510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT COMBINATION TEMPLATE

A. 510(k) Number:

K173725

B. Purpose for Submission:

To obtain a substantial equivalence determination for the NeuMoDx GBS Assay

C. Measurand:

pcsB gene sequence of *Streptococcus agalactiae* chromosome (Group B *Streptococcus*)

D. Type of Test:

Qualitative real-time polymerase chain reaction (PCR) test

E. Applicant:

NeuMoDx Molecular, Inc.

F. Proprietary and Established Names:

NeuMoDx GBS Assay

G. Regulatory Information:

1. Regulation section:

   21 CFR 866.3740: *Streptococcus* spp. serological reagents

2. Classification:

   Class I (non-exempt)

3. Product code:

   NJR: Nucleic Acid Amplification Assay System, Group B Streptococcus, Direct Specimen Test

   OOI: Real-Time Nucleic Acid Amplification System

4. Panel:

   83-Microbiology

H. Intended Use:

1. Intended use(s):

   The NeuMoDx GBS Assay as implemented on the NeuMoDx 288 Molecular System is a qualitative in vitro diagnostic test designed to detect Group B Streptococcus (GBS) DNA from 18-24 hour Lim broth enrichments of vaginal/rectal swabs from pregnant women. The test incorporates automated DNA extraction to isolate the target nucleic acid from the specimen and real-time polymerase chain reaction (PCR) to detect an 88 bp region of the *pcsB* gene sequence in the *Streptococcus agalactiae* chromosome. Results from the NeuMoDx GBS Assay can be used as an aid in determining colonization status in antepartum women.

   The NeuMoDx GBS Assay does not provide susceptibility results. Cultured isolates are needed for performing susceptibility testing as recommended for penicillin-allergic women.

2. Indication(s) for use:

   Same as Intended Use.

3. Special conditions for use statement(s):

   For *in vitro* diagnostic use.

   For prescription use only.

4. Special instrument requirements:

   The NeuMoDx GBS Assay is for use only with the NeuMoDx 288 Molecular System.

I. Device Description:

The NeuMoDx GBS Assay is an automated *in vitro* diagnostic, real-time PCR test for the qualitative detection of Group B *Streptococcus* DNA from Lim broth enrichments of vaginal/rectal swab specimens obtained from pregnant women. The NeuMoDx System automatically extracts the target nucleic acid and amplifies a section of the *pcsB* gene sequence of the GBS chromosome, if present. The NeuMoDx GBS Assay includes a DNA Sample Process Control (SPC1) to monitor for the presence of potential inhibitory substances, as well as system or reagent failures that may be encountered during the extraction and amplification processes. The GBS assay test strip, in combination with required NeuMoDx buffers, extraction plates, wash and release solutions, cartridge, and the

fully automated NeuMoDx 288 Molecular System, utilizes real-time PCR for the amplification of GBS DNA where fluorogenic target-specific TaqMan probes allow for the detection of the amplified GBS DNA.

The test system uses a combination of heat, lytic enzyme and extraction reagents to perform cell lysis, DNA extraction and removal of inhibitors. No operator intervention is necessary once the specimen is loaded onto the NeuMoDx System. After the test is processed, a determination of the presence/absence of GBS DNA in the specimen is automatically made based on the amplification status of GBS and SPC1 using pre-established cut-off criteria. A list of reagents and consumables, including other equipment/materials required, is shown in the package insert.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   BD MAX GBS Assay

