NanoEntek

NESPI-ARH-001EN(V.0.2)

# ADAM- rWBC HT Cell Counting System

***For in vitro diagnostic use***

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**50 Tests – Catalog No. ADHK-050**

For enumeration of residual leukocytes in leukoreduced blood products

Revised 2024


**Manufactured by**

**NanoEntek, Inc.**
851-14, Seohae-ro, Paltan-myeon, Hwaseong-si, Gyeonggi-do, 18531, Korea
Tel. : +82-2-6220-7940  Fax. : +82-2-6220-7999

EC REP

**MT Promedt Consulting GmbH**
Ernst-Heckel-Straße 7, 66386 St. Ingbert, Germany

## 1. INTENDED USE

The ADAM-rWBC Kit is for in vitro quantitative use for enumeration of residual white blood cells (rWBCs) in leukoreduced blood products. The ADAM-rWBC Kit is designed for use with the ADAM-rWBC, ADAM-rWBC2 and ADAM-rWBC HT microscopic cell counter.

## 2. SUMMARY AND EXPLANATION

The presence of white blood cells (WBCs) in blood, platelet and plasma products is associated with an increased incidence of febrile transfusion reactions, transmission of cytomegalovirus, and alloimmunization to HLA antigens in transfusion recipients.[1-3] Leukoreduction, the collection of platelets via apheresis, or post-collection processing with special filters, can lower the WBC count to 5 x $10^6$ per unit or below, thus minimizing complications associated with transfusions.[4,5] The ADAM-rWBC Kit is designed to provide an efficient, sensitive method for enumerating residual WBCs, while eliminating limitations associated with other methods.[6,7]

## 3. PRINCIPLES OF THE PROCEDURE

The ADAM-rWBC utilizes sensitive fluorescence dye staining, LED excitation and CCD detection technologies to make the WBC analysis more accurate and reliable. To count WBCs using ADAM-rWBC, the sample to be tested is mixed with a Propidium Iodide (PI) stain and directly pipetted onto a disposable plastic slide. The slide is then loaded onto a precision stage. The ADAM-rWBC system automatically focuses on the slide, and cells that have been stained are recorded by a sensitive CCD camera. The image results are automatically processed, generating the cell count which is displayed on the front of the device.

## 4. REAGENTS

**The ADAM-rWBC Kit consists of :**

| Contents | Cat. No. |
|---|---|
| r-Solution containing Propidium Iodide (0.04%, w/v), a nucleic acid dye (25 mL) | RDR-50 |
| r-Slide (50 pcs) | RHS-50 |
| Standard Bead Solution (PeakFlow flow cytometry reference beads, Abs 580nm, Em 620nm, 6-diameter) containing 0.1% sodium azide (7 mL) | ADST-001 |

## Precautions

- For *in vitro* diagnostic use. Not for use in therapeutic procedures.
- Reagent should be handled with care to avoid microbial contamination.

### ⚠ WARNING

All biological specimens and materials coming in contact with them are considered biohazards. Handle as if capable of transmitting infection and dispose of with proper precautions in accordance with federal, state, and local regulations. Never pipette by mouth. Wear suitable protective clothing and gloves.

### ⚠ WARNING

The r-Solution reagent contains PI, a suspected mutagen, and a DNA stabilizer known to irritate skin and mucous membranes. Gloves and eye protection should be worn when handling. Avoid contact with eyes, skin, and clothing. Avoid breathing vapors and wash surfaces thoroughly after handling. If contact occurs, flush immediately with water. Consult a physician if contact with eyes occurs.

### ⚠ WARNING

Standard Bead Solution reagent contains sodium azide which is harmful if swallowed. Keep out of reach of children. Keep away from food, drink, and animal feed. Wear suitable protective clothing. If swallowed, seek medical advice immediately and show container or label. Contact with acids liberates very toxic gas. Azide compounds should be flushed with large volumes of water during disposal to avoid deposits in lead or copper plumbing where explosive conditions can develop.

## Storage and Handling

1. Store r-Solution at 2-8℃ when not in use. The shelf-life is one year.
   Do not use after the expiration date shown on the label. Opened r-Solution should be stored at 2-8℃ and used for up to 6 months, but not past the expiration date.

