAIN MENU
    
```



- If the semen quality is poor or there is an error in the manual dosing set-up parameters the following message will be displayed:

BASED ON THE SETTINGS OR SEMEN QUALITY,
 THE SYSTEM CANNOT CALCULATE DOSES.

 PRESS ESC: VIEW SAMPLE QUALITY
 AND/OR CHANGE THE SETTINGS
 BY SELECTING THE MANUAL DOSING OPTION

 PRESS ENTER FOR THE MAIN MENU

- Press the Print button on the keypad to print the semen analysis report and the dosing instructions (if the default is set to auto print, test results will print automatically).

Running Extended/Cooled Samples

- Definitions of EXTENDED vs. COOLED samples for the QwikCheck GOLD EQUINE:
 - **EXTENDED SEMEN** is RAW semen diluted with a commercial extender.
 - **COOLED SEMEN** is EXTENDED semen kept in cooling conditions.
- Select either **TEST EXTENDED** or **TEST COOLED** from the MAIN MENU:

MAIN MENU

 TEST RAW
 TEST EXTENDED
 TEST COOLED
 SET-UP AND SERVICE

- Follow the instructions on the screen below for heating the extended or cooled semen and testing capillary:

NOTE:
Preheat samples prior to testing:
EXTENDED: 2 minutes
COOLED: 4 minutes

HEATING INSTRUCTIONS: **EXTENDED** SEMEN

1. PLACE SEMEN INTO CUP:	1.5 ml
2. PUT CUP INTO HEATER	
3. HEAT SEMEN FOR:	2 MIN
4. HEAT EMPTY CAPILLARY:	> 4 MIN

PRESS ENTER TO CONTINUE

HEATING INSTRUCTIONS: **COOLED** SEMEN

1. PLACE SEMEN INTO CUP:	1.5 ml
2. PUT CUP INTO HEATER	
3. HEAT SEMEN FOR:	4 MIN
4. HEAT EMPTY CAPILLARY:	> 4 MIN

PRESS ENTER TO CONTINUE

- Press ENTER to start the automatic timer for semen warming. Enter the information requested in the next screen:

ENTER SAMPLE DATA: EXTENDED

DATE: 01/04/12	TIME: 08 :15
STALLION ID:	23
SAMPLE #:	12
SEMEN VOLUME:	30.5 ml

ENTER SAMPLE DATA: COOLED

DATE: 01/04/12	TIME: 08 :15
STALLION ID:	23
SAMPLE #:	12
SEMEN VOLUME:	30.5 ml
COOLING TIME:	24 hours

- **STALLION ID:** Enter up to 8 digits
- **SAMPLE #:** Enter up to 10 digits
- **SEMEN VOLUME:** Enter up to 3 integers and 1 decimal point.
- **COOLING TIME** (for cooled semen only): Enter the number of hours the sample was cooled (up to 2 integers).



- The screen below will open automatically:

WAIT FOR BEEP TO BEGIN TESTING
 TIME REMAINING: 01:20
 AUTOCALIBRATION – DO NOT TOUCH UNIT

- When the sample pre-heating is finished, a sample preparation screen will open.
- Follow the instructions on the screen and do not insert the capillary until instructed.
- Wait for a “beep” and the message: **INSERT CAPILLARY INTO CHAMBER** as seen below.

SAMPLE PREPARATION:

1. MIX HEATED SEMEN
2. FILL AND WIPE HEATED CAPILLARY
3. INSERT CAPILLARY INTO CHAMBER

- Insert the testing capillary into the QwikCheck Gold. Testing will begin automatically:

PLEASE WAIT
HEATING CAPILLARY

TESTING

DO NOT MOVE CAPILLARY OR
OPERATE DEVICE DURING TESTING

- After ~50 seconds, a “beep” will indicate that testing is finished and the results will be displayed:

EXTENDED / COOLED SAMPLE #: 12
 STALLION ID: 23
 SEMEN VOLUME: 14.5 ml
 CONC. >50 M/ml
 MOTILITY >50 %
 PROG. MOT. 20-40 %
 VELOC. 62 mic/sec
 # PROG. SPERM/DOSE: 725 M

Note: The Extended and Cooled test results are reported in ranges or as greater than a pre-defined cutoff value.