2. Predicate 510(k) number(s):

   K090191

3. Comparison with predicate:

Similarities		
Item	**Device: NeuMoDx GBS Assay (K173725)**	**Predicate: BD MAX GBS Assay (K090191)**
Intended Use	The NeuMoDx GBS Assay as implemented on the NeuMoDx 288 Molecular System is a qualitative in vitro diagnostic test designed to detect Group B Streptococcus (GBS) DNA from 18-24 hour Lim broth enrichments of vaginal/rectal swabs from pregnant women. The test incorporates automated DNA extraction to isolate the target nucleic acid from the specimen and real-time polymerase chain reaction (PCR) to detect an 88 bp region of the *pcsB* gene sequence in the *Streptococcus agalactiae* chromosome. Results from the NeuMoDx GBS Assay can be used as an aid in determining colonization status in antepartum women. The NeuMoDx GBS Assay does not provide susceptibility results. Cultured isolates are needed for performing susceptibility testing as recommended for penicillin-allergic women.	The BD MAX GBS Assay as implemented on the BD MAX System is a qualitative in vitro diagnostic test designed to detect Group B Streptococcus (GBS) DNA in Lim Broth cultures after incubation for greater than or equal to (>)18 hours, obtained from vaginal-rectal swab specimens from antepartum pregnant women. The test incorporates automated DNA extraction to isolate the target nucleic acid from the specimen and realtime polymerase chain reaction (PCR) to detect a 124 bp region of the *cfb* gene sequence of the *Streptococcus agalactiae* chromosome. Results from the BD MAX GBS Assay can be used as an aid in determining colonization status in antepartum women. The BD MAX GBS Assay does not provide susceptibility results. Cultured isolates are necessary for performing susceptibility testing as recommended for penicillin-allergic women. Subculture to solid media for additional testing when indicated.

Similarities		
Item	**Device: NeuMoDx GBS Assay (K173725)**	**Predicate: BD MAX GBS Assay (K090191)**
Analyte	Group B *Streptococcus* DNA	Group B *Streptococcus* DNA
Specimen Type	Vaginal/Rectal Swab (Enriched in Lim broth 18-24 hrs)	Vaginal/Rectal Swab (Enriched in Lim broth $\geq$ 18 hrs)
Sample Preparation	nucleic acid extraction is automated on the NeuMoDx 288 Molecular System	nucleic acid extraction is automated on the BD MAX System
Assay Format	Amplification: Real-Time PCR Detection	Same
Single-Use Test	Yes	Same
Results Interpretation	Automated	Same
Internal Process Control	Present (process monitor)	Same

Differences		
Item	**Device: NeuMoDx GBS Assay (K173725)**	**Predicate: BD MAX GBS Assay (K090191)**
Platform	NeuMoDx 288 Molecular System	BD MAX System
DNA Target Sequence	*pcsB* gene in *Streptococcus agalactiae*	*cfb* gene in *Streptococcus agalactiae*
Probes	TaqMan	Scorpion

K. Standard/Guidance Document Referenced (if applicable):

- EP07-A2. CLSI: Interference Testing In Clinical Chemistry; Approved Guideline-Second Edition
- EP12-A2. CLSI: User Protocol For Evaluation Of Qualitative Test Performance.
- EP17-A2. CLSI: Evaluation Of Detection Capability For Clinical Laboratory Measurement Procedures; Approved Guideline - Second Edition
- EP25-A. CLSI: Evaluation Of Stability Of In Vitro Diagnostic Reagents; Approved Guideline
- ISO 14971. Second edition (2007) Medical devices - application of risk management to medical devices
- IEC 62304 Edition 1.1 (2015) Medical device software - software life cycle processes
- ISTA 3A (2008) Packaged-products for parcel delivery system shipment 70 kg (150 lb) or less
- 15223-1 (2016) Medical Devices - Symbols To Be Used With Medical Devices Labels, Labeling, And Information To Be Supplied - Part I: General Requirements
- ISO 16142-1. First Edition (2016). Medical Devices - Recognized Essential Principles Of Safety And Performance Of Medical Devices - Part 1: General Essential Principles And Additional Specific Essential Principles For All Non-IVD Medical Devices And Guidance On Selection Of Standards
- MM13-A. CLSI: Collection, Transport, Preparation, And Storage Of Specimens For Molecular Methods; Approved Guideline
- MM03-3rd Edition. CLSI: Molecular Diagnostic Methods For Infectious Diseases; Approved Guideline