2. Store the r-Slides at 0-30℃. Any unused slides should be sealed in the pouch.
   The expiration date is one year before the opening.
   Do not reuse the slide and after the expiration date shown on the label.

3. Store the Standard Bead Solution in the dark at 2-8℃ when not in use. Opened Standard Bead Solution should be stored at 2-8℃ for up to 6 months. Do not use after the expiration date shown on the label.

4. The Standard Bead Solution should not be exposed to light for a long time and is required to be placed on the bench during use.

## 5. INSTRUMENT

**Also refer to the ADAM-rWBC HT Instruction Manual.**

This ADHK-050 model is designed for use on the ADAM-rWBC HT white blood cell counter.

Below is a listing of the accessories:

| | |
|---|---|
| Trays | 5 ea |
| Barcode scanner | 1 ea |

**Instrument Safety Precautions**

1. Always ensure that the power supply input voltage matches the voltage available in your location.
2. To avoid the danger of electric shock, install the instrument per the environmental specifications described in “Technical Specifications”. If the instrument, the adaptor, or power inlet is exposed to water or other material, disconnect the power cord and contact an authorized service person.
3. Do not touch the main plug or power cord with wet hands.
4. This instrument is air-cooled so its surfaces become hot during operation. When installing it, leave a space of more than 10 cm (4 inches) around it.
5. Do not install the instrument on a slant or a place prone to vibrations, which induces the risk of the instrument malfunction or damage of the instrument.
6. Never insert any objects (especially metallic objects) into the air vents of the instrument as this could result in electrical shock, personal injury and instrument damage.
7. Always set the main switch on the power supply unit to OFF before connecting the power cord to the wall outlet.
8. To avoid a potential shock hazard, always connect the grounding terminal of the instrument and that of the wall outlet properly. The power cord should be connected to a grounded, 3-conductor power outlet.
9. Position the instrument so that there is sufficient length for the cable connection.

10. Before moving, set the main switch to “O” (OFF) and unplug the power cord.
11. If the instrument is broken or dropped, disconnect the power cord and contact an authorized service person. Do not disassemble the instrument.
12. Use authorized accessories only.
13. Use instrument only as instructed in this manual and as specified in any documentation associated with its components.

**⚠WARNING**

Cover: Do not remove the cover or dissemble a case. There are no adjustable components inside the instrument. If there is a malfunction, contact the NanoEntek technical service team.

## 6. SPECIMEN COLLECTION AND PREPARATION

Red Blood Cell, platelet and plasma samples must be collected and tested within 48 hours of leukoreduction.

ADAM-rWBC HT testing is not affected by hemolysis or lipemia.

**Stained Sample Stability**

Stained samples (RBC, Platelet and Plasma) may be stored for up to one hour at room temperature prior to use.

**Stored Sample Stability**

**RBC products:** Leukoreduced samples may be stored for up to 48 hours at refrigerated temperature (2~8°C) prior to testing.

**Platelet products:** Leukoreduced samples may be stored for up to 48 hours at room temperature (18~25°C) prior to testing.

**Plasma products:** Leukoreduced samples may be stored for up to 24 hours at refrigerated temperature (2~8°C) prior to testing.

