Summary of Safety and Effectiveness Data

I. General Information

Device Generic Name:	Rapid HIV-1 Antibody Test
Device Trade Name:	INSTI™
Applicant's Name and address:	bioLytical Laboratories, Inc. 1108-13351 Commerce Parkway Richmond, BC, Canada, V6V 2X7 Phone: 604-204-6784 Fax: 604-244-8399 e-mail: info@biolytical.com

Premarket Approval Application (PMA) Number: 090032/0
Date of Notice of Approval to the Applicant:

II. Indications for Use

The **INSTI™ HIV-1 Antibody Test** is a single use, rapid, *in vitro* qualitative immunoassay for the detection of antibodies to Human Immunodeficiency Virus Type 1 (HIV-1) in human venipuncture whole blood, fingerstick blood, or plasma specimens. The test is intended for use by trained personnel in point of care and laboratory situations to aid in the diagnosis of HIV-1 infections. If multiple rapid HIV tests are available, this test is suitable for use in appropriate multi-test algorithms.

III. Device Description

The **INSTI™ HIV-1 Antibody Test** is a manual, visually read, flow-through immunoassay for the qualitative detection of HIV-1 antibodies in human blood obtained from fingerstick or venipuncture, and plasma in as little as 60 seconds. The test consists of a nitrocellulose filtration membrane positioned atop an absorbent material within a plastic cartridge, referred to as the INSTI™ Membrane Unit. The membrane has been spotted with HIV-1 and HIV-2 recombinant proteins, which react with HIV antibodies in the specimen. Although the membrane contains HIV-1 and HIV-2 proteins, the assay has been validated for detection of HIV-1 antibodies only.

The membrane also includes a procedure control. The procedure control consists of a protein-A spot capable of capturing human immunoglobulin G (IgG) antibodies normally present in blood and blood components. The IgG antibodies then react with a chromatic agent contained in the INSTI Color Developer to produce a visual blue spot on the membrane. Since IgG antibodies are present in blood from normal or HIV- positive human specimens, the control spot provides a visual signal when the test is run, indicating that the test was performed correctly and the correct type and volume of specimen was added. If the control spot does not appear, the test is considered invalid.

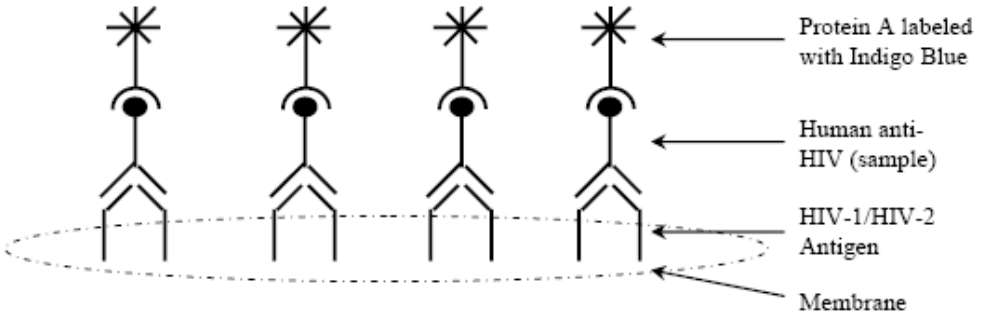
In the case of the test spot, recombinant HIV-1 and HIV-2 proteins bound to the membrane capture HIV-specific antibodies, if present in the specimen. Antibodies captured in the test spot react with a chromatic agent contained in the INSTI™ Color Developer to produce a blue spot on the membrane. The membrane unit is designed to filter, absorb, and retain the test specimen and all the test reagents in such a manner as to limit leakage and exposure of personnel to potentially infectious materials.

Reagents required to conduct a test include Sample Diluent (Solution 1), Color Developer (Solution 2) and Clarifying Solution (Solution 3).The test is performed by adding the fingerstick blood, venipuncture whole blood, or plasma specimen to the vial of Sample Diluent, which lyses the blood cells. This specimen/diluent solution is then poured into the well of the Membrane Unit. HIV antibodies, if present in the specimen, are captured by the HIV proteins on the filtration membrane. Color Developer is then added to the Membrane Unit. The Color Developer reacts with the captured antibodies to generate a distinct blue spot at the location of the control spot and, in the case that HIV antibodies are present in the specimen, a blue spot also appears at the location of the test spot on the membrane. In the final step, the Clarifying Solution is added to the membrane to decrease background color in order to make the control and test spots more distinct.

