**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION**
**DECISION SUMMARY**

**A. 510(k) Number:**

K140957

**B. Purpose for Submission**:

New device

**C. Manufacturer and Instrument Name**:

Applied Spectral Imaging Ltd., GenASIs HiPath IHC Family

**D. Type of Test or Tests Performed**:

Computer-assisted image analysis algorithm for scoring immunohistochemically stained HER2/neu slides, estrogen receptor (ER)/progesterone receptor (PR) slides, and ki67 slides. The image analysis algorithm scoring is confirmed by the pathologist before reporting the scores.

**E. System Descriptions:**

1. Device Description:

The GenASIs HiPath IHC Family, including software is designed to assist the qualified pathologist in the consistent assessment in immohistochemnically stained histologic sections from formalin-fixed, paraffin-embedded breast cancer tissues. The device consists of a slide capture camera, microscope, computer, monitor, keyboard, mouse, image analysis algorithms for specific immunohistochemical markers, and software with a Graphic User Interface (GUI).

The GenASIs HiPath IHC Family is an intranet-based, software application that allows pathology laboratories, to acquire, manage, view, analyze, share, and report test results of pathology specimens. Using the GenASIs HiPath IHC Family software the pathologist can view captured images, add annotations, make measurements, perform image analysis and generate reports.

**Hardware**: A camera based acquisition device designed to captures bright-field microscope digital images of formalin-fixed, paraffin-embedded breast tissues that are suitable for storage and viewing. The device includes a digital slide acquisition camera, X-Y Stage with holder adaptor for loading glass slides on a microscope and a workstation including a computer, keyboard, mouse and monitor.

**Software**: The GenASIs HiPath IHC Family software is designed to complement the

routine workflow of a qualified pathologist in the review of specific immunohistochemically stained FFPE breast histologic slides. It allows the user to select fields of view (FOVs) in the digital image for analysis and provides quantitative data on these FOVs to assist with interpretation. The software makes no independent interpretations of the data and requires competent human intervention for all steps in the analysis process.

2. Principles of Operation:

   The pathologist selects the ROI (Region Of Interest) of the immunohistochemically stained glass slides by using the review microscope. The desired ROI of the glass slide is digitized by an image acquisition system and an Image Analysis scoring algorithm is applied on the digitized image. The image analysis application score is presented to the pathologist on the screen who must verify agreement with the Image Analysis software application score by manually reviewing the slide under the optical microscope.

3. Modes of Operation:

   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:

   Glass slides are identified by slide label.

5. Specimen Sampling and Handling:

   Specimen glass slides are mounted manually on the microscope by the pathologist to be imaged by the camera mounted on the microscope.

6. Calibration:

   Camera White Balance: The image acquisition program automatically adjusts white balance for each image it acquires.

7. Quality Control:

   Performed by the operator as part of the standard operation

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 §864.1860 Immunohistochemistry reagents and kits

2. Classification:

   Class II

3 Product code:

   NQN – Microscope, Automated, Image Analysis, Immunohistochemistry, Operator Intervention, Nuclear Intensity & Percent Positivity

   NOT – Microscope, Automated, Image Analysis, Operator Intervention

4. Panel:

   Pathology (88)

**G. Intended Use:**

1. Indication(s) for Use:

   The GenASIs HiPath IHC Family provides image capture, management, analysis, and viewing of specific immunohistochemically stained slides. It is intended for in vitro diagnostic use as an aid to the pathologist in the display, detection, counting, review and classification of tissues and cells of clinical interest based on particular morphology, color, intensity, size, pattern and shape:

   1. The GenASIs HiPath IHC Family for HER2 (4B5) is for image capture and analysis applications. This particular system is intended for use as an aid to the pathologist in the detection and semi-quantitative measurement of HER2 protein in formalin-fixed, paraffin-embedded breast cancer tissue. This device is an accessory to Ventana Medical Systems, Inc. PATHWAY® anti-HER2/neu (4B5) Rabbit Monoclonal Primary Antibody. The PATHWAY® anti-HER2/neu (4B5) Rabbit Monoclonal Primary Antibody is indicated for use as an aid in the assessment of breast cancer patients for whom HERCEPTIN® (Trastuzumab) treatment is being considered.

NOTE: The GenASIs HiPath IHC Family for HER2 (4B5) image capture and analysis applications are adjunctive computer-assisted methodologies for the qualified pathologist in the acquisition and semi-quantitative measurement of images from microscope glass slides of breast cancer specimens stained for the presence of HER-2/neu receptor protein. The pathologist should verify agreement with the Image Analysis software application score by reviewing the glass slide under the microscope. The accuracy of the test results depends on the quality of the immuohistochemical staining. It is the responsibility of a qualified pathologist to employ appropriate morphological studies and controls as specified in the instructions for the PATHWAY® anti-HER-2/neu (4B5) Rabbit Monoclonal Primary Antibody assay used to assure the validity of the GenASIs HiPath IHC Family for HER2 (4B5) image capture and analysis scores. The actual correlation of PATHWAY® anti-HER-2/neu (4B5) to clinical outcome has not been established.

2. The GenASIs HiPath IHC Family for PR (1E2) is for image capture and analysis applications. This particular system is intended for use as an aid to the pathologist in the detection and qualitative measurement of progesterone receptor (PR) protein in formalin-fixed, paraffin-embedded breast cancer tissue. This device is an accessory to Ventana Medical Systems, Inc. CONFIRMTM anti-Progesterone Receptor (PR) (1E2) Rabbit Monoclonal Primary Antibody assay. The CONFIRMTM anti-Progesterone Receptor (PR) (1E2) Rabbit Monoclonal Primary Antibody assay is indicated for use as an aid in the assessment of breast cancer patients for whom endocrine treatment is being considered (but is not the sole basis for treatment).

Note: The GenASIs HiPath IHC for PR (1E2) image capture and analysis applications are adjunctive computer-assisted methodologies for the qualified pathologist in the acquisition and qualitative measurement of images from microscope glass slides of breast cancer specimens stained for the presence of PR protein. The pathologist should verify agreement with the Image Analysis software application score by reviewing the glass slide under the microscope. The accuracy of the test results depends on the quality of the immunohistochemical staining. It is the responsibility of a qualified pathologist to employ appropriate morphological studies and controls as specified in the instructions for the CONFIRMTM anti-Progesterone Receptor (PR) (1E2) Rabbit Monoclonal Primary Antibody used to assure the validity of the GenASIs HiPath IHC Family for PR (1E2) image capture and analysis scores. The actual correlation of CONFIRMTM anti-PR antibody to clinical outcome has not been established.