- If Concentration falls below 20 M/ml or Progressive Motility below 25%, the following message will be displayed:

EXTENDED / COOLED SAMPLE #: 12
 STALLION ID: 23

POOR QUALITY SAMPLE:

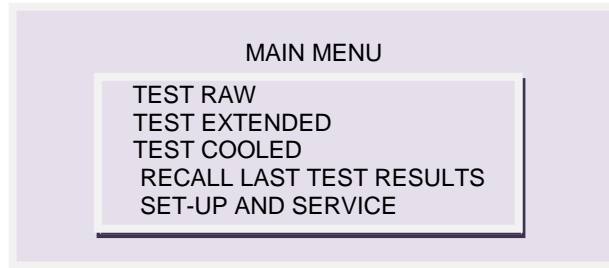
1. CONCENTRATION < 20 M/ml OR
2. PROG. MOTIITY < 25%

- Press the Print button on the QwikCheck GOLD keypad to print a semen analysis report (or wait for the test results to be printed automatically if this default is set).



MAIN MENU:

The MAIN MENU is first displayed when the system is turned on (after calibration). After each test is run, an option to RECALL LAST TEST RESULTS is also displayed in the MAIN MENU:



- **TEST RAW, TEST EXTENDED** and **TEST COOLED** options open the screens for testing the different sample types.
- **RECALL LAST TEST RESULTS:** View the last test results run.
- **SET-UP AND SERVICE:** Default settings and the other features (See Section 5 for details).

Section 5: SET-UP AND SERVICE

Select **SET-UP AND SERVICE** and the following options are available:

LOAD I-BUTTON TESTS

TRANSFER ARCHIVE

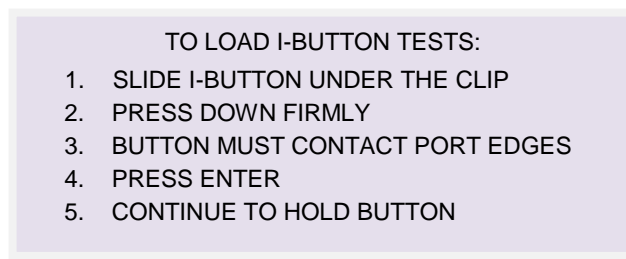
DEFAULT SETTINGS

SERVICE SCREENS

SERVICE PERSONNEL

LOAD I-BUTTON TESTS: The QwikCheck GOLD Equine requires tests be “loaded” using an I-Button. To do this:

- Select: MAIN MENU > SET-UP AND SERVICE > LOAD I-BUTTON TESTS
- Press: ENTER and the screen below will be displayed.
- Follow the instructions on this screen:



- Place the I-Button into the I-Button port by placing it under the spring to be held firmly in place. Make sure it touches both the internal surface and the edges of the port.
- Continue to hold the I-Button in place until the #TESTS ADDED and the TOTAL # OF TESTS REMAINING are displayed on the screen.
- The user is informed when :
 - Less than 10 tests are available.
 - No more tests are available.

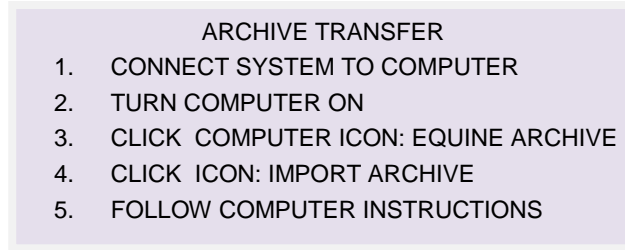
**TRANSFER ARCHIVE: Transfer test results to a PC (computer) Excel file.**

The QwikCheck GOLD Equine archive (which stores ~ 500 tests in memory) can be transferred to a computer Excel file. First, the Equine Archive software provided with the QwikCheck GOLD EQUINE needs to be loaded on the PC by following the instructions provided with the software.

