

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k132082

B. Purpose for Submission:

New device

C. Measurand:

Anti-Deaminated Gliadin-derived Peptide (DGP) Antibodies

D. Type of Test:

Semi-quantitative and qualitative enzyme immunoassay

E. Applicant:

AESKU Diagnostics GmbH & Co.KG

F. Proprietary and Established Names:

AESKULISA® DGP-A
AESKULISA® DGP-G
AESKULISA® DGP-Check

G. Regulatory Information:

1. Regulation section:

21 § CFR 866.5750 – Radioallergosorbent (RAST) immunological test system

2. Classification:

Class II

3. Product code:

MST –Antibodies Gliadin

4. Panel:

Immunology (82)

H. Intended Use:

1. Intended use(s):

AESKULISA DGP-A is an in vitro diagnostic device. This solid phase enzyme immunoassay

employs synthetic, deamidated gliadin-derived peptides for the semiquantitative and qualitative detection of IgA antibodies against deamidated Gliadin-specific peptides (DGP) in human serum. The assay is an aid in the diagnosis of celiac disease (gluten-sensitive enteropathy) and should be used in conjunction with other serological tests and clinical findings.

AESKULISA DGP-G is an in vitro diagnostic device. This solid phase enzyme immunoassay employs synthetic, deamidated gliadin-derived peptides for the semiquantitative and qualitative detection of IgG antibodies against deamidated Gliadin-specific peptides (DGP) in human serum. The assay is an aid in the diagnosis of celiac disease (gluten-sensitive enteropathy) and should be used in conjunction with other serological tests and clinical findings.

AESKULISA DGP-Check is an in vitro diagnostic device. This solid phase enzyme immunoassay employs synthetic, deamidated gliadin-derived peptides for the combined semiquantitative and qualitative detection of IgA and IgG antibodies against deamidated Gliadin-specific peptides (DGP) in human serum. The assay is an aid in the diagnosis of celiac disease (gluten-sensitive enteropathy) and should be used in conjunction with other serological tests and clinical findings.

2. Indication(s) for use:

Same as intended use

3. Special conditions for use statement(s):

Prescription use only

4. Special instrument requirements:

Microplate reader capable of reading absorbance values at 450 nm

I. Device Description:

AESKULISA DGP kits include the following common reagents:

- Sample buffer concentrate (5x), 1x20 mL
- Wash buffer concentrate (50x), 1x20 mL
- Negative/positive/control, containing human serum, BSA and buffer components, 1x1.5mL each
- Cut-off calibrator for qualitative analysis, containing human serum, BSA and buffer components, 1x1.5mL
- Calibrators, (0, 3, 10, 30, 100; 300 U/mL), containing human serum, BSA and buffer components, 6x1.5mL
- Conjugate IgA/IgG – Goat Anti-human Immunoglobulins conjugated to horseradish peroxidase (HRP), 1x15mL
- TMB Substrate - Tetramethylbenzidine (TMB) chromogenic substrate, 1x15 mL
- Stop solution containing 1 M HCl, 1x15mL
- 96-well microtiter plate (12x8 well strips) with breakaway microwell coated with synthetic deaminated gliadin peptides.

J. Substantial Equivalence Information:

1. Predicate device name(s) and 510(k) numbers:

QUANTA Lite™ Gliadin IgA II (k052143)
 QUANTA Lite™ Gliadin IgG II (k052142)
 QUANTA Lite™ Celiac DGP Screen (k062708)

2. Comparison with predicates:

Similarities		
Feature	New devices AESKULISA DGP-A/G/Check	Predicate device INOVA QUANTA Lite Gliadin IgA II/ IgG II/ Celiac DPG Screen
Indications for Use	Aid in the diagnosis of celiac disease and should be used in conjunction with other serological tests and clinical findings	Same
Antigen	Deaminated gliadin peptides	Same
Conjugate	HRP conjugated goat anti-human IgA, IgG or IgA/IgG immunoglobulins	Same
Assay Principle	Indirect noncompetitive enzyme immunoassay	Same
Sample Dilution	1:100	Same
Specimen Type	Serum	Same
Assay Platform	96 well microtiter plates	Same
Incubation times	30-30-30 minutes	Same
Signal detected	Absorbance/optical density (OD) at 450 nm	Same