# 7. PROCEDURE

**Materials Required but Not Provided**

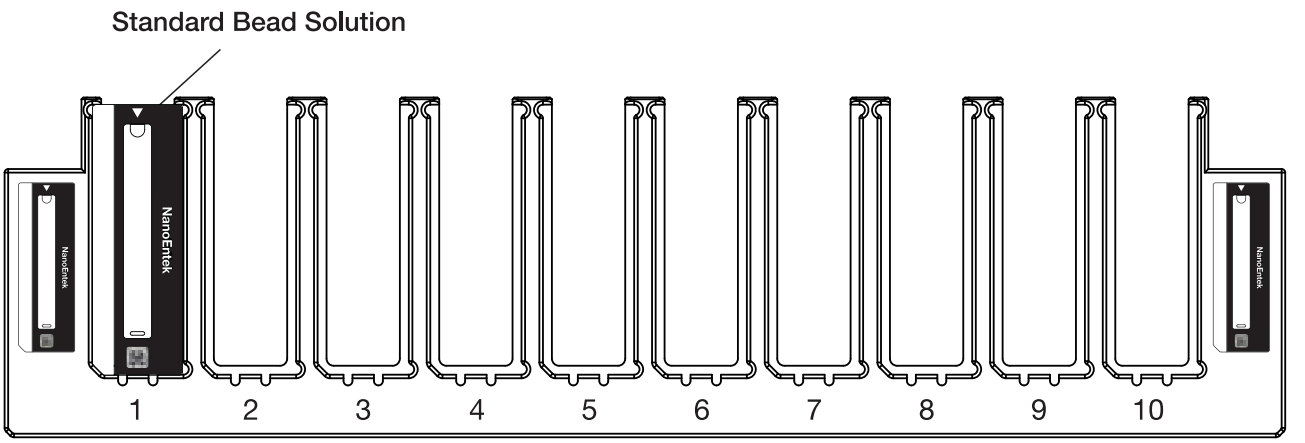
- Micropipette and tips
- Capped test tubes
- Test tube rack
- Timer
- Lint free wipes

ⓘ **NOTE:** *If tips must be wiped, use lint free material only, and discard after a single use. It is also not recommended to use materials that may generate or capture lint as a bench cover in the testing area.*

**Calibration (Standard Bead)**

For the calibration, Standard Bead Solution (with absolute particle count) is used to calibrate the automatic focus of ADAM-rWBC HT. It should be done as soon as the instrument is turned on. This calibration checks the position of the slide stage.
The calibration should be done on a daily basis, at least once a day using the Bead.
Figure 1 shows the position of the r-Slide for calibration test in the Tray.

Figure 1. Position of r-Slide in the Tray for calibration.



1. Click ‘Calibration’ button in the main software.
2. Let the beads come to room temperature for up to 10 minutes before use.
3. Scan the barcodes of the Standard Bead Solution and the r-Slide.
4. Place the scanned empty r-Slide in the Tray.
5. Mix the bead until the substance is distributed evenly.
6. Load 100 µL of the beads onto the r-Slide.
7. Insert the Tray into the Magazine of the ADAM-rWBC HT instrument.
8. Close the door and click ‘Next’ button followed by ‘Run’ button for calibration.

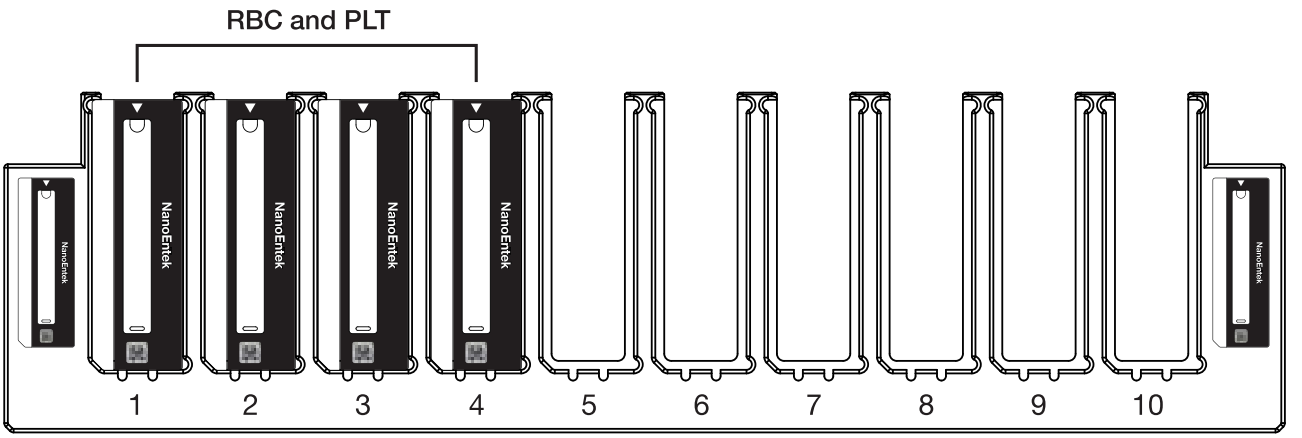
ⓘ **NOTE:** *Please refer to the ADAM-rWBC HT instruction manual for detailed information.*

The instrument is ready to use only if the calibration result is within range as indicated on the individual Standard Bead Solution provided.

