

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

A. 510(k) Number:

k071692

B. Purpose for Submission:

New device

C. Measurand:

IgA and IgG Anti-tissue Transglutaminase (tTG) antibodies

IgA and IgG Anti-Gliadin antibodies

D. Type of Test:

Semi-quantitative

E. Applicant:

TheraTest Laboratories, Inc.

F. Proprietary and Established Names:

TheraTest EL-tTG™ IgA/IgG

TheraTest EL-Glia™ IgA/IgG

G. Regulatory Information:

1. Regulation section:

21 § CFR 866.5660 Multiple autoantibodies immunological test system

21 § CFR 866.5750 Radioallergosorbent (RAST) immunological test system

2. Classification:

Class II

3. Product codes:

MVM, Autoantibodies, Endomysial (Tissue Transglutaminase)

MST, Antibodies, Gliadin

4. Panel:

Immunology (82)

H. Intended Use:

1. Intended use(s):

The TheraTest EL-tTG™ IgA/IgG and TheraTest EL-GLIA™ IgA/IgG Kits are enzyme-linked immunosorbent assay (ELISA) test systems for the semi-quantitative measurement of IgA and IgG anti-tissue transglutaminase (tTG) and anti-gliadin antibodies in human serum. Detection and semi-quantitation of these antibodies is intended to aid the diagnosis of patients with gluten sensitive enteropathies: celiac disease and dermatitis herpetiformis, in conjunction with other clinical findings and laboratory tests.

2. Indication(s) for use:

Same as Intended use.

3. Special conditions for use statement(s):

For prescription only.

4. Special instrument requirements:

Microplate reader: Spectrophotometer with single (450 nm) or dual (450 nm, 620-690 nm reference) wavelength and ELISA plate washer (optional)

I. Device Description:

Each device contains the following: microwell plate with breakaway microwell

coated with either recombinant tTG or purified gliadin antigen; plate frame; assay controls (IgA or IgG positive and IgA or IgG negative); calibrators (IgA or IgG); tTG/ Gliadin goat anti-human IgA conjugate; tTG/ Gliadin goat anti-human IgG conjugate; TMB chromogen; wash buffer (10X) and stop solution.

J. Substantial Equivalence Information:

1. Predicate device name(s):
 ImmuLisa™ Anti-human Tissue Transglutaminase IgA
 ImmuLisa™ Anti-human Tissue Transglutaminase IgG
 ImmuLisa™ Anti-Gliadin Antibody IgA
 ImmuLisa™ Anti-Gliadin Antibody IgG
2. Predicate K number(s):
 k992878 (Anti-hu tTG IgA)
 k040095 (Anti-hu tTG IgG)
 k964341 (Anti-Gliadin IgA)
 k964344 (Anti-Gliadin IgG)
3. Comparison with predicate:

Similarities			
Item	New Device	Predicate Device	
	TheraTest EL-tTG™ IgA/IgG and TheraTest EL-GLIA™ IgA/IgG	ImmuLisa™ Anti-hu tTG Antibody IgA/IgG	ImmuLisa™ Anti-Gliadin Antibody IgG or IgA
Antigen	Human recombinant tTG and purified gliadin	Recombinant human tTG	Purified gliadin
Method	ELISA	Same	Same
Assay Platform	96 well microtiter plates coated with specific antigen	Same	Same
Measurement	Semi-quantitative	Same	Same
Sample	Serum	Same	Same
Sample volume required	10 µL	Same	Same
Microplate Reader	Spectrophotometer	Same	Same
Detection Method	Colorimetric	Same	Same

Differences			
Item	Device	Predicate	
	TheraTest EL-tTG™ IgA/IgG and TheraTest EL-GLIA™ IgA/IgG	ImmuLisa™ Anti-human Tissue Transglutaminase IgA/IgG	ImmuLisa™ Anti-Gliadin Antibody (IgG or IgA)
Intended use	For the semi-quantitative detection of	For the semi-quantitative detection of	For the semi-quantitative detection of IgA

Differences			
Item	Device	Predicate	
	IgA/IgG antibodies to human recombinant tTG and purified gliadin in human serum	IgA/IgG antibodies to recombinant human tTG in serum	or IgG antibodies to gliadin in human serum
Indications for Use	Aid in the diagnosis of IgA/IgG celiac disease and dermatitis herpetiformis	<p>Anti-hu tTG IgA: Aid in the diagnosis of celiac disease and dermatitis herpetiformis</p> <p>Anti-hu tTG IgG: Aid in the diagnosis of celiac disease in patients with IgA deficiency</p>	Aid in the diagnosis of celiac disease and dermatitis herpetiformis
Positive and Negative Control	To be diluted 1:101 with 1X Wash buffer	Pre-diluted human serum. Ready to use.	Pre-diluted human serum. Ready to use.
Calibrator	One calibrator	Set of four calibrators	Set of four calibrators
Conjugate	Goat anti-human IgA (α chain specific) and IgG (Fc γ specific) horseradish peroxidase	Anti-human IgA/IgG alkaline phosphatase conjugate	Anti-human IgA or IgG alkaline phosphatase conjugate
Serum Dilution	1:101	1:51	1:51
Assay wash step	3X wash twice	4X wash twice	4X wash twice
Incubation	30-30-15 (IgA and IgG)	60-30-30 (IgA) 30-30-30 (IgG)	30-30-15 (IgA and IgG)
Reading after Stop solution	Within 30 min	Within 1 hour	Within 1 hour
Result Interpretation	≥ 20 U/mL: Normal 21-25: Equivocal >25 : Abnormal	< 20 EU/mL: Neg 20-25: Borderline >25 : Positive	Children: AGA-IgA: <23 EU/mL: Neg >23 : Positive AGA-IgG: <28 EU/mL: Neg >28 : Positive