L. Test Principle:

Samples are fully processed on the NeuMoDx System using the NeuMoDx GBS Test Strip reagents. After incubation of inoculated selective broth for 18-24 hours at 37 °C in ambient air or 5% CO_2, an aliquot of the enriched Lim broth culture is mixed with lysis buffer to begin lysis of the sample. The NeuMoDx System automatically extracts the target nucleic acid and amplifies a section of the *pcsB* gene sequence of the GBS chromosome, if present. Briefly, the released nucleic acids are captured by magnetic affinity microspheres. The microspheres, with the bound nucleic acids, are loaded into the NeuMoDx Cartridge where the unbound, non-DNA components are removed with NeuMoDx WASH Solution. The bound DNA is eluted using NeuMoDx RELEASE Solution. The NeuMoDx System then uses the released DNA to rehydrate dried assay reagents containing all the elements necessary for amplification of the GBS-specific target. The dried PCR reagents also contain the components required to amplify a section of the SPC1 sequence to enable simultaneous amplification and detection of both target and control DNA sequences. After reconstitution of the dried PCR reagents, the NeuMoDx System dispenses the prepared PCR-ready mixture into one PCR chamber (per specimen) of the NeuMoDx Cartridge. Amplification and detection of the control and target DNA sequences occur in PCR chamber. The chamber and the cartridge are designed to contain the amplicon following real-time-PCR to reduce contamination risk post-amplification. The NeuMoDx System monitors the fluorescent signal emitted by the TaqMan probes at the end of each amplification cycle and presents the test result. When amplification is complete, the NeuMoDx System analyzes the data and reports a final result (POSITIVE / NEGATIVE / INDETERMINATE / UNRESOLVED). No operator intervention is necessary once the specimen is loaded onto the NeuMoDx System.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   a. Precision/Reproducibility:

   Precision Study

   A Precision Study was conducted to evaluate the within-site variability over 12 days with three different NeuMoDx 288 Molecular instruments, two operators, and two lots of reagents. Two runs per day were conducted on each instrument where panel members were tested in triplicate. One GBS strain was used to prepare panel members, which included a true negative, low negative (>100 fold dilution of 1X LoD), moderate negative (>10 fold dilution of 1X LoD), low positive (1-2X LoD), and moderate positive (3-4X LoD) sample. Panel members were prepared in pooled, negative clinical remnant Lim broth specimens. For each run performed, a positive and negative external control were processed in addition to the panel samples. A total of 72 runs and 1224 tests were performed for the study (including the external controls). The percent agreement with the expected result for low positive, moderate positive, and true negative samples was >95%. As shown in Table 1, the positive rates for the low negative and moderate negative samples were reported as 1.4% and 17.6%, respectively. Table 2 is a summary of the coefficient of variance (%CV) and standard deviation (SD) for Ct values. All values met acceptance criteria for the study.

Table 1. GBS Positivity Rates Across Multiple Instruments

Sample	% Positive for GBS			
	Instrument A	**Instrument B**	**Instrument C**	**Combined**
MP (1600 CFU/ml)	100% (72/72)	100% (72/72)	100% (72/72)	100% (216/216)
LP (600 CFU/ml)	100% (72/72)	95.8% (69/72)	97.2% (70/72)	97.7% (211/216)
MN (40 CFU/ml)	22.2% (16/72)	13.9% (10/72)	16.7% (12/72)	17.6% (38/216)
LN (4 CFU/ml)	2.8% (2/72)	0% (0/72)	1.4% (1/72)	1.4% (3/216)
TN (0 CFU/ml)	0% (0/72)	0% (0/72)	0% (0/72)	0% (0/216)

Table 2. Summary of the SD and % CV for the Ct values for the Within-Site Precision Study

Sample	Instrument	GBS C_t, Avg	GBS C_t, SD	GBS C_t, %CV	SPCl C_t, Avg	SPCl C_t, SD	SPCl C_t, %CV
MP	A	31.91	0.65	2.0%	31.15	0.35	1.1%
	B	31.91	0.56	1.8%	31.27	0.44	1.4%
	C	31.92	0.62	1.9%	31.35	0.40	1.3%
LP	A	34.15	0.68	2.0%	31.41	0.34	1.1%
	B	34.23	0.68	2.0%	31.52	0.44	1.4%
	C	34.40	0.74	2.2%	31.47	0.32	1.0%
MN	A	N/A			31.49	0.29	0.9%
	B				31.55	0.28	0.9%
	C				31.57	0.38	1.2%
LN	A	N/A			31.39	0.33	1.0%
	B				31.54	0.40	1.3%
	C				31.59	0.32	1.0%
TN	A	N/A			31.45	0.28	0.9%
	B				31.53	0.49	1.6%
	C				31.61	0.50	1.6%