Differences		
Feature	New devices	Predicate devices
Antigen used	Synthetic, deaminated gliadin peptide	Purified gliadin peptides
Calibrators	6 calibrators: 0; 3; 10; 30; 100; 300 (U/mL) 1 cut-off calibrator	None
Controls	Positive Control	Low Positive Control High Positive Control
Assay Cut-off DGP-A & DGP-G DGP-Check	12-18 U/mL equi zone 16-24 U/mL equi zone	20 U/mL 20 U/mL

Differences		
Feature	New devices	Predicate devices
Result Interpretation		
Qualitative	Based on the cutoff calibrator Negative: < 0.8xOD cutoff Equivocal: 0.8xOD cut-off ≤OD patient ≤ 1.2 x OD cut-off Positive: > 1.2 x OD cutoff	No qualitative interpretation
Semi-quantitative: IgA or IgG	≤12 U/mL= Negative 12-18 U/mL=Equivocal >18 U/mL=Positive	≤ 20 U/mL= Negative 20-30 U/mL=Weak Positive >30 U/mL= Moderate-High Positive
IgA/IgG	≤16 U/mL= Negative 16-24 U/mL=Equivocal >24 U/mL=Positive	≤ 20 U/mL= Negative 20-30 U/mL=Weak Positive >30 U/mL= Moderate-High Positive

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

CLSI EP06-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

CLSI EP07-A2: Interference Testing in Clinical Chemistry, Approved Guideline – Second Edition

CLSI EP9-A2: Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Second Edition

CLSI EP17-A2: Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline

CLSI C28-A3c: Defining, Establishing, and verifying Reference Intervals in the Clinical Laboratory

CLSI EP25-A: Evaluation of Stability of In Vitro Diagnostic Reagents

CLSI H18-A4: Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests

L. Test Principle:

The AESKULISA® DGP assays are solid phase indirect noncompetitive immunoassays. The antigen (deaminated gliadin specific peptides) is coated to 96 well microtiter plates. Antibodies specific for DGP present in the diluted patient sera bind to the synthetic gliadin antigen immobilized on wells of a microtiter plate. The formed antigen-antibody complex then interacts with the enzyme-labeled anti-human antibody conjugate that is added to each well and reacts with a chromogenic substrate to develop a colored reaction product. A quantitative determination of antibody concentration is obtained by absorbance measurement of the colored solution using a spectrophotometric microtiter plate reader. The intensity of color formation from the chromogen is a

function of the amount of conjugate bound to the antigen-antibody complex and thus is proportional to the initial concentration of antibodies in the patient sera.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

For the semi-quantitative claim, studies were performed with six sera samples with analyte levels across the reportable range of each assay. Each sample was tested in 8 replicates in 5 runs in 5 days with 1 reagent lot (n = 40). Lot-to-lot precision study was performed with the same samples. The same antigen but a different lot of conjugate was used for each of the three lots. The results are summarized in the tables below:

AESKULISA DGP-A (assay range 1.74-300 U/mL; 12-18 U/mL equivocal)

Mean (U/mL)	Intra-assay		Inter-assay		Lot-to-Lot	
	SD	%CV	SD	%CV	SD	%CV
2.7	0.3	11.2	0.6	21.0	0.5	17.6
12.9	0.9	6.9	1.4	10.5	1.2	9.1
16.7	1.6	9.6	1.8	10.7	n/d	n/d
49.9	2.4	4.7	5.3	10.6	4.3	8.2
97.2	5.2	5.4	9.9	10.2	8.8	9.0
209.3	12.2	5.7	29.7	14.2	24.4	10.8

AESKULISA DGP-G (assay range 2.09-300 U/mL; 12-18 U/mL equivocal)

Mean (U/mL)	Intra-assay		Inter-assay		Lot-to-Lot	
	SD	%CV	SD	%CV	SD	%CV
3.2	0.4	11.3	0.7	21.5	0.2	7.6
12.1	1.2	10.4	1.8	14.8	1.4	12.4
19.5	1.8	9.0	2.0	10.0	n/d	n/d
24.3	2.8	11.7	3.2	13.0	2.0	9.1
86.2	8.0	9.3	9.3	10.8	5.8	7.2
256.9	21.7	8.5	30.4	11.8	22.7	8.6