## Quality Control (Control Material)

The package insert will include instructions to test external quality control (RBC Low level and High level; PLT low level and High level) at initial operation and after long-term non-use, using ‘Control’ mode. The quality control material can run togetherin a single Tray. Figure 2 shows an example of the r-Slides for quality control material inserted in the Tray. The slides do not have to be in this order as long as they are inserted in the slot 1 through 4.

Figure 2. Position of the r-Slides in the Tray for quality control material.



1. Click ‘Control’ button in the main software.
2. Let the quality control materials come to room temperature for up to 15 minutes before use.
3. Scan the barcode of the r-Solution.
4. Sequentially scan the barcodes of each pair of control material and r-Slide. Scan the control material tube first and then the empty r-Slide.
5. Place the scanned empty r-Slide in the Tray.
6. Gently invert the control material until mixed thoroughly.
7. Insert 400 µL of r-Solution and 100 µL of quality control material into a clean test tube.
8. Mix the tube well.
9. Load 100 µL of the mixed sample onto the r-Slide.
10. Insert the Tray into the Magazine of the ADAM-rWBC HT instrument.
11. Close the door and click ‘Next’ button followed by ‘Run’ button for measurement.

ⓘ **NOTE:** *Please refer to the ADAM-rWBC HT instruction manual for detailed information.*

The instrument is ready to use only if the quality control material result is within range as indicated on the data sheet provided with quality control material.

## Sample Preparation and Testing

1. Click 'Sample' button the main software.
2. Enter the ‘Folder Name’, if necessary.
3. Select sample type. For each test, select only one sample type (RBC or PLT or Plasma).
4. Place the Tray(s) to be used on a flat table (Fig. 3).
5. Sequentially scan the barcodes of each pair of blood sample and r-Slide. Scan the blood sample first, and then the empty r-Slide.
6. Enter ‘Optional ID’ and ‘Volume’, if necessary.
7. Place the scanned empty r-Slide in the Tray.
8. Repeat step from 5 to 7 until all of the r-Slides for testing are well positioned.

**NOTE:** *Please refer to the ADAM-rWBC HT instruction manual for detailed information.*

Figure 3. An example of empty Tray.

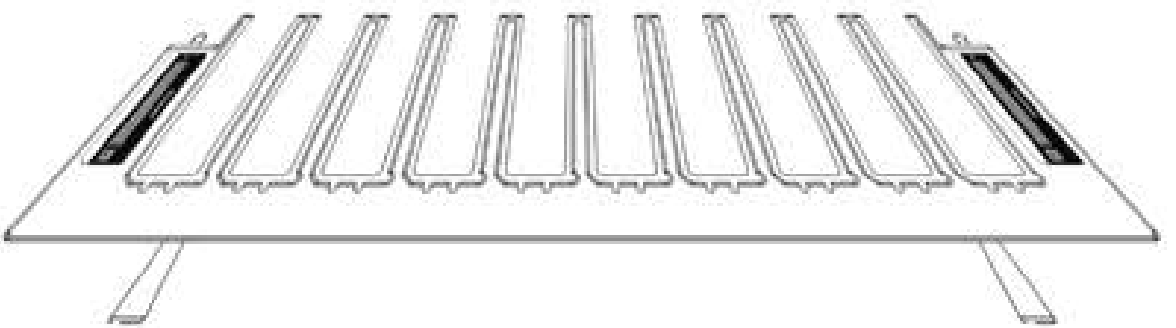
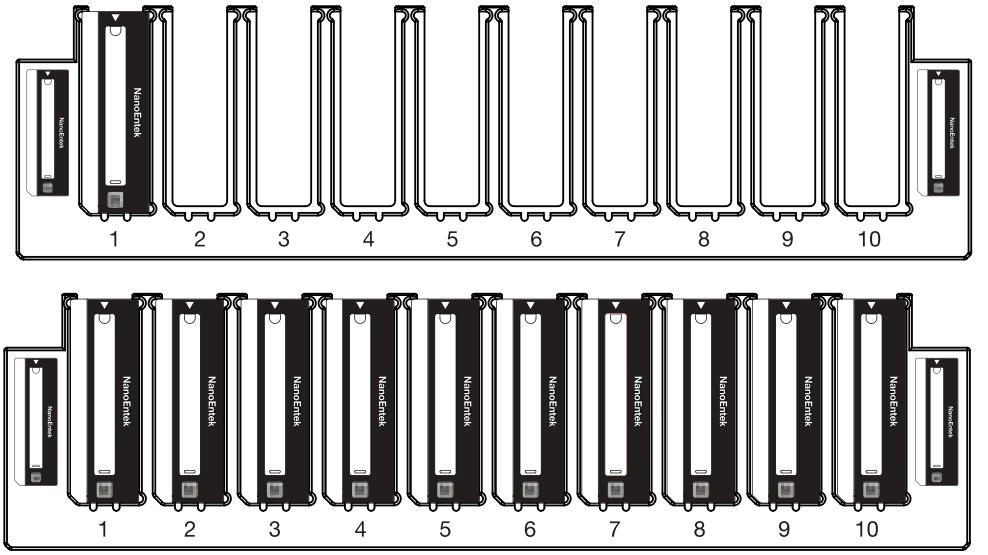