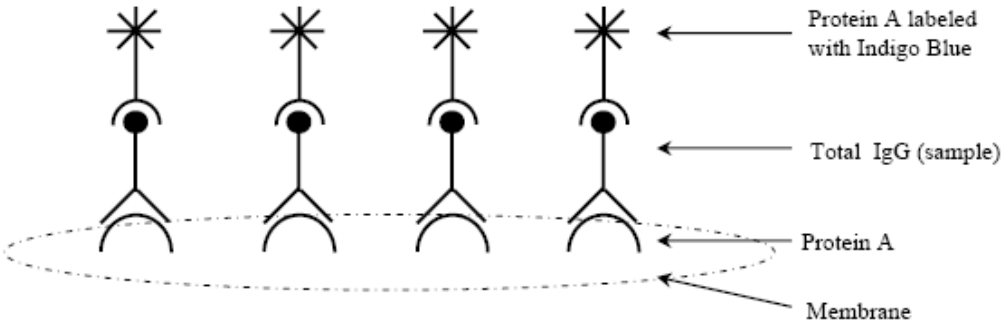
The INSTI™ is packaged as either a test kit pouch containing one test with support materials for single use, or as individual components for 24 tests with or without support material.

INSTI™ HIV-1 Assay Design

TEST SPOT



CONTROL SPOT



IV. Restrictions:

- Sale of the INSTI™ HIV-1Antibody Test is restricted to clinical laboratories
  - That have an adequate quality assurance program, including planned activities to provide adequate confidence that requirements for quality will be met; and
  - Where there is assurance that operators will receive and use instructional materials.
- The INSTI™ HIV-1 Antibody Test is approved for use only by an agent of a clinical laboratory.
- Test subjects must receive the “Subject Information” brochure prior to specimen collection and appropriate counselling when test results are provided.
- The INSTI™ HIV-1 Antibody Test is not approved for use to screen donors of blood, plasma, cells or tissues.

V. Warnings:

For *in vitro* diagnostic use only

1. **Read the entire Package Insert prior to beginning the test procedure. Complete conformance with the test procedure is necessary to ensure accurate results.**
2. **Before performing testing, operators must read and become familiar with Universal Precautions for Prevention of Transmission of Human Immunodeficiency Virus, Hepatitis B Virus and other Blood-borne Pathogens in Health-Care Settings.**
3. **Do not use the Membrane Unit if the foil pouch has been previously opened or if the packaging integrity of any component has been compromised. Once the Membrane Unit has been opened, it must be used immediately.**
4. **Sodium azide is present at 0.1% in all assay reagents. Sodium azide may react with lead or copper plumbing to form highly explosive metal azides. If products containing sodium azide are discarded into a drain, flush with large amounts of water to prevent azide build-up. Check with local regulatory agencies to determine at what concentration sodium azide may cause a product to be regulated as hazardous waste.**
5. **The performance characteristics of the INSTI™ HIV-1 Antibody Test have not been established for body fluids other than venipuncture whole blood, fingerstick blood, and plasma. Insufficient data are available to interpret tests performed on other body fluids, pooled blood plasma, or products made from such pools.**
6. **If the test kit is exposed to temperatures outside of the ambient temperature (15° – 30°C, 59° – 86°F), ensure it is brought to the ambient temperature range of 15° – 30°C, 59° – 86°F before performing testing. Use the INSTI™ Controls to ensure kit performance.**
7. **Patients that are receiving highly active antiretroviral therapy (HAART) may have undetectable levels of antibody to HIV-1 and give a false Non-Reactive INSTI™ HIV-1 Antibody Test result.**
8. **Specimens from patients with multiple myeloma may result in false Non-Reactive or invalid results with the INSTI™ HIV-1 Antibody Test.**
9. **Patients with elevated hemoglobin levels may test false Non-Reactive with the INSTI™ HIV-1 Antibody Test**.

VI. Limitations:

1. The INSTI™ HIV-1 Antibody Test must be used in accordance with the instructions in this package insert to obtain accurate results.
2. In some instances, specimens may exhibit longer than normal flow times (from the time the Sample Diluent specimen mixture is poured in the membrane well to the time the Clarifying Solution has fully flowed through the membrane). This is due to variable factors such as cellular components, especially with whole blood. As long as the contents from all three INSTI™ solution bottles completely flow through the membrane, regardless of flow time, the test can be properly interpreted according to the Interpretation of Results section of this package insert. **In occasional instances of long flow times, a faint result in the form of a ring may appear at the test spot location. This should be considered as a Reactive result.** In these instances, a venous blood specimen should be drawn in an anticoagulant blood collection tube, and forwarded to a laboratory for HIV confirmatory testing. **If any of the solutions completely stop flowing into the Membrane Unit, the procedure must be halted, a new specimen collected, and the procedure re-started from the beginning with fresh INSTI™ components.**
3. For a Reactive result, the intensity of the test spot does not necessarily correlate with the titer of antibody in the specimen.
4. The test is approved by FDA for use with fingerstick whole blood, venipuncture whole blood, and plasma specimens only. Other specimen types have not been evaluated and may give an incorrect result.
5. Use of other anticoagulants not listed in the Specimen Collection and Testing Procedure for Venipuncture Whole Blood and Plasma section of this package insert has not been evaluated and may give incorrect results.