AESKULISA DGP-Check (assay range 1.84-300 U/mL; 16-24 U/mL equivocal)

Mean (U/mL)	Intra-assay		Inter-assay		Lot-to-Lot	
	SD	%CV	SD	%CV	SD	%CV
11.5	1.2	10.8	1.4	12.2	1.3	12.4
18.9	1.2	10.3	1.4	12.2	1.29	12.4
28.8	3.6	12.5	2.3	12.2	2.1	11.0
64.0	5.2	8.2	3.8	13.0	3.3	11.0
128.0	8.9	7.1	18.5	14.4	10.3	8.1

For the qualitative claim, the same 6 samples were evaluated. Negative samples were 100% negative and positive samples were 100% positive. Results of the equivocal and positive samples close to the upper limit of the equivocal zone are summarized below:

Sample concentration	negative	Equivocal	positive
DGP-A (12-18U/mL equivocal zone)			
12.9 U/mL	30.0%	70.0%	0.0%
16.7 U/mL	0.0%	75.0%	25.0%
DGP-G (12-18U/mL equivocal zone)			
12.1 U/mL	42.5%	57.5%	0.0%
19.5 U/mL	0.0%	20.0%	80.0%
24.3 U/mL	0.0%	5.0%	95.0%
DGP-Check (16-24U/mL equivocal zone)			
18.9 U/mL	15.0%	85.0%	0.0%
28.8 U/mL	5.0%	95.0%	0.0%

b. Linearity/assay reportable range:

Three positive serum samples were serially diluted using a negative serum sample and tested in duplicates. A calculation of the percent recovery and a regression analysis were performed on the results for each assay. The samples with $> \pm 10\%$ recovery from the expected value were typically highly diluted samples. The results are summarized below:

Dilution range U/mL	Slope (95%CI)	Y-intercept (95%CI)	R ²
DGP-A			
331.9 - 35.6	1.016 (0.91 to 1.13)	-12.7 (-40.19 to 14.78)	0.988
89.6 - 5.3	1.015 (0.88 to 1.15)	5.24 (-18.63 to 8.16)	0.983
13.9 - 1.3	0.974 (0.803 to 1.14)	-0.706 (-2.31 to 0.89)	0.97
Combined	y = -1.2987 + 1.0040x R ² 0.9958		
DGP-G			
313.9 - 42.3	0.979 (0.89 to 1.06)	2.32 (-14.66 to 19.3)	0.993
119.2 - 13.7	0.927 (1.107 to 1.11)	-3.37 (-10.20 to 3.47)	0.992
16.7 - 1.2	1.08 (0.93 to 1.23)	-1.06 (-2.70 to 0.58)	0.980
Combined	y = -0.5213 + 0.9905x R ² 0.9969		

Dilution range U/mL	Slope (95% CI)	Y-intercept (95% CI)	R ²
DGP-Check			
Dilution range U/mL	Slope (95% CI)	Y-intercept (95% CI)	R ²
392.5-26.9	0.988 (0.88 to 1.09)	2.926 (-19.51 to 25.37)	0.989
156.5- 11.2	1.044 (0.958 to 1.13)	-5.27 (-10.18 to -0.37)	0.993
14.0-1.3	1.002 (0.904 to 1.10)	-0.479 (-1.35 to 0.39)	0.991
Combined	y = -2.8031 + 0.9827x R ² 0.9941		

The DGP-A claimed linear range is 1.74 – 300 U/mL

The DGP-G claimed linear range is 2.1 – 300 U/mL

The DGP-Check claimed linear range is 1.84 – 300 U/mL

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

Traceability:

There are no international standards for the gliadin antibodies. The results are reported in arbitrary units (U/mL). In order to have traceability with newly made calibrators, each new lot of calibrators is assayed on an already cleared kit and compared to the exiting calibrators.

Calibrators and Controls:

The calibrators are human serum in standard buffer. There are 6 levels (A-F) with assigned values from 0-300 U/mL. Calibrators E to Cal B are generated by diluting Cal F according to the dilution scheme shown in table below. The cut-off calibrator for the qualitative analysis is derived from Cal F by diluting Cal F to 15 U/mL for IgA and IgG and 20 U/mL for IgA/G.

