

510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION DECISION SUMMARY

A. 510(k) Number:

K151133

B. Purpose for Submission:

Device modification for addition of reticulocytes parameters (Ret #, Ret%, CRC, IRF) to the complete blood count (CBC) and differential (DIFF) on the ABX PENTRA 80 hematology analyzer and changing the device name to PENTRA XLR.

C. Manufacturer and Instrument Name:

HORIBA ABX SAS; PENTRA XLR

D. Type of Test or Tests Performed:

Quantitative test for white blood cell (WBC), red blood cell (RBC), hemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), red cell distribution (RDW), platelet (PLT), mean platelet volume (MPV), percent lymphocytes (LYM%), percent monocytes (MON%), percent neutrophils (NEUT%), absolute neutrophils (NEUT#), absolute lymphocytes (LYM#), absolute monocytes (MON#), absolute eosinophils (EOS#), percent eosinophils (EOS%), absolute basophils (BAS#), percent basophils (BAS%), reticulocyte absolute value (RET#), reticulocyte percentage (RET%), corrected reticulocyte count (CRC), and immature reticulocyte fraction (IRF).

E. System Descriptions:

1. Device Description:

The PENTRA XLR is a device modification to the ABX PENTRA 80 (K024002), which consists in the addition of the measurement of reticulocytes (RET) parameters to the existing Complete Blood Count (CBC) and Differential (DIFF) counts already performed by the analyzer.

The additional RET mode, based on the use of thiazol orange reagent and fluorescence detection by optical bench, is independent from the modes already existing on the ABX PENTRA 80: the CBC and the CBC+DIFF modes. There have been no modifications to the existing mechanical and analytical portions of the original device.

The existing fundamental scientific technology for the analyzer has not changed. Hematological parameters for complete blood count and differential leucocyte count,

the reagents and controls, measuring principles, and the principles of operation are the same as previously cleared by the FDA.

The parameters already available on the ABX PENTRA 80 are unchanged:

- CBC parameters: WBC, RBC, HGB, HCT, MCV, MCH, MCHC, RDW, PLT, MPV
- DIFF parameters: LYM#, LYM%, MON#, MON%, NEU#, NEU%, EOS#, EOS%, BAS#, BAS%

The additional reticulocytes parameters are the following:

RET#: reticulocyte absolute value

RET%: reticulocyte percentage

CRC: corrected reticulocyte count

IRF: immature reticulocyte fraction

RBC parameter is also reported in the RET mode.

Software changes reflect routine updates to the cycle operation, increased user interface options, and the support of the reticulocyte counting.

2. Principles of Operation:

The PENTRA XLR performs hematology analyses using the following methods:

- RBC / PLT: Impedance
- WBC: Impedance
- HGB: Spectrophotometry
- DIFF: Impedance with DHSS (Double Hydrodynamic Sequential System) for Cytometry and Cytochemistry
- MCV: Calculated
- RET: Fluorescence and Impedance

The principles of operation for the parameters already existing on the ABX PENTRA 80 are unchanged and are, therefore, not explained again. The principle of operation for the reticulocyte parameters is described below.

The instrument samples 35 µL of blood and injects 10 µL of it into the DIL1 chamber. It then mixes the sample with 1.7 mL of ABX Diluent. Then the instrument samples 28 µL of diluted blood and mixes it with 2.5 mL of ABX Fluocyte. This reagent contains a fluorescent stain which is specific to nucleic acids: thiazol orange.

The solution is warmed at 35°C for 50 seconds. The stain molecules enter through the cell membrane and fix the ribonucleic acid molecules. This binding gives an increase of the fluorescence (enhancement: ~3000x). After 50 seconds, the solution is transferred to the laser optical bench to be measured.

The laser optical bench simultaneously measures the fluorescence of the cells passing through the measuring point into the flowcell and the volume by impedance. A cell passing through the flowcell gives two types of information:

- the size of the cell measured by resistivity (Cell Impedance Signal (CIS));
- the fluorescence signal (Orthogonal Fluorescence Light (OFL)).

The fluorescence is collected using:

- a lens focused on the optical flowcell and located at 90° from the laser beam;
- an interferential filter specific to the thiazol orange stain selecting only the fluorescent wavelength;
- an avalanche photodiode.