6. Reading the test results after more than 5 minutes has elapsed following the addition of Clarifying Solution might produce erroneous results.
7. The INSTI™ HIV-1 Antibody Test detects antibodies to HIV-1 and is useful as an aid in the diagnosis of infection with HIV-1. Because a variety of factors may cause non-specific reactions, a patient found to be reactive using the INSTI™ HIV-1 Antibody Test should have a blood specimen drawn for laboratory-based confirmatory testing. A person who has antibodies to HIV is presumed to be infected with the virus and appropriate counseling and medical evaluation should be offered. The presence of HIV antibodies indicates past exposure to HIV but is not a diagnosis of AIDS, which can only be made by a physician. However, a non-reactive test does not rule out past exposure to HIV.
8. Patients that are receiving highly active antiretroviral therapy (HAART) may have undetectable levels of antibody to HIV-1 and give a false Non-Reactive INSTI™ HIV-1 Antibody Test result.
9. Specimens from patients with multiple myeloma may result in false Non-Reactive or invalid results with the INSTI™ HIV-1 Antibody Test.
10. Patients with elevated haemoglobin levels may test false Non-Reactive with the INSTI™ HIV-1 Antibody Test.
11. A person who has antibodies to HIV-1 is presumed to be infected with the virus, except a person who has participated in an HIV vaccine study may develop antibodies to the vaccine and may or may not be infected with HIV. Clinical correlation is indicated with appropriate counseling, medical evaluation, and possibly additional testing to decide whether a diagnosis of HIV infection is accurate.

VII. Alternative Practices and Procedures

The detection of HIV antibodies in human subjects is primarily conducted with laboratory based assays that use blood, serum, plasma and oral fluids. The majority of these tests use principles similar to the INSTI™ test device. Peptides, viral lysates or recombinant proteins are used to capture specific antibodies in a patient sample on a solid phase. The detection of these captured antibodies is accomplished with indicator systems including enzymes, colloids, or chemiluminescence components usually conjugated to antibody or antigen preparations. Once the test is complete the assays provide qualitative results usually interpreted as reactive, non-reactive or indeterminate (equivocal). Interpretation of laboratory assay results is usually conducted with an instrument. INSTI™ differs from these assays in two ways:1) INSTI™ results are interpreted visually rather than with an instrument. 2) INSTI™ uses a dye-based indicator system which is typically not used in laboratory assays.
Other commercially available HIV rapid test devices have been approved for use in point of care settings, and incorporate *immunochromatography* (lateral flow) or *immunofiltration* (flow through) principles in their design. INSTI™ differs from these assays in two ways: 1) INSTI™ uses a proprietary blue dye indicator system, producing a distinctive blue spot whereas most other rapid tests use colloidal gold, producing red colour results. 2) INSTI™ results can be interpreted within 60 seconds of adding the patient sample to the Membrane Unit, compared to 10-20 minutes for other rapid HIV tests.

VIII. Potential Adverse Effects of the Device on Health

No known adverse effects have been found with the INSTI™ HIV-1 Antibody Test in any study or application of the test to date. Although the risk of false Reactive or false Non-Reactive results with the INSTI™ test is small, as demonstrated by clinical studies, the potential for such inaccurate results exists. The risk of incorrect results is minimized by following the procedures and instructions outlined in the package insert.

IX. Summary of Pre-Clinical Studies

The following are brief summaries of the non-clinical laboratory studies that have been conducted to assess the performance of the INSTI™ HIV-1 Antibody Test:

Reactivity With HIV-1 Seroconversion Panels

Twenty three seroconversion panels were tested and the INSTI™ results were compared with licensed or approved anti-HIV EIA s. The results of this study are shown in Table 1. In this study, the INSTI™ HIV-1 Antibody Test demonstrated the ability to detect HIV-1 antibodies during seroconversion similar to FDA licensed or approved HIV EIAs.

Table 1
Comparison of the INSTI™ HIV-1 Antibody Test and Licensed or Approved Anti-HIV EIA Tests Using Seroconversion Panels.