Once the archive transfer software is loaded on a PC, the QwikCheck GOLD EQUINE archive can be transferred by following the instructions below (and on-screen).

TRANSFER ALL TESTS – not during testing

- Select: **MAIN MENU > SET-UP AND SERVICE > TRANSFER ARCHIVE** and press **ENTER** to view the screen below:

**TRANSFER INDIVIDUAL TESTS – on-line during testing**

- Connect the QwikCheck GOLD EQUINE system to the computer that is loaded with the EQUINE ARCHIVE software. Make sure a printer is connected to the computer.
- Set the QwikCheck GOLD EQUINE defaults. Go to: **MAIN MENU > SET-UP AND SERVICE > DEFAULT SETTINGS** and press **ENTER**.
- The default setting must be set to the following:
 - **AUTO PRINTING: YES**
 - **# LABELS TO PRINT 1**
- Test results will now be transferred to the computer EXCEL file after each test. And, each test results will be printed individually when the test/dosing process is completed.

DEFAULT SETTINGS: To set the system and sample defaults go to:

MAIN MENU > SET-UP AND SERVICE > DEFAULT SETTINGS and enter:

- **DATE/TIME:** Enter local date and time.
- **DATE FORMAT:** Select the format DD/MM/YY or MM/DD/YY using the right/left arrows on the keypad.
- **AUTO PRINTING YES/NO:** Select YES/NO to automatically print test results to an external label printer (recommended setting: YES).
- **# LABELS TO PRINT 1/2:** Select the number of labels required.
- **CONC STD:** Select "1" for Neubauer (default) or "2" for Nucleocounter.

SERVICE SCREENS: To view three service screens go to:

MAIN MENU > SET-UP AND SERVICE > SERVICE SCREENS

- **SERVICE DATA SCREEN:** Service information for technical support.
- **SELF-TEST DATA SCREEN:** Internal system information. Two types of data are displayed, (1) Self-Test or (2) Internal Data (which shows values after running a sample).
- **ALGORITHM DATA SCREEN:** This screen displays algorithm calculations.

SERVICE PERSONNEL: For technical service (requires a password).**NOTE:**

The Archive Transfer software will only import a test once. It will 'look' for duplicates and eliminate them from the transfer process.

NOTE:

Be sure to set the **# LABELS TO PRINT** to '1' or duplicate records will be imported.



Section 6: Troubleshooting

Failed Self-Test:

FAILED SELF-TEST
TURN OFF SWITCH ON REAR PANEL
CLEAN TESTING CHAMBER
THEN RESTART SYSTEM

IF SYSTEM FAILS AGAIN,
CALL TECHNICAL SUPPORT

- Check to see that there is no testing capillary in the measurement compartment.
- Remove the QwikCheck GOLD Equine from sources of electronic noise and/or vibration (centrifuge, etc.).
- Clean the measurement compartment (refer to the Appendix section).
- Reboot the QwikCheck GOLD Equine 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 the system warming-up and Self-Test.
- Call for technical support if this message is displayed again. Prepare for technical support by printing a copy of the QwikCheck GOLD Equine Self-Test/Service Data:
 - Press **SERVICE** key and open SERVICE SCREENS > SCREEN #2: SELF-TEST DATA.
 - Press **PRINT** button and the Service Data will be printed by the label printer.

Electronic Noise:

ELECTRONIC NOISE
TURN OFF MAIN SWITCH ON REAR PANEL
REACTIVATE UNIT

IF PROBLEM PERSISTS,
CALL FOR TECHNICAL SUPPORT

- Check that there is no testing capillary in the measurement compartment.
- Remove QwikCheck GOLD Equine from sources of electronic noise and/or vibration (centrifuge, etc.).
- Clean the measurement compartment (refer to the Appendix section) and then:
 - Activate **MAIN MENU** and re-run the test.
- If this message is displayed again, reboot the QwikCheck GOLD Equine:
 - Turn the system **OFF** then **ON** at the main switch on the rear panel.
 - Press the front panel **ON/OFF** key to begin the system warming-up and Self-Test.
 - From MAIN menu: Re-run the test.
 - 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 and open SERVICE SCREENS > SCREEN #2: SELF-TEST DATA.