The reticulocytes matrix is generated from two measurements: CIS and OFL of cells according to the X and Y axes respectively. Mature red blood cells without RNA show little or no fluorescent signal. They are located at the bottom of the matrix and horizontally distributed according to their MCV and RDW. Reticulocytes are separated from the red blood cells by their fluorescence which is proportional to the RNA content and their immaturity. The most fluorescent elements, which are saturated at the top of the matrix, are the most immature. Erythroblasts may also be found in this area.

3. Modes of Operation:

The PENTRA XLR instrument can operate in:

- CBC mode (Complete Blood Count)
- DIFF mode (CBC + WBC Differential)
- RET mode (Reticulocytes + RBC)

Sampling modes:

The PENTRA XLR operates in:

- Automatic sampling (auto mixer and auto loader) for CBC and DIFF modes only or
- Manual sampling (STAT mode) for CBC, DIFF and RET modes.

Therefore, for the RET mode, only the manual sampling (STAT mode) is available.

Does the applicant's device contain the ability to transmit data to a computer, webserver, or mobile device?

Yes ☒X☐ or No ☐

Does the applicant's device transmit data to a computer, webserver, or mobile device using wireless transmission?

Yes ☐ or No ☒X☐

4. Specimen Identification:

Tube sample ID can be identified manually or by double barcode label reading.

5. Specimen Sampling and Handling:

PENTRA XLR has both open tube sampling and closed tube sampling with cap-piercing mechanism. For the open tube sampling the cap is removed before placing tube into the tube holder and closing the door of the analyzer. Under the closed tube sampling, the blood collection tube is placed directly in the analyzer without removing the cap. The User Manual provides a list of tubes that can be used with PENTRA XLR. Both K₂EDTA and K₃EDTA anticoagulants are acceptable.

6. Calibration:

Calibration is performed by a HORIBA ABX SAS representative during specific situations such as installation, maintenance or service intervention using ABX Minocal.

The RET measurement is adjusted by a HORIBA medical technician upon the PENTRA XLR installation and according to procedure described in the User Manual.

7. Quality Control:

The RET parameters are controlled with ABX MINOTROL RETIC, cleared under K943336 as Retic CONTROL-H Hematology Control by R&D Systems, Inc.

8. Software:

FDA has reviewed applicant's Hazard Analysis and Software Development processes for this line of product types:

Yes ☒ X ☐ or No ☐

F. Regulatory Information:

1. Regulation section:

21 CFR 864.5220, Automated differential cell counter

2. Classification:

Class II

3. Product code:

GKZ, Counter, Differential Cell

4. Panel:

Hematology (81)

G. Intended Use:

1. Indication(s) for Use:

The PENTRA XLR is a quantitative multi-parameter, automated hematology analyzer for in vitro diagnostic use in clinical laboratories to identify and enumerate the following parameters: WBC, RBC, HGB, HCT, MCV, MCH, MCHC, RDW, PLT, MPV, LYM (#, %), MON (#, %), NEU (#, %), EOS (#, %), BAS (#, %), as well as the Reticulocyte parameters RET (#, %), CRC, and IRF in K₂EDTA and K₃EDTA anticoagulated venous whole blood samples from patients ≥ 18 years of age.

2. Special Conditions for Use Statement(s):

For prescription use only.

H. Substantial Equivalence Information:

1. Predicate Device Name(s) and 510(k) numbers:

ABX PENTRA DX 120; K050719, K991839, K990311

2. Comparison with Predicate Device:

Similarities		
Item	Device	Predicate
Manufacturer	HORIBA ABX SAS	Same
Type of product	Automated blood cell counter	Same
Diagnostic Parameters	RET (#, %), IRF (%), CRC (%),	RET (#, %), IRF (%), MRV (fL), CRC (%), RET L (%), RET M (%), RET H (%)
Sample types	K ₂ and K ₃ EDTA anti-coagulated whole blood	Same
RET Principles	Fluorescence and Impedance	Same
RET Reagents	ABX Fluocyte	Same