Figure 4. Position of r-Slide in the Tray for sample test.

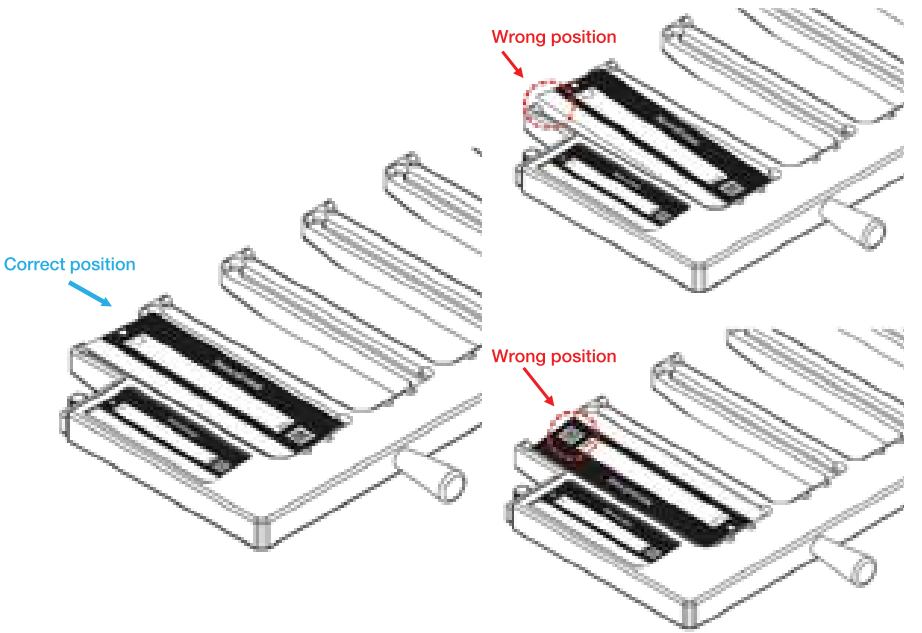



※ *Up to 50 samples (in the 5 Trays) can be performed at once.*

**⚠WARNING**

The r-Slide(s) should be inserted in the same direction as the example attached in the Tray.

Figure 5. Position of r-Slide in the Tray.




※ *The barcode must be located at the bottom of the Tray.*

Due to stained sample stability, there should be no delay between any of the following steps: sample processing, r-Slide loading, Tray insertion into the Magazine and starting of the counting process.

9. Add 400 µL of r-Solution to each test tube using a pipette.
10. Carefully add 100 uL of well mixed sample into a test tube.
    - RBC or platelet : 100 µL sample + 400 µL r-Solution
    - Plasma: 100 µL sample + 100 µL r-Solution

**⚠WARNING**

[Mix ratio error]

Please mix the sample & reagent with the correct ratio. The ADAM-rWBC HT does not provide the mixing ratio.

