COBAS MINOS® STEL

Automated Hematology Analyzer

OPERATOR'S MANUAL

Manufactured by: ABX, Montpellier, France for:

Roche Diagnostic Systems
a subsidiary of Hoffmann - La Roche, Inc.

Roche Diagnostic Systems, Inc.
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Any references in this manual to ABX Service should now be refered to :
Roche Response Center
1-800-526-1247

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SECTION I

Introduction

SECTION I - INTRODUCTION

MINOS STE-L

INTENDED USE

The MINOS STE-L System is an in-vitro, quantitative, automated hematology analyzer for use in clinical laboratories. It is intended for use in the quantitation of the following hematologic parameters:

WBC	[White Blood Cell (Leukocyte) Count]
RBC	[Red Blood Cell (Erythrocyte) Count]
HGB	[Hemoglobin Concentration]
HCT	[Hematocrit]
MCV	[Mean Corpuscular Volume]
MCH	[Mean Corpuscular Hemoglobin]
MCHC	[Mean Corpuscular Hemoglobin Concentration]
PLT	[Platelet Count]
LYM #	[Lymphocyte Count (absolute number)]
LYM %	[Lymphocyte Percentage]

The MINOS STE-L System directly measures the following hematology parameters:

WBC, RBC, HGB, HCT, PLT, LYM %

The data obtained from the direct measurements, above, are used to automatically calculate the remaining hematology parameters:

MCV, MCH, MCHC, LYM #

PRINCIPLES OF OPERATION:

A sample of 25 uL whole blood is required to determine the 10 hematologic parameters. The whole blood is automatically diluted in a buffered, isotonic saline solution, which prepares the sample for WBC, RBC, and PLT analyses.

Leukocytes (WBC), WBC Histogram, and Hemoglobin (HGB) concentration are measured from the more concentrated WBC dilution. Erythrocytes (RBC), RBC Histogram, Hematocrit (HCT), Platelets (PLT), and Platelet Histogram are measured from the RBC dilution.

The principle used in the particle-counting technique is impedance. The principle requires the blood sample to be diluted in an electrolyte solution (e.g. sodium chloride). Two electrodes are immersed in the electrolytic solution and a small current of electricity is passed from one (*outside*) electrode to the other (*inside*) electrode. The two electrodes are separated by a small aperture[2] through which the electrolyte solution, the electrical current, and the particles to be counted flow. Since blood cells do not conduct electricity, their passage through the aperture will cause a change (*a pulse*) in the electrical current. The size of the pulse is directly proportional to the volume of the particle generating the change in voltage (see **Figure I-1**). The pulse is further amplified by electronic circuitry, and thresholds are established to allow for counting and accurate sizing by the microprocessor.

During the RBC count, pulses that are counted as red blood cells are subsequently analyzed in an analog integrator, where the heights of the individually counted pulses are added together, and the hematocrit is determined at the end of the counting cycle.

The platelet count is determined concurrently with the RBC count. A unique electronic circuit board is dedicated to sorting and analyzing the platelet pulses, to permit the accurate counting of platelets, as well as the specialized analyses ultimately used in the determination of the Mean Platelet Volume (MPV). Proprietary circuits used in the **MINOS System** incorporate a moving upper threshold to eliminate interference in the platelet count by microcytic red blood cells (and/or schistocytes), and to warn the operator of potentially invalid results when the presence of Giant Platelets, platelet aggregates, microcytes, and/or schistocytes are present in sufficient quantities to create interference.

The above data are also used in the generation of the WBC, RBC, and PLT Histograms.

Hemoglobin is determined by a modified Drabkin (Cyanmethemoglobin) technique. The lysing reagent added to the WBC dilution disrupts the erythrocyte membrane, liberating the hemoglobin.[3] Hemoglobin is chemically converted to cyanmethemoglobin (a stable pigment) and is measured at 540 nanometers (*nm*).

The remaining hematologic parameters are calculated, by the microprocessors, from the above measurements.

[2] **The WBC dilution bath uses a 100 micron diameter aperture. The RBC dilution bath uses a 50 micron diameter aperture.**

[3] **Hemoglobin is chemically converted to methemoglobin and then to cyanmethemoglobin prior to spectrophotometric analysis.**

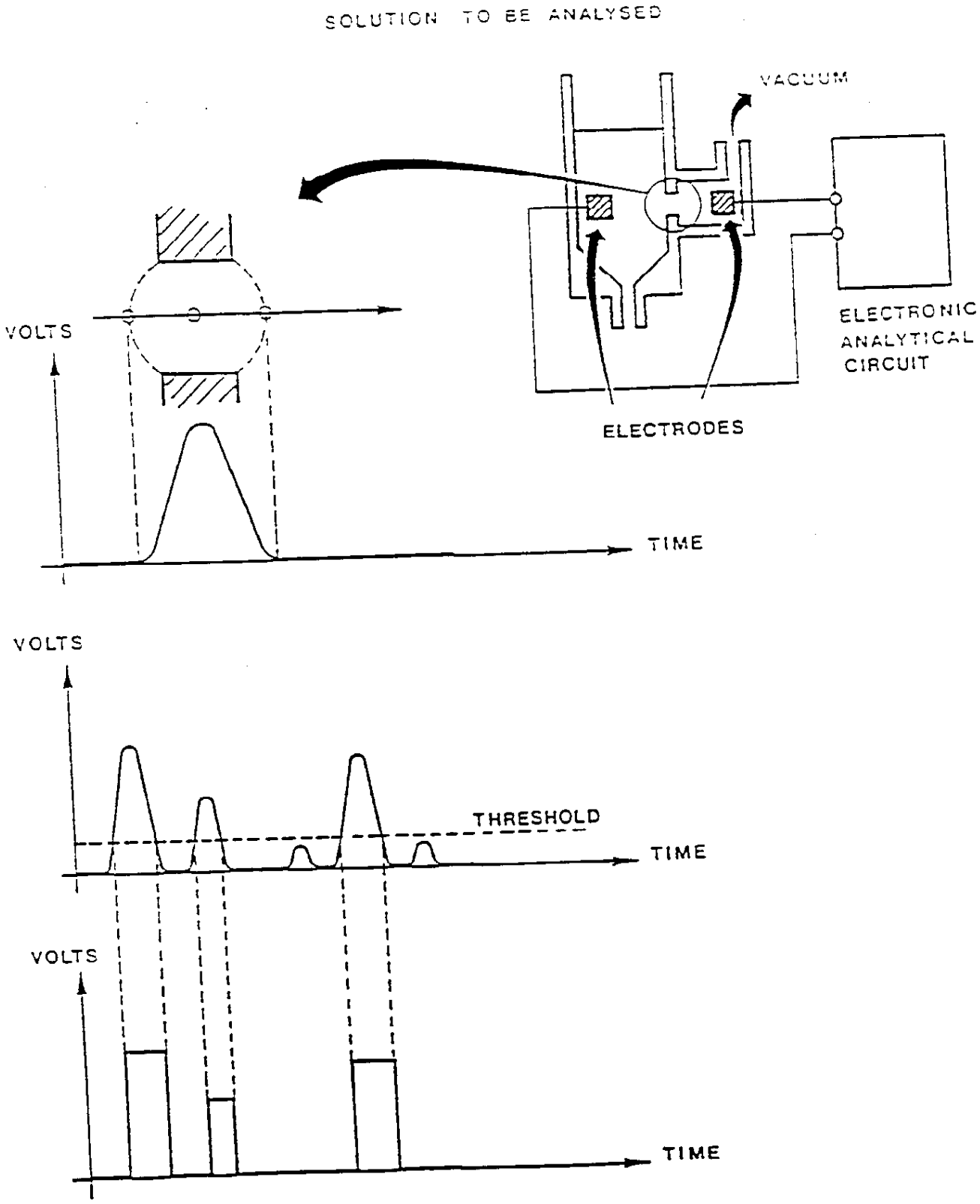
SOLUTION TO BE ANALYSED
VACUUM
ELECTRONIC
ANALYTICAL
CIRCUIT
ELECTRODES
VOLTS
TIME
VOLTS
THRESHOLD
TIME
VOLTS
TIME

Figure I-1

OPERATIONAL CYCLES

A. DILUTION CYCLE

The well-mixed EDTA[4] anticoagulated blood sample is held beneath the **sample needle**, and the cycle is activated by pressing the **ASPIRATE SAMPLE** key. 25 uL of the blood sample is aspirated. After aspiration is completed, the sample needle is automatically removed from the blood sample, and the external surface is cleaned and dried by a stream of fresh diluent and air.

The sample needle carriage is automatically moved to a position above the **mixing chamber**, where the needle lowers and the 25 uL of blood sample is dispensed along with 5.0 mL of diluent.[5] Carefully-controlled air bubbles are generated to insure proper mixing of the diluted sample.

Once the diluted sample has been thoroughly mixed, the sample needle aspirates 25 uL of the diluted (1:200 dilution) sample. After aspiration is completed, the sample needle is automatically removed from the diluted sample, and the external surface is cleaned and dried by a stream of fresh diluent and air. The sample needle is automatically raised and moved to a position above the **RBC chamber**, where it lowers and the 25 uL of diluted sample is dispensed along with 2.5 mL of diluent.[6] Again, carefully controlled air bubbles are generated to insure proper mixing of the diluted sample. The secondary dilution is automatically dispensed into the **WBC chamber**,[7] causing the erythrocytes to lyse, and initiating the cyanmethemoglobin reaction.

B. MEASUREMENT CYCLE

Each of the two (**WBC & RBC**) dilution chambers contains electrode blocks having an external and internal electrode separated by an aperture. The WBC aperture is 100 microns in diameter, and the RBC aperture is 50 microns in diameter. During the measurement cycle, the diluted blood sample is pulled through the apertures by the vacuum in the **waste chamber**.

The MINOS STE-L captures and processes the pulses of the particles passing through the individual apertures for exactly eight (8) seconds, then repeats the process for an additional eight seconds. Between the first and second counting cycles, both apertures are cleaned by a back flushing technique to eliminate clogging of the orifices.

During the above counting cycles, a small pump is used to aspirate 1.0 mL of the WBC dilution for spectrophotometric analysis, for the determination of hemoglobin concentration.

[4] **Ethylenediamine tetraacetic acid**

[5] **The 25 uL of blood sample mixed with 5.0 mL of diluent creates a 1:200 dilution ratio.**

[6] **The 25 uL of the 1:200 dilution and the 2.5 mL of diluent create a 1:20,000 dilution ratio.**

[7] **The addition of the 1.0 mL of lysing reagent to the WBC mixing chamber creates a final dilution ratio of 1:240.**

Prior to reading the WBC dilution, the **spectrophotometric chamber** is rinsed with clean diluent; this fluid serves as a reference blank (reference zero). Once the absorbance of the WBC dilution has been determined, the solution is aspirated into the **waste chamber**, and the **spectrophotometric chamber** is rinsed with clean diluent.

After the two counting cycles (8 seconds each) are completed, the data is analyzed by the microprocessor. If the two counts compare favorably to the predetermined expected limits,[8] the counting cycle is completed. However, if the two counts exceed the expected limits, a third count (preceded by an aperture-cleaning back flush) is automatically initiated by the instrument, and the two closest counts will be taken. If, after the third count, no two counts are in agreement, a **sample rejection** will occur. In such case, only the lowest value will be reported; however, *** will be printed on the report form.

During data acquisition and reduction, the microprocessor automatically calculates for coincidence and calibration coefficients.

C. CLEANING (RINSE) CYCLE

At the conclusion of the measurement cycle, the **MINOS System** automatically cleans (rinses) **all** tubing and chambers that have come into contact with the blood sample and/or the diluted blood sample, prior to accepting the next blood specimen. At the end of the cleaning cycle, the sample needle is automatically moved into position and lowered to accept the next blood specimen.

D. RESULTS

During the cleaning cycle, results from the sample analysis are displayed on the front panel of the **MINOS System** and on the monitor, and are printed by the printer and/or sent directly to the laboratory's assigned computer via the RS232 computer interface.

E. DEFINITION OF HEMATOLOGIC PARAMETERS

The following list of parameters are measured or calculated directly by the **MINOS STE-L System**:

WBC: White Blood Cells (Leukocytes)

The number of white blood cells, per unit volume of whole blood, assayed in 10^3/uL (10^9/L [SI])[9]. N.B. This parameter also includes any nucleated red blood cells (NRBC), if present.

RBC: Red Blood Cells (Erythrocytes)

The number of red blood cells, per unit volume of whole blood, assayed in 10^6/uL (10^{12}/L[SI]).

[8] A third counting cycle will be initiated if the difference between the first and the second count is greater than 5% for WBC, RBC, and HGB, and greater than 10% for PLT.

[9] SI = International Units

HGB: Hemoglobin

Hemoglobin concentration/unit. The unit of measure is in g/dL (mmol/L [SI]).

HCT: Hematocrit

The percent volume of RBCs in whole blood. The unit of measure is % (% [SI]).

MCV: Mean Cell Volume

The mean (*average*) cell (corpuscular) volume of RBCs, in cubic microns (u^3).[10]

MCH: Mean Corpuscular Hemoglobin

The Mean Corpuscular (cell) Hemoglobin is the average weight of hemoglobin contained in each RBC. The MCH is measured in picograms (pg).[11]

MCHC: Mean Corpuscular Hemoglobin Concentration

The *average* concentration of hemoglobin per RBC measured in g/dL (mmol/L [SI]).