Similarities		
Item	Device	Predicate
RET Methodology	ABX Fluocyte contains a fluorescent stain which is specific to nucleic acids: thiazol orange. The stain molecules enter through the cell membrane and fix the ribonucleic acid molecules. The binding gives an increase of fluorescence. The laser optical bench simultaneously measures the fluorescence of the cells passing through the measuring point into the flowcell, and volume by absorbance. The size of the cell is measured by resistivity, the scattered light (FSL) is measured approximately 200 μ S after the aperture measurement, the fluorescence signal (OFL) is measured simultaneously with the FSL.	Same
RET Calibrators	None	Same
RET Controls	ABX Minotrol Retic	Same
RET Technical parameters	MFI	Same
	PIC	Same

Differences		
Item	Device	Predicate
Intended Use	The PENTRA XLR is a quantitative multi-parameter, automated hematology analyzer for in vitro diagnostic use in clinical laboratories to identify and enumerate the following parameters: WBC, RBC, HGB, HCT, MCV, MCH, MCHC, RDW, PLT, MPV, LYM#, LYM%, MON#, MON%, NEU#, NEU%, EOS#, EOS%, BAS#, BAS%, as well as the Reticulocyte parameters RET#, RET%, CRC, and IRF in K2EDTA and K3EDTA anticoagulated venous whole blood samples from patients ≥ 18 years of age.	The ABX PENTRA DX 120 Hematology Analyzer is an automated hematology analyzer providing complete blood count (CBC), differential leucocyte count (DIFF) as well as reticulocyte count (RET) and nucleated red blood cell count (NRBC) for the in vitro diagnostic use in clinical laboratories. The clinical use of the reticulocyte count, specifically the immature reticulocyte fraction (IRF) is to monitor erythropoietic activity in patients.
Specimen volume	RET mode: 35 μ L	Manual cycle: 130 μ L Automatic cycle: 200 μ L
Throughput	80 samples / hour	120 samples / hour

I. Special Control/Guidance Document Referenced (if applicable):

- CLSI EP05-A2: Evaluation of Precision Performance of Quantitative Measurement Methods – 2004
- CLSI EP06-A: Evaluation of the Linearity of Quantitative Measurement Procedure: A Statistical Approach – 2003
- CLSI EP07-A2: Interference Testing in Clinical Chemistry – 2005
- CLSI EP09-A3: Measurement Procedure Comparison and Bias Estimation Using Patient Samples – 2013
- CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures – 2012
- CLSI EP28-A3c: Defining, Establishing and Verifying Reference Intervals in the Clinical Laboratory – 2008
- CLSI H26-A2: Validation, Verification and Quality Assurance of Automated Hematology Analyzers – 2010
- CLSI H44-A2: Methods for reticulocyte Counting (Automated Blood Cell Counters, Flow Cytometry, and Supravital Dyes); Approved Guideline – Second Edition - 2004
- IEC61010-1, IEC61010-2-081, IEC61010-2-101, UL61010-1, CAN/CSA-C22.2 No.61010-1-12, CAN/CSA-C22.2 No. 61010.2.081-04, CAN/CSA-C22.2 No. 61010-2-10104: Safety requirements for electrical equipment for measurement, control, and laboratory use
- EN61326-1, EN61326-2-6: Electrical equipment for measurement, control and laboratory use - EMC requirements
- EN60825-1: Safety of Laser products – Part 1: Equipment classification and requirements
- ISO14971: Medical devices – Application of risk management to medical devices

FDA Guidances Followed:

- Guidance for Industry and FDA Staff: Format for Traditional and Abbreviated 510(k)s - 2005
- Final Guidance for Industry and FDA: Class II Special Controls Guidance Document: Premarket Notifications for Automated Differential Cell Counters for Immature or Abnormal Blood Cells – 2001
- Guidance for Industry and FDA Staff: Guidance for the Content of Premarket Submissions for Software Contained in Medical Devices – 2005
- Guidance for Industry, FDA Reviewers and Compliance on Off-The-Shelf Software Use in Medical Devices – 1999
- Guidance for Industry and Food and Drug Administration Staff on the Content of Premarket Submissions for Management of Cybersecurity in Medical Devices - 2014

J. Performance Characteristics:

1. Analytical Performance:

a. Method Comparison

A total of 376 venous whole blood specimens (collected in K₂EDTA) were analyzed at three test sites in the US. Different instruments and operators were used at each site. Each of the samples was analyzed in duplicate on the PENTRA XLR and on the predicate ABX PENTRA DX 120. Each replicate from the PENTRA XLR was compared to the mean value of duplicates from the ABX PENTRA DX 120. The study population included subjects ≥ 18 years of age.