Calibrator	Assigned Units (U/mL)	Dilution Range
Cal F	300	
Cal E	100	1:3 dil of F
Cal D	30	1:3 dil of E
Cal C	10	1:3 dil of D
Cal B	3	1:3 dil of C
Cal A	0	Buffer (contains no sera)

Calibrator curve is acquired by fitting the values of 6 calibrators using a 4 parameter Lin-Log fit.

The negative control contains negative human serum in buffer. The positive control is from Cal F by diluting Cal F to 45-50 U/mL.

Kit Stability:

Shelf life stability was tested at real-time. Three lots of the assay kit, including all

components of the kit, were stored at 2-8°C. At different time points (0, 6 months, 12 months and 18 months), three samples with low, medium and high level of the analyte were tested in duplicate in one assay run. The results were compared to the results of the same samples tested at time = 0. The results support an unopened/shelf-life of 18 months at 2-8°C.

Open vial stability - 4 samples – high negative, cut-off, low positive and high positive (approximately 10, 15, 30-70 and >100 U/mL) – were tested on different days on DGP-A, G and Check. The recovery results after 1 month were 96% to 112.7%.

Transport stability – the kit was assayed with 4 samples (negative, low, medium, high); each sample was tested in duplicate after being for 2 weeks at 37°C and compared to t=0. The stability results are summarized below:

Stability	DGP-A/DGP-G/DGP-Check
Kit shelf life (2-8°C)	18 months
Transport stability (37°C)	2 weeks
Reconstituted buffers	1 month
Open kit stability (2-8°C)	28 days
Serum samples	Up to 4 freeze/thaw cycles

d. Detection limit:

The Limit of Blank (LoB) was based on 60 determinations of a blank sample and was estimated as the 95% percentile of the distribution. The Limit of Detection (LoD) was calculated according to the equation: the LoB + c_{β} x SD where SD, the standard deviation, was determined by assaying eight samples with analyte level near the lower limit of the reportable range in eight replicates. The results are summarized below:

	LoB (U/mL)	LoD (U/mL)
AESKULISA DGP-A	0.53	1.69
AESKULISA DGP-G	0.30	1.83
AESKULISA DGP-Check	0.20	1.44

e. Analytical specificity:

Interference:

Interferences were assessed by testing 4 serum samples with analyte concentrations across the assay range. Each sample was spiked with interfering substances and tested in duplicates. The data demonstrated that the assay was not affected by high levels of Rheumatoid Factor (400 IU/mL), Bilirubin (20 mg/dL), Triglyceride (3000 mg/dL), and Hemoglobin (800 mg/dL). The data is summarized in the table below:

Interfering substance (concentration)	Percent Recovery (%)		
	DGP-A	DGP-G	DGP-Check
Rheumatoid Factor (400 IU/mL)	93.4-105.9	99.4-107.8	96.9-113.9
Bilirubin (20 mg/dL)	87.4-112.6	98.4-117.7	102.5-110.5
Triglyceride (3000 mg/dL)	89.2-101.9	93.9-97.3	87.3-107.2
Hemoglobin (800 mg/dL)	91.5-95.5	95.6-101.6	86.9-105.7

Cross-reactivity:

Cross-reactivity was tested using samples from other autoimmune conditions. The samples and results are summarized in the table below:

Diagnosis	Crossreactivity		
	DGP-A pos >18 U/mL	DGP-G pos >18 U/mL	DGP-Check pos >24 U/mL
Autoimmune Hepatitis	0/5 (0%)	0/5 (0%)	0/6 (0%)
CCP	0/5 (0%)	0/5 (0%)	0/5 (0%)
CCP; Rheumatoid Factor	0/11 (0%)	0/11 (0%)	0/11 (0%)
Crohns Disease	ND	ND	0/7 (0%)
Healthy	0/99 (0%)	0/99 (0%)	1/99 (1%)
non-DH/CD controls (unspecified, diagnosis unknown)	1/56 (1.8%)	3/56 (5.4%)	3/56 (5.4%)
Rheumatoid Arthritis	0/18 (0%)	0/18 (0%)	0/18 (0%)
SLE	0/13 (0%)	0/13 (0%)	0/23 (0%)
Vasculitis	0/2 (0%)	0/2 (0%)	0/3 (0%)
Total	1/209 (0.5%)	3/209 (1.4%)	4/228 (1.8%)

f. Assay cut-off:

The assay cut-off was established by measuring serum samples from 160 healthy donors and 139 subjects with other autoimmune diseases expected to be negative for DPG antibodies. These samples were not used in the clinical evaluation study. The cut-off was determined from the mean ± 3 standard deviations. The results are shown in the table below:

	Negative	Equivocal	Positive
DGP-A	< 12 U/mL	12–18 U/mL	> 18 U/mL
DGP-G	< 12 U/mL	12–18 U/mL	> 18 U/mL
DGP-Check	< 16 U/mL	16-24 U/mL	> 24 U/mL

2. Comparison studies:

a. Method comparison with predicate device

For the method comparison study, 431 serum samples were tested by both the new and predicate devices (see table below). Patients with Celiac Disease (CD) were diagnosed by serology and confirmed with biopsy. Patients with Dermatitis Herpetiformis (DH) were diagnosed based on skin biopsy and serology.

Disease Group	Number of Samples
Celiac Disease (CD)	79
CD IgA Def	16
CD suspect	41
CD suspect IgA Def	2
Dermatitis Herpetiformis (DH)	65
Total	203

Disease Controls	Number of Samples
Autoimmune Hepatitis	6
CCP	5
CCP; Rheumatoid Factor	11
Crohns Disease (DGP-Check only)	7
Rheumatoid Arthritis	18
SLE	23
Vasculitis	3
Non DH-CD (diagnosis unspecified/unknown)	56
Total	129
Healthy Controls	99
Total	431

For the comparison study calculations, samples outside of the reportable range of both devices and the 99 normal samples were excluded. For DGP Check testing, 12 diluted samples were included to cover the assay range. Results are summarized below:

AEKULISA DPG -A

DGP-A		INOVA (U/mL)		
		Positive (>20)	Negative (≤20)	Total
AESKULISA DGP-A Test (U/mL)	Positive (>18)	88	16*	104
	Equivocal (18-12)	7**	20	27
	Negative (<12)	13**	64	77
	Total	108	100	208

*10 CD, 2 DH, 3 suspected of CD, 1 non-DH/CD

**16 CD, 1 suspected of CD, 2 DH, 1 non-DH/CD

DGP-A		INOVA (U/mL)		
Equivocal as negative		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive >18	88	16	104
	Negative ≤18	20	84	104
	Total	108	100	208

Positive agreement: 81.5% (88/108) (95% CI: 73.1 – 87.7%)

Negative agreement: 84.0% (84/100) (95% CI: 75.6 – 89.9%)

Total agreement: 82.7% [(88+84)/208] (95% CI: 77.0 – 87.2%)

DGP-A		INOVA (U/mL)		
Equivocal as positive		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive ≥12	95	36	131
	Negative <12	13	64	77
	Total	108	100	208

Positive % agreement: 88.0% (95/108) (95% CI: 80.5 - 92.8%)
 Negative % agreement: 64.0% (64/100) (95% CI: 54.2 - 72.7%)
 Total % agreement: 76.4% [(95+64)/208] (95% CI: 70.2 - 81.7%)

AESKULISA DGP-G

DGP-G		INOVA (U/mL)		
		Positive (>20)	Negative (≤20)	Total
AESKULISA DGP-G (U/mL)	Positive (>18)	136	13*	149
	Equivocal (18-12)	8**	13	21
	Negative (<12)	0	48	48
	Total	144	74	218

*7 CD, 4 DH, 2 non-DH/CD

**2 CD, 3 suspected of CD, 3 DH

DGP-G		INOVA (U/mL)		
Equivocal as negative		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive >18	136	13	149
	Negative ≤18	8	61	69
	Total	144	74	218

Positive % agreement: 94.4% (136/144) (95% CI: 89.4 – 97.2%)

Negative % agreement: 82.4% (61/74) (95% CI: 72.2 – 89.4%)

Total % agreement: 90.4% [(136+61)/218] (95% CI: 85.7 – 93.6%)

DGP-G		INOVA (U/mL)		
Equivocal as positive:		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive ≥12	144	26	170
	Negative <12	0	48	48
	Total	144	74	218

Positive % agreement: 100% (144/144) (95% CI: 97.4 – 100.0%)

Negative % agreement: 64.9% (48/74) (95% CI: 53.5 – 74.8%)