3. The GenASIs HiPath IHC Family for ER (SP1) is for image capture and analysis applications. The particular system is intended for use as an aid to the pathologist in the detection and qualitative measurement of ER (SP1): protein in formalin-fixed, paraffin-embedded breast cancer tissue. This device is an accessory to the Ventana Medical Systems, Inc. CONFIRMTM anti-Estrogen Receptor (ER) (SP1) Rabbit Monoclonal Primary Antibody. The Ventana Medical Systems, Inc. CONFIRMTM anti-Estrogen Receptor (ER) (SP1) Rabbit Monoclonal Primary Antibody is indicated for use as an aid in the assessment of ER status in breast cancer patients (but is not the sole basis for treatment).

Note: The GenASIs HiPath IHC Family for ER (SP1) image capture and analysis applications are adjunctive computer-assisted methodologies for the qualified pathologist in the acquisition and qualitative measurement of images from microscope glass slides of breast cancer specimens stained for the presence of ER protein. The pathologist should verify agreement with the Image Analysis software application score by reviewing the glass slide under the microscope. The accuracy of the test results depends on the quality of the immuohistochemical staining. It is the responsibility of a qualified pathologist to employ appropriate morphological studies and controls as specified in the instructions for the CONFIRMTM anti-Estrogen Receptor (ER) (SP1) Rabbit Monoclonal Primary Antibody to assure the validity of the GenASIs HiPath IHC Family for ER (SP1) image capture and analysis scores. The actual correlation of CONFIRMTM anti-Estrogen Receptor (ER) (SP1) Rabbit Monoclonal Primary Antibody to clinical outcome has not been established.

4. The GenASIs HiPath IHC Family for Ki67 (30-9) is for image capture and analysis applications. The particular system is intended for use as an aid to the pathologist in the detection and qualitative measurement of Ki67 (30-9): protein in formalin-fixed, paraffin-embedded breast cancer tissue. This device is an accessory to the Ventana Medical Systems, Inc. CONFIRMTM anti-Ki67 (30-9) Rabbit Monoclonal Primary Antibody assay. The Ventana Medical Systems, Inc. CONFIRMTM anti-Ki67 (30-9) assay is indicated for use in assessing the proliferative activity of breast cancer tissue. When used with this assay, the GenASIs HiPath IHC Family for Ki67 (30-9) is indicated for use as an aid in the assessment of Ki-67 status in breast cancer patients (but is not the sole basis for treatment).

Note: The GenASIs HiPath IHC Family for Ki67 (30-9) image capture and analysis applications are adjunctive computer-assisted methodologies for the qualified pathologist in the acquisition and qualitative measurement of images from microscope glass slides of breast cancer specimens stained for the presence of Ki67 protein. The pathologist should verify agreement with the Image Analysis software application score by reviewing the glass slide under the microscope. The accuracy of the test results depends on the quality of the immunohistochemical staining. It is the responsibility of a qualified pathologist to employ appropriate morphological studies and controls as specified in the instructions for the CONFIRMTM anti-Ki67 (30-9) Rabbit Monoclonal Primary Antibody assay to assure the validity of the GenASIs HiPath IHC Family for Ki67 (30-9) image capture and analysis scores. The actual correlation of CONFIRMTM anti-Ki67 (30-9) Rabbit Monoclonal Primary antibody assay to clinical outcome has not been established.

2. Special Conditions for Use Statement(s):

- For prescription use only.
- Results acquired using this product must always be reviewed by a qualified Pathologist by reviewing the slides under the microscope.

**H. Substantial Equivalence Information:**

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

   Virtuoso™ System by Ventana Digital Pathology, K111543 (HER2), K111869 (PR), K130515 (ER) and K111755 (Ki67)

2. Comparison with Predicate Device:

| **Similarities** | | |
|---|---|---|
| Item | Device | Predicate |
| Intended Use | The GenASIs HiPath IHC Family provides image capture, management, analysis, and viewing. | Same |
| Intended Use | Intended for *in vitro* diagnostic use as an aid to the pathologist in the display, detection, counting, review and classification of tissues and cells of clinical interest based on particular morphology, color, intensity, size, pattern and shape. Intended for use as an aid to the pathologist in the detection and qualitative or semi-quantitative measurement of HER2, PR, ER and Ki67 (30-9) protein in formalin-fixed, paraffin-embedded normal and neoplastic tissue. Indicated as an aid to pathologists in the assessment of HER2, PR, ER and Ki67 (30-9) status in breast cancer patients | Same |
| ROI Selection | Image and Region Of Interest (ROI) selected by the pathologist | Same |
| Scoring Algorithm | Based cells classification, an overall score for the image is computed using the numbers of stained cells, non-stained cells, and total cells for the calculations. | Same |
| Main SW processes | • Control of man-machine Interface<br>• Slide Acquisition/ Capture<br>• Handling the images display, storage, communication and reporting.<br>• Image manipulation<br>• Calibration and Quality Control | Same |

| Similarities | | |
|---|---|---|
| Item | Device | Predicate |
| | • allows the user to select fields of view (FOVs)<br>• Provides quantitative data on these FOVs to assist with interpretation | |

| Differences | | |
|---|---|---|
| Item | Device | Predicate |
| Specimen Identification | Identified by slide label. | Identified by slide label or barcode. |
| Slide loading | Manually by pathologist | Multi-slide automatic loading |
| Calibration | Camera White Balance. The image acquisition program automatically adjusts white balance for each image it acquires. | Tests must be run with nine custom slides calibrated for the module which can be obtained from the sponsor. |
| Quality Control | Performed by the operator as part of the standard operation procedures. | Performed by the operator before releasing the images to the pathologist for review. Slides with sub-optimal images will be rescanned |
| Slide acquisition/ Capture | CCD high resolution color camera and racks for loading the glass slides.<br>Images to be processed selected via microscope's eyepiece. | Digital slide bright field scanner (Ventana iScan) and racks for loading glass slides.<br>Images to be processed selected from the screen digital read image. |
| Device Components | Microscope, CCD color camera, PC, keyboard, Mouse, Color Monitor, X-Y stage and rack for loading 1 glass slide. | Digital slide scanner, PC, keyboard, Mouse, color Monitor, racks for loading up to 160 glass slides. |
| Illumination Method | Xenon Lamp | Integrated LED (part of the Slide scanner) |
| ROI Selection | Image and Region Of Interest (ROI) selected by the pathologist using the microscope's eyepiece. | Region of Interest (ROI) selected by the pathologist using the digital read image on the screen. |

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

1. Guidance for Industry "Guidance for Submission of Immunohistochemistry Applications to the FDA", dated June 3 1998.
2. CLSI I/LA28-A2Q:2009 - Quality assurance for design control and implementation of immunohistochemistry assays; approved guidelines-second edition. (InVitro Diagnostics).
3. Guidance for Industry and FDA Staff: "Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests"; March 2007.
4. "Guidance for the Content of Premarket Submission for Software Contained in Medical Device", CDRH, May 2005.
5. Guidance for Industry, Clinical Investigators, Institutional Review Boards and Food and Drug Administration Staff, Doc. 1776 dated November 2013

## J. Performance Characteristics*:*

1. Analytical Performance:

*a. Accuracy*:

The GenASIs HiPath IHC Family for ER (SP1)

Table 1 presents frequency distribution of the study cases and final outcome by manual and GenASIs HiPath IHC Family reading by site and pooled over all three sites.