Individual site and combined results for RBC, RET%, RET#, IRF and CRC passed the acceptance criteria at all the pre-determined medical decisions points when sample results analyzed by the PENTRA XLR were compared to the same sample analyzed by the predicate device. These findings support the claim that the PENTRA XLR candidate device and the ABX PENTRA DX 120 predicate device are substantially equivalent.

Summary of Method Comparison Correlation Data – All sites								
Parameter	Rep	N	R ² obtained	R ² claim	Intercept		Slope	
					Intercept	95% CI	Slope	95% CI
RBC (10 ⁶ /mm ³)	Rep-1	347	1.00	>0.95	-0.13	-0.16 ; -0.10	1.04	1.03 ; 1.04
	Rep-2	350	1.00	>0.95	-0.14	-0.17 ; -0.11	1.04	1.03 ; 1.05
RET%	Rep-1	326	0.99	>0.95	-0.050	-0.12 ; 0.01	1.062	1.04 ; 1.08
	Rep-2	326	0.99	>0.95	-0.02	-0.06 ; 0.05	1.030	1.00 ; 1.05
RET# (10 ⁶ /mm ³)	Rep-1	348	0.96	>0.95	-0.001	-0.003 ; 0.001	1.047	1.022 ; 1.074
	Rep-2	348	0.97	>0.95	0.000	-0.001 ; 0.002	1.004	0.986 ; 1.033
IRF	Rep-1	319	0.49	n/a	0.053	0.041 ; 0.066	0.582	0.532 ; 0.644
	Rep-2	319	0.46	n/a	0.046	0.033 ; 0.059	0.590	0.532 ; 0.666
CRC	Rep-1	288	0.98	n/a	-0.032	-0.10 ; 0.03	1.112	1.09 ; 1.14
	Rep-2	288	0.99	n/a	0.000	-0.05 ; 0.04	1.070	1.05 ; 1.09

b. Precision/Reproducibility:

Repeatability

Repeatability was performed using a minimum of 10 normal and 11 abnormal fresh whole blood samples (six low level and five high level concentration of RBC and reticulocytes) collected into tubes containing K₂EDTA anticoagulant, at three different clinical sites. Each sample was run 12 consecutive times on the PENTRA XLR in a single day and all runs were completed within 8 hours of sample collection. The results obtained were in the specifications. RBC results are consistent with ABX PENTRA 80 claim.

Summary of Repeatability Results, Site 1													
		Low Level			Normal Level				High Level				
		Mean	CV%			Mean	CV%			Mean	CV%		
Parameter			Min	Max	Mean		Min	Max	Mean		Min	Max	Mean
RBC($10^6/\text{mm}^3$)		2.49	0.49	1.94	0.92	4.62	0.41	1.18	0.65	7.21	0.47	1	0.63
Ret%		N/A	N/A	N/A	N/A	1.59	3.44	10.38	6.83	14.27	2.3	6.98	4.17
RET#	$10^6/\text{mm}^3$	N/A	N/A	N/A	N/A	0.05	4.41	11.2	8.39	0.21	2.38	8.86	5.43
	cells/ μL	N/A	N/A	N/A	N/A	51501	4.41	11.2	8.39	209353	2.38	8.86	5.43
CRC		N/A	N/A	N/A	N/A	1.64	4.35	11.28	7.76	4.83	2.5	8.86	5.34
IRF		0.084	0.009†	0.03†	0.02†	0.189	5.411	16.004	11.482	0.40	3.04	8.44	4.62

Summary of Repeatability Results, Site 3													
		Low Level			Normal Level				High Level				
		Mean	CV%			Mean	CV%			Mean	CV%		
Parameter			Min	Max	Mean		Min	Max	Mean		Min	Max	Mean
RBC($10^6/\text{mm}^3$)		2.33	0.51	1.96	1.25	4.86	0.54	0.99	0.73	6.46	0.95	0.98	0.96
Ret%		N/A	N/A	N/A	N/A	1.54	4.26	10.31	7.17	7.67	2.75	5.14	3.93
RET #	$10^6/\text{mm}^3$	N/A	N/A	N/A	N/A	0.054	4.34	12.40	8.02	0.186	3.13	5.25	4.16
	cells/ μL	N/A	N/A	N/A	N/A	54000	4.33	12.40	8.02	186000	3.13	5.25	4.16
CRC		N/A	N/A	N/A	N/A	1.25	4.43	10.99	7.06	4.12	3.10	5.29	4.18
IRF		0.085	0.01†	0.02†	0.01†	0.22	5.17	17.11	11.78	0.34	3.33	8.85	5.84