Panel	Relative Bleed Day	INSTI™	EIA #1	EIA #2	EIA #3	EIA #4	EIA #5
PRB 904	0	NR	NR	NR	NR	NR	NR
	21	NR	NR	NR	NR	NR	NR
	49	NR	NR	NR	NR	NR	NR
	92	R	RR	RR	RR	RR	RR
	99	R	RR	RR	RR	RR	RR
PRB 910	0	NR	NR	NR	NR	NR	NR
	14	NR	NR	NR	NR	NR	NR
	26	R	RR	RR	RR	RR	RR
	28	R	RR	RR	RR	RR	RR
	32	R	RR	RR	RR	RR	RR
	35	R	RR	RR	RR	RR	RR
	40	R	RR	RR	RR	RR	RR
PRB 914	O	R	RR	RR	NR	NR	NR
	4	R	RR	RR	NR	RR	NR
	7	R	RR	RR	NR	RR	NR
	25	R	RR	RR	RR	RR	RR
	31	R	RR	RR	RR	RR	RR
PRB 916	0	NR	NR	NR	NR	NR	NR
	4	NR	NR	NR	NR	NR	NR
	9	NR	NR	NR	NR	NR	NR
	15	NR	NR	NR	NR	NR	NR
	30	R	RR	RR	RR	RR	RR
	35	R	RR	RR	RR	RR	RR
PRB 919	0	NR	NR	NR	NR	NR	NR
	9	R	RR	RR	NR	NR	NR
	11	R	RR	RR	NR	RR	NR
PRB 924	0	NR	NR	NR	NR	NR	NR
	2	NR	NR	NR	NR	NR	NR
	8	NR	NR	NR	NR	NR	NR
	10	NR	NR	NR	NR	NR	NR
	26	NR	NR	NR	NR	NR	NR
	33	R	NR	RR	NR	NR	NR
	35	R	RR	RR	NR	NR	NR
	40	R	RR	RR	NR	NR	RR
PRB 925	0	NR	NR	NR	NR	NR	NR
	10	NR	NR	NR	NR	NR	NR
	18	NR	NR	NR	NR	NR	NR
	22	NR	NR	NR	NR	NR	NR
	44	R	RR	RR	NR	NR	NR
	49	R	RR	RR	RR	RR	RR

PRB 926	0	NR	NR	NR	NR	NR	NR
	2	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	9	NR	NR	NR	NR	NR	NR
	27	R	RR	RR	RR	RR	RR
	32	R	RR	RR	RR	RR	RR
PRB 927	0	NR	NR	NR	NR	NR	NR
	28	NR	NR	RR	NR	NR	NR
	33	R	RR	RR	RR	RR	RR
	35	R	RR	RR	RR	RR	RR
	40	R	RR	RR	RR	RR	RR
PRB 928	0	NR	NR	NR	NR	NR	NR
	111	R	NR	RR	NR	NR	NR
	120	R	RR	RR	RR	RR	RR
	125	R	RR	RR	RR	RR	RR
	130	R	RR	RR	RR	RR	RR
PRB 929	0	NR	NR	NR	NR	NR	NR
	4	NR	NR	NR	NR	NR	NR
	14	NR	NR	NR	NR	NR	NR
	18	NR	NR	NR	NR	NR	NR
	21	NR	NR	NR	NR	NR	NR
	25	R	NR	RR	NR	NR	NR
	28	R	NR	RR	NR	RR	RR
PRB 934	0	NR	NR	NR	NR	NR	NR
	7	R	RR	RR	NR	NR	RR
	11	R	RR	RR	NR	RR	RR
PRB 935	0	NR	NR	NR	NR	NR	NR
	10	NR	NR	NR	NR	NR	NR
	16	NR	NR	NR	NR	NR	NR
	21	NR	NR	NR	NR	NR	NR
	24	NR	NR	NR	NR	NR	NR
	28	NR	NR	NR	NR	NR	NR
	43	R	RR	RR	RR	NR	RR
PRB 937	0	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	9	NR	NR	NR	NR	NR	NR
	14	NR	NR	NR	NR	NR	NR
	16	NR	NR	NR	NR	NR	NR
	21	NR	NR	RR	NR	NR	NR
PRB 938	0	NR	NR	NR	NR	NR	NR
	3	NR	NR	NR	NR	NR	NR
	9	R	NR	RR	NR	NR	NR
PRB 940	0	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	11	R	NR	RR	NR	NR	NR
	15	R	NR	RR	NR	RR	NR
	18	R	RR	RR	NR	RR	RR
	22	R	RR	RR	NR	RR	RR
	25	R	RR	RR	RR	RR	RR
	29	R	RR	RR	NR	RR	RR
PRB 941	0	NR	NR	NR	NR	NR	NR
	4	NR	NR	NR	NR	NR	NR
	9	NR	NR	NR	NR	NR	NR