**Table 1 ER Results: Manual versus GenASIs HiPath IHC 3x3 Frequency Distribution**

| Center / Scan Outcome | | Manual Outcome | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|
| | | Negative (<1%) | | Around cutoff (1% to 20%) | | Positive (>20%) | | | |
| | | N | % | N | % | N | % | N | % |
| HaEmek | Negative (<1%) | 46 | 100.0 | 0 | 0 | 0 | 0 | 46 | 31.1 |
| | Around cutoff (1% to 20%) | 0 | 0 | 8 | 88.9 | 0 | 0 | 8 | 5.4 |
| | Positive (>20%) | 0 | 0 | 1 | 11.1 | 93 | 100.0 | 94 | 63.5 |
| S&W | Negative (<1%) | 53 | 100.0 | 1 | 7.7 | 0 | 0 | 54 | 37.2 |
| | Around cutoff (1% to 20%) | 0 | 0 | 12 | 92.3 | 0 | 0 | 12 | 8.3 |
| | Positive (>20%) | 0 | 0 | 0 | 0 | 79 | 100.0 | 79 | 54.5 |
| Spartanburg | Negative (<1%) | 52 | 98.1 | 0 | 0 | 0 | 0 | 52 | 38.8 |
| | Around cutoff (1% to 20%) | 1 | 1.9 | 9 | 81.8 | 0 | 0 | 10 | 7.5 |
| | Positive (>20%) | 0 | 0 | 2 | 18.2 | 70 | 100.0 | 72 | 53.7 |
| Overall | Negative (<1%) | 151 | 99.3 | 1 | 3.0 | 0 | 0 | 152 | 35.6 |
| | Around cutoff (1% to 20%) | 1 | 0.7 | 29 | 87.9 | 0 | 0 | 30 | 7.0 |
| | Positive (>20%) | 0 | 0 | 3 | 9.1 | 242 | 100.0 | 245 | 57.4 |
| Total | | 152 | 100.0 | 33 | 100.0 | 242 | 100.0 | 427 | 100.0 |

Table 2 includes ER 2x2 accuracy statistics for each site and pooled over all three sites,

along with the corresponding PPA, NPA, and OA, and their 95% two-sided Exact Binomial confidence intervals.

**Table 2 ER 2x2 accuracy statistics for each site and pooled over all three sites**

| ER Frequency Distribution of Agreement | | Manual Analysis | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 148) | | Site 2 (n = 145) | | Site 3 (n = 134) | | Overall (n = 427) | |
| | | Neg | Pos | Neg | Pos | Neg | Pos | Neg | Pos |
| **Analyze with HiPath System** | Neg (<1%) | 46 | 0 | 53 | 1 | 52 | 0 | 151 | 1 |
| | Pos (≥1%) | 0 | 102 | 0 | 91 | 1 | 81 | 1 | 274 |
| | Total | 46 | 102 | 53 | 92 | 53 | 81 | 152 | 275 |
| % Overall agreement (95% CI) | | 100% (98% - 100%) | | 99% (96% - 100%) | | 99% (96% - 100%) | | 100.0% (98% - 100%) | |
| Negative Agreement (95% CI) | | 100% (92% - 100%) | | 100% (93% - 100%) | | 98% (90% - 100%) | | 99% (96% - 100%) | |
| Positive Agreement (95% CI) | | 100% (96% - 100%) | | 99% (94% - 100%) | | 100% (96% - 100%) | | 100% (98%-100%) | |

The lower limit of 95% confidence intervals for pooled overall centers results is 98%, 96% and 98% for overall, negative and positive agreements respectively. They are above the pre-specified acceptance criteria of 90%.

The GenASIs HiPath IHC Family for PR (1E2)

Table 3 presents frequency distribution of the study cases and final outcome by manual and GenASIs HiPath IHC Family reading by site and pooled over all three sites.

**Table 3 PR Results: Manual versus GenASIs HiPath IHC 3x3 Frequency Distribution**

| Center / Scan Outcome | | Manual Outcome | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|
| | | Negative (<1%) | | Around cutoff (1% to 10%) | | Positive (>10%) | | | |
| | | N | % | N | % | N | % | N | % |
| HaEmek | Negative (<1%) | 51 | 96.2 | 0 | 0 | 0 | 0 | 51 | 42.5 |
| | Around cutoff (1% to 10%) | 2 | 3.8 | 9 | 69.2 | 0 | 0 | 11 | 9.2 |
| | Positive (>20%) | 0 | 0 | 4 | 30.8 | 54 | 100.0 | 58 | 48.3 |
| S&W | Negative (<1%) | 57 | 100.0 | 1 | 5.6 | 0 | 0 | 58 | 43.3 |
| | Around cutoff (1% to 10%) | 0 | 0 | 16 | 88.9 | 1 | 1.7 | 17 | 12.7 |
| | Positive (>10%) | 0 | 0 | 1 | 5.6 | 58 | 98.3 | 59 | 44.0 |
| Spartanburg | Negative (<1%) | 53 | 98.1 | 0 | 0 | 0 | 0 | 53 | 40.5 |
| | Around cutoff (1% to 10%) | 1 | 1.9 | 16 | 88.9 | 0 | 0 | 17 | 13.0 |
| | Positive (>10%) | 0 | 0 | 2 | 11.1 | 59 | 100.0 | 61 | 46.6 |
| Overall | Negative (<1%) | 161 | 98.2 | 1 | 2.0 | 0 | 0 | 162 | 42.1 |
| | Around cutoff (1% to 10%) | 3 | 1.8 | 41 | 83.7 | 1 | 0.6 | 45 | 11.7 |
| | Positive (>10%) | 0 | 0 | 7 | 14.3 | 171 | 99.4 | 178 | 46.2 |
| Total | | 164 | 100.0 | 49 | 100.0 | 172 | 100.0 | 385 | 100.0 |

Table 4 includes PR 2x2 accuracy statistics for each site and pooled over all three sites, along with the corresponding PPA, NPA, and OA, and their 95% two- sided Exact Binomial confidence intervals.