Summary of Repeatability Results, Site 4													
		Low Level			Normal Level				High Level				
		Mean	CV%			Mean	CV%			Mean	CV%		
Parameter			Min	Max	Mean		Min	Max	Mean		Min	Max	Mean
RBC($10^6/\text{mm}^3$)		2.77	0.70	2.07	1.20	4.90	0.67	2.24	1.14	6.19	0.55	1.35	0.88
Ret%		N/A	N/A	N/A	N/A	1.54	4.43	9.85	6.83	5.77	2.45	4.16	3.21
RET #	$10^6/\text{mm}^3$	N/A	N/A	N/A	NA	4.77	11.36	7.75	0.134	2.28	6.77	4.55	4.16
	cells/ μL	N/A	N/A	N/A	NA	4.77	11.36	7.75	134000	2.28	6.77	4.55	4.16
CRC		N/A	N/A	N/A	N/A	1.34	4.53	9.15	6.77	3.34	2.34	5.18	4.01
IRF		0.109	0.009†	0.03†	0.019†	0.220	7.756	18.036	11.812	0.344	4.545	8.518	6.686

Summary of Repeatability Results, 3 Sites Combined													
		Low Level				Normal Level				High Level			
		Mean	CV%			Mean	CV%			Mean	CV%		
Parameter			Min	Max	Mean		Min	Max	Mean		Min	Max	Mean
RBC($10^6/\text{mm}^3$)		2.53	0.49	2.07	1.08	4.78	0.41	2.24	0.85	6.70	0.47	1.35	0.78
Ret%		N/A	N/A	N/A	N/A	1.56	3.44	11.06	7.31	10.72	2.3	6.98	3.89
RET#	$10^6/\text{mm}^3$	N/A	N/A	N/A	N/A	0.054	4.34	12.41	8.05	0.18	2.28	8.86	4.98
	cells/ μL	N/A	N/A	N/A	N/A	54000	4.34	12.41	8.05	180000	2.28	8.86	4.98
CRC		N/A	N/A	N/A	N/A	1.44	4.35	11.28	7.36	4.36	2.34	8.86	4.82
IRF		0.091	0.009†	0.030†	0.018†	0.209	5.17	18.03	11.67	0.36	3.04	8.86	5.70

† = values reported are SD

Reproducibility

Reproducibility was assessed on four PENTRA XLR instruments at four different sites, one operator per site. At each site; high, normal, and low levels of a single lot of control material (Minotrol Retic for the Reticulocyte parameters, Difftrol for the RBC parameter) were run in duplicate, twice each day, during a minimum of 25 days.

Standard deviation and CV% were calculated for each measurand and the results obtained were within specifications. RBC results are consistent with ABX PENTRA 80 claim.

RBC Precision ABX Minotrol Retic Combined sites ($10^6/\text{mm}^3$)

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	3.4	0.045	1.32	0.028	0.82	0.041	1.21	0.070	2.06	0.097	2.85
Level 2	400	3.27	0.029	0.89	0.031	0.95	0.036	1.10	0.065	1.99	0.085	2.60
Level 3	400	3.31	0.036	1.09	0.025	0.76	0.045	1.36	0.067	2.02	0.092	2.78

RBC Precision ABX Difftrol Combined sites ($10^6/\text{mm}^3$)

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level L	300	0.02	0.84	0.02	0.63	0.01	0.51	0.02	0.97	0.04	1.52	0.02
Level N	300	0.03	0.70	0.01	0.30	0.03	0.62	0.03	0.64	0.06	1.17	0.03
Level H	300	0.04	0.66	0.02	0.30	0.03	0.64	0.03	0.49	0.06	1.08	0.04