PLT: Platelets

The number of platelets per unit volume of whole blood measured in 10^3/uL (10^9/L [SI]).

LYM #: The number of lymphocytes per unit volume of whole blood, measured in 10^3/uL (10^9 [SI]).

LYM %: The ratio of the number of lymphocytes to the total number of leukocytes, measured in percent (%).

[10] Femtoliter = 10^{-15}; 1 femtoliter (fl) = $1u^3$.

[11] Picogram = 10^{-12}g (10^{-15} mol [SI]).

*See 43 and 44 in Bibliography

SECTION II

Installation Procedures and Special Requirements

SECTION II

INSTALLATION PROCEDURES and SPECIAL REQUIREMENTS

SPECIFICATIONS

A. Electrical Input

The ABX system should be supplied from an **independent, protected** circuit. It should not be installed next to a source of noise or in the same power line as a centrifuge, refrigerator, heavy duty motors, etc.....

Power Supply:

MINOS STE-L: 120 Volts (91 - 129 Volts), 60 Hz.

Consumption:

MINOS STE-L: Start-up = 250 W; Routine Operation = 150 W

B. Ambient Operating Temperature

MINOS STE-L: 59° - 98°F (15° - 37°C)

C. Humidity

Up to 95%, without condensation.

D. Dimensions:

MINOS STE-L:

Analyzer:	Height	14.5 in. (37.0 cm)
	Width	16.75 in. (40.0 cm)
	Depth	17.0 in. (43.0 cm)
Printer:	Height	3.23 in. (8.2 cm)
	Width	9.50 in. (24.0 cm)
	Depth	6.93 in. (17.6 cm)

E. Weight

MINOS STE-L:

Analyzer: 71 lbs. (32 kg)

Printer: 41 lbs. (2 kg)

F. Space and Accessibility

1. Easy access to the side door of the unit.
2. Sufficient room for work space around system.

G. Waste Disposal

Automatic

H. Recommended Diluent

MINOTON LMG

I. Recommended Lysing Reagent

MINOLYSE LMG

J. Recommended Cleaning Reagents

MINOTERGE
MINOCLAIR

K. Blood Sample Requirements

MINOS STE-L System: 25 uL

L. Dilutions

WBC: (WBC & HGB)1:240

RBC: (RBC & PLT) 1:20,000

M. Aperture Sizes

WBC: 100 Microns in diameter

RBC: 50 Microns in diameter

N. Hemoglobin

Wavelength: 540 nm
Band Width: 60 nm

O. Start-up

Automatic. Time required to perform analyses from turn-on of the instrument: **5 minutes.**

Time required to perform analyses from stand-by mode: **Immediate.**

P. Assay (operating) Cycle Time:

1 minute per sample.

Q. Data Storage:

The memory buffer stores all the hematological parameter results for up to 350 patient samples.

R. Carry-Over:

WBC:	Less than 0.5%
RBC:	Less than 0.5%
HGB:	Less than 0.5%
HCT:	Less than 0.5%
PLT:	Less than 0.5%

SYSTEM DESCRIPTION (GENERAL)

The MINOS STE-L System is comprised of the following:

EQUIPMENT:

- MINOS STE-L Automated Hematology Analyzer
- Roll PRINTER

REAGENTS:

- MINOTON LMG Diluent
- MINOLYSE LMG Lysing Reagent
- MINOTERGE Cleaning Detergent
- MINOCLAIR Cleaning Bleach

INSTALLATION OF THE MINOS STE-L SYSTEM

A. System Elements

1. **MINOS STE-L** Automated Hematology Analyzer
2. Roll PRINTER

B. Unpacking the Instrument

1. Place the shipping container on the floor.
2. Open the container CAREFULLY!
3. Remove the instrument, and place it on a **clean** laboratory workbench.
4. Be certain that the area selected is not in direct sunlight. Where possible, the area should be well-ventilated and maintained at a constant temperature. Allow a space of 4 to 8 inches (10 to 20 cm) behind the instrument, for proper ventilation.

C. Power Fuse

Insert the two 1.25 Amp fuses. The fuse housings are located in the rear of the **MINOS STE-L** instrument above the main power plug receptacle (See **Figure II-1**).

D. Printer Cable

Insert one end of the printer cable into the rear of the printer. Insert the other end of the cable into the appropriately labeled receptacle, located in the rear of the **MINOS STE-L** instrument.

E. Reagent/Waste Connections (See **Figure II-1**)

The input connectors for the reagents are located in the rear of the **MINOS STE-L** instrument. In the center of the connector panel there is a column of four connectors. Attach one end of the appropriately labeled tubing to the correct fitting:

#1 (Top) **MINOTON LMG** diluent. Connect the other end of the tubing to the **MINOTON LMG** diluent container.
#2 **MINOTERGE** detergent. Connect the other end of the tubing to the **MINOTERGE** detergent bottle.
#3 **MINOLYSE LMG** lyse. Connect the other end of the tubing to the **MINOLYSE LMG** lysing reagent bottle.
#4 (Bottom [waste]). Connect the other end of the tubing into a drain receptacle or empty waste container.

To the right of the four-connector column is the connector for **MINOCLAIR** bleach. Attach one end of the appropriately labeled tubing to this fitting. Attach the other end of the tubing to the **MINOCLAIR** bleach bottle.

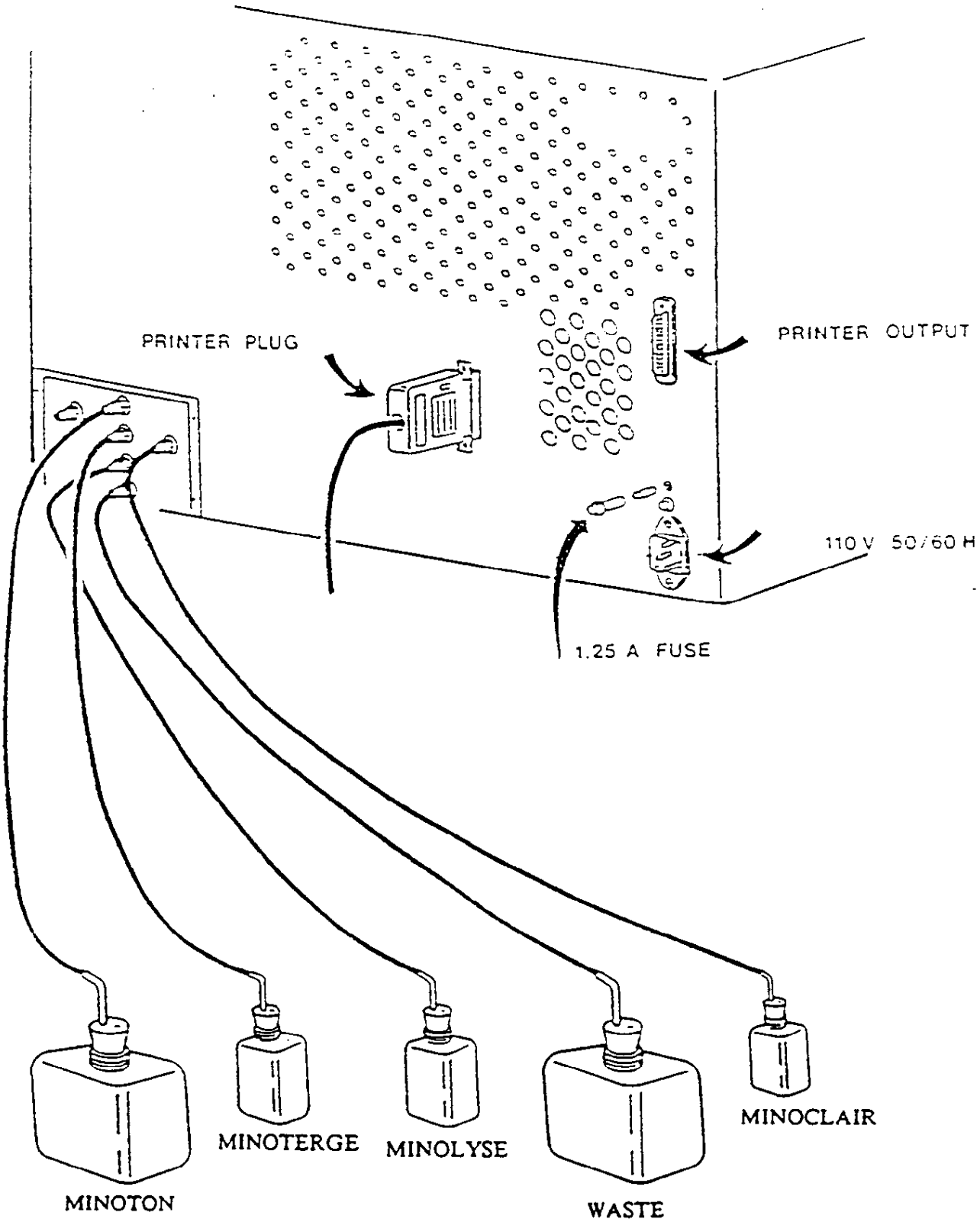
PRINTER PLUG
PRINTER OUTPUT
110 V 50/60 H
1.25 A FUSE
MINOTON
MINOTERGE
MINOLYSE
WASTE
MINOCLAIR

Figure II-1

The waste connector is located at the bottom of the row of four connectors. Attach the appropriately labeled tubing to this fitting.

NOTE: ALL REAGENTS (MINOTON LMG, MINOLYSE LMG, MINOTERGE, and MINOCLAIR) MUST BE AT THE SAME LEVEL AS THE INSTRUMENT (i.e. at benchtop level). The waste, however, may be located below the level of the MINOS STE-L instrument level.

F. Electrical Connections

The female end of the power cord plugs directly into the back of the MINOS STE-L System. The male receptacle is located in the rear of the instrument, below the two fuses. The male end of the power cord may be inserted directly into any 120 V/60Hz wall outlet. For maximum performance, it is recommended to use a dedicated line.

SYSTEM DESCRIPTION (DETAILED)

The **MINOS STE-L System** is composed of these standard items:

A) ANALYZER
B) PRINTER

The following is a description of each of the key components in the system.

A. Analyzer

The front panel of the **MINOS STE-L** Hematology Analyzer is made of impervious, flexible plastic for easy cleaning. Each key located on the front panel is touch-activated. In addition, each key is mounted with a visible indicator lamp and gives an audible "beep" when activated.

1. POWER ON/OFF SWITCH

The **power on/off switch** is used for switching the **MINOS STE-L** system "on" and "off." When the instrument is turned "on" the red **power on/off switch** will be lighted. When the instrument is turned "off" the light will be turned off also.

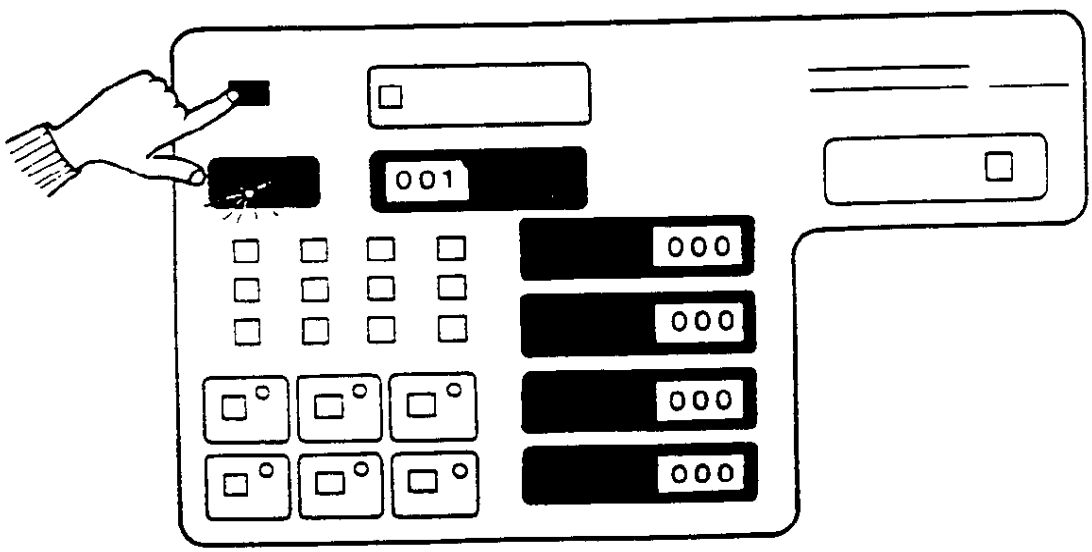

Figure II-3

It is recommended that the **MINOS STE-L System** be left in the "power-on" state. When the system is not being used, it will automatically switch into the **standby** mode and be ready for immediate sample analysis, when necessary. When the pneumatics have reached their proper operating condition, the red pressure indicator light will turn green.