Reproducibility Study

The reproducibility of the NeuMoDx GBS Assay was evaluated by testing the assay on three NeuMoDx 288 Molecular instruments with one operator at each of the three sites (one internal, two-external sites) for five days. A blinded reproducibility panel was prepared from one GBS strain in negative clinical Lim broth matrix and tested in replicates of five at different concentrations. A true negative sample was also included as a panel member. Across three sites, a total of 75 replicates were tested per panel member with the NeuMoDx GBS Assay. The overall percent agreement with the expected result (i.e. negative for true negative and positive for moderate positive and low positive samples) was >95% for GBS samples tested at low positive and moderate positive concentrations. The true negatives yielded expected results 100% of the time. A total of (4) Indeterminates were observed for an initial rate of 1.3%. Two samples yielded a valid result upon repeat testing. The remaining two samples yielded an Indeterminate result a second time before a valid result was obtained for the two samples. No Unresolved samples were reported in this study. The results from the site-to-site Reproducibility Study for the NeuMoDx GBS Assay are presented in Table 3 below. Also as part of this study, there were 30 sets of GBS external control samples tested. All controls were valid and produced the expected results.

Table 3. Summary of the Agreement Analysis Across all Sites

Sample	**Agreement (observed result/expected)**			
	Site 1	**Site 2**	**Site 3**	**All Sites (%)**
Moderate Positive (MP)	25/25	25/25	25/25	75/75 (100%)
Low Positive (LP)	24/25	25/25	24/25	73/75 (97.3%)
Low Negative (LN)	25/25	25/25	24/25	74/75 (98.7%)
True Negative (TN)	25/25	25/25	25/25	75/75 (100%)

The Reproducibility/Precision Studies are acceptable.

b. Linearity/assay reportable range:

Not Applicable

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Controls
For the studies conducted, the positive external control was prepared from AcroMetrix's GBS Positive Control added to commercially sourced Lim broth at a concentration determined to yield an average Ct value of between 27 – 29. Lim broth was used for the negative external control.

A total of 65 external positive/negative control tests were processed on the NeuMoDx System during the GBS clinical study. Only one Indeterminate was reported, which when repeated, gave a valid correct result. One positive/negative set was excluded due to the use of expired consumables. All remaining external controls gave the expected results.

Reagent Stability
Stability studies were performed to assess the in-use stability of reagents and the shelf-life of the packaged NeuMoDx GBS test strips and Lysis Buffer 4.

- *In Use Stability*
  Wash solution, release solution, GBS Test Strips, and extraction plates were stored (~236 days) according to real-time stability study protocol and tested at various time points (Day 8, Day 15, Day 22, and Day 32) at different temperatures (-20°C, ambient temperature, or 28°C). Overall testing was performed with two lots of reagents, 3 primary storage conditions, and 4 time points where 624 samples in total were tested in the study. At time 0 (after ~236 day storage), reagents stored under the primary conditions of -20°C, ambient temperature, and 28°C over 6 months exhibited a GBS positivity rate of 100%, 98.7%, and 100%, respectively, for positive samples. After removing from their primary storage conditions and aged under the simulated in-use condition for 32 days, the GBS positive rate for positive samples from all groups was 100%. All negative samples generated the expected results at time 0 and after 32 days. Only three Indeterminates were generated during the course of the study.

  A similar study with conducted with Lysis Buffer 4. The goal was to determine the in-use shelf life of Lysis Buffer 4 which had been aged for 6 months (time points 15, 22, 29 days tested). Overall, these results showed each storage temperature group after 29 days in-use storage had GBS positive detection rates of 100% with no difference in positive detection rate between time 0 and 29 day. One Unresolved result was reported during the study. The in-use stability of Lysis Buffer 4 met the acceptance criteria as reported by NeuMoDx for the test system.

- *Real-Time Shelf Life*
  Wash solution, release solution, GBS Test Strips, and extraction plates were stored (~236 days) according to real-time stability study protocol. Briefly, 27 Positive Samples, and 3 Negative Samples were processed using the aforementioned GBS reagents stored either at -20°C, ambient temperature, or 28°C. The -20°C set served as the t=0 control and was the standard against which the ambient temperature set and 28°C set were compared. Ninety samples per storage condition were tested with 270 samples in total across all 3 storage conditions. The GBS positive samples contained GBS at 1000 CFU/ml in Lim broth, while the negative samples consisted of commercial Lim broth. Three Indeterminate results were reported. The results showed a 100% GBS positive rate for the -20°C and 28°C storage condition. At ambient temperature, a 98.7% positive rate was reported with one false negative. This met the acceptance criteria as reported by NeuMoDx. These reagents were determined to have a minimum 7 month stability for the NeuMoDx 288 Molecular System. In a separate study, Lysis Buffer 4 was determined to have a minimum 6-month stability for GBS DNA extractions (only one Unresolved reported at -20°C as reported by NeuMoDx for the test system).