RET% Precision ABX Minotrol Retic Combined sites

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	2.11	0.131	6.21	0.000	0.00	0.019	0.90	0.266	12.61	0.297	14.08
Level 2	400	5.4	0.188	3.48	0.038	0.70	0.046	0.85	0.360	6.67	0.411	7.61
Level 3	400	10.51	0.236	2.25	0.135	1.28	0.105	1.00	0.522	4.97	0.598	5.69

RET# Precision ABX Minotrol Retic Combined sites (10⁶/mm³)

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	0.072	0.005	6.94	0.000	0.00	0.001	1.39	0.009	12.50	0.010	13.89
Level 2	400	0.176	0.006	3.41	0.002	1.14	0.002	1.14	0.012	6.82	0.014	7.95
Level 3	400	0.348	0.009	2.59	0.005	1.44	0.006	1.72	0.018	5.17	0.021	6.03

RET# Precision ABX Minotrol Retic Combined sites (Cells/μL)

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	71806	4550	6.94	0	0.00	1326	1.39	9011	12.50	10182	13.89
Level 2	400	176377	6399	3.41	2083	1.14	2192	1.14	12487	6.82	14354	7.95
Level 3	400	348204	9017	2.59	4871	1.44	6260	1.72	17783	5.17	21459	6.03

CRC Precision ABX Minotrol Retic Combined sites

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	1.27	0.079	6.22	0.000	0.00	0.032	2.52	0.151	11.89	0.173	13.62
Level 2	400	3.44	0.126	3.66	0.035	1.02	0.069	2.01	0.218	6.34	0.263	7.65
Level 3	400	6.2	0.160	2.58	0.085	1.37	0.139	2.24	0.275	4.44	0.357	5.76

IRF Precision ABX Minotrol Retic Combined sites

All Sites	N	Mean	Within-Run		Between-Run		Between-Day		Between-Site		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Level 1	400	0.439	0.032	7.29	0.000	0.00	0.000	0.00	0.039	8.88	0.051	11.62
Level 2	400	0.565	0.017	3.01	0.008	1.42	0.005	0.88	0.023	4.07	0.031	5.49
Level 3	400	0.58	0.013	2.24	0.006	1.03	0.012	2.07	0.023	3.97	0.029	5.00

c. Linearity:

Commercial linearity kits were used to perform the linearity studies. The expected values of the kit samples were considered the “true values”. Each level was run in replicates of four (n=4) as recommended by the kit supplier. For each level, the four replicate results were plotted versus the theoretical value. The findings of the polynomial regression analysis indicate that the PENTRA XLR exhibits linearity across the claimed range.

The Analytical Measuring Range (AMR) is defined as the range comprised between the Analytical Limit or Limit of Quantitation and the High Linearity Limit determined for each parameter. The claimed AMR are provided below:

Parameter	AMR on PENTRA XLR
RBC (10 ⁶ /mm ³)	0.2 – 8
RET%	0.7 – 27
RET# (10 ⁶ /mm ³)	0.01 – 0.5

RBC results are consistent with ABX PENTRA 80 claim.

d. Carryover:

The potential for sample carryover was tested in duplicate on the PENTRA XLR instrument using alternating high and low concentrations samples according to the process below:

- Process a sample in RET mode with “low values” three times (*low 1, low 2, low 3*)
- Then process a sample with a “high values” three times (*high 1, high 2, high 3*)
- Finally process again the sample with “low values” three times (*low 4, low 5, low 6*)

The percentage of carryover is calculated using the formula below:



All carry-over results are within specifications for the PENTRA XLR. RBC results are consistent with ABX PENTRA 80 claim.

	Carry-over Limit (%CV)
RBC ($10^6/\text{mm}^3$)	<2%
RET%	<1%
RET# ($10^6/\text{mm}^3$)	<1%

e. Interfering Substances:

The interference effect is evaluated following two methodologies.

By addition - evaluating the effect of potentially interfering substances added to the sample of interest:

A potential interfering substance is added to a sample and the bias relative to a control portion of the sample is evaluated ("paired-difference testing"). This bias was compared to the acceptance criteria.

For all tests performed with the following potential interferents: urea, bilirubin, lipemia, and hemolysis, the bias remained below the acceptable limit, therefore, no significant interference was observed.