 - Press **PRINT** button and the Service Data will be printed by the label printer.

Remove Capillary:

REMOVE CAPILLARY
FOLLOW ON-SCREEN INSTRUCTIONS

If the testing capillary has been left in the measurement chamber after testing a sample and prior to testing the next sample this screen will be displayed.



Appendix I: Semen Sample Preparation

EQUIPMENT REQUIRED:

- 10-ml plastic container
- Pipette
- QwikCheck GOLD Equine capillary
- On-board QwikCheck GOLD EQUINE Heater
- 20-micron Nylon Filter to filter samples with debris

NOTE:

Semen containing a lot of debris should be filtered with a 20-micron mesh filter prior to testing.

RAW/EXTENDED/COOLED SEMEN SAMPLES:

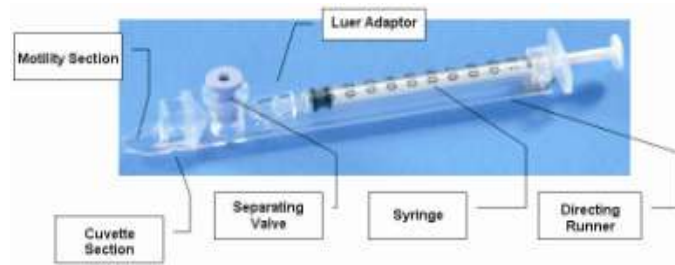
SAMPLE PREPARATION

1. Place QwikCheck GOLD Equine empty capillaries and 10-ml plastic containers in the heating unit.
2. Distribute a 1.5-ml aliquot of semen into 10-ml container.
3. Close the plastic container and pre-heat the semen to 37°C (98.6°F) for 2 minutes (RAW/EXTENDED room temperature samples) or for 4 minutes (COOLED samples).
4. Gently but thoroughly mix the sample for 10 seconds.
5. The sample is now ready for testing.
6. Fill a pre-heated to 37°C (98.6°F) QwikCheck GOLD Equine testing capillary following the instructions in the Appendix II: Capillary Filling Instructions: Raw/Extended/Cooled Specimens.

How to quickly observe samples for debris

- Place 10 µl of sample onto a standard slide, cover it with 22 mm x 22 mm coverslip and observe under the microscope for the presence of debris.
- If debris (big aggregates of epithelial cells) is observed, use a 20-micron nylon mesh to filter ~ 2 ml of sample (place the filter on a 10-ml plastic container and place the semen on the filter using a Pasteur pipette).

Appendix II: Capillary Filling Instructions: Raw/Extended/Cooled Specimen



1. Push the syringe piston in fully. Place only the thin part of the capillary into the bottom of the 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 below any surface bubbles (Figure 1). Continue to aspirate the sample until it appears in the Luer adaptor.
3. Hold the capillary in a vertical position and visually confirm that the sample has completely filled the thin section and the cuvette section and appears in the Luer adaptor (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).
6. Visually confirm that the capillary chambers are still full after wiping. If some of the sample has been depleted, a meniscus will be visible in the thin section of the capillary. If this is evident, push very slightly on the piston to re-fill the thin capillary section.
7. Slowly and carefully push-in the separating valve until it is level with the plastic. The capillary is now ready for testing (Figure 4).
8. Insert the capillary into the QwikCheck GOLD Equine (Figure 5)



Figure 1



Figure 2



Figure 3



Figure 5



Figure 4

APPENDIX III: Cleaning the Capillary/Slide Compartment

When to clean:

Daily or after every 10-15 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

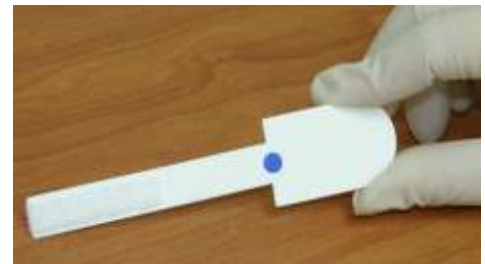


Figure 1

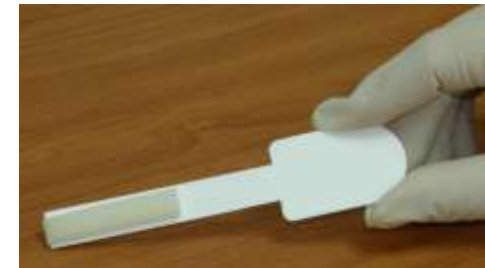


Figure 2



Figure 3



Figure 4



Figure 5

CLEANING: STEP 1

1. **TURN OFF** the QwikCheck GOLD Equine device.