	18	R	NR	RR	NR	NR	NR
	21	R	RR	RR	RR	NR	NR
	25	R	RR	RR	RR	RR	NR
PRB 943	0	NR	NR	NR	NR	NR	NR
	5	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	12	NR	NR	NR	NR	NR	NR
	14	NR	NR	RR	NR	NR	NR
	19	R	NR	RR	NR	NR	NR
	21	R	RR	RR	NR	NR	NR
PRB 944	0	NR	NR	NR	NR	NR	NR
	2	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	9	NR	NR	NR	NR	NR	NR
	14	R	NR	RR	NR	NR	NR
	16	R	RR	RR	NR	NR	NR
PRB 945	0	NR	NR	NR	NR	NR	NR
	3	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	13	NR	NR	RR	NR	NR	NR
	15	R	NR	RR	NR	NR	NR
	20	R	RR	RR	NR	NR	RR
PRB 947	0	NR	NR	NR	NR	NR	NR
	9	R	NR	RR	NR	NR	NR
	11	R	NR	RR	NR	NR	NR
	20	R	RR	RR	RR	RR	RR
PRB 950	0	NR	NR	NR	NR	NR	NR
	18	NR	NR	NR	NR	NR	NR
	21	NR	NR	NR	NR	NR	NR
	28	R	NR	RR	NR	RR	NR
PRB 952	0	NR	NR	NR	NR	NR	NR
	7	NR	NR	NR	NR	NR	NR
	10	NR	NR	NR	NR	NR	NR
	14	NR	NR	RR	RR	NR	NR
	17	R	RR	RR	RR	NR	NR
	21	R	RR	RR	RR	NR	RR

NR = Non Reactive; R = Reactive; RR = Repeatedly Reactive

Reactivity With HIV-1 Low Titer Panel

One HIV-1 low titer panel was tested with three production lots of INSTI™ and results were compared to FDA licensed or approved HIV assays. The results of this study are shown in Table 2. In this study, the INSTI™ HIV-1 Antibody Test was capable of detecting low levels of antibodies to HIV-1 similar to or better than FDA licensed or approved EIA's.

Table 2
Comparison of the INSTI™ HIV-1 Antibody Test and Licensed or Approved Anti-HIV EIA Tests Using a Low Titer HIV-1 Antibody Panel.

PANEL	MEMBER	INSTI™[1]	EIA #1	EIA #2	EIA #3

PRB 108	1	R	RR	RR	RR
	2	NR	NR	NR	NR
	3	R	RR	RR	RR
	4	R	RR	RR	RR
	5	R	RR	RR	RR
	6	R	RR	RR	RR
	7	R	RR	RR	RR
	8	R	RR	RR	RR
	9	R	RR	RR	NR
	10	R	RR	NR	NR
	11	R	RR	RR	RR
	12	R	RR	NR	NR
	13	R	RR	NR	NR
	14	R	RR	NR	NR
	15	R	RR	RR	RR

NR = Non Reactive; R = Reactive; RR = Repeatedly Reactive
[1]Identical INSTI™ results were obtained across the three production lots tested

Potentially Interfering Medical Conditions and Substances

To assess the impact of unrelated medical conditions or potentially interfering substances on the sensitivity and specificity of the INSTI™ HIV-1 Antibody Test, 195 serum/plasma specimens from a cross section of medical conditions unrelated to HIV-1 infection and 217 specimens with potentially interfering substances were tested unspiked and spiked with an HIV-1 positive specimen to give a low level of reactivity in the INSTI™ HIV-1 Antibody Test. The results are shown in **Tables 3 and 4**. In addition, a study was performed to assess the potential effect of common blood tube anticoagulants on assay sensitivity. Venipuncture blood was collected from 13 volunteer subjects in each of 3 tubes containing one of three anticoagulants (EDTA, sodium heparin, and sodium citrate). A total of13 specimens for each anticoagulant type were spiked with an HIV-1 positive specimen to give a low level of reactivity in the INSTI™ HIV-1 Antibody Test. Aliquots of the spiked specimens were stored refrigerated (2° – 8°C) and at ambient temperature (20° – 24°C) and tested at day 3 and day 7 over a 7 day period. There was no effect of the anticoagulants on sensitivity or specificity with specimens held up to 7 days at 2° - 24°C. The results are shown in **Tables 3 and 4**.

Table 3

INSTI™ HIV-1 Antibody Test reactivity with HIV-1 spiked specimens from individuals with potentially interfering medical conditions (n=195) and with HIV-1 spiked specimens containing potentially interfering substances (n=217).

Medical Condition (n=195)	No. of Specimens	INSTI™ Reactive	INSTI™ Nonreactive
Toxoplasmosis	20	20	0
Rheumatoid Factor	20	20	0
Multiple Myeloma	10	5[1]	0
Syphilis	30	30	0
SLE	5	5	0
Rubella	20	20	0
Cytomegalovirus	20	20	0
Epstein Barr Virus	20	20	0
HTLV-I/II panel	15	15	0
Hepatitis B Virus	20	20	0
Hepatitis A Virus	15	15	0
Potentially Interfering Substances (n=217)			
Icteric	20	20	0

Elevated Bilirubin (≥8.0mg/dL)	19	19	0
Lipemic	20	20	0
Visual Hemolysis	5	5	0
Elevated Triglyceride (≥292mg/dL)	19	19	0
Elevated Hemoglobin (>12g/100mL)	20	19[2]	1
Elevated Albumin (11.5-13.0g/dL)	15	15	0
EDTA	13	13	0
Sodium Heparin	13	13	0
Sodium Citrate	13	13	0
Bacterially Contaminated	60	60	0

[1]Up to 5 specimens from individuals with multiple myeloma produced invalid INSTI™ results depending on the INSTI™ kit lot tested.
[2]Of the 20 specimens from individuals with elevated hemoglobin, one tested false Non-Reactive in 2 out of 3 INSTI™ kit lots.