- *On-Board Specimen Stability*
  A Specimen Stability Study was performed to provide evidence for the stability of the enriched Lim broth specimens once on-board the instrument. A set of 23 GBS positive and 23 negative clinical samples with different collection dates was tested, then left on the worktable for 24 hours before re-testing with the NeuMoDx GBS Assay. All 23 GBS negative samples gave negative results at both t=0 and t=24 time points, resulting in 100% concordance between the two testing time points for GBS negative samples. At t=0, all 23 GBS positive samples gave positive results. However, at t=24 time point, one GBS positive sample returned a negative result (22/23 positive). According to NeuMoDx, the reference laboratory confirmed the one sample had high Ct value ($C_t > 33$) compared to other samples and no growth in culture.

d. Detection limit:

Limit of Detection Determination

A Limit of Detection (LoD) study was performed to evaluate the analytical sensitivity of the NeuMoDx GBS Assay using one representative strain of GBS (ATCC BAA-611, Serotype V). Preliminary LoD concentrations were determined by diluting the strain to five levels in five independent clinical negative pools. For each concentration level, 60 replicates were tested across two instruments. A unique lot of NeuMoDx GBS Test Strips, extraction plates, and Lysis Buffer was tested on each system. The LoD was determined to be 500 CFU/ml (Table 4).

Table 4. Limit of Detection

% GBS Detected at the indicated CFU/ml				
0	**100**	**200**	**500**	**1000**
0%	58% (35/60)	88% (53/60)	100% (60/60)	100% (60/60)

Analytical Reactivity

The analytical reactivity (inclusivity) of the NeuMoDx GBS Assay was evaluated with a panel of 12 GBS strains covering 10 serotypes (Ia, Ib, II, III, Ic, IV, VI, VII, VIII, IX), as well as one non-hemolytic and one clinical strain. Out of the twelve strains, two GBS strains (serotypes Ia and Ib) did not yield a 100% detection rate at 1-2X LoD (800 CFU/ml). Sample GBS strain concentrations for these two strains were increased until all replicates were positive. Serotype Ia and Ib strains achieved 100% positivity at 1500 CFU/ml and 1000 CFU/ml, respectively.

e. Analytical specificity (Cross-Reactivity):

A study was conducted with the NeuMoDx GBS Assay to determine the cross-reactivity of 136 microorganisms (or nucleic acid) representing various non-GBS groups of Streptococci, along with other bacteria, parasites, fungi, and viruses normally common to the urogenital and digestive tract. Bacteria and yeasts were tested at $\geq 10^6$ CFU/ml. Viruses and parasites were tested at the highest available concentration. To simplify testing, pools were prepared with 5-6 organisms in triplicate and spiked into negative GBS clinical matrix before testing with the NeuMoDx GBS Assay. A total of 30 pools were evaluated that included a total of 116 bacteria, 12 viruses, and 7 fungi.

All pools showed negative results, except for Pool 1 and Pool 17. One (out of three replicates) was positive for GBS in each pool. Therefore, organisms from these two pools were tested in triplicate individually in negative GBS clinical matrix—*Streptococcus pyogenes*, *Streptococcus salivarius*, *Streptococcus sanguinis*, *Moraxella catarrhalis*, and *Neisseria gonorrhoeae*. Individually, none of the organisms generated a positive result on the NeuMoDx GBS Assay. See Table 5 for a list of organisms tested during the Cross-Reactivity Study.