By comparison - evaluating the bias of individual specimen:

Representative patient specimens (with known or potential reticulocyte interferents, as given in CLSI H44-A2 guidance document) and control samples (without interferent) are run in duplicate in comparison to a comparative measurement procedure (ABX PENTRA DX 120). Then the bias versus comparative measurement values was plotted for each

specimen group. Both measurement procedures had 10 to 20 samples in each group to demonstrate sufficient precision. A comparable effect was observed on the PENTRA XLR and the reference device for the interferences from: abnormal RBC, basophilic stippling, cold agglutinins, giant platelets, hemolysis, Howell-Jolly bodies, leukocyte fragments, nucleated erythrocytes, Pappenheimer bodies, paraproteins, platelet clumps, platelet/erythrocyte coincidence.

Additionally, Heinz Bodies, parasites and autofluorescence are potential interferents on WBC and Monocytes counts that could not be tested, but that are well described in literature.

2. Other Supportive Instrument Performance Data Not Covered Above:

a. Specimen Stability Studies

Ten whole venous blood specimens (collected in K₂EDTA) were analyzed on the PENTRA XLR at one site in US. Following the collection (T₀), each specimen was divided in half, with one half of the sample stored at ambient temperature (24°C) and the other stored under refrigerated conditions (4°C). Testing for stability was performed at 1, 2, 3, 4, 6, 8, 24, 48 and 72 hours after T₀. When sufficient volume is available, each time point is tested in duplicate. The acceptance criteria for sample stability is given as an acceptable maximum bias of the value at T with the value at T₀. All data passed specifications. RBC results are consistent with ABX PENTRA 80 claim.

Parameters	Sample stability when stored refrigerated (2-8°C)	Sample stability when stored at room temperature (20-24°C)
RET%	48 hours	24 hours
RET# (10 ⁶ /mm ³)	48 hours	24 hours
CRC	48 hours	24 hours
IRF	48 hours	24 hours
RBC (10 ⁶ /mm ³)	48 hours	48 hours

b. Comparability between CBC and RET modes for RBC

A total of 107 normal and pathological blood specimens were analyzed on two PENTRA XLR instruments at one site in France. The specimens used in this study were venous blood specimens collected in K₂EDTA. Each subject specimen was analyzed in duplicate on both instruments in RET mode and CBC mode. Each replicate from the RET mode was compared to the mean value of the replicates from the CBC mode.

Bias was estimated at three points for RBC parameter: the low end of the distribution of observations, the mid-point, and the high end of the distribution. Bias was estimated separately for each replicate. Acceptance criteria were met at all levels. These findings support the claim that CBC and RET modes give comparable RBC results as measured on the PENTRA XLR hematology analyzer.

		Mean	Mean			Intercept		Slope	
	Rep	Ret mode	CBC mode	N	R ²	Intercept	95% CI	Slope	95% CI
PXLN N°1	Rep-1	4.45	4.48	106	0.999	-0.064	-0.099 ; -0.030	1.007	1.000 ; 1.014
	Rep-2	4.45	4.47	106	0.999	-0.02	-0.040 ; 0.014	1.000	0.992 ; 1.004
PXLN N°2	Rep-1	4.33	4.37	104	0.999	-0.032	-0.050 ; -0.005	0.997	0.990 ; 1.002
	Rep-2	4.36	4.39	104	0.999	-0.04	-0.053 ; -0.006	1.000	0.992 ; 1.006
All	Rep-1	4.39	4.43	210	0.999	-0.04	-0.065 ; -0.024	1.000	0.996 ; 1.006
	Rep-2	4.40	4.43	212	0.999	-0.03	-0.040 ; -0.006	1.000	0.994 ; 1.003

c. Anticoagulant Comparison Study - K₂EDTA vs. K₃EDTA whole blood:

A total of 90 normal and pathological blood specimens were analyzed on the PENTRA XLR at two sites in the US. Different instruments and operators were used at each site. The specimens used in this study were venous blood specimens that were prospectively collected for this study specifically. Each subject provided blood collected in both K₂EDTA and K₃EDTA. Each of the samples was analyzed in duplicate on the PENTRA XLR.