2. Use a **BLUE DOT** fibrous material capillary (Fig. 1).
3. Moisten with **ONE** drop of cleaning fluid, shaking off excess fluid. **DO NOT USE MORE THAN 1 DROP**
4. Insert into the measurement compartment - fibrous material facing up. Move back and forth a few times. Repeat with the material facing down.
5. Use a sponge-tipped drying capillary to dry the same compartment (Fig. 2-3).

CLEANING: STEP 2

1. Insert the brush (bristle-side down – Fig. 4) into the measurement chamber of the QwikCheck GOLD Equine device (Fig. 5).
2. Pull the brush out of the chamber while sweeping or "dusting off" the lens (you will feel a step or shelf at the back and top of the chamber – this is the top of the lens).
3. Switch QwikCheck GOLD Equine system ON and observe Self-Test results. The analyzer should now PASS the Self-Test. If not, repeat cleaning procedure with the brush.

Appendix IV: Capillary Washing Instructions

Testing Capillary



Reposition the blue valve with the jig



Remove the plunger



Reassemble the capillary

Sample collection cups

FOR ANIMAL SYSTEMS ONLY!!

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

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%

Step 3: Remove all liquid from the testing capillary:

- Pump the syringe plunger a couple of times to remove liquids.

Step 4: Capillary Washing – Follow this order:

- **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.**
- After final washing, remove the plunger from the syringe.

Step 5: Drying the Capillaries:

- Place the capillaries:
 - On a flat surface to dry overnight.
 - In a commercial desiccator—follow manufacturer's instructions.
 - In an oven on low heat for a few hours.

Step 6: Final Preparation/Inspection:

- Replace the plunger into the syringe.
- Discard capillaries with debris, cracks or broken parts.
- Place a dot on the capillary with a water proof marker after each washing cycle. Discard after 10 uses (after nine washings).

Step 7: Capillary Reassembly:

- Confirm (using the jig) that the blue stopper valve is in the correct position.
- Check syringe by aspirating air in and out twice.

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 V: Glossary of Terms

	Parameter	QwikCheck GOLD Equine Terms	Definition
Sample Data	Serial number	SN	Serial Number of the QwikCheck GOLD Equine
	Date & time	DATE/TIME	The date and time the test was performed
	Stallion ID	STALLION ID	The identifying number of the stallion being tested
	Sample number	SAMPLE #	The number assigned to the semen sample
	Semen volume	SEMEN VOLUME	Raw, extended or cooled semen volume expressed in ml
Test Results	Sperm Concentration	CONC.	Sperm concentration expressed in millions/ml
	Motility	MOTILITY	Percent of motile spermatozoa
	Progressive Motility	PROG. MOT.	Percent of progressively motile spermatozoa
	Normal Morphology	MORPHOLOGY	Percent of morphologically normal spermatozoa
	Velocity	VELOC.	The average velocity of the motile spermatozoa in the sample, in microns per second
	Total Sperm Number	SPERM #	Total number of spermatozoa in the ejaculate
	Total Motile Sperm	MOTILE	Total number of motile spermatozoa in the ejaculate
	Total Progressively Motile Sperm	PROG.	Total number of progressively motile spermatozoa in the ejaculate



Appendix VI: QwikCheck GOLD Equine System Specifications

Dimensions: 29 X 24 X 20 cm (length X width X height); Weight: 4.15 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

Display

- Operational backlight LCD (16 lines x 40 characters)

Printer

- External label printer
- Thermostatic label paper with ~30 characters per line (Brother)

Keypad

- **Operational keys:** ON/OFF, TEST, PRINT, SERVICE, DELETE, ENTER, four cursor buttons, ESC, numeric buttons (0-9)

Front Panel

- LCD operational display
- Measurement compartment
- Multi-button keypad

Rear/Side Panel

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

Specimen Testing Supplies

- **Measurement capillary:** Disposable, multi-use plastic testing capillary (purchase from manufacturer).