Total % agreement: 88.1% [(144+48)/218] (95% CI: 83.1 – 91.7%)

AESKULISA DPG -Check:

DGP-Check		INOVA (U/mL)		
		Positive (>20)	Negative (≤20)	Total
AESKULISA DGP-Check (U/mL)	Positive (>24)	122	8*	130
	Equivocal (16-24)	14**	5	19
	Negative (<16)	7**	60	67
	Total	143	73	216

*6 CD, 1 suspected of CD, 1 non-DH/CD

**14 equivocal [7 CD (diluted); 6 suspected of CD; 1 DH]

**7 negative (2 CD; 2 suspected of CD suspect; 3 DH)

DGP-Check Equivocal as negative		INOVA (U/mL)		
		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive >24	122	8	130
	Negative ≤24	21	65	86
	Total	143	73	216

Positive % agreement: 85.3% (122/143) (95% CI: 78.6% – 90.2%)

Negative % agreement: 89.0% (65/73) (95% CI: 79.8% – 94.3%)

Total % agreement: 86.6% [(122+65)/216] (95% CI: 81.4 – 90.5%)

DGP-Check Equivocal as positive		INOVA (U/mL)		
		Positive >20	Negative ≤20	Total
AESKU (U/mL)	Positive ≥16	136	13	149
	Negative <16	7	60	67
	Total	143	73	216

Positive % agreement: 95.1% (136/143) (95% CI: 90.2% – 97.6%)

Negative % agreement: 82.2% (60/73) (95% CI: 71.9% – 89.3%)

Total % agreement: 90.7% [(136+60)/216] (95% CI: 86.1 – 93.9%)

b. Matrix comparison:

Not applicable. Each assay uses only serum as the matrix.

3. Clinical studies:

a. Clinical Sensitivity and Specificity:

Serum samples used in the method comparison studies were also used to determine sensitivity and specificity of each assay. The following samples were excluded from the calculations: 43 samples with suspected CD and CD IgA deficient diagnosis and 99 healthy controls. For the DGP-A assay, the 16 CD samples with IgA deficiency were also excluded. For the DGP-Check, the 12 diluted samples were also excluded.

The tables below summarize the groups of diagnosed patients evaluated by each assay and the calculated specificity and sensitivity results with equivocal as positive or negative:

AESKULISA DGP-A:

DGP-A Equivocal as negative:	Diagnosis		
	Positive	Negative	Total
Positive >18 U/mL	110	1	111
Negative ≤18 U/mL	34	109	143
Total	144	110	254

Sensitivity: 76.4% (110/144) (95% CI: 68.8 - 82.6)

Specificity: 99.1% (109/110) (95% CI: 95.0 - 99.8)

Overall Agreement: 86.2% [(110+109)/254] (95% CI: 81.4 – 89.9)

DGP-A	Diagnosis		
	Positive	Negative	Total
Equivocal as positive			
Positive ≥ 12 U/mL	123	23	146
Negative < 12 U/mL	21	87	108
Total	144	110	254

Sensitivity: 85.4% (123/144) (95% CI: 78.7 – 90.3)

Specificity: 79.1% (87/110) (95% CI: 70.6 – 85.6)

Overall Agreement: 82.7% [(123+187)/254] (95% CI: 77.6 – 86.8)

DGP-A	AESKU		
Disease Group	POS (>12)	POS (>18)	Total
CD	60(75.9%)	51(64.6%)	79
DH	63(96.9%)	59(90.8%)	65
SLE	7(53.8%)	0(0%)	13
Vasculitis	0(0%)	0(0%)	2
Rheumatoid Arthritis	2(11.1%)	0(0%)	18
CCP	1(20.0%)	0(0%)	5
CCP; Rheumatoid Factor	1(9.1%)	0(0%)	11
Autoimmune Hepatitis	0(0%)	0(0%)	5
non-DH/CD controls (diagnosis not specified)	12(21.4%)	1(1.8%)	56
Total	146(57.5%)	111(43.7%)	254

AESKULISA DGP-G :

DGP-G	Diagnosis		
	Positive	Negative	Total
Equivocal as negative			
Positive > 18 U/mL	150	3	153
Negative ≤ 18 U/mL	10	107	117
Total	160	110	270