2. REAGENT & CLEANING KEYS
(bottom left)

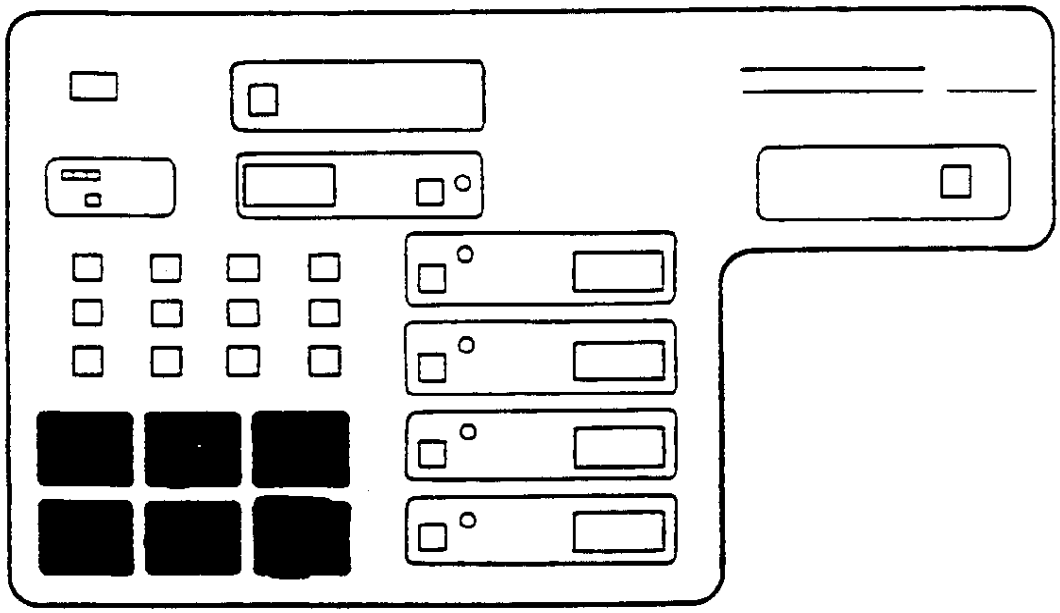

Figure II-4

a. **DILUENT RINSE** Key: This key is used to prime the diluent fluid lines after changing the **MINOTON LMG** diluent container. The key is also used for automatically rinsing the instrument lines with clean diluent.

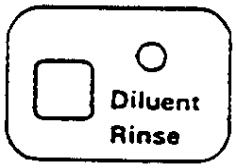

Figure II-5

b. **DETERGENT RINSE Key:** This key is used to activate the automatic detergent **(MINOTERGE)** cleaning cycle. At the end of this cycle, the instrument is automatically switched into the "standby" mode. The automatic detergent cleaning cycle is used at the end of each eight-hour working shift (when applicable).

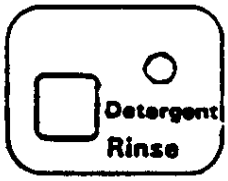

Figure II-6

c. **LYSE PRIME** Key: This key is used to prime the lysing reagent lines after changing the **MINOLYSE LMG** lyse container.

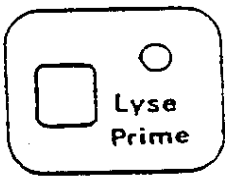

Figure II-7

d. **DRAIN CHAMBER key:** This key is used to drain the **WBC** and **RBC counting chambers**, as well as the **waste chamber.**

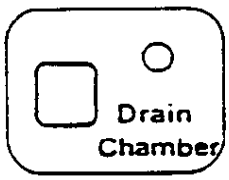

Figure II-8

e. **BACK FLUSH Key:** This key is used to force fluids back through the **WBC** and **RBC Apertures.** The **Back Flush** cycle is activated to clear any debris that may be obstructing either or both aperture(s).

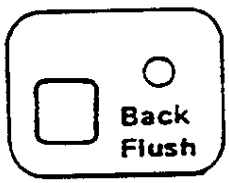

Figure II-9

f. **AUTOMATIC CLEANING Key:** This key automatically cycles **MINOCLAIR** (bleach reagent) and **MINOTERGE** (detergent) through the system, cleaning the **WBC** and **RBC Apertures** and **hemoglobin spectrophotometric chamber** as well as the associated tubings, followed by a complete rinse with **MINOTON LMG.** The duration of this cycle is five minutes.

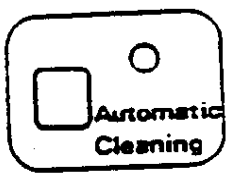

Figure II-10

3. NUMERIC KEYS
(Middle, Left)

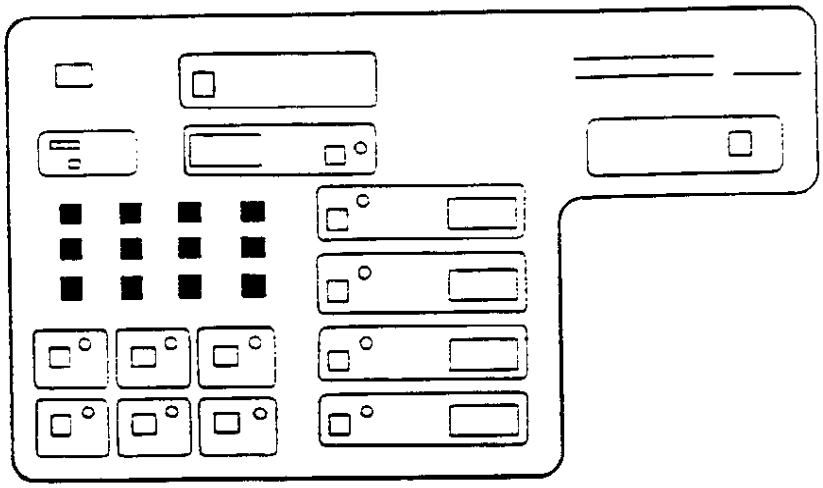

Figure II-11

The 12 number keys (0 - 9, + & -) are used to enter the patient identification code, the laboratory accession number, and to enter the required value for a selected parameter during instrument calibration.

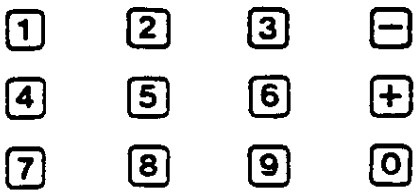

Figure II-12

4. PATIENT-IDENTIFICATION & CALIBRATION KEYS and DISPLAY WINDOWS
(Top Center and Right)

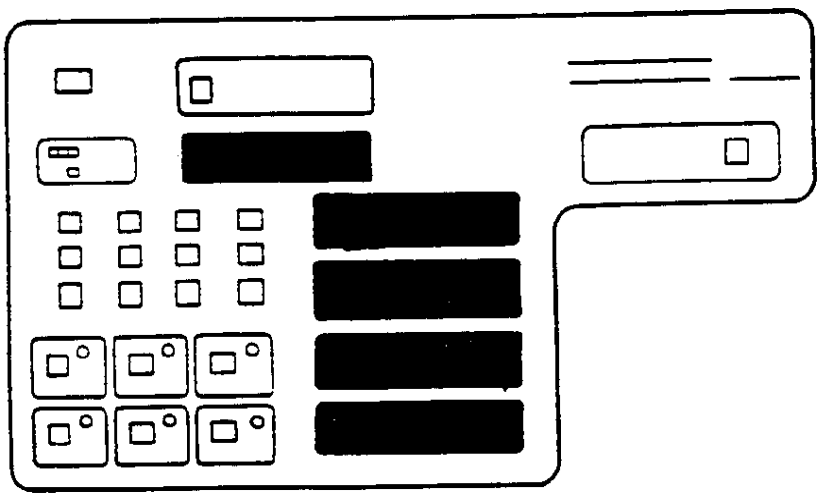

Figure II-13

These keys and related displays are used during calibration of the **MINOS STE-L System**. The keys, identified below, are used in conjunction with the **Numeric Keys (#3 above, Figure II-11)**, during calibration of the instrument.

a. **PLATELETS Key and Display Window:** This key is used during the calibration of the **platelet** parameter. The display window is used to display the **patient identification number**, the **calibration value**, or the **platelet results** from a blood sample analysis.

Figure II-14

b. **WHITE BLOOD CELLS Key and Display Window:** This key is used during the calibration of the **White Blood Cell** (WBC) parameter. This display window is used to display the **calibration value**, or the **WBC results** from a blood sample analysis.

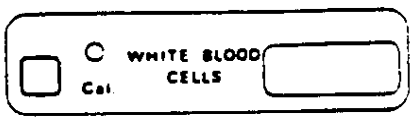

Figure II-15

c. **RED BLOOD CELLS Key and Display Window:** This key is used during the calibration of the **Red Blood Cell** (RBC) parameter. This display window is used to display the **calibration value**, or the **RBC results** from a blood sample analysis.

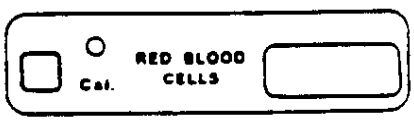

Figure II-16

d. **HEMOGLOBIN Key and Display Window:** This key is used during the calibration of the **hemoglobin** parameter. The display window is used to display the **calibration value**, or the **hemoglobin results** from a blood sample analysis.

Figure II-17

e. **HEMATOCRIT Key and Display Window:** This key is used during the calibration of the **hematocrit** parameter. The display window is used to display the **calibration value**, or the **hematocrit results** from a blood sample analysis.

Figure II-18

5. DATA & FUNCTION KEYS
(Top Center)

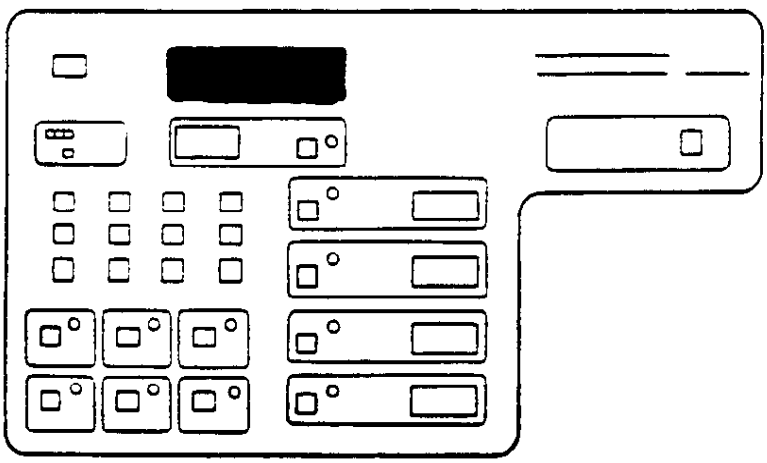

Figure II-19

a. **SPECIAL FUNCTIONS Key:** This key is used to access the various sub-routines described in the **OPERATING INSTRUCTIONS** Section of this manual.

Figure II-21

6. **ASPIRATE SAMPLE Key**
(Top Right)

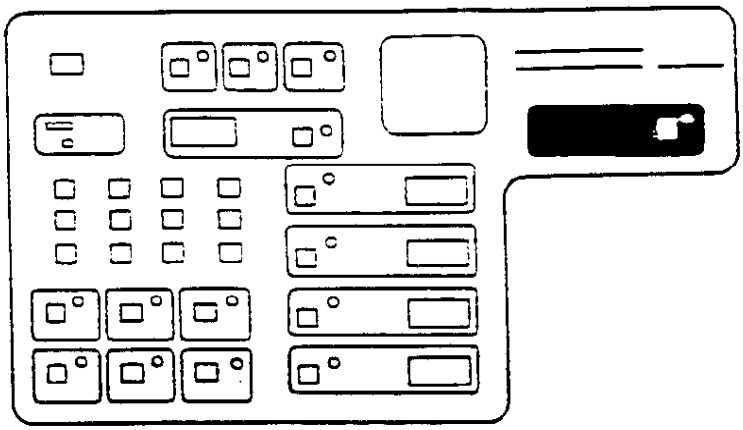

Figure II-21

This key is used to activate the **sample analysis cycle** of the MINOS STE-L System.