Table 5. Cross-Reactivity Organisms Included in Panel Pools

Streptococcus pyogenes	*Salmonella enterica* (serovar Minnesota)	*Cryptococcus neoformans*
Streptococcus salivarius	*Alcaligenes faecalis*	*Candida glabrata*
Streptococcus sanguinis	*Staphylococcus saprophyticus*	*Achromobacter xerosis*
Moraxella (Branhamella) *catarrhalis*	*Eikenella corrodens*	*Rhodospirillum rubrum*
Neisseria gonorrhoeae	*Enterococcus avium*	*Neisseria subflava*
Streptococcus pyogenes	*Micrococcus luteus*	*Pseudomonas putida*
Streptococcus mitis	*Citrobacter freundii*	*Bacillus subtilis*
Lactococcus lactis;sbsp lacis	*Gemella haemolysans*	*Corynebacterium xerosis*
Listeria monocytogenes	*Kingella kingae*	*Mycobacterium smegmatis*

Morganella morganii	*Rahnella aquatilis*	*Legionella pneumophila*
Plesiomonas shigelloides	*Bacillus cereus*	*Moraxella lacunata*
Proteus vulgaris	*Aeromonas hydrophila*	*Streptomyces griseus*
Salmonella enterica (serovar Typhi)	*Enterobacter cloacae*	*Gardnerella vaginalis*
Staphylococcus aureus	*Brevibacterium linens*	*Clostridium perfringens*
Staphylococcus epidermidis	*Candida parapsilosis*	*Peptostreptococcus anaerobius*
Streptococcus mutans	*Lactobacillus brevis*	*Bifidobacterium adolescentis*
Yersinia enterocolitica	*Deinococcus radiodurans*	*Derxia gummosa*
Providencia stuartii	*Pseudomonas protegens*	*Veillonella parvula*
Pseudomonas aeruginosa	*Acinetobacter calcoaceticus*	*Mycoplasma pneumoniae*
Acinetobacter lwoffii	*Lactobacillus acidophilus*	*Bacteroides fragilis*
Proteus mirabilis	*Vibrio parahaemolyticus*	*Acinetobacter baumannii*
Klebsiella pneumoniae	*Corynebacterium genitalium*	*Corynebacterium, strain* HFH0082
Aerococcus viridans	*Enterococcus faecalis*	*Enterobacter aerogenes*
Enterococcus faecium	*Salmonella enterica*	*Klebsiella oxytoca*
Neisseria lactamica	*Lactobacillus jensenii*	*Escherichia coli*
Neisseria meningitidis	*Lactobacillus delbrueckii*	*Streptococcus canis*
Streptococcus pneumoniae	*Serratia marcescens*	*Streptococcus dysgalactiae*
Kingella denitrificans	*Candida albicans*	*Streptococcus oralis*
Haemophilus influenzae	*Candida tropicalis*	*Streptococcus uberis*
Neisseria perflava	*Chromobacterium violaceum*	*Streptococcus suis*
Moraxella osloensis	*Candida krusei*	CMV[a]
Neisseria meningitidis Sero C	*Saccharomyces cerevisiae*	EBV (HHV-4)[b]
Neisseria meningitidis Sero A	*Corynebacterium urealyticum*	HSVl[a]
Streptococcus anginosus (Grp C)	MRSA	HSV2[a]
Streptococcus bovis	*Chlamydia trachomatis*	VZV (HHV 3)[a]
Streptococcus intermedius	*Bifidobacterium breve*	HPV-16[a]
Neisseria meningitidis M158 group D	*Mobiluncus mulieris*[a]	JC virus[a]
Neisseria flavescens	*Propionibacterium acnes*	BK virus[b]
Streptococcus parasanguinis	*Campylobacter jejuni*	HHV-6A[b]
Lactobacillus casei	*Hemophilus ducreyi*	HHV-6B[b]
Lactobacillus lactis	*Mycoplasma hominis*	HHV-7 [b]
Haemophilus influenza type B	*Mycoplasma genitalium*	HHV-8[b]
Salmonella newport	*Trichomonoas vaginalis*	
Shigella flexneri	*Pseudomonas fluorescens*	
Shigella sonnei	*Enterococcus dispar*	
Enterococcus durans	*Ureaplasma urealyticum*	
Enterococcus sp.*(ATC 202155)*	*Chlamydia pneumoniae*[a]	

[a]*Tested:10 ng/ml*
[b]*Tested:>10^6 cp/ml*

f. Interfering Studies:

Microbial Interference

The impact of non-target organisms on the detection of GBS by the NeuMoDx GBS Assay was assessed in a separate study. One strain of GBS (1200 CFU/ml, which is 2-3X LoD) was spiked into each pool of non-target organisms previously evaluated during the Cross-Reactivity Study and prepared negative clinical Lim broth matrix. Each pool was tested in triplicate. Only one replicate of Pool 17 gave a negative result. Three new samples of Pool 17 were tested in the presence of GBS to confirm results of initial testing. All 3 samples returned positive results after re-testing. The results of the study suggest that the NeuMoDx GBS Assay can detect GBS in the presence of high levels of non-target organisms associated with vaginal/rectal swab specimens.