- **I-Button:** Required to run tests (purchase from manufacturer)

Operating System

- **Control:** Keypad
- **Analysis Time:** 60 seconds pre-heating + 45 seconds testing
- **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°C (98.6°F).
- **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

Operational Temperature and Humidity

- System is operational at 15-40°C.
- **NOTE:** QwikCheck GOLD Equine 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 10-15 tests (refer to User Guide – "Cleaning Instructions").

Manufacturer Recommendations

- Operate the QwikCheck GOLD Equine system 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

Auto printing: YES



Appendix VII: QwikCheck GOLD Equine ARCHIVE SOFTWARE Installation Instructions

Overview of the Archive application

The QwikCheck™ Gold Equine Archive software is used to export test data to an EXCEL file and to print a test report from a printer connected to the computer. It runs automatically when the QwikCheck™ Gold Equine starts-up and continues to run as long as the computer is on and connected to the QwikCheck system.

Minimum PC Requirements

Hardware

- 1 GHz or higher CPU.
- 256 Megabytes of RAM.
- CD-ROM compatible drive.
- RS232 communication port (serial).

Software

- Windows XP Service pack 2 or higher.
- MS Excel 2003 or higher.

Installation Instructions:

1. Connect the QwikCheck™ GOLD Equine to a PC USB port using the RS232 to USB cable provided. **Note the port # of the USB hub used.**
2. Close all programs and reboot the computer.
3. Insert the "QwikCheck™ Gold Archive Equine" CD into the PC CD-ROM. The program disk will start automatically.
4. If a "Welcome to the QwikCheck™ Gold Archive Equine Setup Wizard" screen is displayed, immediately skip to # 10 below.
5. Click: **ACCEPT** when "Windows Installer 3.1" screen is displayed.
6. Click: **ACCEPT** when ".NET Framework 3.5" screen is displayed and the file will be automatically copied.
7. Click: **YES** when informed: "Setup must reboot before proceeding". This process may take a few moments.
8. Click: **NEXT** when the "Welcome to the QwikCheck™ Gold Archive Equine Setup Wizard" screen is displayed.
9. A new screen: "Select Installation Folder" will be displayed (not shown here).
 - a. Click: **NEXT** to accept the location for where to locate the software, or **BROWSE** to select another location on the computer.
 - b. Click: **EVERYONE**.
 - c. Click: **NEXT** to proceed.
10. Click: **NEXT** when the "Confirm Installation" screen is displayed.
11. When the "Installing QwikCheck™ Gold Archive Equine" screen is displayed (not shown here), the installation is in process.
12. Click: **OK** when the "Warning message" is displayed.
13. Click: **CLOSE** when the "Installation Complete" screen is displayed.
14. **Re-boot** the computer to complete the process. The software is now ready to set-up. Proceed to the SETTINGS section below.





Software Activation

The QwikCheck™ Gold Archive Equine operates automatically when the QwikCheck™ GOLD Equine and the PC are on. An icon is visible in the taskbar and can be clicked to maximize the archive application window.

Alternative ways to launch the program are as follows:

- **Click** on the shortcut icon on the computer desktop.
- **Click: START>ALL PROGRAMS>QwikCheck GOLD ARCHIVE EQUINE > QwikCheck Gold Archive Equine**

Settings

1. Open the "QwikCheck™ Gold Archive Equine" software using one of the SOFTWARE ACTIVATION methods above.
2. **Click:** The "Settings" icon to set up the defaults.
3. **Click:**
 - **Select Port:** Choose the PC serial port for the "QwikCheck™ Gold Equine".
 - **Printer Name:** Click: **CHOOSE** to select the default printer.