Figure II-22

B. PRINTER

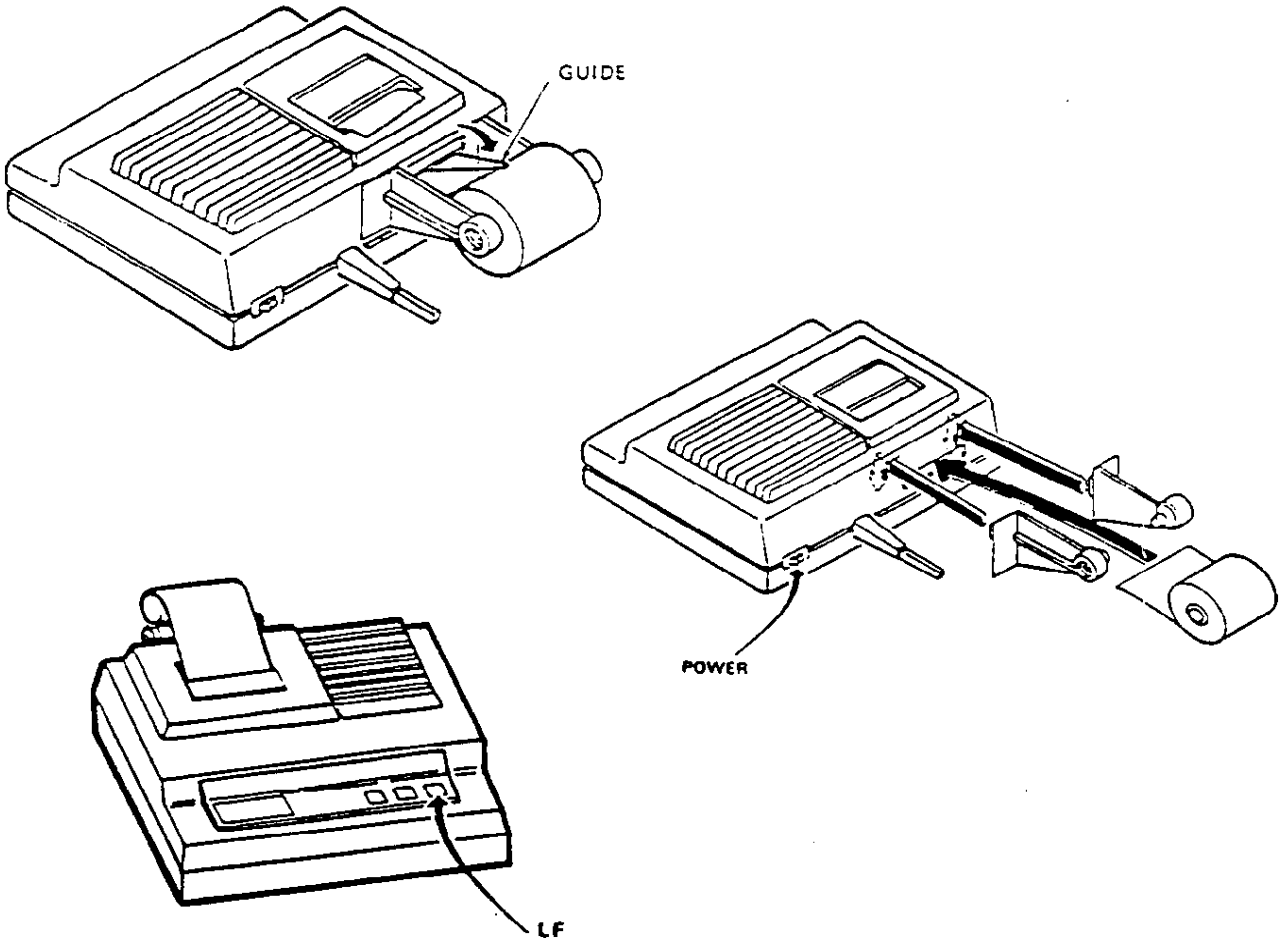

Figure II-23

1. **POWER ON/OFF Switch.** The **POWER ON/OFF** switch is located on the back of the printer. When the switch is turned to the **ON** position, the **POWER** Light Emitting Diode (LED) will be activated.

2. **LF Key.** The **LINE FEED (LF)** key is used to advance the paper one line at a time. It is used for form alignment.

3. **SEL Key.** The **Select (SEL)** key is used to place the printer "ON-LINE". When activated, it allows data from the **MINOS STE-L** to be automatically printed by the printer.

4. **DS Key.** The **De-Select (DS)** key is used to take the printer "OFF-LINE". No results will be printed when this key has been activated.

START UP AND CALIBRATION

The MINOS STE-L System has been calibrated at the factory prior to shipment. However, after installation, and before assaying any patient blood samples, it is mandatory that instrument calibration be re-verified.

A. START-UP

1. Prior to turning the **MINOS STE-L System ON**, verify the following:

   a. A sufficient level of **MINOTON LMG** diluent is connected to the instrument.

   b. A sufficient level of **MINOLYSE LMG** lysing reagent is connected to the instrument.

   c. A sufficient level of **MINOTERGE** detergent is connected to the instrument.

   d. A sufficient level of **MINOCLAIR** bleach reagent is connected to the instrument.

   e. The waste line is connected to the instrument and the opposite end of the line is securely placed in a drain or empty waste container.

   f. All pneumatic/reagent lines are properly connected.

   g. All electrical plugs are properly connected.

2. The MINOS STE-L System is turned **ON** in the following sequence:

   1st: ANALYZER power switch is turned **ON**.
   2nd: **PRINTER** power switch is turned **ON**.

   When the analyzer is turned **ON**, the instrument will automatically proceed through a number of internal self-checks to insure proper instrument functioning.

3. **Instrument Priming** is necessary when the instrument is initially installed or if it has been shut down in distilled water for extended storage. Once the instrument has been primed, this procedure can be eliminated from the daily start-up routine.

   a. Press the **DILUENT RINSE** key once; the red LED will light. The cycle initiates a diluent priming and rinse by sending 5 mL of **MINOTON LMG** diluent from the dispenser into the **mixing** and **RBC chambers.** The diluent is then transferred from the **mixing chamber** to the **WBC chamber.**

   b. Press the **ASPIRATE SAMPLE** key once. The LED will light.

All chambers are emptied via the **waste chamber**, after which the cycle is repeated. During the process above, diluent is transferred to the **hemoglobin chamber**.

The cycle is repeated a total of four times to insure that total priming of all the relevant tubing has occurred.

c. Press the **DETERGENT RINSE** key once and the LED will light. This cycle proceeds in the same manner as described in **b**, above; however, the instrument utilizes **MINOTERGE** detergent rather than diluent.

d. Press the **LYSE PRIME** key once and the LED will light. The instrument will automatically activate the lyse pump seven times (1 mL of **MINOLYSE LMG** lyse reagent per activation).

N.B.: Never initiate two complete LYSE PRIME cycles back to back (doing so will cause excessive foaming in the waste chamber.) ALWAYS **run at least one ASPIRATE SAMPLE cycle between two LYSE PRIME cycles.**

e. Press the **DILUENT RINSE KEY.**

f. Press the **ASPIRATE SAMPLE** key (LED will light) a second time. Check for proper operation of the aspirating needle, the proper filling of the **WBC and RBC chambers**, and the proper emptying of the **WBC and RBC chambers.** The cycle is completed when the LED on the **ASPIRATE SAMPLE** Key goes out.

B. Background Counts

A background count must be obtained following the daily start-up procedure and prior to assaying any patient blood samples. To obtain the background count of the instrument, simply press the **ASPIRATE SAMPLE** key; the instrument will perform the normal operation cycles automatically. At the conclusion of the counting cycle, the instrument will print out the results of the background counts.

The following is a list of acceptable background counts:

WBC:	**00.0 - 00.3**
RBC:	**0.00 - 0.03**
HGB:	**00.0 - 00.3**
PLT:	**000 - 005**

The **HCT** value may be ignored during the background counting cycle.

If the values obtained from the background count exceed the acceptable limits above, perform one **DILUENT RINSE** cycle. Repeat the background count. If the values obtained still exceed the acceptable limits, perform an **AUTOMATIC CLEAN** cycle, followed by one **DILUENT RINSE** cycle. Repeat the background count. If the background count is acceptable, proceed with the start-up procedure. If the background count remains unacceptable, contact **ABX Inc. Field Service** for assistance.

C. CALIBRATION

Calibration of the MINOS STE-L System is not necessary if the results of a full series of quality control assays demonstrate that the instrument values are within the specified ranges for all parameters, for all three levels of control (abnormal low, normal, and abnormal high).

The MINOS STE-L System retains all the calibration coefficients in its internal memory. Additionally, each of the directly measured parameters may be calibrated individually, as described below. The three erythrocyte indices (MCV, MCH, and MCHC) are calculated automatically by the microprocessor.

When Calibration of the MINOS STE-L System is necessary, the following points should be observed prior to beginning the calibration procedure.

- The instrument must be clean and achieving acceptable background counts.
- Instrument reproducibility (precision) must be demonstrated.

1. Control Material Calibration

   a. Calibrate the instrument according to the NORMAL control mean values.
   b. Check the LOW and HIGH control values.
   c. Check, if possible, with a known whole blood sample.

2. Calibration

   a. Calibrate the instrument according to the instructions provided in the calibration kit.

C-1. WHOLE BLOOD CALIBRATION PROCEDURE

Whole blood calibration is necessary for proper calibration of the **WBC, RBC, HGB, HCT, and PLT parameters.** The remaining parameters - MCV, MCH, MCHC do not require whole blood calibration. Accurate verification of the MINOS STE-L System calibration is accomplished by using normal whole blood and the following procedure:

1. Selection of Whole Blood Specimens

Obtain 20 normal, fresh whole blood specimens. The specimens designated must have normal hematologic parameter values, including normal morphology of all formed elements, and the donors must not be receiving any medication. Collection of the blood specimens, in evacuated tubes, must be performed correctly.

N.B.: Blood collection tubes used for the donation of the blood samples employed for calibration of the instrument must contain the same salt of EDTA as is used for the routine collection of patient blood sample for analysis.

2. **Pre-Calibration Values**

Each of the 20 specimens must be assayed three times on the MINOS STE-L System. Determine the average value for each specimen, for each of the directly measured parameters (WBC, RBC, HGB, HCT, and PLT). The averaging is performed by totaling the value of the three separate assays and dividing the subsequent total by three.

N.B.: The MPV parameter is calibrated by the ABX Inc. Field Service Engineer, using latex beads.

3. **Reference Values**

a. WBC Reference Values

WBC reference values are obtained by performing three semi-automated WBC counts on each of the 20 specimens, above. This will require that three separate dilutions be prepared for each specimen.

Total the three assays for each specimen and divide the resulting total by three to obtain the average value per specimen.

b. RBC Reference Values

RBC reference values are obtained by performing three semi-automated RBC counts on each of the 20 specimens, above. This will require that three separate dilutions be prepared for each specimen.

Total the three assays for each specimen and divide the resulting total by three to obtain the average value per specimen.

c. HGB Reference Values

The reference values for the hemoglobin parameter are obtained by strict adherence to the following procedure:

- Prepare a duplicate sample from each of the 20 whole blood specimens, using the **Drabkin method** of hemoglobin determination.
- Using a spectrophotometer and a newly prepared standard curve, determine the hemoglobin concentration of all the prepared samples.
- Total the hemoglobin concentration values obtained for each pair of duplicate samples and divide the result by two (2) to obtain the average hemoglobin concentration for each blood specimen.

d. HCT Reference Values

The HCT reference values are obtained by preparing three (3) hematocrit centrifuge tubes for each of the 20 whole blood specimens. Preparation requires that the hematocrit tubes be filled with well-mixed whole blood and sealed at the opposite end of the tube. Place the properly filled and sealed hematocrit tubes into a **hematocrit centrifuge** (refer to the manufacturer's operator's manual for the proper operation of the instrument).

Following centrifugation, determine the hematocrit value for each of the three tubes, for each of the 20 whole blood specimens. For each individual specimen, total the hematocrit values obtained for each of the three tubes and divide the total by three (3) to obtain the average hematocrit value per specimen.

e. PLT Reference Values

The platelet reference values are obtained using the phase-contrast microscopy technique. Prepare three (3) separate dilutions per whole blood specimen. Total the three platelet count results obtained for each specimen and divide the total by three (3) to obtain the average platelet value for each specimen.

4. Calibration Verification & Adjustment

The pre-calibration average values obtained from each measured parameter on the MINOS STE-L System must be compared to the reference average values obtained for each parameter. The percent difference in values obtained, if any, is determined by the following formula:

$$\frac{\text{Ref(Av)}}{100} : \frac{\text{Pre-Cal(Av)}}{X}$$

Where:

Ref(Av) = Reference Results (Average)
Pre-Cal(Av) = Pre-Calibration Results (Average)

Percent Difference = 100 - X

Having determined the percent difference, if any, the Operator may implement the following calibration corrections on the **MINOS STE-L System**, to regulate the individual parameters +/- to achieve the required Reference Assay Value.

C-2. COMMERCIAL CONTROL PROCEDURE:

When the following procedure is performed properly, this method is easier than the whole blood calibration procedure.

1. Take a normal blood control that has been assayed on the **MINOS STE-L System.**
2. Allow the vials to stand at room temperature for approximately 15 minutes.
3. Mix by rapid inversion until red blood cells are completely resuspended. DO NOT MIX MECHANICALLY.
4. Run the normal control two (2) times. Record the value for each parameter on the worksheet.
5. Calculate the mean value (average) for each parameter.
6. Compare mean value to the assigned values (use the tolerance limits given with the control).
7. If the mean value is not within the tolerance limits, adjust the instrument to the assigned values following the calibration procedure.
8. After calibration, check **LOW** and **HIGH Control** values by repeating steps 2 through 6.

C-3. COMMERCIAL CALIBRATOR PROCEDURE:

This method can be used the same way as the normal control above (steps 1 through 7). To make sure that your calibrator is still reliable, run three (3) levels of control (LOW, NORMAL and HIGH).

CALIBRATION PROCEDURE:

Find which of the 20 blood samples has its mean values per parameter closest to the *grand mean* of all 20 samples on the STE-L System. Assay it twice. While the second assay values are displayed, follow instructions a through f below. Enter the grand mean Reference Assay Value. **NOTE: You will enter the assigned values for the commercial control or calibrator procedure.**

a. Determine the Existing Calibration Coefficients

Press the SPECIAL FUNCTIONS key. Enter 991 on the numeric key pad. The existing calibration coefficients will be printed out.

b. WBC Calibration Adjustment

Press the WHITE BLOOD CELLS Cal. key. Three dashes (- -.-) will appear in the display area. Enter the reference value via the numeric key pad (NOTE: 3 digits must be entered [e.g. 081 = 08.1]).