Interfering Substances (Endogenous/Exogenous Substances)

This study was conducted to evaluate the potential interference of a panel of 26 endogenous and exogenous substances with the NeuMoDx GBS Assay (Table 6). Potentially interfering substances were added to pooled clinical negative Lim broth samples containing either 1200 CFU/ml (2-3X LoD) or 4000 CFU/ml (8X LoD) of GBS. Six samples yielded an Indeterminate result, but upon re-testing, produced valid results. No interference with the detection of GBS was observed with the panel of endogenous/exogenous substances. Results of the Interference Study are acceptable.

Table 6. List of Potentially Interfering Substances Tested

Exogenous/Endogenous Substances	Concentrations Tested
Monistat Cream	4.2 mg/ml
Yeast Gard (Douche)	17.1 mg/ml
Metamucil Fiber Supplement	1.8 mg/ml
Exlax (Chocolate Pieces)	20 mg/ml
Philips Milk of Magnesia	2.3 mg/ml
Pepto Bismol	20.4 mg/ml
Kaopectate	24.1 mg/ml
Dulcolax Suppositories	19.4 mg/ml
Fleet Enema	19.8 mg/ml
Preparation H Cream	8.1 mg/ml
Vagisil Powder	0.9 mg/ml
Norforms Suppositories	20.3 mg/ml
FDS Deodorant Spray	3.7 mg/ml
New Mama Bottom Spray	10.6 mg/ml
K-Y Jelly	5.9 mg/ml
McKesson gel	20.3 mg/ml
Contraceptive Foam	1.2 mg/ml
Moisturizing Lotion	7 mg/ml
Neutrogena Body Oil	12.5 mg/ml
Gold Bond Powder	0.4 mg/ml
Human Amniotic Fluid	10% V/V
Human Whole Blood	10% V/V
Human Urine	43% V/V
Human Fecal Sample	22.5 mg/ml
Mucus	22.9 mg/ml
Human Genomic DNA	1.5 µg/ml

g. Carry-Over/Cross-Contamination

A Carry-Over and Cross Contamination Study for the NeuMoDx GBS Assay was conducted to verify that no cross-contamination occurs on the test system with alternating GBS positive (n=53) and negative samples (n=52), as well as across multiple runs. Samples were tested in an alternating pattern with high positive samples (1 x 10^7 CFU/ml) and negative samples. In the checkerboard-design test, both the Indeterminate and Unresolved rates were determined to be 0.95% (1/105). A full run of high positive GBS samples (n=104) followed by a negative run (n=104) were tested to assess the across-run cross contamination. In the second study, (2) Indeterminates were generated to yield a 0.96% Indeterminate rate. No Unresolved results were reported in the second study. No carry-over/cross-contamination was observed in both studies. The overall percent agreement was 100% for valid positive and negative samples.

h. Assay cut-off:

NeuMoDx stated that the assay settings were chosen based on an in-house study to maximize the sensitivity and specificity of the NeuMoDx GBS Assay, which was subsequently used in the clinical evaluation of 1193 clinical specimens at 3-sites. In addition, the assay cut-offs were verified using 2 instruments and a set of 30 clinical negative Lim broth samples and 30 positive samples (clinical negative Lim broth spiked with GBS cells at/near the LoD). All samples were valid and generated the expected results. The cut-off values for a positive result for both the internal process control and GBS were set as follows (Table 7):

Table 7. Results Interpretation for the NeuMoDx GBS Assay

Result	**GBS C_t**	**Sample Process Control (SPC1) C_t**
Positive	9 < **C_t** < 37 And EP > 3000	N/A
Negative	N/A **OR** C_t < 9 **OR** > 37	25 < **C_t** < 35 **And** EP > 2000
Indeterminate	N/A SYSTEM ERROR NOTED	N/A SYSTEM ERROR NOTED
Unresolved	Not detected	Not detected

2. Comparison studies:

a. Method comparison with predicate device:

Not Applicable

b. Matrix comparison:

Not Applicable

3. Clinical studies:

a. Clinical Sensitivity:

NeuMoDx conducted a prospective multi-center study at three geographically diverse sites. Vaginal/rectal swab specimens were collected from pregnant women at 35-37 weeks gestation for routine standard of care screening purposes. Vaginal/rectal specimen swabs were cultured in Lim broth from 18-24 hrs at 35-37°C according to established CDC guidelines. Residual de-identified Lim broth cultures were used for testing with the NeuMoDx GBS Assay. Of the total 1250 specimens enrolled and tested during the study, (57) were shown to be non-compliant for the following reasons:

- (48)-Protocol deviation: Lim broth samples were frozen
- (9)-No reference culture result (equivocal culture)

A total of 1193 specimens were compliant and provided valid results for evaluation from the three participating sites.

For the Reference Culture Method, Lim broth cultures were subcultured to non-selective blood agar for up to 48 hours and inspected for organisms suggestive of GBS. Suspected colonies were gram stained and the Gram-positive cocci colonies were tested for catalase production. Gram positive cocci, catalase negative colonies were further characterized using streptococcal grouping latex agglutination to determine the presence of GBS.

The NeuMoDx GBS Assay was performed according to the package insert. The overall initial invalid rates (Indeterminate and Unresolved) was 0.8% (11/1314) for the clinical study. The final non-reportable result rate was 0% for the NeuMoDx GBS Assay, as all samples yielded valid results upon re-testing. Results of the NeuMoDx GBS Assay were compared to a Reference Culture Method as shown in Table 8. Site specific performance is shown in Table 9.

Table 8. Clinical Performance Data for the NeuMoDx GBS Assay vs Reference Culture Method (All Sites)

NeuMoDx GBS Assay	**Reference Culture Method**		
	Positive	Negative	Total
Positive	253	37[a]	290
Negative	8[b]	895	903
Total	261	932	1193
Sensitivity: 96.9% (253/261), 95% CI (94.1%-98.4%) Specificity: 96.0% (895/932), 95% CI (94.6%-97.1%)			

[a]20 of 37 FP samples were also reported as GBS Positive by another FDA-cleared GBS molecular assay.
[b]7 of 8 FN samples were also reported as GBS Negative by another FDA-cleared GBS molecular assay.

Table 9. Clinical Performance by Site

Site	n	Sensitivity	Specificity	Prevalence*
A	351	92.4% (73/79) [95% CI: 84.4-96.5]	96.7% (263/272) [95% CI:93.8-98.3]	22.5% (79/351)
B	400	98.4% (62/63) [95% CI: 91.5-99.7]	94.4% (318/337) [95% CI: 91.4-96.4]	15.8% (63/400)
C	442	99.2% (118/119) [95% CI: 95.4-99.9]	97.2% (314/323) [95% CI: 94.8-98.5]	26.9%(119/442)
Total	1193	96.9% (253/261) [95% CI: 94.1-98.4]	96.0% (895/932) [95% CI: 94.6-97.1]	21.9% (261/1193)

*By Reference Method

*b. Clinical specificity**:***

See above

c. Other clinical supportive data (when a. and b. are not applicable):

Not Applicable

4. Clinical cut-off**:**

Not Applicable

5. Expected values/Reference range:

In the investigational study with the NeuMoDx GBS Assay on the NeuMoDx 288 Molecular System, the overall GBS prevalence rate was determined to be 24.3% (290/1193) by the test system and 21.9% (261/1193) by the Reference Culture Method.

N. Instrument Name:

NeuMoDx 288 Molecular System

O. System Descriptions:

1. 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_____

2. Software:

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

   Yes ____X____ or No _____

3. Specimen Identification:

   Specimens are identified by scanning a barcode or by manual entry.

4. Specimen Sampling and Handling:

   Swab specimens are cultured in Lim broth for 18-24 hours after which a pipette is used to transfer Lim broth culture to a barcoded specimen (daughter) tube. Consumables and specimen tubes are loaded into carriers for the system before initiating testing.

5. Calibration:

   As specified in this Operator's Manual, only NeuMoDx certified technicians are authorized to perform installation, maintenance and service of the NeuMoDx 288 Molecular System. The System is calibrated on-site as part of the installation procedure, as well as during preventive maintenance.

6. Quality Control:

   Refer to Section M(1)(c) for information on the internal and external controls associated with the NeuMoDx GBS Assay and their performance.

P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:

Not Applicable

Q. Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

R. Conclusion:

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