 - **Auto Print:** Mark: "✓" to print automatically after each test.
 - **Export to Excel:** Mark: "✓" to export test results to EXCEL. If the "✓" is marked, defaults need to be selected:
 - a. **Date Format:** select the date format for all exported data.
 - b. Set the File Name: Click: **OPEN** and select the data export file. To name a new file to export to, select a folder where the file will be located, and write the new name in the "File Name" row. The file will be created automatically.



Appendix VIII: QwikCheck GOLD Equine Product Performance Data

Performance Data Summary:

The performance of the QwikCheck GOLD Equine Sperm Analyzer is summarized in the text, tables and graphs below. All values concerning sperm concentration measurements are expressed as 10⁹ sperm cells per milliliter (M/ml). Motility values are expressed as a percent (%). Unless otherwise noted, all testing was performed using raw, extended and cooled equine semen samples. Manufacturers claims are generally lower than actual performance data. Please also note that Sensitivity & Specificity are clinical screening parameters that demonstrate the accuracy of device. Sensitivity & Specificity results are based on the cutoffs established by Society of Theriogenology.

Each QwikCheck GOLD Equine device is biologically calibrated against two reference systems at MES A-Tech laboratory.

Abbreviations:

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

Table 1. Dynamic Range

Sample Type	Concentration M/ml	% Motility	% Progressive Motility
Raw	0-500	0-100	0-100
Extended	0-50, >50	0-50, >50	0-40, >40
Cooled	0-50, >50	0-50, >50	0-40, >40

Sensitivity, specificity, precision and correlation to manual method established in the in-house and field clinical trials using equine semen samples

Clinical claims:

Sensitivity

- Concentration: 90%
- Motility: 90%
- Prog. Motility: 90%

Specificity

- Concentration: 90%
- Motility: 90%
- Prog. Motility: 80%

Precision (Intra-device CVs)

- Conc.: 3%
- Motility: 3%
- Prog. Motility: 7%

Precision (Inter-device CVs)

- Conc.: 10%
- Motility: 10%
- Prog. Motility: 10%

Accuracy (regression coefficients of the dilution trend line)

- Conc.: 0.9
- MSC: 0.9

Table 2. Sensitivity/Specificity

QwikCheck GOLD Equine vs. Microscope	Sensitivity %	Specificity %	% False Positive	% False Negative
Sperm Concentration	96.4	100.0	0	2.2
Motility	95.0	96.3	2.1	2.1
Progressive Motility	100.0	90.0	4.3	0

Table 3. Precision: QwikCheck GOLD Equine intra- and inter-device variability

Semen Parameters	Intra-device CV, %	Inter-device CV, %
Sperm Concentration	2.0	7.0
Motility	0.3	7.2
Prog. Motility	5.6	8.6



Correlation to Manual Method

- Concentration: 0.9
- Motility: 0.9
- Prog. Motility: 0.8

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:

The QwikCheck GOLD Equine system was compared to the microscope based on WHO'99 manual guidelines. The QwikCheck GOLD Equine automated readings for sperm concentration, motility and progressive motility were compared to microscopic results. The manual sperm concentration measurements were performed using a cell counting chamber according to the **manufacturer's instructions**. A microscope and standard slide were used to manually assess motility. **The protocols were based on WHO'99 manual and MES guidelines.** The clinical trials were conducted at the Medisoos vet clinic. A total of 201 raw, extended and cooled semen samples were analyzed.

Accuracy: Dilution plots.

The accuracy of the QwikCheck GOLD Equine system was assessed by diluting equine semen and analyzing the resulting sperm concentrations. Raw stallion semen was gradually diluted with commercial transparent extender. Dilutions provided varying motile and total sperm concentrations. Semen samples were tested using the QwikCheck GOLD Equine device and the results were plotted. Linear trend lines were established for Concentration and MSC (calculated by $CONC. \times Motility / 100$) vs. expected values.

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, debris and seminal plasma is used.
- Low noise and high electronic resolution hardware components and compensation circuits ensure analytical specificity optimization.