Bias was estimated at three points for each reticulocyte parameter: the low end of the distribution of observations, the mid-point, and the high end of the distribution. Bias was estimated separately for each replicate. Acceptance criteria were met for all measurands at all levels. These findings support the claim that K₂EDTA and K₃EDTA specimens give comparable results as measured on the PENTRA XLR hematology analyzer.

d. Determination of limit of blank, lower limits of detection and quantitation:

Analytical Limits at Low Level

Limit of Blank (LoB) and Limit of Detection (LoD) studies cannot be performed for reticulocyte parameters, because it is not possible to have result when there is no or not enough RBC in the sample. A verification of the LoB and LoD has been done for the RBC parameter.

Analytical sensitivity has been determined for the Reticulocyte parameters according to CLSI H44-A2 guideline. A verification of the LoQ according to CLSI EP17-A2 guideline has been done for the RBC parameter.

Limit of Blank (LoB)

Plasma samples, obtained by centrifugation of normal samples, were used as blank

samples, in order to be as close as possible as the blood sample matrix. To estimate the LoB, a total of 60 repeated measurements of different plasma are run in the same series (six different samples run 10 times). This test is performed on two PENTRA XLR instruments with two reagents lots. RBC results on PENTRA XLR met specifications and are consistent with ABX PENTRA 80 claim.

LoB obtained from 60 repeated measurements of six different plasma samples, are:

Measurand	LoB
RBC	$0 \times 10^6/\text{mm}^3$

Limit of Detection (LoD)

A set of six samples with very low parameter concentration are run 10 times over several days. To estimate the LoD, 60 results and calculate the pooled standard deviation (SDs). This test was performed on two PENTRA XLR instruments with two reagents lots. RBC results on PENTRA XLR met specifications and are consistent with ABX PENTRA 80 claim.

LoD obtained from 10 runs of 6 low samples on each instrument are:

Measurand	LoD
RBC	$0.01 \times 10^6/\text{mm}^3$

Analytical sensitivity - Limit of Quantitation (LoQ)

For Reticulocyte parameters:

To estimate the analytical sensitivity for reticulocyte parameters, a range of samples in low concentrations is prepared. Seven levels are prepared and run four times each. Testing was performed on three PENTRA XLR instruments using four reagents lots.

The analytical sensitivity data are considered acceptable when the distribution of the residuals is randomly distributed and r^2 higher than 0.98. The lowest value obtained in agreement with the acceptance criteria is the sensitivity limit and will be used as low limit for measuring range.

Analytical sensitivity limits obtained are:

Measurand	Analytical sensitivity
RET%	0.7%
RET#	$0.01 \times 10^6/\text{mm}^3$

For RBC parameter:

To estimate the limit of quantitation, several ranges of linearity in low concentrations were prepared. Between three to six samples per level were prepared and run at least five

times each, 40 replicates per level. The LoQ data are considered acceptable when the %Total-error is smaller than the desired total error for each measurand. This test is performed on two PENTRA XLR instruments with two reagents lots. RBC results on PENTRA XLR met specifications and are consistent with ABX PENTRA 80 claim.

LoQ obtained from at least 40 runs of 4 samples by level are:

Measurand	LoQ
RBC	$0.24 \times 10^6/\text{mm}^3$

e. Reference Intervals:

A total of 242 (122 female and 120 male) normal adult samples (whole blood samples collected in K₂EDTA) were analyzed in duplicate on the PENTRA XLR at two test sites in the US. The nonparametric data analysis method was used, depending only on the ranks of the reference data arranged in order of increasing size. Per EP28-A3, the reference interval is determined to be between and including the lower and upper reference limits, which enclose 95% of the values from the reference population subjects. Confidence intervals for the reference limit were calculated using a 90% probability (90% CI).

For each gender, a reference interval has been defined as described in the following table.

PENTRA XLR Reference Interval	FEMALES (N=122)		MALES (N=120)	
	LOW	HIGH	LOW	HIGH
RET%	0.70	2.35	0.70	2.06
RET# ($10^6/\text{mm}^3$)	0.022	0.106	0.024	0.106
CRC	0.44	2.27	0.51	2.15
IRF	0.018	0.191	0.033	0.232

These intervals are given in the labeling of the PENTRA XLR. However, expected values will vary with sample population and/or geographical location. Horiba highly recommends that each laboratory establish its own normal ranges based upon its local population.

K. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

L. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.