Table 4

INSTI™ HIV-1 Antibody Test specificity with specimens from individuals with potentially interfering medical conditions (n=195) and with specimens containing potentially interfering substances (n=217).

Medical Condition (n=195)	No. of Specimens	INSTI™ Reactive	INSTI™ Nonreactive
Toxoplasmosis	20	0	20
Rheumatoid Factor	20	0	20
Multiple Myeloma	10	0	5[1]
Syphilis	30	0	30
SLE	5	0	5
Rubella	20	0	20
Cytomegalovirus	20	0	20
Epstein Barr Virus	20	0	20
HTLV-I/II panel	15	0	15
Hepatitis B Virus	20	0	20
Hepatitis A Virus	15	0	20
Potentially Interfering Substances (n=217)			
Icteric	20	0	20
Elevated Bilirubin (≥8.0mg/dL)	19	0	19
Lipemic	20	0	20
Visual Hemolysis	5	0	5
Elevated Triglyceride (≥292mg/dL)	19	0	19
Elevated Hemoglobin (>12g/100mL)	20	0	20
Elevated Albumin (11.5-13.0g/dL)	15	0	15
EDTA	13	0	13
Sodium Heparin	13	0	13
Sodium Citrate	13	0	13
Bacterially Contaminated	60	0	60

[1]Up to 5 specimens from individuals with multiple myeloma produced invalid INSTI™ results depending on the INSTI™ kit lot tested. A related statement is listed in the **Warnings** and **Limitations** sections of the Package Insert.

Reproducibility

The reproducibility of the INSTI™ HIV-1 Antibody Test was tested at 3 laboratory sites using 3 lots of the INSTI™ HIV-1 Antibody Test on 3 separate days with 9 operators (3 per site). A panel of 5 blind-coded contrived plasma specimens, consisting of 4 HIV-1 antibody positive (one strong positive and three low positive)and 1 HIV antibody

negative specimen was tested at each site. A total of 405 tests were conducted, 135 at each site, with a total of 81 tests per panel specimen. The overall reproducibility of the INSTI™ HIV-1 Antibody Test was 405/405 = 100%.

Detection of Anti HIV-1 non-B Subtypes

To assess the sensitivity of the INSTI™ HIV-1 Antibody Test for detection of antibodies to non-B subtypes of HIV-1, a total of 207 serum/plasma specimens collected from individuals from various geographic regions who were infected with non-B subtypes of HIV-1 were tested. Of these 207 specimens, a total of 206 were Reactive with the INSTI™ HIV-1 Antibody Test, for an overall sensitivity of 99.5% (95% CI = 97.3-99.9%). One subtype A specimen tested false Non-Reactive. The INSTI™ results and HIV-1 non-B subtype listings are presented in **Table 5** below.

Table 5

Sensitivity of the INSTI™ HIV-1 Antibody Test for Detection of Antibodies to HIV-1 Non-B Subtypes

HIV Subtype	Number of Specimens	INSTI™ Reactive
A	28	27
C	57	57
D	22	22
E	7	7
F	9	9
G	10	10
H	2	2
J	4	4
K	1	1
O	23	23
AE	11	11
AG	31	31
CRF06	1	1
CRF11	1	1
TOTAL	207	206

X. Summary of Clinical Studies

SENSITIVITY

A sensitivity study was performed in 14 clinical trial sites using freshly obtained matching fingerstick whole blood, EDTA whole blood, and EDTA plasma specimens collected from 1076 individuals known to be infected with HIV-1. Additionally, matching fingerstick whole blood, EDTA whole blood, and EDTA plasma specimens were collected from 782 previously unscreened individuals from populations at high risk for HIV-1 from which 22 were confirmed seropositive by an FDA licensed test. For the 1098 total HIV-1 positives, results for fingerstick whole blood, venipuncture whole blood and plasma are shown in **Tables 6, 7, 8 and 9.**

Table 6

Detection of Antibody to HIV-1 in Fingerstick Whole Blood Specimens from Seropositive Individuals and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Reactive	INSTI™ Non-Reactive	INSTI™ Invalid[1]	Approved Test Repeatedly Reactive	Approved Test Non-Reactive	True Positive[2]
Known HIV-	1075	1074	1	0	1075	0	1075

1 Positive							
High Risk	782	22[3]	756[3]	4	22	760	22
TOTAL	1857	1096	757	4	1097	760	1097

[1] Invalid results were not included in the calculation of sensitivity. The 4 specimens which gave invalid results on INSTI™ were Non-Reactive on the approved test.
[2] Confirmed by licensed HIV-1 Western Blot
[3] Of the 22 true positive specimens, 1 was Non-Reactive on INSTI™ (false Non-Reactive). One other specimen was false Reactive on INSTI™.