**Table 4 PR 2x2 accuracy statistics for each site and pooled over all three sites**

| PR Frequency Distribution of Agreement | | Manual Analysis | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 120) | | Site 2 (n = 134) | | Site 3 (n = 131) | | Overall (n = 385) | |
| | | Neg | Pos | Neg | Pos | Neg | Pos | Neg | Pos |
| Analyze with HiPath System | Neg (<1%) | 51 | 0 | 57 | 1 | 53 | 0 | 161 | 1 |
| | Pos (≥1%) | 2 | 67 | 0 | 76 | 1 | 77 | 3 | 220 |
| | Total | 53 | 67 | 57 | 77 | 54 | 77 | 164 | 221 |
| % Overall agreement (95% CI) | | 98% (94% - 100%) | | 99% (96% - 100%) | | 99% (96% - 100%) | | 99% (97% - 100%) | |
| Negative Agreement (95% CI) | | 96% (87% - 100%) | | 100% (94% - 100%) | | 98% (90% - 100%) | | 98% (95% - 100%) | |
| Positive Agreement (95% CI) | | 100% (95% - 100%) | | 99% (93% - 100%) | | 100% (95% - 100%) | | 100% (98%-100%) | |

The lower limit of 95% confidence intervals for pooled overall centers results is 97%, 95% and 98% for overall, negative and positive agreements respectively. They are above the pre-specified acceptance criteria of 90%.

The GenASIs HiPath IHC Family for Ki67 (30-9)

Table 5 presents Ki67 2x2 accuracy statistics for each site and pooled over all three sites, along with the corresponding PPA, NPA, OA and their 95% two- sided Exact Binomial confidence intervals. A 10% threshold is used for the definition of negative and positive classes.

**Table 5 Ki67 2x2 accuracy statistics for each site and pooled over all three sites**

| Ki67 Frequency Distribution of Agreement | | Manual Analysis | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 110) | | Site 2 (n = 137) | | Site 3 (n = 126) | | Overall (n = 373) | |
| | | Neg | Pos | Neg | Pos | Neg | Pos | | |
| **Analyze with HiPath System** | Neg (≤10%) | 44 | 2 | 38 | 3 | 50 | 1 | 132 | 6 |
| | Pos (>10%) | 4 | 60 | 2 | 94 | 4 | 71 | 10 | 225 |
| | Total | 48 | 62 | 40 | 97 | 54 | 72 | 142 | 231 |
| % Overall agreement (95% CI) | | 95% (89% - 98%) | | 96% (92% - 99%) | | 96% (91% - 99%) | | 96% (93% - 98%) | |
| Negative Agreement (95% CI) | | 92% (80% - 98%) | | 95% (83% - 99%) | | 93% (82% - 98%) | | 93% (87% - 97%) | |
| Positive Agreement (95% CI) | | 97% (89% - 100%) | | 97% (91% - 99%) | | 99% (93% - 100%) | | 97% (94%-99%) | |

The lower limit of 95% confidence intervals for pooled results overall centers is 93%, 87% and 94% for overall, negative and positive agreements respectively.

The GenASIs HiPath IHC Family for HER2 (4B5)

The following tables (6-8) present frequency distribution of agreement between manual count and the system output where the outcome is summarized in three ways—two-level outcome (positive / negative), three-level outcome (positive / equivocal / negative) and four-level outcome (positive / equivocal / negative class 1+ / negative class 0). The distribution is given by center and overall. The corresponding PPA, NPA and OA, and their 95% two- sided Exact Binomial confidence intervals are included.

**Table 6 HER2/neu Two-level Outcome Frequency Distribution of Agreement**

| HER2/neu – 2x2 Frequency Distribution of Agreement | | Manual Analysis | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 119) | | Site 2 (n = 104) | | Site 3 (n = 134) | | Overall (n = 357) | |
| | | 0&1+ | 2+&3+ | 0&1+ | 2+&3+ | 0&1+ | 2+&3+ | | |
| **Analysis with HiPath System** | 0&1+ | 55 | 3 | 49 | 1 | 81 | 1 | 185 | 5 |
| | 2+&3+ | 1 | 60 | 0 | 54 | 3 | 49 | 4 | 163 |
| | Total | 56 | 63 | 49 | 55 | 84 | 50 | 189 | 168 |
| % Overall agreement (95% CI) | | 97%(N=119) (92% - 99%) | | 99%(N=104) (95% - 100%) | | 97%(N=134) (93% - 99%) | | 97%(N=357) (95% - 99%) | |
| Negative Agreement (95% CI) | | 98%(N=56) (90% - 100%) | | 100%(N=49) (93% - 100%) | | 96%(N=84) (90% - 99%) | | 98%(N=189) (95% - 99%) | |
| Positive Agreement (95% CI) | | 95% (N=63) (87% - 99%) | | 98%(N=55) (90% - 100%) | | 98%(N=50) (89% - 100%) | | 97%(N=168) (93%-99%) | |

**Table 7 HER2/neu Three-level Outcome Frequency Distribution of Agreement**

| HER2/neu – 3x3 Frequency Distribution of Agreement | | Manual Analysis | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 119) | | | Site 2 (n = 104) | | | Site 3 (n = 134) | | | Overall (n = 357) | | |
| | | 0&1+ | 2+ | 3+ | 0&1+ | 2+ | 3+ | 0&1+ | 2+ | 3+ | 0&1+ | 2+ | 3+ |
| **Analysis with HiPath System** | Neg(0&1+) | 55 | 3 | 0 | 49 | 1 | 0 | 81 | 1 | 0 | 185 | 5 | 0 |
| | Equi (2+) | 1 | 31 | 1 | 0 | 25 | 1 | 3 | 23 | 1 | 4 | 79 | 3 |
| | Pos (3+) | 0 | 4 | 24 | 0 | 0 | 28 | 0 | 0 | 25 | 0 | 4 | 77 |
| | Total | 56 | 38 | 25 | 49 | 26 | 29 | 84 | 24 | 26 | 189 | 88 | 80 |
| % Overall agreement (95% CI) | | 92% (86% - 96%) | | | 98% (93% - 100%) | | | 96% (92% - 99%) | | | 96% (93% - 97%) | | |
| Negative Agreement (95% CI) | | 98% (90% - 100%) | | | 100% (93% - 100%) | | | 96% (90% - 99%) | | | 98% (95% - 99%) | | |
| Equivocal Agreement (95% CI) | | 82% (66% - 92%) | | | 96% (80% - 100%) | | | 96% (79% - 100%) | | | 90% (81% - 95%) | | |
| Positive Agreement (95% CI) | | 96% (80% - 100%) | | | 97% (82% - 100%) | | | 96% (80% - 100%) | | | 96% (89%-99%) | | |