11. Load 100 µL of mixed sample/reagent onto the r-Slide.

    **Note:** *Reverse pipetting is recommended.*

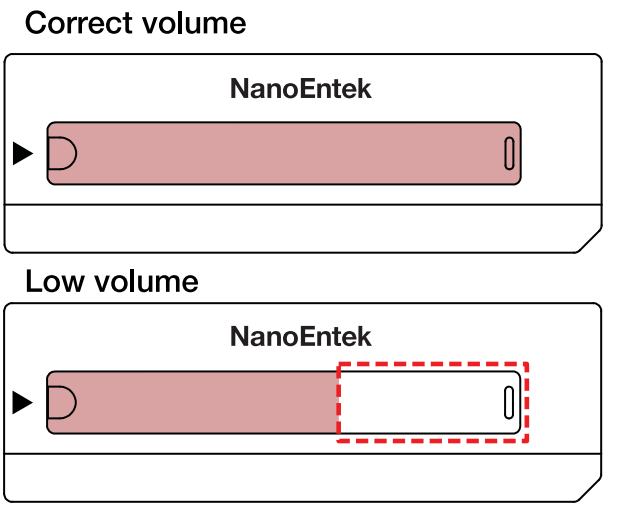
**⚠WARNING**

Avoid bubbles which may negatively affect the result.

**⚠WARNING**

[Sample loading error]
Make sure to load the correct volume (100 µL) of the sample onto the r-Slide. The ADAM-rWBC HT will not detect low or high sample volume.

Figure 6. Example of correct volume and low volume loaded onto the r-Slide.



12. Insert the first Tray at the bottom of the ADAM r-WBC HT Magazine.
13. Click 'Next' in the main software.
14. After finishing Tray(s) insertion, make sure that the software displays the correct number and placement of inserted Tray(s) with blue color.
15. Close the door.
16. Click ‘Run’ button to start.

## 8. RESULTS

The calculated number of WBCs per µL will be displayed automatically.

**[Error message – Bad Sample]**
Figure 7. In case of bad sample after test.


In case this error message appears in the results, discard the error sample, prepare a new stained sample and retest. If the error occurs repeatedly, please obtain a new whole sample and start again.

## 9. PERFORMANCE CHARACTERISTICS

**All testing was conducted at the NanoEntek laboratory.**

**Linearity**

Linearity was assessed using 2 clinical samples (RBCs and platelets) at 7 concentrations.

Results were observed to be linear within the 1 – 100 cells/µL range.

## Method Comparison- Accuracy

Method comparison was conducted comparing the ADAM-rWBC HT to the ADAM-rWBC. 300 CPDA-1 RBC and 300 CPDA-1 Platelet clinical samples per three (3) operator/ instrument were tested (N=900 total for each sample type).

And 150 randomly produced clinical plasma samples were tested for method comparison comparing the ADAM-rWBC HT to the ADAM-rWBC.

Figure 8. RBC.

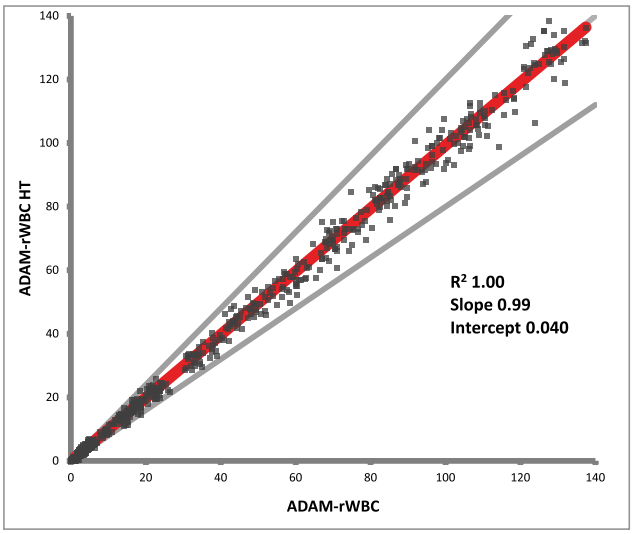



Figure 9. Platelet.