Of the 1076 known HIV-1 positive individuals, one did not provide a fingerstick specimen. Of the 1075 fingerstick specimens collected from the known HIV-1 positive patients that were repeatedly Reactive by an FDA approved test, 1074 gave a Reactive result with INSTI™. Within the high risk group, 22 specimens were confirmed seropositive by an FDA licensed test and, of those, 21 were Reactive with INSTI™. One specimen was false Non-Reactive on INSTI. One additional fingerstick specimen from the high risk population was INSTI false Reactive (see Table 7 below). The overall sensitivity of the INSTI™ HIV-1 Antibody Test in **fingerstick whole blood** specimens for the confirmed HIV-1 positives from the combined high risk and known HIV-1 positive populations was calculated to be 1095/1097= 99.8% (95% CI = 99.3% - 99.9%). The rate of invalid tests was 4/1857 (0.2%).

Table 7

Comparison of Results for Fingerstick Whole Blood Specimens from Seropositive Individuals and Individuals at High Risk of HIV Infection

		Approved Test Result		
INSTI™ Test Result		Reactive	Non-Reactive	TOTAL
	Reactive	1095	1[1]	1096
	Non-Reactive	2[2]	755	757
	Invalid	0	4	4
	TOTAL	1097	760	1857

[1] The one specimen that gave a false Reactive result on INSTI™ was from an individual at high risk for HIV infection.
[2] Of the two false Non-Reactive specimens on INSTI™, one was from an individual known to be infected with HIV-1 and one was from an individual at high risk for HIV infection.

Table 8

Detection of Antibody to HIV-1 in Venipuncture Whole Blood Specimens from Seropositive Individuals and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Reactive	INSTI™ Non-Reactive	INSTI™ Invalid	Approved Test Repeatedly Reactive	Approved Test Non-Reactive	True Positive[1]
Known HIV-1 Positive	1076	1075	1	0	1076	0	1076
High Risk	782	22	760	0	22	760	22

TOTAL	1858	1097	761	0	1098	760	1098

[1]Confirmed by licensed HIV-1 Western Blot

Of the 1076 known HIV-1 positive EDTA whole blood specimens, 1075 gave Reactive results with INSTI™. Within the high risk group, 22 EDTA whole blood specimens were confirmed seropositive by an FDA licensed test and these same 22 were Reactive with INSTI. The overall sensitivity of the INSTI™ HIV-1 Antibody Test in **venipuncture whole blood** specimens for the confirmed HIV-1 positives from the combined high risk and known HIV-1 positive populations was calculated to be 1097/1098 = 99.9% (95% CI = 99.5% - 100%).

Table 9

Detection of Antibody to HIV-1 in Plasma Specimens from Seropositive Individuals and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Reactive	INSTI™ Non-Reactive	INSTI™ Invalid	Approved Test Repeatedly Reactive	Approved Test Non-Reactive	True Positive[1]
Known HIV-1 Positive	1076	1075	1	0	1076	0	1076
High Risk	782	22	760	0	22	760	22
TOTAL	1858	1097	761	0	1098	760	1098

[1]Confirmed by licensed HIV-1 Western Blot

Of the 1076 known HIV-1 positive EDTA plasma specimens, 1075 gave Reactive results with INSTI™. Within the high risk group, 22 plasma specimens were confirmed seropositive by an FDA licensed test and these same 22 were Reactive with INSTI™. The overall sensitivity of the INSTI™ HIV-1 Antibody Test in **plasma** specimens for the confirmed HIV-1 positives from the combined high risk and known HIV-1 positive populations was calculated to be 1097/1098 = 99.9% (95% CI = 99.5% - 100%).

SPECIFICITY

A specificity study was performed in the same 14 clinical trial sites using freshly obtained matching fingerstick whole blood, EDTA whole blood, and EDTA plasma specimens collected from 1410 low or unknown risk and high risk individuals. Of the 1388 individuals identified as HIV negative using an approved comparator assay, 2 did not provide a fingerstick specimen. Of the remaining 1386 fingerstick specimens, 1376 gave a Non-Reactive result with INSTI™, and 4 were invalid. Within the high risk group, 22 specimens were confirmed seropositive by Western Blot and of those, 21 were Reactive with INSTI™; an additional 1/782 high risk specimens was INSTI™ false Reactive. Of the 1388 matching EDTA whole blood and plasma specimens, 1388 gave Non-Reactive results with INSTI™. Results are shown in **Tables 10, 11, 12, 13 and 14**.