**Table 8 HER2/neu Four-level Outcome Frequency Distribution of Agreement**

| HER2/neu - 4x4 Frequency Distribution of Agreement | | Manual Analysis | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Site 1 (n = 119) | | | | Site 2 (n = 104) | | | | Site 3 (n = 134) | | | | Overall (n = 357) | | | |
| | | 0 | 1+ | 2+ | 3+ | 0 | 1+ | 2+ | 3+ | 0 | 1+ | 2+ | 3+ | 0 | 1+ | 2+ | 3+ |
| **Analysis with HiPath System** | 0 | 19 | 0 | 0 | 0 | 13 | 9 | 0 | 0 | 24 | 1 | 0 | 0 | 56 | 10 | 0 | 0 |
| | 1+ | 6 | 30 | 3 | 0 | 1 | 26 | 1 | 0 | 1 | 55 | 1 | 0 | 8 | 111 | 5 | 0 |
| | 2+ | 0 | 1 | 31 | 1 | 0 | 0 | 25 | 1 | 0 | 3 | 23 | 1 | 0 | 4 | 79 | 3 |
| | 3+ | 0 | 0 | 4 | 24 | 0 | 0 | 0 | 28 | 0 | 0 | 0 | 25 | 0 | 0 | 4 | 77 |
| | Total | 25 | 31 | 38 | 25 | 14 | 35 | 26 | 29 | 25 | 59 | 24 | 26 | 64 | 125 | 88 | 80 |
| % Overall agreement (95% CI) | | 87% (80% - 93%) | | | | 88% (81% - 94%) | | | | 95% (90% - 98%) | | | | 90% (87% - 93%) | | | |
| Class 0 Agreement (95% CI) | | 76% (55% - 91%) | | | | 93% (66% - 100%) | | | | 96% (80% - 100%) | | | | 88% (77% - 94%) | | | |
| Class 1+ Agreement (95% CI) | | 97% (83% - 100%) | | | | 74% (57% - 88%) | | | | 93% (84% - 98%) | | | | 89% (82% - 94%) | | | |
| Class 2+ (Equivocal) Agreement (95% CI) | | 82% (66% - 92%) | | | | 96% (80% - 100%) | | | | 96% (79% - 100%) | | | | 90% (81% - 95%) | | | |
| Class 3+ (Positive) Agreement (95% CI) | | 96% (80% - 100%) | | | | 97% (82% - 100%) | | | | 96% (80% - 100%) | | | | 96% (89%-99%) | | | |

*b. Precision/Reproducibility*:

For the repeatability and reproducibility tests a total of 40 slides were used for each application. The slides were chosen in a way that they will span the entire clinically meaningful range. Per each sample, attention was given to the relevant categories, so that there will be enough samples per each category.

The following quantities of slides per category were used (Table 9):

**Table 9 Quantities of slides per category**

| Sample type | Negative (<1%) | Around cutoff (1% - 20%) | Positive (>20%) |
|---|---|---|---|
| **ER** | 15 | 8 | 17 |

| Sample type | Negative(<1%) | Around cutoff (1% - 10%) | Positive (>10%) |
|---|---|---|---|
| **PR** | 15 | 10 | 15 |

| Sample type | Negative (<10%) | Positive(≥10%) |
|---|---|---|
| **Ki67** | 23 | 17 |

| Sample type | Negative 0 | Negative 1+ | Equivocal 2+ | Positive 3+ |
|---|---|---|---|---|
| **HER2/neu** | 10 | 10 | 10 | 10 |

For each application the precision was tested for manual outcomes and for system outcomes separately.

<u>The GenASIs HiPath IHC Family for ER (SP1)</u>

Each ER slide was evaluated three times on a same system/day. The following tables describe the agreement between these three runs:

**Table 10 Test: ER with 2 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| Run 3 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| Run 3 | Run 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

Each ER slide is re-evaluated three times on non-consecutive days (at least 5 days apart) on a same system. The following tables describe the agreement between these different days:

**Table 11 Test: ER with 2 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| Day 3 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| Day 3 | Day 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

Each ER slide is re-evaluated three times using different systems (at least 7 days apart). The following tables describe the agreement between these different systems:

**Table 12 Test: ER with 2 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| System 3 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

| System 3 | System 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 0 | 0 | 15 | 37.5 |
| Positive | 0 | 0 | 25 | 62.5 | 25 | 62.5 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

**Table 13 Between-Run, Between-Day and Between-Systems Precision (ER)**

**Between-Run Precision (Within-Day, Within-System):**

| Runs | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 100.0% (100.0%, 100.0%)* | 100.0% |
| 1 vs 3 | 100.0% | 100.0% |
| 2 vs 3 | 100.0% | 100.0% |

**Between-Day Precision (Within-System):**

| Days | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 100.0% (100.0%, 100.0%)* | 100.0% |
| 1 vs 3 | 100.0% | 100.0% |
| 2 vs 3 | 100.0% | 100.0% |

**Between-Systems Precision:**

| Systems | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 100.0% (100.0%, 100.0%)* | 100.0% |
| 1 vs 3 | 100.0% | 100.0% |
| 2 vs 3 | 100.0% | 100.0% |

The GenASIs HiPath IHC Family for PR (1E2)

Each PR slide was evaluated three times on a same system/day. The following tables describe the agreement between these three runs:

**Table 14 Test: PR with 2 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 17 | 42.5 | 0 | 0 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| Run 3 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 17 | 42.5 | 0 | 0 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| Run 3 | Run 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 17 | 42.5 | 0 | 0 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

Each PR slide is re-evaluated three times on non-consecutive days (at least 5 days apart) on the same system. The following tables describe the agreement between these different days:

**Table 15 Test: PR with 2 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 0 | 0 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| Day 3 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 0 | 0 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| Day 3 | Day 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 1 | 2.5 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 15 | 37.5 | 25 | 62.5 | 40 | 100 |

Each PR slide is re-evaluated three times using different systems (at least 7 days apart). The following tables describe the agreement between these different systems:

**Table 16 Test: PR with 2 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 0 | 0 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| System 3 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 0 | 0 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

| System 3 | System 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg<1% Pos≥1% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 0 | 0 | 16 | 40.0 |
| Positive | 0 | 0 | 24 | 60 | 24 | 60.0 |
| Total | 16 | 40.0 | 24 | 60 | 40 | 100 |

**Table 17 Between-Run, Between-Day and Between-Systems Precision (PR)**

**Between-Run Precision (Within-Day, Within-System):**

| Runs | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 100.0% | 100.0% |
| 1 vs 3 | 100.0% | 100.0% |
| 2 vs 3 | 100.0% | 100.0% |

**Between-Day Precision (Within-System):**

| Days | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 97.9% (93.2%, 100.0%) | 97.1% (90.6%, 100.0%) |
| 1 vs 3 | 97.9% (92.5%, 100.0%) | 97.1% (90.0%, 100.0%) |
| 2 vs 3 | 95.8% (89.8%, 100.0%) | 93.8% (83.9%, 100.0%) |

**Between-Systems Precision:**

| Systems | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 97.9% (92.9%, 100.0%) | 97.1% (90.9%, 100.0%) |
| 1 vs 3 | 97.9% (93.2%, 100.0%) | 97.1% (91.2%, 100.0%) |
| 2 vs 3 | 100.0% | 100.0% |

The GenASIs HiPath IHC Family for Ki67 (30-9)

Each Ki67 slide was evaluated three times on a same system and on a same day. The following tables describe the agreement between these three runs:

**Table 18 Test: Ki67 with 2 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 16 | 40.0 | 1 | 2.5 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

| Run 3 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 14 | 35.0 | 3 | 7.5 | 17 | 42.5 |
| Positive | 2 | 5.0 | 21 | 52.5 | 23 | 57.5 |
| Total | 16 | 42.5 | 24 | 60 | 40 | 100 |

| Run 3 | Run 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 2 | 5.0 | 17 | 42.5 |
| Positive | 2 | 5.0 | 21 | 52.5 | 23 | 57.5 |
| Total | 17 | 42.5 | 23 | 57.5 | 40 | 100 |

Each Ki67 slide is re-evaluated three times on non-consecutive days (at least 5 days apart) on a same system. The following tables describe the agreement between these different days:

**Table 19 Test: Ki67 with 2 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 1 | 2.5 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

| Day 3 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 1 | 2.5 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

| Day 3 | Day 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 1 | 2.5 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

Each Ki67 slide is re-evaluated three times using different systems (at least 7 days apart). The following tables describe the agreement between these different systems:

**Table 20 Test: Ki67 with 2 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 15 | 37.5 | 1 | 2.5 | 16 | 40.0 |
| Positive | 1 | 2.5 | 23 | 57.5 | 24 | 60.0 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

| System 3 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 16 | 40 | 1 | 2.5 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

| System 3 | System 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg≤10% Pos>10% | N | % | N | % | N | % |
| Negative | 16 | 40 | 1 | 2.5 | 17 | 42.5 |
| Positive | 0 | 0 | 23 | 57.5 | 23 | 57.5 |
| Total | 16 | 40 | 24 | 60 | 40 | 100 |

**Table 21 Between-Run, Between-Day and Between-Systems Precision (Ki67)**

**Between-Run Precision (Within-Day, Within-System):**

| Runs | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 97.9% (93.8%, 100.0%) | 97.1% (90.0%, 100.0%) |
| 1 vs 3 | 89.4% (79.7%, 97.4%) | 84.9% (70.8%, 96.2%) |
| 2 vs 3 | 91.3% (81.8%, 98.1%) | 88.2% (75.0%, 97.5%) |

**Between-Day Precision (Within-System):**

| Days | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 95.8% (88.4%, 100.0%) | 93.8% (82.6%, 100.0%) |
| 1 vs 3 | 95.8% (90.2%, 100.0%) | 93.8% (84.9%, 100.0%) |
| 2 vs 3 | 95.8% (89.3%, 100.0%) | 93.8% (84.0%, 100.0%) |

**Between-Systems Precision:**

| Systems | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 95.8% (88.4%, 100.0%) | 93.8% (81.8%, 100.0%) |
| 1 vs 3 | 97.9% (93.5%, 100.0%) | 97.1% (91.2%, 100.0%) |
| 2 vs 3 | 97.9% (92.9%, 100.0%) | 97.1% (90.5%, 100.0%) |

## The GenASIs HiPath IHC Family for HER2 (4B5)

Each HER2/neu slide was evaluated three times on a same system and on a same day. The following tables describe the two-level, three-level and four-level agreements between these three runs.

**Table 22 Test: HER2/neu with 2 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| Neg 0,1+ | Negative | | Positive | | All | |
| Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 0 | 0 | 19 | 47.5 |
| Positive | 1 | 2.5 | 20 | 50.0 | 21 | 52.5 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| Run 3 | Run 1 | | | | | |
|---|---|---|---|---|---|---|
| Neg 0,1+ | Negative | | Positive | | All | |
| Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 1 | 2.5 | 20 | 50 |
| Positive | 1 | 2.5 | 19 | 47.5 | 20 | 50 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| Run 3 | Run 2 | | | | | |
|---|---|---|---|---|---|---|
| Neg 0,1+ | Negative | | Positive | | All | |
| Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 1 | 2.5 | 20 | 50 |
| Positive | 0 | 0 | 20 | 50.0 | 20 | 50 |
| Total | 21 | 47.5 | 21 | 52.5 | 40 | 100 |

Each HER2/neu slide is re-evaluated three times on non-consecutive days (at least 5 days apart) on a same system. The following tables describe the agreement between these different days:

**Table 23 Test: HER2/neu with 2 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 20 | 50.0 | 0 | 0 | 20 | 50.0 |
| Positive | 0 | 0 | 20 | 50.0 | 20 | 50.0 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| Day 3 | Day 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 0 | 0 | 19 | 47.5 |
| Positive | 1 | 2.5 | 20 | 50.0 | 21 | 52.5 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| Day 3 | Day 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 0 | 0 | 19 | 47.5 |
| Positive | 1 | 2.5 | 20 | 50.0 | 21 | 52.5 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

Each HER2/neu slide is re-evaluated three times using different systems (at least 7 days apart). The following tables describe the agreement between these different systems:

**Table 24 Test: HER2/neu with 2 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 0 | 0 | 19 | 47.5 |
| Positive | 1 | 2.5 | 20 | 50.0 | 21 | 52.5 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| System 3 | System 1 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 20 | 50 | 1 | 2.5 | 21 | 52.5 |
| Positive | 0 | 0 | 19 | 47.5 | 19 | 47.5 |
| Total | 20 | 50 | 20 | 50 | 40 | 100 |

| System 3 | System 2 | | | | | |
|---|---|---|---|---|---|---|
| | Negative | | Positive | | All | |
| Neg 0,1+ Pos 2+ 3+ | N | % | N | % | N | % |
| Negative | 19 | 47.5 | 2 | 5.0 | 21 | 52.5 |
| Positive | 0 | 0 | 19 | 47.5 | 19 | 47.5 |
| Total | 19 | 47.5 | 21 | 52.5 | 40 | 100 |

**Table 25 Between-Run, Between-Day and Between-Systems Precision (HER2/neu)**

**Between-Run Precision for two-level (Within-Day, Within-System):**

| Runs | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 97.6% (91.2%, 100.0%) | 97.5% (90.6%, 100.0%) |
| 1 vs 3 | 95.0% (87.9%, 100.0%) | 95.0% (87.2%, 100.0%) |
| 2 vs 3 | 97.6% (92.5%, 100.0%) | 97.5% (91.7%, 100.0%) |

**Between-Day Precision for two-level (Within-System):**

| Days | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 100% (100%, 100%) | 100% (100%, 100%) |
| 1 vs 3 | 97.6% (92.3%, 100.0%) | 97.5% (92.1%, 100%) |
| 2 vs 3 | 97.6% (92.1%, 100.0%) | 97.5% (91.7%, 100%) |

**Between-Systems Precision for two-level:**

| Systems | Average Positive Agreement (95% two-sided CI) | Average Negative Agreement (95% two-sided CI) |
|---|---|---|
| 1 vs 2 | 97.6% (92.1%, 100.0%) | 97.5% (91.7%, 100%) |
| 1 vs 3 | 97.5% (91.3%, 100.0%) | 97.6% (91.3%, 100%) |
| 2 vs 3 | 95.2% (88.5%, 100.0%) | 95.2% (88.6%, 100%) |

**Table 26 Test: HER2/neu with 3 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 0 | 0 | 0 | 0 | 19 | 47.5 |
| Equivocal ( 2) | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Run 3 | Run 1 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 1 | 2.5 | 0 | 0 | 20 | 50.0 |
| Equivocal ( 2) | 1 | 2.5 | 9 | 22.5 | 0 | 0 | 10 | 25.0 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Run 3 | Run 2 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 1 | 2.5 | 0 | 0 | 20 | 50.0 |
| Equivocal ( 2) | 0 | 0 | 10 | 25.0 | 0 | 0 | 10 | 25.0 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 19 | 47.5 | 11 | 27.5 | 10 | 25.0 | 40 | 100.0 |

**Table 27 Test: HER2/neu with 3 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | | All | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 20 | 50.0 | 0 | 0 | 0 | 0 | 20 | 50.0 |
| Equivocal ( 2) | 0 | 0 | 10 | 25.0 | 0 | 0 | 10 | 25.0 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Day 3 | Day 1 | | | | | | All | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 0 | 0 | 0 | 0 | 19 | 47.5 |
| Equivocal ( 2) | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Day 3 | Day 2 | | | | | | All | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 0 | 0 | 0 | 0 | 19 | 47.5 |
| Equivocal ( 2) | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 19 | 47.5 | 11 | 27.5 | 10 | 25.0 | 40 | 100.0 |

**Table 28 Test: HER2/neu with 3 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 0 | 0 | 0 | 0 | 19 | 47.5 |
| Equivocal ( 2) | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| System 3 | System 1 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 20 | 50.0 | 1 | 2.5 | 0 | 0 | 21 | 52.5 |
| Equivocal ( 2) | 0 | 0 | 9 | 22.5 | 0 | 0 | 9 | 22.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 20 | 50.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| System 3 | System 2 | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Negative (0 or 1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % |
| Negative ( 0 or 1+) | 19 | 47.5 | 2 | 5.0 | 0 | 0 | 21 | 52.5 |
| Equivocal ( 2) | 0 | 0 | 9 | 22.5 | 0 | 0 | 9 | 22.5 |
| Positive ( 3) | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 19 | 47.5 | 11 | 27.5 | 10 | 25.0 | 40 | 100.0 |

**Table 29 Between-runs, between-days and between-systems precision for three-level**

**Between-Run Precision for Three-level (Within-Day, Within-System):**

| Runs | Average positive Agreement (95% two sided CI) | Average Negative Agreement (95% two-sided CI) | Average Equivocal Agreement (95% two-sided CI) |
|---|---|---|---|
| 1 vs 2 | 100% (100%, 100%) | 97.5% (91.7%, 100.0%) | 95.5% (84.6%, 100.0%) |
| 1 vs 3 | 100% (100%, 100%) | 95.0% (86.3%, 100.0%) | 90.0% (68.6%, 100.0%) |
| 2 vs 3 | 100% (100%, 100%) | 97.5% (91.7%, 100.0%) | 95.5% (83.3%, 100.0%) |

**Between-Day Precision for Three-level (Within-System):**

| Days | Average positive Agreement (95% two sided CI) | Average Negative Agreement (95% two-sided CI) | Average Equivocal Agreement (95% two-sided CI) |
|---|---|---|---|
| 1 vs 2 | 100% (100%, 100%) | 100% (100%, 100.0%) | 100% (100%, 100.0%) |
| 1 vs 3 | 100% (100%, 100%) | 97.5% (92.1%, 100.0%) | 95.5% (83.3%, 100.0%) |
| 2 vs 3 | 100% (100%, 100%) | 97.5% (91.7%, 100.0%) | 95.5% (83.3%, 100.0%) |

**Between-Systems Precision for Three-level:**

| Systems | Average positive Agreement (95% two sided CI) | Average Negative Agreement (95% two-sided CI) | Average Equivocal Agreement (95% two-sided CI) |
|---|---|---|---|
| 1 vs 2 | 100% (100%, 100%) | 97.5% (92.1%, 100.0%) | 95.5% (85.7%, 100.0%) |
| 1 vs 3 | 100% (100%, 100%) | 97.6% (91.7%, 100.0%) | 95.0% (81.3%, 100.0%) |
| 2 vs 3 | 100% (100%, 100%) | 95.2% (88.6%, 100.0%) | 90.9% (78.6%, 100.0%) |