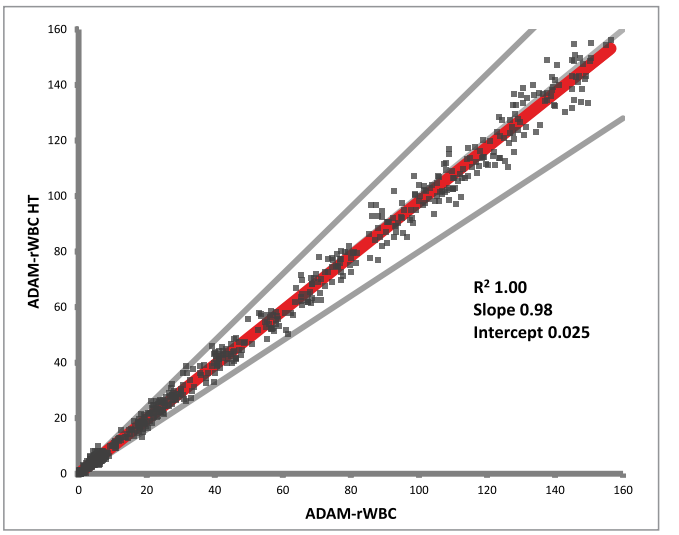



Figure 10. Plasma

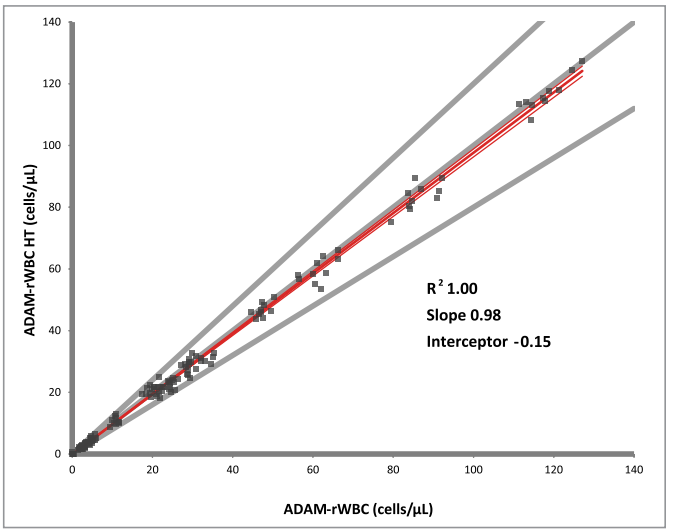


Table 1. Method Comparison Bias.

| Bias | N | Parameter | Estimate | 95% CI | SE |
|---|---|---|---|---|---|
| **RBC** | | | | | |
| WBC <5 cells/μL | 379 | Mean difference | 0.0016 | -0.0440 to 0.0754 | 0.0304 |
| WBC ≥5 cells/μL | 521 | Mean relative difference | -1.94% | -2.626% to -1.256% | 0.35% |
| **Platelet** | | | | | |
| WBC <5 cells/μL | 318 | Mean difference | 0.1008 | 0.03183 to 0.16986 | 0.03508 |
| WBC ≥5 cells/μL | 582 | Mean relative difference | 1.89% | 1.102% to 2.677% | 0.40% |
| **Plasma** | | | | | |
| WBC <5 cells/μL | 46 | Mean difference | -0.157 | -0.3248 to 0.0114 | 0.0835 |
| WBC ≥5 cells/μL | 104 | Mean relative difference | -2.98 | -4.54% to -1.414% | -0.788% |

## Precision

For the study, three operators & instruments have operated using 5 target concentrations as follows:

- 0-1, 5-10, 20-30, 50-60 and 80-100 cells/μL
- It was conducted in 2 days and the operator conducted 50 replicates (testing) per sample twice each day.