Sensitivity: 93.8% (150/160) (95% CI: 88.9 – 96.6)

Specificity: 97.3% (107/110) (95% CI 92.3 – 99.1)

Overall Agreement: 95.2% [(150+107)/270] (95% CI: 91.9 – 97.2)

DGP-G	Diagnosis		
	Positive	Negative	Total
Equivocal as positive:			
Positive ≥ 12 U/mL	159	31	190
Negative < 12 U/mL	1	79	80
Total	160	110	270

Sensitivity: 99.4% (159/160) (95% CI: 96.6 – 99.9)

Specificity: 71.8% (79/110) (95% CI: 62.8 – 79.4)

Overall Agreement: 88.2% [(159+79)/270] (95% CI: 83.8 – 91.5)

DGP-G	AESKU		
	POS >12	Pos >18	Total
CD	79(100%)	75(94.9%)	79
CD IgA Def	16(100%)	16(100%)	16
DH	64(98.5%)	59(90.8%)	65
SLE	9(69.2%)	0(0%)	13
Vasculitis	0(0%)	0(0%)	2
Rheumatoid Arthritis	5(27.8%)	0(0%)	18
CCP	1(20%)	0(0%)	5
CCP; Rheumatoid Factor	4(36.4%)	0(0%)	11
Autoimmune Hepatitis	4(80%)	0(0%)	5
non-DH/CD controls	8(14.3%)	3(5.4%)	56
Total	190(70.4%)	153(56.7%)	270

AESKULISA DGP -Check:

DGP-Check	Diagnosis		
	Positive	Negative	Total
Equivocal as negative			
Positive >24 U/mL	151	3	154
Negative ≤24 U/mL	9	126	135
Total	160	129	289

Sensitivity: 94.4% (151/160) (95% CI: 89.7 – 97.0)

Specificity: 97.7% (126/129) (95% CI 93.4 – 99.2)

Overall Agreement: 95.9% [(151+126)/289] (95% CI: 92.9 – 97.6)

DGP-Check	Diagnosis Group		
	Positive	Negative	Total
Equivocal as positive:			
Positive ≥16 U/mL	152	13	165
Negative <16 U/mL	8	116	124
Total	160	129	289

Sensitivity: 95.0% (152/160) (95% CI: 90.5 – 97.5)

Specificity: 89.9% (116/129) (95% CI: 83.5 – 94.0)

Overall Agreement: 92.7 %[(152+116)/289] (95% CI: 89.2 – 95.2)

DGP-Check	AESKU		
	POS (≥16)	POS (>24)	Total
Disease Group			
CD	77(97.5%)	77(97.5%)	79
CD IgA Def	15(93.8%)	15(93.8%)	16
DH	60(92.3%)	59(90.8%)	65
SLE	7(30.4%)	0(0%)	23
Vasculitis	0(0%)	0(0%)	3
Crohns Disease	2(28.6%)	0(0%)	7
Rheumatoid Arthritis	0(0%)	0(0%)	18
CCP	0(0%)	0(0%)	5

DGP-Check	AESKU		
Disease Group	POS (≥ 16)	POS (> 24)	Total
CCP; Rheumatoid Factor	1 (9.1%)	0(0%)	11
Autoimmune Hepatitis	0(0%)	0(0%)	6
non-DH/CD controls	3 (5.4%)	3(5.4%)	56
Total	165(57.1%)	154(53.3%)	289

b. Other clinical supportive data (when a. is not applicable):

Not applicable

4. Clinical cut-off:

See assay cut-off.

5. Expected values/Reference range:

The expected value in the general population is negative. A study of 133 apparently healthy blood donors was performed on all three AESKULISA kits. The samples included 69 female, 64 male; 116 in the age group of 16-45, 17 in the age group of 46+). The results are summarized in the table below:

Results	Healthy Samples		
	DGP-A	DGP-G	DGP-Check
Max	13.7 IU/mL	18.2 IU/mL	28.4 IU/mL
Mean \pm SD	3.1 \pm 1.8 IU/mL	3.2 \pm 2.2 IU/mL	8.3 \pm 4.4 IU/mL
95% CI	97.2 - 100.0%	95.9 - 99.9%	95.9 - 99.9%

N. Proposed Labeling:

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

O. Conclusion:

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