Limitations of method:

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

Table 4: Correlation to Manual Method

Semen Parameters	Correlation coefficients
Sperm Concentration, M/ml	0.996
Motility, %	0.956
Progressive Motility, %	0.892

Fig. 1. Method comparison: Regression plot of QwikCheck GOLD Equine Sperm Concentration in Raw equine semen vs. manual results

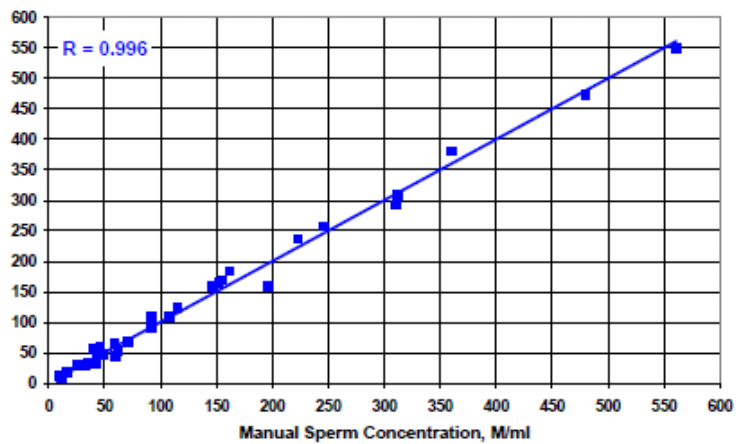
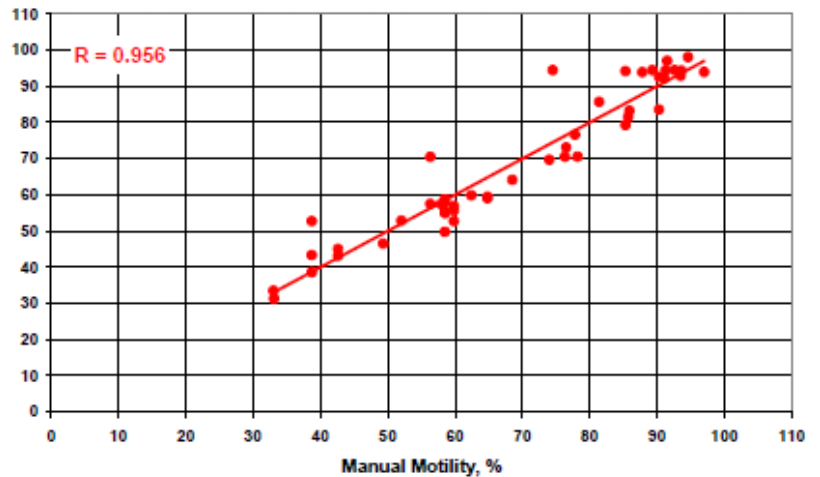


Fig. 2. Method comparison: Regression plot of QwikCheck GOLD Equine Motility in Raw equine semen vs. manual results



Performance parameters:

- Sensitivity and specificity were calculated using ROC analysis. Cutoffs normally used for sperm concentration and motility were used for the calculation of sensitivity, specificity, false positive and false negative parameters (Table 2).
- Precision of the QwikCheck GOLD Equine device was estimated by calculation of the intra-device and inter-device coefficients of variation (CV) of duplicate measurements (Table 3). CV is calculated according to the formula:
$$CV = SD / MEAN \times 100$$
The lower CV, the higher precision of the method.
- Correlation to manual method was established by calculating correlation coefficients (Table 4, Fig. 1-2).
- The accuracy of the QwikCheck GOLD Equine system was determined by the regression coefficients of the dilution trendlines (Fig. 3).

Conclusions:

- The QwikCheck GOLD Equine Sperm Analyzer demonstrated high levels of sensitivity, specificity and correlation to the manual method.
- The QwikCheck GOLD Equine system is precise and accurate with low coefficients of variation for all semen parameters assessed (<10%).
- The QwikCheck GOLD Equine device can be used for semen quality assessment and QC of equine semen samples.

Fig. 3. Regression plot of QwikCheck GOLD Equine Conc. & MSC in equine semen vs. expected values