Table 10

Performance of the INSTI™ HIV-1 Antibody Test on Fingerstick Whole Blood Specimens from Individuals Presumed to be HIV Negative and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Non-Reactive	INSTI™ Reactive	INSTI™ Invalid[1]	Approved Test Non-Reactive	Approved Test Reactive	True Negative[2]
Low or Unknown Risk	626	620	6	0	626	0	626
High Risk	782	756	22[3]	4	760	22	760

TOTAL	1408	1376	28	4	1386	22	1386

[1] Invalid results were not included in the calculations of specificity. The 4 specimens which gave invalid results on INST™ were Non-Reactive on the approved test.
[2] Reactives were confirmed by licensed HIV-1 Western Blot and excluded from the calculation of specificity.
[3] Of the 22 INSTI™ Reactive specimens, one was Non-Reactive by the approved test, ie. INSTI™ false Reactive.

A total of 7 INSTI™ false Reactive results (1 from the high risk group, 6 from the low or unknown risk group) were obtained from the 1382 specimens from HIV-negative individuals that produced valid INSTI™ results (see Tables 11 and 12 below). From Table 10, the overall specificity of the INSTI™ HIV-1 Antibody Test in **fingerstick whole blood** specimens from the combined high risk and low or unknown risk populations, minus the invalid results, was calculated to be 1375/1382 = 99.5% (95% CI = 99.0% - 99.8%). From Table 11, the specificity in the high risk populations, minus the invalid results, was calculated to be 755/756 = 99.9% (95% CI = 99.3% - 100%). From Table 12, the specificity of the INSTI™ HIV-1 Antibody Test from low or unknown risk populations was calculated to be 620/626 = 99.0% (95% CI = 97.9% - 99.6%).

Table 11

Comparison of Results for Fingerstick Whole Blood Specimens from Individuals at High Risk of HIV Infection

		Approved Test Result		
INSTI™ Test Result		Reactive	Non-Reactive	TOTAL
	Reactive	21	1	22
	Non-Reactive	1	755	756
	Invalid	0	4	4
	TOTAL	22	760	782

Table 12

Comparison of Results for Fingerstick Whole Blood Specimens from Low and Unknown Risk Individuals Presumed to be Negative for HIV Infection

		Approved Test Result		
INSTI™ Test Result		Reactive	Non-Reactive	TOTAL
	Reactive	0	6	6
	Non-Reactive	0	620	620
	TOTAL	0	626	626

Table 13

Performance of the INSTI™ HIV-1 Antibody Test on Venipuncture Whole Blood Specimens from Individuals Presumed to be HIV Negative and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Non-	INSTI™ Reactive	INSTI™ Invalid	Approved Test Non-	Approved Test	True Negative[1]

		Reactive			Reactive	Reactive	
Low or Unknown Risk	628	628	0	0	628	0	628
High Risk	782	760	22	0	760	22	760
TOTAL	1410	1388	22	0	1388	22	1388

[1] Reactives were confirmed by licensed HIV-1 Western Blot and excluded from the calculation of specificity.

The overall specificity of the INSTI™ HIV-1 Antibody Test in **venipuncture whole blood** specimens from the combined HIV negative high risk and low or unknown risk populations, was calculated to be 1388/1388 = 100% (95% CI = 99.7% - 100%).

Table14

Performance of the INSTI™ HIV-1 Antibody Test on Plasma Specimens from Individuals Presumed to be HIV Negative and Individuals at High Risk of HIV Infection

Test Group	Total Specimens	INSTI™ Non-Reactive	INSTI™ Reactive	INSTI™ Invalid	Approved Test Non-Reactive	Approved Test Reactive	True Negative[1]
Low or Unknown Risk	628	628	0	0	628	0	628
High Risk	782	760	22	0	760	22	760
TOTAL	1410	1388	22	0	1388	22	1388

[1] Reactives were confirmed by licensed HIV-1 Western Blot and excluded from the calculation of specificity.

The overall specificity of the INSTI™ HIV-1 Antibody Test in **plasma** specimens from the combined HIV negative high-risk and low or unknown risk populations, was calculated to be 1388/1388 = 100% (95% CI = 99.7% - 100%).

XI. Conclusions Drawn from Studies

Risk/Benefit Analysis
Rapid tests for identification of antibodies to HIV, particularly if conducted in point of care settings, provide significant advantages for the testing population. The risk of false Reactive or false Non-Reactive results with the INSTI™ test is small, as demonstrated by clinical studies. This very low risk of false results must be weighed against the benefits of being tested to the patient and to public health surveillance and prevention initiatives.

Safety
No significant adverse events were observed in any of the clinical studies conducted. All operators conducted testing in accordance with instructions for use of the INSTI™ device and training provided.

Effectiveness
The accuracy of the INSTI™ HIV-1 Antibody test for all specimen types studied (fingerstick whole blood, venipuncture whole blood, plasma) is greater than or equal to 99.5% with the lower boundary of the 95% confidence interval (CI) greater than or equal to 99.0% for all sample types. This meets the FDA established requirements for approval of a rapid HIV test device.