**Table 30 Test: HER2/neu with 4 Levels Reference, Variance Source: Between-Runs (Within-Day, Within-System)**

| Run 2 | Run 1 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 2 | 5.0 | 0 | 0 | 0 | 0 | 12 | 30.0 |
| Negative ( 1+) | 0 | 0 | 7 | 17.5 | 0 | 0 | 0 | 0 | 7 | 17.5 |
| Equivocal ( 2+) | 0 | 0 | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Run 3 | Run 1 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Negative ( 1+) | 0 | 0 | 9 | 22.5 | 1 | 2.5 | 0 | 0 | 10 | 25.0 |
| Equivocal ( 2+) | 0 | 0 | 1 | 2.5 | 9 | 22.5 | 0 | 0 | 10 | 25.0 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Run 3 | Run 2 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Negative ( 1+) | 2 | 5.0 | 7 | 17.5 | 1 | 2.5 | 0 | 0 | 10 | 25.0 |
| Equivocal ( 2+) | 0 | 0 | 0 | 0 | 10 | 25.0 | 0 | 0 | 10 | 25.0 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 12 | 30 | 7 | 17.5 | 11 | 27.5 | 10 | 25.0 | 40 | 100.0 |

**Table 31 Test: HER2/neu with 4 Levels Reference, Variance Source: Between-Days (Within-System)**

| Day 2 | Day 1 | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Negative ( 1+) | 0 | 0 | 10 | 25.0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Equivocal ( 2+) | 0 | 0 | 0 | 0 | 10 | 25.0 | 0 | 0 | 10 | 25.0 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Day 3 | Day 1 | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 2 | 5.0 | 0 | 0 | 0 | 0 | 12 | 30.0 |
| Negative ( 1+) | 0 | 0 | 7 | 17.5 | 0 | 0 | 0 | 0 | 7 | 17.5 |
| Equivocal ( 2+) | 0 | 0 | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| Day 3 | Day 2 | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | All | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 2 | 5.0 | 0 | 0 | 0 | 0 | 12 | 30.0 |
| Negative ( 1+) | 0 | 0 | 7 | 17.5 | 0 | 0 | 0 | 0 | 7 | 17.5 |
| Equivocal ( 2+) | 0 | 0 | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

**Table 32 Test: HER2/neu with 4 Levels Reference, Variance Source: Between-Systems**

| System 2 | System 1 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 1 | 2.5 | 0 | 0 | 0 | 0 | 11 | 27.5 |
| Negative ( 1+) | 0 | 0 | 8 | 20.0 | 0 | 0 | 0 | 0 | 8 | 20.0 |
| Equivocal ( 2+) | 0 | 0 | 1 | 2.5 | 10 | 25.0 | 0 | 0 | 11 | 27.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| System 3 | System 1 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Negative ( 1+) | 0 | 0 | 10 | 25.0 | 1 | 2.5 | 0 | 0 | 11 | 27.5 |
| Equivocal ( 2+) | 0 | 0 | 0 | 0 | 9 | 22.5 | 0 | 0 | 9 | 22.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 10 | 25.0 | 40 | 100.0 |

| System 3 | System 2 | | | | | | | | All | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Negative (0) | | Negative (1+) | | Equivocal (2+) | | Positive (3+) | | | |
| | N | % | N | % | N | % | N | % | N | % |
| Negative ( 0) | 10 | 25.0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 |
| Negative ( 1+) | 1 | 2.5 | 8 | 20.0 | 2 | 5.0 | 0 | 0 | 11 | 27.5 |
| Equivocal ( 2+) | 0 | 0 | 0 | 0 | 9 | 22.5 | 0 | 0 | 9 | 22.5 |
| Positive ( 3+) | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 25.0 | 10 | 25.0 |
| Total | 11 | 27.5 | 8 | 20.0 | 11 | 27.5 | 10 | 25.0 | 40 | 100.0 |

**Table 33 Between-runs, between-days and between-systems precision for four-level**

**Between-Runs Precision for Four-level (Within-Day, Within-System):**

| Runs | Average Class 0 Agreement (95% two-sided CI) | Average Class 1 Agreement (95% two-sided CI) | Average Class 2 Agreement (95% two-sided CI) | Average Class 3 Agreement (95% two-sided CI) |
|---|---|---|---|---|
| 1 vs 2 | 91.7 (80.0%, 100.0%) | 85.0% (68.8%, 100.0%) | 95.5% (83.3%, 100.0%) | 100% (100%, 100%) |
| 1 vs 3 | 100% (100%, 100.0%) | 90.0% (70.8%, 100.0%) | 90% (71.4%, 100.0%) | 100% (100%, 100%) |
| 2 vs 3 | 91.7% (79.2%, 100.0%) | 85.0% (70.0%, 100%) | 95.5% (83.3%, 100.0%) | 100% (100%, 100%) |

**Between-Days Precision for Four-level (Within-System):**

| Days | Average Class 0 Agreement (95% two-sided CI) | Average Class 1 Agreement (95% two-sided CI) | Average Class 2 Agreement (95% two-sided CI) | Average Class 3 Agreement (95% two-sided CI) |
|---|---|---|---|---|
| 1 vs 2 | 100% (100%, 100.0%) | 100% (100%, 100%) | 100% (100%, 100%) | 100% (100%, 100%) |
| 1 vs 3 | 91.7% (80.8%, 100.0%) | 85.0% (68.8%, 100.0%) | 95.5% (85.7%, 100.0%) | 100% (100%, 100%) |
| 2 vs 3 | 91.7% (80.0%, 100.0%) | 85.0% (68.2%, 100%) | 95.5% (85.7%, 100.0%) | 100% (100%, 100%) |

**Between-Systems Precision for Four-level:**

| Systems | Average Class 0 Agreement (95% two-sided CI) | Average Class 1 Agreement (95% two-sided CI) | Average Class 2 Agreement (95% two-sided CI) | Average Class 3 Agreement (95% two-sided CI) |
|---|---|---|---|---|
| 1 vs 2 | 95.5% (85.0%, 100.0%) | 90% (77.3%, 100.0%) | 95.5% (85%, 100.0%) | 100% (100%, 100%) |
| 1 vs 3 | 100% (100%, 100%) | 95.5% (85.7%, 100.0%) | 95% (84.6%, 100.0%) | 100% (100%, 100%) |
| 2 vs 3 | 95.5% (85.7%, 100.0%) | 86.4% (71.4%, 100%) | 90.9% (78.6%, 100.0%) | 100% (100%, 100%) |

c. *Linearity*:

Not applicable.

d. *Carryover*:

Not applicable.

e. *Interfering Substances*:

Not applicable.

2. Other Supportive Instrument Performance Data Not Covered Above:

None

**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.