Press the WHITE BLOOD CELLS Cal. key once again to exit the calibration mode. The new calibration coefficient is automatically stored in the system and presented in the display area.

c. RBC Calibration Adjustment

Press the RED BLOOD CELLS Cal. key. Three dashes (-.- -) will appear in the display area. Enter the reference value via the numeric key pad (NOTE: 3 digits must be entered [e.g. 315 = 3.15]).

Press the RED BLOOD CELLS Cal. key once again to exit the calibration mode. The new calibration coefficient is automatically stored in the system and presented in the display area.

d. HGB Calibration Adjustment

Press the HEMOGLOBIN Cal. key. Three dashes (- -. -) will appear in the display area. Enter the reference value via the numeric key pad. (NOTE: 3 digits must be entered [e.g. 102 = 10.2]).
Press the HEMOGLOBIN Cal. key once again to exit the calibration mode. The new calibration coefficient is automatically stored in the system and presented in the display area.

e. HCT Calibration Adjustment

Press the HEMATOCRIT Cal. key. Three dashes (- -.-) will appear in the display area. Enter the reference value via the numeric key pad (NOTE: 3 digits must be entered [e.g. 281 = 28.1]).

Press the **HEMATOCRIT Cal.** key once again to exit the calibration mode. The new calibration coefficient is automatically stored in the system and presented in the display area.

f. PLT Calibration Adjustment

Press the **PLATELETS Cal.** key. Three dashes (- - -) will appear in the display area. Enter the reference value via the numeric key pad (NOTE: 3 digits must be entered [e.g. 045 = 045]).

Press the **PLATELETS Cal.** key once again to exit the calibration mode. The new calibration coefficient is automatically stored in the system and presented in the display area.

Verification of any calibration adjustment must be made, if changes were made to any or all of the above calibration coefficients. Whole-blood verification of the calibration may be accomplished by the following procedure.

- Assay any blood specimens of known reference values on the **MINOS STE-L System**. Compare the grand average result for each parameter (the average result per parameter of all specimens) to the grand average result for each parameter obtained (Reference Assay Values). The **MINOS STE-L System** is considered to be **properly calibrated** if the difference in results does not exceed the following guidelines:

WBC	**±**	**0.3**
RBC	**±**	**0.05**
HGB	**±**	**0.3**
HCT	**±**	**1.0**
PLT	**±**	**8.0**

If any of the above parameters exceed the specified maximum limits, meticulously review all the procedures for that particular parameter. If no error in the procedure (or in the mathematical calculations) has been identified, call your **ABX Inc. Field Service Engineer** for assistance. If, however, all the above parameters fall within the stated limits, the **MINOS STE-L System** is considered properly calibrated. Following proper calibration, the **MINOS STE-L System** is ready for assaying patient blood samples (see **SECTION IV, OPERATING INSTRUCTIONS).**

SECTION III

Specimen Collection and Preparation

SECTION III

SPECIMEN COLLECTION and PREPARATION

25 uL[1] of a whole blood sample is automatically aspirated[2] by the **MINOS STE-L System.** Both venous and capillary blood samples (collected in properly anticoagulated micro-sample collection devices) are accurately processed by the **MINOS STE-L System.**

WHOLE BLOOD

Whole blood specimens are collected by venipuncture using EDTA (a salt of **ethylenediaminetetraacetic acid**) as the proper anticoagulant for hematological samples. The Operator is referred to **PROCEDURES FOR THE COLLECTION OF DIAGNOSTIC BLOOD SPECIMENS BY VENIPUNCTURE**, publication H3-A, published by the National Committee for Clinical Laboratory Standards (NCCLS), for detailed information on the collection of whole blood specimens.

For Capillary Blood Samples refer to **PROCEDURES FOR THE COLLECTION OF DIAGNOSTIC BLOOD SPECIMENS BY SKIN PUNCTURE**, publication H4-A, published by the National Committee for Clinical Laboratory Standards (NCCLS). Only 25 uL of the capillary blood sample is required for *whole blood analysis.*

WARNING

Fresh, human blood samples may be a source of infectious agents. The preparation and transportation of blood specimens may constitute a hazard if proper handling precautions are not followed. **Good laboratory practice** dictates that instructions for the handling of blood specimens and for the cleaning of contaminated surfaces be available for routine use. Such procedures should be observed at all times.

ANTICOAGULANT of CHOICE

The salts of ethylenediaminetetraacetic acid (EDTA), namely K_2, K_3, and Na_2, are the primary anticoagulants of choice and routinely available for most clinical hematology laboratory procedures. Heparin, Sodium Citrate, Ammonium Oxalate, and Potassium Oxalate are additional anticoagulants used for specialized procedures; however, these anticoagulants are NOT intended for use in conjunction with the **MINOS STE-L System.**

INTERFERING SUBSTANCES

See **SECTION V: KNOWN INTERFERING SUBSTANCES.**

[1] uL = microliter.

SPECIMEN STORAGE

Whole blood specimens collected in EDTA should be analyzed on the **MINOS STE-L** System within four (4) hours, from time of collection. Specimens that **require PLATELET counts SHOULD** be kept at room temperature and should not be refrigerated. Specimens **not** requiring a **PLATELET** count may be stored at 4°C (39°F) for up to 24 hours.

For the lymphocyte percentage and absolute number, the best results are obtained when specimens are analyzed between 15 minutes and 6 hours from time of collection.

SECTION IV

Operating Instructions

SECTION IV

OPERATING INSTRUCTIONS

GENERAL

The following **OPERATING INSTRUCTIONS** are presented to enable optimum performance of the **MINOS STE-L System.**

MATERIALS REQUIRED

1. **MINOS STE-L Analyzer**

2. **MINOS STE-L Printer**

3. **Reagents**

    a. MINOTON LMG Diluent
    b. MINOLYSE LMG Lysing Reagent
    c. MINOTERGE Detergent
    d. MINOCLAIR Cleaning Reagent

MATERIAL REQUIRED, BUT NOT INCLUDED

1. Whole blood control material (3-level)

    a. Abnormal Low
    b. Normal
    c. Abnormal High

2. Mixer for blood tubes

3. Test tube rack

QUALITY CONTROL

An effective quality control program must be established for the assaying of human whole blood samples on the **MINOS STE-L System.** Good clinical laboratory practice requires strict adherence to the quality control program to ensure the quality of the assaying procedure.

ABX Inc. recommends that a quality control program be established to monitor the **MINOS STE-L System.** The program established should include three levels of control:

1. Abnormal Low
2. Normal
3. Abnormal High

There are commercially-available control materials that provide the three levels of control. Specific handling procedures for the use of the control material are established by the individual manufacturers and are detailed in the specific product insert sheets.

ABX Inc. further recommends establishing the use of Levy-Jennings charts to monitor the daily quality control values. When using the Levy-Jennings charts, any value (for any parameter) falling outside the ± 2SD (Standard Deviation) must be investigated. The **ABX Inc. Field Service Department** should be notified if the operator is unable to establish the cause for and/or correct any problem causing the quality control material to fall outside the ± 2SD limits.

Quality control materials should be assayed at least once during each work shift, prior to assaying any patient blood samples. The quality control materials are assayed using the same method used for blood samples analysis.

DAILY START-UP PROCEDURE

Prior to turning on the **MINOS STE-L System**, check for the following conditions:

1. All electrical plugs are properly connected to 120V outlets.

2. All reagent lines are properly connected to the appropriate reagents.

3. The waste line is properly positioned in the waste drain (or container).

4. A sufficient level of reagents is available to complete the assays:

   a. **MINOTON LMG** Diluent
   b. **MINOLYSE LMG** Lysing Reagent
   c. **MINOTERGE** Detergent
   d. **MINOCLAIR** Cleaning Reagent

TURNING SYSTEM ON

The **MINOS STE-L System** is turned **ON** by following the sequence listed below:

1st : **MINOS STE-L** Analyzer power switch is turned **ON.**

2nd : **PRINTER** power switch is turned **ON,** followed by the **SEL** key.

When the **MINOS STE-L System** is first turned **ON**, the **Vacuum** and **Pressure** indicator lamps will be **red**, indicating that the instrument has not reached proper vacuum/pressure for operating the analyzer. Within a few seconds, however, the **Vacuum** indicator, and then the **Pressure** indicator, will turn **green**, indicating readiness.

> **NOTE:** Until both the **Vacuum** and **Pressure** indicator lamps have turned **green**, the system will <u>not</u> accept any samples for analysis.

When the system is first turned **ON**, the analyzer performs a series of internal quality control checks to make certain that all operating systems within the instruments are functioning properly.

REAGENT PRIMING (No Reagents in the System)

This procedure is followed during initial set-up of the **MINOS STE-L System**, or when reagent containers are changed. The procedure ensures that all reagent lines will be primed with the appropriate reagents, and cleared of any air or air bubbles.

1. **DILUENT PRIME:**

   Press the **DILUENT RINSE** key. The instrument will automatically perform several priming cycles and fill the appropriate lines and chambers with **MINOTON LMG** diluent. It will also perform an automatic sample cycle (background) with no data transmitted to the printer or monitor.

2. **DETERGENT PRIME:**

   Press the **DETERGENT RINSE** key. The instrument will automatically perform several priming cycles and fill the appropriate lines and chambers with **MINOTERGE** detergent.

3. **LYSE PRIME:**

   Press the **LYSE PRIME** key. The instrument will automatically perform several priming cycles and fill the appropriate lines and chambers with **MINOLYSE LMG** lysing reagent. Since this particular reagent causes considerable foaming, an additional **DILUENT RINSE** is required to clear the system (Step #4, below).

   NOTE: <u>Never</u> run two <u>LYSE PRIME</u> cycles in immediate succession, since excessive foaming of the reagent will result. <u>Always</u> run at least one ASPIRATE SAMPLE in between.

4. **DILUENT RINSE:**

   Press the **DILUENT RINSE** key. The instrument will automatically perform several cycles, rinsing the appropriate chambers with clean **MINOTON LMG** diluent, removing the excess foam, and will perform an automatic sample cycle (background) with no data transmitted to the printer or monitor.

5. **SAMPLE CYCLE:**

   Press the **ASPIRATE SAMPLE** key. The instrument will automatically perform one complete assay cycle.

SYSTEM FLUSH (Assumes ALL reagents have been primed) - Normal Operation:

Press the **DILUENT RINSE** key. The instrument will automatically perform several cycles, clearing the appropriate lines and chambers with **MINOTON LMG.**

N.B.: **During normal operation, the MINOS STE-L System will be shut-down with MINOTERGE detergent in the appropriate lines and chambers.**

BACKGROUND COUNTS

The background count must be determined prior to any blood sample analyses. A background count is initiated by pressing the **ASPIRATE SAMPLE** key. the instrument will complete one entire assay cycle and display the results, as well as print out the results. During usual operating conditions the following are the values that must not be exceeded during a background cycle:

WBC	±	00.3
RBC	±	0.03
HGB	±	00.3
HCT	±	00.3
PLT	±	005

If, for any reason, the values obtained exceed the above limits, perform a **DILUENT RINSE** cycle followed by another background count. If the limits are still exceeded, perform an **Automatic Cleaning** cycle by pressing the **AUTOMATIC CLEANING** key. Following the Automatic Cleaning cycle, perform a **DETERGENT RINSE** cycle, a **DILUENT RINSE** cycle, and another **Background Count.**

CALIBRATION

See SECTION II: **Installation Procedures and Special Requirements, "Start-up and Calibration".**

PRE-SETTING (SPECIAL FUNCTIONS)

The **MINOS STE-L System** has several Operator options available, called **SPECIAL FUNCTIONS.** These options may be initiated prior to the actual assaying of the blood specimens. To initiate any of the **SPECIAL FUNCTIONS,** press the **SPECIAL FUNCTIONS** key. When this key is pressed, a "beep" will be heard, and the displays for **PLATELETS, WHITE BLOOD CELLS, RED BLOOD CELLS, HEMOGLOBIN,** and **HEMATOCRIT** will go blank; only the decimal points will remain lighted.

After pressing the **SPECIAL FUNCTIONS** key, enter any one of the following **three-digit codes** on the numeric keypad:

■ 990 - CHANGES THE DATE

Enter 990 on the number keys. Two dashes will appear in the **RBC**, **HGB**, and **HCT** display areas. Enter the date as follows:

2 digits for the **MONTH**.

2 digits for the **DAY**.

2 digits for the **YEAR**.

NOTE: After each of the two-digit pair is entered, the instrument automatically advances to the next data entry level.

After entering the complete date, press the **SPECIAL FUNCTIONS** key once again to exit.

NOTE: After the date has been changed, all stored data will be erased from memory when an **ASPIRATE SAMPLE** cycle is run.

■991 - **Prints out Calibration Coefficients**

Enter **991** on the number keys. The printer will automatically print out the calibration coefficients stored in the instrument's memory.

■992 - **Prints out internal operating program version number.**

Enter **992** on the number keys. The printer will automatically print out the version number of the operating program.

■ REPRINTING THE RESULTS OF A SINGLE SAMPLE ASSAY.

Press the **SPECIAL FUNCTIONS** key. Enter the required patient number on the number keys. The specified patient result will be automatically printed out on the printer.

MEASUREMENT OF WHOLE BLOOD SAMPLES

The MINOS STE-L System stores up to 350 patient assay results for a given date. The results are stored even if the System is disconnected from electricity. Whenever the date is changed, however, the patient assay results that have been stored in memory are automatically erased when the next sample is run.