Table 2. RBC

| Sample | Mean value | N | Repeatability | | Between Day | | Between Run | | Between instrument | | Reproducibility | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | SD | % CV | SD | % CV | SD | % CV | SD | % CV | SD | % CV |
| 0-1 | <1 | 600 | 0.20 | NA | 0.00 | NA | 0.00 | NA | 0.26 | NA | 0.33 | NA |
| 5-10 | 7.94 | 600 | 0.94 | 11.81% | 0.12 | 1.50% | 0.00 | 0.00% | 0.48 | 6.00% | 1.06 | 13.34% |
| 20-30 | 21.39 | 600 | 1.68 | 7.87% | 0.00 | 0.00% | 0.00 | 0.00% | 0.57 | 2.70% | 1.78 | 8.31% |
| 50-60 | 52.15 | 600 | 2.65 | 5.09% | 0.00 | 0.00% | 0.00 | 0.00% | 2.11 | 4.00% | 3.39 | 6.50% |
| 80-100 | 97.02 | 600 | 4.21 | 4.34% | 0.59 | 0.60% | 0.00 | 0.00% | 2.69% | 2.80% | 5.03 | 5.19% |

Table 3. Platelet

| Sample | Mean value | N | Repeatability | | Between Day | | Between Run | | Between instrument | | Reproducibility | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | SD | % CV | SD | % CV | SD | % CV | SD | % CV | SD | % CV |
| 0-1 | <1 | 600 | 0.22 | NA | 0.05 | NA | 0.00 | NA | 0.05 | NA | 0.24 | NA |
| 5-10 | 8.14 | 600 | 0.81 | 9.95% | 0.00 | 0.00% | 0.00 | 0.00% | 0.15 | 1.90% | 0.82 | 10.12% |
| 20-30 | 21.02 | 600 | 1.41 | 6.71% | 0.00 | 0.00% | 0.00 | 0.00% | 0.67 | 3.20% | 1.56 | 7.43% |
| 50-60 | 54.35 | 600 | 2.36 | 4.35% | 0.00 | 0.00% | 0.00 | 0.00% | 1.89 | 3.50% | 3.02 | 5.56% |
| 80-100 | 100.08 | 600 | 3.80 | 3.80% | 0.73 | 0.70% | 0.00 | 0.00% | 1.45% | 1.40% | 4.14 | 4.13% |

**Interfering Substances**

ADAM-rWBC HT testing is not affected by the use of lipemic or hemolyzed samples.

## 10. LIMITATIONS

It is important to follow these Directions for Use and the ADAM-rWBC HT Instruction manual. Our kit is made specifically for use with the ADAM-rWBC HT instrument - only use provided reagents and materials. Do not use reagents or slides beyond the expiration date. Do not use previously used r-Slides.

## REFERENCES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2. de Graan-Hentzen YC, Gratama JW, Mudde GC,et al. Prevention of primary cytomegalovirus infection in patients with hematologic malignancies by intensive white cell depletion of blood products. *Transfusion*. 1989;29:757-760.
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4. Venglen-Tyler V, ed. Leukoreduction of RBC and platelet units. *American Association of Blood Banks*. 1996; 722-725.
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6. Rubella P, Porretti L, Bertolini F, et al. White cell-reduced red cells prepared by filtration: a critical evaluation of current filters and methods for counting residual white blood cells. *Transfusion*. 1993;33:128-133.
7. Vachula M, Simpson SJ, Martinson JA, et al. A flow cytometric method for counting very low levels of white cells in blood and blood components. *Transfusion*. 1993;33:262-267.


CE IVD

# Glossary of Symbols

| | |
|---|---|
|  | Caution, warning,<br>Consult accompanying documents |
| REF | Catalogue number/Reference number |
|  <br>www.nanoentek.com/eifu.php | Consult Instructions for Use<br>An electronic instructions for use (eIFU) indicator (website address) may accompany the symbol when used to indicate an instruction to consult an eIFU. |
| LOT | Lot number/Batch number |
|  | Use by<br>YYYY-MM-DD or YYYY-MM |
|  | Manufacturer |
| EC REP | Authorized representative in the European Community |
| CE | CE marking |
| IVD | *In vitro* diagnostic medical device |
|  | Temperature limitation |
|  | Contains sufficient for <n> tests |
|  | Do not reuse |
|  | Do not use if package is damaged |
| Rx Only | For prescription use only<br>CAUTION: Federal (U.S.) law restricts this device to saleby or on order of a physician. |
| US Corporation | US Corporation |