1. Prime the **MINOS STE-L System** with a blood sample.

2. Enter the patient number on the number key pad. Be certain to enter three digits, e.g. *PATIENT #1 = 001*. If the blood samples are sequentially numbered, the system will automatically increment the patient number by one digit. However, non-sequential numbering systems require that the new patient number be entered manually prior to sample analysis.

3. Place the well mixed whole blood sample under the aspiration needle. Raise the blood sample until the aspiration needle is well into the blood sample.

4. Press the **ASPIRATE SAMPLE** key. The **MINOS STE-L System** will automatically aspirate 25 uL of the whole blood sample. The assay in process light will turn **ON** and stay on during the analysis. While this light is on, no additional blood sample can be processed.

Following aspiration, the needle will automatically rise out of the blood sample.

> **CAUTION: DO NOT REMOVE THE BLOOD SAMPLE UNTIL THE ASPIRATING NEEDLE IS COMPLETELY REMOVED FROM THE BLOOD SAMPLE.**

The **MINOS STE-L System** will automatically process the blood sample. At the end of analysis, the assay in process light will turn **OFF**, indicating that the system is **ready** to accept the next blood sample.

5. The results of the analysis are automatically stored in memory and printed on the printer.

> NOTE: If a blood sample is repeated using the same PATIENT #, only the last analysis is stored in the memory of the system. To store both the original and the repeat assay results it is recommended that a code be established for repeat samples; e.g. all 300 series numbers are repeat assays.

6. The system will automatically go into **STANDBY** mode if the instrument has not been used for the eight to nine minutes. While in **STANDBY**, the **MINOS STE-L System** shuts down the pressure and vacuum pumps; however, the system takes only a few seconds to return to the **READY** mode and begin sample analysis again.

SAMPLE ALERTS

During the processing of data from blood sample analysis, the **MINOS STE-L System** examines specific characteristics of the blood analyzed. Once the data is analyzed and reduced by the microprocessor, the results are printed out on the Printer.

Along with the assay results, an **ALERT (Flag)** may be printed next to the parameter in question. The following is a list of the **ALERTS** and their definitions.

Alert	Definition
***	This ALERT next to the WBC, RBC, HCT, or PLT parameter indicates that the system performed three counts on the same sample (dilutions), but that all three counts differed significantly - outside the system's established limits. **When this ALERT appears, the analysis should be repeated.**
S	This ALERT next to the Test Number and preceding the result of a particular parameter indicates that the system performed three counts on the sample (dilutions), and two of the three counts were valid - within the system's established limits.
DIL*	This ALERT next to the WBC or HCT parameter indicates that the linear range of the system, for the particular parameter, has been exceeded. Such blood samples must be diluted, and the diluted sample re-assayed. EXAMPLE: Add 0.5 mL of well-mixed whole blood to 0.5 mL of MINOTON LMG diluent. Mix well. Assay the dilution on MINOS STE-L System. Multiply the results by two (2). If the ALERT still appears, additional dilutions will be required. Be certain to multiply the results by the dilution factor used.
***MIC	This ALERT next to the PLT parameter indicates the presence of microcytic red blood cells (microcytes) in the PLATELET MEASUREMENT ZONE.
SCH	This ALERT next to the PLT parameter indicates the presence of Giant Platelets (Macrothrombocytes), platelet aggregates, schistocytes, and/or microcytic red blood cells (microcytes) in the PLATELET MEASUREMENT ZONE.
***SCL	This ALERT next to the PLT parameter indicates the presence of cellular fragments, debris, etc. in the 2 to 3 cubic micron zone. When this ALERT occurs, a second assay should be performed on the same blood sample. If the alert is still present, press the **AUTOMATIC CLEANING** button. After the cleaning cycle has been completed, repeat the assay on the same blood sample. If the ALERT persists, a manual platelet count should be performed.

L1 This ALERT, appearing below the results of the lymphocyte %, #, indicates the presence of abnormal cells (smaller than normal lymphocytes). These may be platelet aggregates and/or nucleated red blood cells (NRBC). Confirmation is made through thorough examination of the stained blood film.

L2 This ALERT, appearing below the results of the lymphocyte %, #, indicates the presence of an increased number of mononuclear cells, and no clear distinction can be made between the mononuclear and granulocytic populations. The area between 130 to 160 um^3 indicates the presence of lymphoblasts, myelocytes, atypical lymphs, or basophils.

GENERAL MAINTENANCE OF THE SYSTEM

Refer to **SECTION VII** for important information regarding maintenance before, during, and after assaying.

The MINOS STE-L System is equipped with automatic, microprocessor-controlled cleaning commands. For the Operator, this means "press a key", and the cleaning is automatically controlled.

1. **DAILY MAINTENANCE**

a. Each 50 Samples:

An **AUTOMATIC CLEANING** cycle will be ran after every 50 samples *automatically.*

b. End of Assaying/End of Work Shift:

Press the **AUTOMATIC CLEANING** key if you run less **than** 50 samples a day.

Press the **DETERGENT RINSE** key. Leave the instrument in this condition until needed again, e.g. the next work shift or the next work day.

2. **WEEKLY MAINTENANCE:**

Press the **DETERGENT RINSE** key. Leave the instrument in this condition for as long as possible (preferably overnight) but for at least four (4) hours.

NOTE: An **AUTOMATIC CLEANING** cycle will be ran after every 50 samples *automatically.* This will eliminate the need for **DETERGENT RINSE** cycles as long as monthly maintenance is followed.

3. **MONTHLY MAINTENANCE:**

a. Press the **DRAIN CHAMBER** key. This drains the **RBC, WBC,** and **waste chambers.**

b. Fill a 5 mL or 10 mL syringe with **MINOCLAIR.**

c. Fill the **mixing chamber** with **MINOCLAIR** almost to the top.

d. Fill the **RBC chamber** with **MINOCLAIR** almost to the top.

e. Press pinch valves #8 and #81 at the same time. This will transfer the **MINOCLAIR** from the **mixing chamber** to the **WBC chamber.**

f. Press pinch valves #16 and #17 at the same time, and hold for 5 to 10 seconds. Release and repeat again. This will transfer the **MINOCLAIR** from the **RBC mixing chamber** to the **WASTE chamber.**

g. Press the **DRAIN CHAMBER** key on the front panel.

h. Press the **DILUENT RINSE** key on the front panel.

i. Press the **ASPIRATE SAMPLE** key and check the background counts.

NOTE: This procedure takes five (5) minutes.

4. **ADDITIONAL MAINTENANCE:**

The **MINOS STE-L System** does not, except for the procedures above, require any Operator-initiated maintenance on the system. During the Warranty Period - or under FULL Service Contract (after the one year warranty period) - the **ABX Inc. Field Service Engineer** will routinely service, clean, and replace worn parts, and install any system updates, once every four (4) months.

SECTION V

Performance Characteristics

SECTION V

PERFORMANCE CHARACTERISTICS

PERFORMANCE SPECIFICATIONS

The performance characteristics specified in this manual apply only to the **MINOS STE-L** System. Furthermore, the specifications apply only when the systems have been properly maintained - as described in this manual - using **MINOTON LMG** diluent, **MINOLYSE LMG** lysing reagent, **MINOTERGE** detergent, and **MINOCLAIR** cleaning reagent.

Precision:

Precision of the instrument is defined as the coefficient of variation (CV). The data presented is based on a minimum of 30 determinations of the same blood sample.

PARAMETER	%CV
WBC, @ 10.0 X 10^9/L	Less than 2.0%
RBC, @ 5.00 X 10^{12}/L	Less than 2.0%
HGB, @ 15.0 g/dL	Less than 1.0%
HCT, @ 45.0 %	Less than 2.0%
PLT, @ 300 X 10^9/L	Less than 5.0%
LYM, @ 10.0 X 10^9/L	Less than 5.0%

Accuracy:

The **MINOS STE-L System** is Operator-adjustable; consequently, adjustments made to the system permit agreement to predetermined *reference values*.

Linearity:

Linearity is defined as the operating range limit of the system, per parameter. The linearity limits, per parameter, are listed below. To obtain comparable results, multiple readings must be taken at each data position in order to eliminate any statistical variances.

PARAMETER	LINEARITY	RANGE LIMITS
WBC (X10^9/L)	0.1 - 80.0	0.2 or 3% (whichever is greater)
RBC (X10^{12}/L)	0.5 - 9.0	0.03 or 2%(whichever is greater)
HGB (g/dL)	2.0 - 25.0	0.2 or 2%(whichever is greater)
HCT (%)	10.0 - 75.0	3%
PLT (X10^9/L)	20 - 1,800	5%

Expected Values:

WBC	4.8 - 10.8	X 10^9/L
RBC	4.20 - 6.20	X 10^{12}/L
HGB	11.8 - 17.5	g/dL
HCT	35.0 - 52.0	%
MCV	82.0 - 98.0	u^3
MCH	28.0 - 32.0	pg
MCHC	31.0 - 35.9	g/dL
PLT	140 - 350	X 10^9/L
LYM%	20.0 - 45.0	%
LYM#	1.0 - 4.0	X 10^9/L

PERFORMANCE CHARACTERISTICS

The data described in this section were obtained using a typical **MINOS STE-L System**, maintained in accordance with the operating instructions described in this manual.

Precision:

The precision study was performed on a fresh blood sample. Forty-one (41) consecutive assays were performed and the results are listed below.

PARAMETER	MEAN	%CV
WBC	6.6 X 10^9/L	1.59%
RBC	5.20 X 10^{12}/L	1.02%
HGB	14.9 g/dL	0.66%
HCT	43.6 %	1.28%
MCV	83.9 u^3	0.51%
MCH	28.7 pg	1.20%
MCHC	34.2 g/dL	1.38%
PLT	252 X 10^9/L	2.90%
%L	34.8%	4.53%
#L	2.27 X 10^9/L	3.46%

Accuracy:

The accuracy study was performed on 120 fresh blood samples, from both hospitalized patients and outpatients. The study compared the results of the **MINOS STE-L System** versus the results of the hospital's Coulter S-Plus IV System[1]. The results of the directly measured parameters are listed below.

PARAMETER	r	RANGE
WBC	0.9977	0.4 to 26.9
RBC	0.9921	2.43 to 5.44
HGB	0.9953	5.1 to 16.9
HCT	0.9845	17.0 to 49.0
PLT	0.9710	23 to 996

[1] Coulter and S-PLUS IV are registered trademarks of Coulter Electronics, Inc., Hialeah, FL.

SECTION VI

Operational Precautions, Limitations, and Hazards

SECTION VI

OPERATIONAL PRECAUTIONS, LIMITATIONS, and HAZARDS

The information included in this section summarizes Operator and procedural alerts that are contained in other sections of this Operator's Manual. Also included are **additional** operational precautions.

LIMITATIONS

The **MINOS STE-L System** measures the hematologic characteristics of human blood samples, collected in EDTA (see **SECTION V: Performance Characteristics**, under **Precision** and **Accuracy**.) Any result outside the stated linearity and/or accuracy limits requires verification by other methods.

When results obtained are outside the operational limits of the **MINOS STE-L System**, successful measurement of the abnormal blood specimen may be achieved by preparing additional dilutions of the sample.

EXAMPLE:

To prepare a blood sample found to have an abnormally high result, add 0.5 mL of the blood to 0.5 mL of **MINOTON LMG** diluent, mix well, and re-assay. Multiply the subsequent result by (2).

> NOTE: When a blood sample is diluted and assayed on the **MINOS STE-L System**, the **WBC, RBC, HGB, HCT, PLT, LYM#**, parameter must be multiplied by the dilution factor. The **MCV, MCH, MCHC, LYM%**, parameter *should not* be multiplied by the dilution factor.

REAGENTS

Precise operation of the **MINOS STE-L System** requires the use of reagents that have not exceeded their date of expiration, and that remain uncontaminated. All reagents used for the **MINOS STE-L System** must be examined prior to use and even during use for obvious precipitation (particulate matter) and/or microbial growth. Also, any changes in the color of the reagents (namely **MINOTON LMG** diluent and **MINOLYSE LMG** lysing reagent) may cause erroneous results, especially in the hemoglobin parameter. **If any of the above conditions exist do not use the reagent.**

> NOTE: The presence of any particulate matter and/or the presence of microbial growth may lead to falsely elevated counts.

The reagents for the **MINOS STE-L System - MINOTON LMG** diluent, **MINOLYSE LMG** lysing reagent, **MINOTERGE** detergent, and **MINOCLAIR** cleaning reagent - have been devised specifically for use in this system. The use of any reagents in the **MINOS STE-L System** other than those listed above may cause erroneous results and/or damage to certain components.

CLEANING

In SECTION IV: Operating Instructions, specific start-up, shutdown, daily and weekly cleaning methods are listed. The cleaning procedures identified are mandatory for the proper use and operation of the **MINOS STE-L System. FAILURE TO EXECUTE ANY OF THESE RECOMMENDED PROCEDURES MAY RESULT IN DECREASED RELIABILITY OF THE SYSTEM.**

BLOOD SPECIMENS

Verification of any abnormal blood specimens should be performed using **reference methods** and other standard laboratory procedures for the conclusive verification of results. The sections below list **known** limitations of automated blood cell counters which use the principle of impedance.

KNOWN INTERFERING SUBSTANCES

WBC (White Blood Cells [Leukocytes]):

Evaluated WBC results that exceed the linearity limits of the system will require dilution of the blood sample. Re-assaying the diluted sample may help to obtain the correct assay value.

Immature nucleated red blood cells (**NRBC**) will be counted in the WBC (White Blood Cell) parameter. If the number of nucleated red blood cells is sufficient to activate an **L1** ALARM, such interference will be detected. However, the differential white blood cell count - performed on the stained blood film - will reveal the presence of NRBC's. Following the differential white blood cell count, the WBC assay value must be corrected for the presence of nucleated red blood cells.

The formula utilized for correcting the WBC parameter, when nucleated red blood cells are present, is:

$$\text{CORRECT WBC} = \frac{\text{Uncorrected WBC X 100}}{\text{100 + \# of NRBC/100 WBC}}$$

In particular rare instances, the erythrocytes in the blood sample may not completely lyse. These non-lysed red blood cells may be detected on the WBC Histogram as an elevated baseline on the left side (leading edge) of the lymphocyte population. Non-lysed erythrocytes will cause a falsely elevated WBC count.

RBC (Red Blood Cells [Erythrocytes]):

The red blood cell dilution contains all the formed elements in the blood: erythrocytes, leukocytes, and platelets. During the counting of the erythrocytes (red blood cells), platelets are not counted, since their size falls below the minimum threshold. Leukocytes (white blood cells), on the other hand, are included in the RBC count. However, since the normal ratio between red blood cells and white blood cells is so extreme, the influence of the WBC on the RBC count is negligible. In rare cases where the WBC is extremely high, the RBC count may need to be corrected, especially if the RBC count is extremely low.

Agglutinated red blood cells may cause a falsely decreased RBC count. Blood samples containing the agglutinated red blood cells may be identified by observing abnormal MCH and MCHC values, as well as by examination of the stained blood film.

HGB (Hemoglobin):

Turbidity of the blood sample due to any number of physiologic and/or therapeutic factors may produce falsely elevated HGB results. To obtain accurate hemoglobin results when increased turbidity of the blood sample occurs, determine the cause of the turbidity and follow the appropriate method below:

1. Elevated WBC: An extremely elevated WBC will cause excessive light scatter. In these cases use reference (manual) methods. The diluted sample should be centrifuged, and the supernatant fluid measured on a spectrophotometer.

2. Elevated Lipids: Elevated lipids in the blood sample will give the plasma a "milky" appearance. Accurate hemoglobin determinations can be achieved by using reference (manual) methods and a plasma blank.

Increased turbidity may also be seen in cases where the red blood cells are resistant to lysing. This condition will cause a falsely elevated HGB result, but may be detected by observing the abnormal MCH, MCHC values, and the increased baseline on the leading edge of the WBC Histogram.

Erroneous hemoglobin results will cause the results of the MCH and MCHC to be erroneous as well.

HCT (Hematocrit):

Red blood cell agglutination may produce an erroneous HCT value. Red blood cell agglutination may be detected by observing the abnormal MCH and MCHC values, as well as by examination of the stained blood film. In such cases, manual methods may be required to obtain an accurate HCT value.

MCV (Mean Corpuscular Volume):

Red blood cell agglutination may produce an erroneous MCV value. Red blood cell agglutination may be detected by observing the abnormal MCH and MCHC values, as well as by examination of the stained blood film. In such cases, manual methods may be required to obtain an accurate MCV value.

Excessive large platelets and/or the presence of an excessively high WBC count may interfere with the accurate determination of the MCV value. In such cases, careful examination of the stained blood film may reveal the error.

MCH (Mean Corpuscular Hemoglobin):

The MCH is a function of the HGB value and the RBC count. The limitations listed for the HGB and RBC will have an effect on the MCH and may cause erroneous values.

MCHC (Mean Corpuscular Hemoglobin Concentration):

The MCHC is a function of the HGB and HCT values. The limitations listed for the HGB and HCT will have an effect on the MCHC and may cause erroneous values.

PLT (Platelets):

Very small erythrocytes (microcytes), erythrocytic fragments (schistocytes), and white blood cell fragments may interfere with the proper counting of platelets. Blood samples containing agglutinated erythrocytes may trap platelets, causing an erroneously low PLT count. The presence of agglutinated erythrocytes may be detected by observation of abnormal MCH and MCHC values and by careful examination of the stained blood film.

Giant Platelets in excessive numbers may cause an erroneously low PLT count since these large platelets exceed the upper threshold for the platelet parameter and are not counted.

Reference (manual) methods may be necessary to obtain an accurate PLT count.

LYM# (Lymphocyte Count [Absolute]):

The lymphocyte count is derived from the WBC count. The presence of nucleated red blood cells (NRBC), certain parasites, and erythrocytes that are resistant to lysis may interfere with an accurate LYM# count.

Limitations listed for the WBC count pertain to the LYM# as well.

LYM % (Lymphocyte Percentage):

The lymphocyte percent is a function of the WBC count and the number of lymphocytes. The presence of nucleated red blood cells (NRBC), certain parasites, and erythrocytes that are resistant to lysis may interfere with an accurate LYM% count.

Limitations listed for the WBC count pertain to the LYM% as well.

PRECAUTIONS

General

1. All abnormal blood specimens must be thoroughly investigated and the results verified.

2. Excessively elevated WBC, RBC, and PLT counts may result in carryover to the next blood specimen. When abnormally elevated counts are encountered in one specimen, it is recommended that the next blood specimen be assayed twice on the **MINOS STE-L System**, and that the results of the second assay be used in the reporting of the hematology values.

3. It is the responsibility of the Operator to establish and maintain a log that correlates individual test numbers with the appropriate blood specimens.

4. It is the responsibility of the Operator to be familiar with the Operating Procedures of the **MINOS STE-L System**. Failure to operate the system properly may result in damage to the components.

5. The Operator must observe all results for ALARMS and verify the results of the assay.

6. It is the responsibility of the Operator not to use any chemicals that are not specifically recommended for use with the **MINOS STE-L System**. Failure to use only those reagents specified for use on the **MINOS STE-L System** may result in damage to the components.

7. DO NOT place any reagents for use with the **MINOS STE-L System** above or below the level of the instrument. All reagents must be at the same level as the instrument.

8. Replacement fuses of the same value must be used in the **MINOS STE-L System** components.

9. The **MINOS STE-L System** is intended for **IN VITRO DIAGNOSTIC USE ONLY**. Only venous or capillary whole blood, properly collected in EDTA, should be used.

SECTION VII

Service and Maintenance

SECTION VII

SERVICE and MAINTENANCE

GENERAL MAINTENANCE OF THE SYSTEM

The MINOS STE-L System is equipped with automatic, microprocessor-controlled cleaning commands. For the Operator, this means "press a key", and the cleaning is automatically controlled.

1. **DAILY MAINTENANCE**

a. Each 50 Samples:

- An **AUTOMATIC CLEANING** cycle will be ran after every 50 samples *automatically*.

b. End of Assaying/End of Work Shift:

- Press the **AUTOMATIC CLEANING** key if you run **less than** 50 samples a day.
- Press the **DETERGENT RINSE** key. Leave the instrument in this condition until needed again, e.g. the next work shift or the next work day.

2. **WEEKLY MAINTENANCE:**

Press the **DETERGENT RINSE** key. Leave the instrument in this condition for as long as possible (preferably overnight) but for at least four (4) hours.

NOTE: An **AUTOMATIC CLEANING** cycle will be ran after every 50 samples *automatically*. This will eliminate the need for **DETERGENT RINSE** cycles as long as monthly maintenance is followed.

3. MONTHLY MAINTENANCE:

a. Press the **DRAIN CHAMBER** key. This drains the **RBC, WBC, and waste chambers.**

b. Fill a 5 mL or 10 mL syringe with **MINOCLAIR.**

c. Fill the **mixing chamber** with **MINOCLAIR** almost to the top.

d. Fill the **RBC chamber** with **MINOCLAIR** almost to the top.

e. Press pinch valves #8 and #81 at the same time. This will transfer the **MINOCLAIR** from the **mixing chamber** to the **WBC chamber.**

f. Press pinch valves #16 and #17 at the same time, and hold for 5 to 10 seconds. Release and repeat again. This will transfer the **MINOCLAIR** from the **mixing chamber** to the **WASTE chamber.**

g. Press the **DRAIN CHAMBER** key on the front panel.

h. Press the **DILUENT RINSE** key on the front panel.

i. Press the **ASPIRATE SAMPLE** key and check the background counts.

NOTE: This procedure takes five (5) minutes.

4. **ADDITIONAL MAINTENANCE:**

The **MINOS STE-L System** does not, except for the procedures above, require any additional Operator-initiated maintenance on the system. During the Warranty Period - or under FULL SERVICE Service Contract (after the one year Warranty Period)- the **ABX Inc. Field Service Engineer** will routinely service, clean, and replace worn parts, and install any system updates, once every four (4) months.

STE-L TROUBLESHOOTING

PNEUMATIC, MECHANICAL AND ELECTRICAL PROBLEMS

Most problems should be correctable with the help of the following guide and accompanying diagrams. If the problem is not solved with one of the solutions given in this manual, call **ABX Inc. Field Service.**

NO POWER:

1) Is the ON/OFF switch **ON**?
   - If no, turn it **ON**.
   - If yes, go to #2.

2) Is the instrument plugged into the wall outlet?
   - If no, plug cord into wall outlet.
   - If yes go to #3.

3) Is current going to wall outlet?
   - If no, call your maintenance department.
   - If yes, go to #4.

4) Check the two fuses located in the rear of the system. If the fuse(s) is(are) defective, replace with fuses of the same rating that are located in your ABX maintenance kit.

If, after checking all those steps, there is still no power in the system, call **ABX Inc. Field Service.**

NO ASPIRATION OF THE BLOOD SAMPLE:

1) Check for the correct operation of the 25 uL sampling syringe.
   - Does it operate smoothly?
     - If yes, go to #2.
     - If no, apply a small amount of grease to the needle and move it up and down (power OFF) until you feel that the syringe is moving smoothly.
     - If still not operating smoothly, call **ABX Inc. Field Service.**

2) Check the correct operation of the pinch valve #24.
   - Is the pinch valve working properly on sample cycle?
     - If yes, go to the next step.
     - If no, replace this pinch valve with the equivalent valve located in your ABX maintenance kit.

   - Is the corresponding tubing pinched closed?
     - If no, go to #3.
     - If yes, remove the tube from the pinch valve, roll it between your fingers and place it properly back into the valve.

3) Check for blockage in the sample needle (29 in pneumatic diagram).
- Is the sample needle blocked?
  - If no, go to the next step.
  - If yes, remove the tubing at the top side of the sample needle and flush the inside (using syringe) with MINOCLAIR until you can see the MINOCLAIR going through the tube. Place tubing back into the valve. Check the operation by running one blank cycle.

- Is the tubing in the corresponding line blocked?
  - If yes, pinch that part of the tubing which may contain a clog and run the system again.
  - If no, call **ABX Inc. Field Service.**

INCORRECT DISTRIBUTION OF THE 5 mL OF DILUENT (Minoton LMG)

1) - Is the dispenser (15 in pneumatic diagram) working properly during the sample cycle?
  - If yes, go to #2.
  - If no, turn the power **OFF**, move the dispenser up and down by hand. If there is difficulty doing so, remove the dispenser, replace the O-ring located on the inside and apply a thin film of grease around the O-ring. Make certain to properly replace the O-ring in the dispenser operating the system.

2) - Is there an air leak in the diluent system?
  - If no, go to #3.
  - If yes, try to locate the leak by identifying air bubbles in the tubing. Otherwise, call **ABX Inc. Field Service.**

3) - Is the pinch valve #5 working properly during a sample cycle?
  - If yes, go to #4.
  - If no, replace the valves (after ensuring that the tubing is not pinched) with the equivalent valve from your ABX maintenance kit.

4) - Is pinch valve #6 working properly during the sample cycle?
  - If no, replace the valve (after ensuring that the tubing is not pinched) with the equivalent valve that is in your ABX maintenance kit.
  - If yes, call **ABX Inc. Field Service.**

TRANSFER BETWEEN THE MIXING CHAMBER AND WBC CHAMBER

1) - Drain the chambers by pressing the **DRAIN CHAMBERS** key.

2) - Make a manual transfer by simultaneously pressing pinch valves #8 and #81.

3) - If the dilution does not drain from the **mixing chamber** to the **WBC chamber**, perform the following:

   a. Check the red restrictor (red plastic) located to the right of pinch valve #81 and replace (if necessary) with the equivalent spare located in your ABX maintenance kit.

b. Check that the tubing in the pinch valves is not pinched. If the tubing is pinched, roll it between your fingers and replace it properly in the valve.

c. Verify that there is no clot in the transfer tubing. If a clot is present, remove it by pressing that part of the tubing with your fingers. Verify proper operation by performing one blank cycle.
If, after the above check-up, the problem is still occurring, call **ABX Inc. Field Service.**

NO (REGULATED) VACUUM

1) - Is the vacuum **LED** (on the front panel) in the red zone (left side)?
- If no, go to the next step.
- If yes, do the following:

a. Push the **DRAIN CHAMBERS** key;

- If this operation occurs properly, the system will perform the cycles.
- If the chambers are incompletely drained, increase the vacuum (via the vacuum regulator knob, 23 in the view pneumatic) by a maximum of one turn.
- If you are still having problems, call **ABX Inc. Field Service.**

NO RBC OR WBC CHAMBER DRAINING

1) - Is the vacuum **LED** on the front panel in the red zone (left side)?
- If yes, go back to the above [**NO (REGULATED) VACUUM**].
- If no, check the following steps

a. Are pinch valves #20 (drains **RBC chamber**) and #21 (drains **WBC chamber**) working properly?
- If yes, go to the next step.
- If no, replace them, if necessary, and repeat the cycle.

b. Check that the tubing corresponding to the above two pinch valves is not clamped. If it is, remove it and roll the tubing between your fingers. Return the tubing to its proper location.

c. If you are still having some problems, call **ABX Inc. Field Service.**

NO DRAINING OF THE WASTE CHAMBER

Drain the **waste chamber** manually by pressing valve #14 and **then** valve #15

1) - Does drainage occur?
- If yes, go to #2.
- If no, do the following steps:

a. Check the draining tube (waste) at the rear of the system.
b. Check that the tubing inside pinch valves #14 and #15 is not pinched. If it is pinched, remove the tubing from its location, roll it between your fingers and return the tubing to its proper location.

2) - Are pinch valves #14 and #15 working properly?
- If no, replace the one which does not work, and try the system again.
- If the problem still occurs, call **ABX Inc. Field Service.**

NO LYSE TRANSFER:

1) - Is MINOLYSE bottle empty?
- If yes, replace the empty bottle with a full bottle of **MINOLYSE LMG** and prime the lyse.
- If no, go to the next step.

2) - Is pinch valve #12 operational during cycle?
- If yes, go to next step
- If no, replace it with the spare one in your maintenance kit.

3) - Is lyse pump moving during cycle?
- If no, call **ABX Inc. Field Service.**
- If the problem still occurs, call **ABX Inc. Field Service.**

INCORRECT RESULTS ON THE MEASURED PARAMETERS

The **Blood Cell Counter** is a system that measures the **WBC - RBC - HGB - HCT** and **PLT** parameters, directly.

Should technical problems arise, it is extremely important that the parameter(s) giving problems is (are) clearly identified.

PROBLEM ON WBC AND HGB:

1) - Is the sample needle working properly?
- If yes, go to #2.
- If no, go back to **NO BLOOD ASPIRATION.**

2) - Is the diluent dispenser working properly?
- If yes, go to #3.
- If no, go back to **DISTRIBUTION OF 5 mL OF DILUENT.**

3) - Is the first dilution being correctly transferred from the **mixing chamber** to the **WBC chamber**?
- If yes, go to #4.
- If no, go back to **TRANSFER BETWEEN THE MIXING CHAMBER AND WBC CHAMBER.**

4) - Is the lyse level in the bottle adequate?
- If yes, go to #5.
- If no, change the bottle and press the **LYSE PRIME** key.

5) - Is the lyse pump sending 1 mL of lyse to the **WBC chamber**?
- If yes, go to #6.
- If no, go back to **NO LYSE TRANSFER.**

6) - Are there any air bubbles in the lyse pump?
- If yes, press the **LYSE PRIME** key, and go back to #4 if you still have air bubbles.
- If no, call **ABX Inc. Field Service.**

PROBLEM ON WBC ONLY

1) - Press the **BACK FLUSH** key and re-run a cycle.
- If the problem is still present, go to #3.

2) - Press the **AUTOMATIC CLEANING** key and re-run the cycle.
- If the problem is still present, go to #5.

3) - Is the WBC calibration coefficient correct?
(retrieve the valve using the **SPECIAL FUNCTIONS** key).
- If yes, go to #5.
- If no, recalibrate the WBC coefficient.

4) - Is pinch valve #17 working properly, i.e., open during the counting cycle?
- If yes, go to #6.
- If no, replace this pinch valve with the spare one located in your ABX maintenance kit.

5) - Is the tubing in pinch valve #17 clamped?
- If no, go to #7.
- If yes, remove the tubing, roll it between your fingers and return it to its proper location.

6) - Put concentrated bleach in the **WBC chamber**, leave it for few minutes, then press on the **BACK FLUSH** key. After you perform an **AUTOMATIC CLEANING**, re-run a sample cycle.
- If the problem is still present, call **ABX Inc. Field Service.**

PROBLEM ON HGB ONLY:

1) - Perform an **AUTOMATIC CLEANING** and re-run a cycle.
- If the problem is still present, go to #2.

2) - Is the calibration coefficient correct?
- If yes, go to #3.
- If no, recalibrate the **HGB** coefficient and check that the displayed value is 12.0 to 15.0 for a displayed result (final value) of 13.0 to 14.0 g/dL.

3) - Is the lyse pump working properly?
- If yes, go to #4.
- If no, go back to **NO LYSE TRANSFER.**

4) - Is the pinch valve #18 working properly?
- If yes, go to #5.
- If no, replace it with the spare one in your ABX maintenance kit.

5) - Is the **HGB** lamp lit when the power of the system is **ON**?
- If no, call **ABX Inc. Field Service.**

PROBLEM ON RBC AND HCT

1) - Is the bubbling in the **mixing chamber** sufficient?
- If yes, go to #2.
- If no, fill the **mixing chamber** manually with **MINOTON LMG**, press on pinch valve #9 and turn the bubbling regulator (**6** on pneumatic view) clockwise until you achieve strong bubble action.
- If there is no bubble action, replace the red restrictor in the tubing line coming out of pinch valve #9 and try the bubble flow again.

2) - Is the position of the sample needle in the **mixing chamber** correct?
(It should be between the edge and the center of the **mixing chamber**, close to the bottom).
- If yes, go to #4.
- If no, try to set it or call **ABX Inc. Field Service.**

3) - Is the 25 uL sampling syringe working correctly?
- If yes, go to #4.
- If no, go back to **NO BLOOD SUCTION.**

4) - Is the dispenser working properly?
- If yes, go to #5.
- If no, go back to **DISTRIBUTION OF 5 mL DILUENT INCORRECT.**

5) - Is the first dilution properly transferred from the **mixing chamber** to the **WBC chamber**?
- If yes, go to #6.
- If no, go back to **TRANSFER FROM MIXING CHAMBER TO WBC CHAMBER.**

6) - Is the diluent (**MINOTON LMG**) level in the container sufficient?
- If yes, go to #7.
- If no, change it with a new container and press the key **DILUENT RINSE.**

7) - Perform an **AUTOMATIC CLEANING**, re-run a cycle. If the problem persists, call **ABX Inc. Field Service.**

PROBLEM ON RBC ONLY

1) - Are there pulses on the **CRT** during the count cycle?
(the normal initial counts should display about 100 to 120 for a final value of 4.00 to 5.00 $x10^6/mm^3$).
- If yes, go to #4.
- If no, go to #2.

2) - Press the **BACK FLUSH** key and re-run a cycle.
   - If the problem is still present, go to #3.

3) - Press the **AUTOMATIC CLEANING** key and re-run a cycle.
   - If the problem is still present go to #5.

4) - Is the calibration coefficient correct?
   (retrieve the value using the **SPECIAL FUNCTIONS** key).
   - If yes, go back to #2.
   - If no, recalibrate the **RBC** coefficient.

5) - Is pinch valve #16 working properly, i.e., open on count cycle?
   - If yes, go to #6.
   - If no, replace it with the spare in your ABX maintenance kit.

6) - Is the tubing in pinch valve #16 clamped?
   - If no, go to #7.
   - If yes, remove it, roll it between your fingers and place it back in the proper location.

7) - Is the sample needle working properly?
   - If yes, go to #8.
   - If no, go back to **NO BLOOD SUCTION.**

8) - Is the rinse needle pump (25 on pneumatic view) working properly?
   - If yes, go to #9.
   - If no, call **ABX Inc. Field Service.**

9) - Is pinch valve #7 working properly?
   - If yes, go to #10.
   - If no, replace it with the spare in your ABX maintenance kit.

10)- Is the tubing in pinch valve #7 clamped?
   - If yes, remove it, roll it between your fingers and return the tubing to its proper location.
   - If no, call **ABX Inc. Field Service.**

PROBLEM ON HCT ONLY

1) - Press the **BACK FLUSH** key and re-run a cycle.
   - If the problem still persists, go to #2.

2) - Is the calibration coefficient correct?
   (retrieve it using the **SPECIAL FUNCTIONS** key).
   - If yes, go to #3.
   - If no, recalibrate the **HCT** coefficient.

3) - Check for a normal initial count: the value is about 15.0 to 17.0 for a result between 45.0 and 52.0 %.
   - If this is not the case, go to #4.

4) - Press the AUTOMATIC CLEANING key, and re-run a cycle by checking #3 again.
   - If the problem persists, call **ABX Inc. Field Service.**

PROBLEM ON PLT:

1) - Press the **BACK FLUSH** key and re-run your sample.
   - If the problem persists, go to #2.

2) - Is the sample needle position in the **mixing chamber** correct?
   - If yes, go to #3.
   - If no, go back to section #2 of **PROBLEM ON RBC AND HCT.**

3) - Is the **PLT** calibration coefficient correct?
   - If yes, call **ABX Inc. Field Service.**
   - If no, recalibrate it and try your sample again.

SECTION VIII

Bibliography & Appendices

SECTION VIII

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APPENDIX A

HEMATOCRIT - related corrective factor for platelet counts using Platelet-Rich Plasma (PRP).

METHOD OF PRP (PLATELET-RICH PLASMA) MEASUREMENT:

1. Prepare a PRP on the blood sample with the MICROC alert (i.e. by sedimentation).
2. Run a cycle on the PRP (with sample needle in plasma only).
3. Multiply the PRP platelet result by the Hematocrit Factor (F. Hct). This factor is read off the table below, using the true HCT from the original full blood count results.
4. Compare the a newly calculated PLT count with the original.

If they are similar, then the original PLT count was acceptable.
If they are significantly different, then an alternative PLT measurement must be made (i.e. by manual method).

Hematocrit = HCT%	Hematocrit Factor = F Hte	Hematocrit = HCT%	Hematocrit Factor = F Hte
10	0,90	41	0,44
11	0,89	42	0,42
12	0,87	43	0,41
13	0,86	44	0,40
14	0,85	45	0,39
15	0,84	46	0,38
16	0,83	47	0,37
17	0,82	48	0,36
18	0,80	49	0,35
19	0,79	50	0,34
20	0,78	51	0,33
21	0,77	52	0,32
22	0,75	53	0,31
23	0,74	54	0,30
24	0,73	55	0,29
25	0,71	56	0,28
26	0,70	57	0,27
27	0,68	58	0,26
28	0,66	59	0,25
29	0,65	60	0,24
30	0,63	61	0,23
31	0,61	62	0,22
32	0,60	63	0,21
33	0,58	64	0,20
34	0,56	65	0,19
35	0,54	66	0,18
36	0,52	67	0,17
37	0,50	68	0,16
38	0,48	69	0,15
39	0,47	70	0,14
40	0,45		

APPENDIX B

I. WBC SCREENING OR ONE-PART DIFFERENTIAL COUNT

A. Technology

Lyse effect

The lyse reacts as a differentiator on the cytoplasmic membrane. In contact with the lyse, the lymphocyte membrane shrinks completely down to the nucleus.

The best result is obtained with a slow lyse reaction along with a perfectly-timed flow of the diluent. It is then very important to have a perfect transfer time for the lyse as well as for the blood dilution from the **mixing chamber** to the **WBC chamber**.

Volumetric Studies

After the lysing effect, the **MINOS STE-L** analyses, during the first eight seconds, each pulse recorded through the aperture, and files it in 256 channels of memory, according to its threshold.

Abnormal cells will, of course, be included in some parts of the distribution curve.

In order to achieve the best possible measurement, the following regions have been placed according to the actual locations of the abnormal cells.

NUMBER OF CELLS

B. WBC Flags

Flags will alert the user to abnormal WBCs in the sample.

FLAG L1

This flag detects a higher cell count than expected in the 30 to 60 um^3 range. Abnormal cells found mostly in this range could be erythroblasts or platelet aggregates.

This flag corresponds to the number of cells counted in the first 5 channels, divided by the total number of lymphocytes. The average rate is 8 to 10 percent. The threshold is adjustable by the user. The adjustment is made by pressing the **SPECIAL FUNCTION** key and then entering the code 997 on the number keys.

FLAG L2

Monitoring the range from 130 to 150 um^3, this flag allows the detection of cells such as lymphoblasts, myelocytes, atypical lymphocytes, and of high numbers of basophils.

This flag is adjustable by the user. It corresponds to the number of cells counted in channels 30 to 40, divided by the total number of granulocytes. The adjustment is performed the same way as with Flag L1. The range is between 15 and 18 percent.

III. ADJUSTMENT OF FLAGS

The adjustment of flag L2 is important for the detection of the number of abnormal cells. Individual laboratories should adjust the level of flags according to the pathology most often expected in their lab.

This flag should not be considered thoroughly reliable, as cell membrane resistance varies from sample to sample (i.e., with individual patients, different pathologies, etc.).

In fact, from the above defined parameters, abnormal cells might be detected in areas other than those expected (i.e.: myelocytes in L2, eosinophils, but also in L2).