Differences			
Item	Device	Predicate	
			Adults: AGA-IgA and AGA-IgG: <20 EU/mL: Neg >20: Positive

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

Not applicable

L. Test Principle:

The TheraTest EL-tTG™ IgA/IgG or The TheraTest EL-Gliadin™ IgA/IgG Test System is a solid phase enzyme immunoassay in a 96-well plate format for the measurement of IgA and IgG antibodies against tissue transglutaminase or gliadin. Wells are coated with human recombinant tissue transglutaminase or purified gliadin, and incubated with Specimens, Calibrators, Positive and Negative Controls. During the incubation, anti-tTG or anti-gliadin antibodies present in the test sample are bound to the solid phase antigen. The wells are subsequently washed, and isotype-specific horseradish-peroxidase labeled anti-human immunoglobulin antibody (enzyme conjugate) is added. After incubating the wells with the enzyme conjugate, unbound labeled antibody is removed by washing. A chromogenic substrate solution is then added to the wells, and the presence of antibodies to tissue transglutaminase is detected by a color change produced by the conversion of the substrate by the enzyme. The reaction is stopped, and the intensity of the color, which is proportional to the amount of the bound antibody, is read by an ELISA reader. The absorbance value in the blank well (incubated with Specimen Diluent) is subtracted from the values obtained with Specimens, Calibrators and Controls.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Intra-assay studies on anti-tTG IgA/IgG and anti-Gliadin IgA/IgG:

Three specimens (high, moderate and near cut-off) were assayed for a total of 20 times and two runs per day. The mean units for anti-tTG IgA ranged from 26 to 187 units and the mean units for anti-tTG IgG ranged from 29 to 83 units. The percent CV for anti-tTG IgA ranged from 2.8% – 10.9% and for anti-tTG IgG, 2.9% – 5.3%. The mean units for anti-Gliadin IgA ranged from 29 to 154 units and the mean units for anti-Gliadin IgG ranged from 23 to 88 units. The percent CV for anti-Gliadin IgA ranged from 4.0% – 7.6% and for anti-Gliadin IgG 2.4% – 10.8%.

Inter-assay studies on anti-tTG IgA/IgG and anti-Gliadin IgA/IgG:

Three specimens (high, moderate and near cut-off) were assayed for a total of 20 different times in one or two runs. The mean units for anti-tTG IgA ranged from 30 to 193 units and for anti-tTG IgG ranged from 27 to 83 units. The percent CV for anti-tTG IgA ranged from 3.9% – 10.1% and 7.8% – 10.7%

for anti-tTG IgG. The mean units for anti-Gliadin IgA ranged from 34 to 134 units and the mean units for anti-Gliadin IgG ranged from 29 to 65 units. The percent CV for anti-Gliadin IgA ranged from 8.4% – 10.1% and 9.0% – 10.2% for anti-Gliadin IgG.

- b. *Linearity/assay reportable range:*
Not applicable.
- c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
There are no reference standards for tTG and gliadin. The results are reported in arbitrary units.
- d. *Detection limit:*
Not applicable.
- e. *Analytical specificity:*
Interference by endogenous substances: No data provided. The package insert states that grossly hemolyzed, lipemic, icteric or microbially contaminated serum should not be used in this assay.
Crossreactivity with other autoantibodies: Testing was performed with 56 sera consisting of 40 inflammatory bowel disease (IBD) and 16 thyroid disease. The positive samples were: one IBD with anti-tTG IgA, one IBD with anti-tTG IgG, two IBD with anti-gliadin IgA, one IBD with anti-gliadin IgG and one thyroid disease with anti-gliadin IgG.
- f. *Assay cut-off:*
The cut-off value of >25 U/mL was established based on testing 100 blood bank donors. There were 51 females and 49 males with a median age of 29 years and a range from 16 to 70 years. The race distribution was 14% Hispanic, 8% Black, 4% Asian, and 74% other. The 98th percentile was used to establish the upper limit of normal for IgA and IgG Anti-tTG and the 97.5th and 94th percentile were used to establish for IgA and IgG Anti-gliadin respectively.

2. Comparison studies:

- a. *Method comparison with predicate device:*
Testing was performed on 106 samples (42 from IgA sufficient CD patients, 14 from IgA deficient patients and 50 healthy donors). The comparative study on anti-tTG IgA had a 100% Positive Percent Agreement (40/40) (95% CI: 91%-100%); 97% Negative Percent Agreement (64/66) (95% CI: 89%-100%); and 98% Overall Agreement (104/106) (95% CI: 93%-100%). Refer to table below:

		ImmuLisa™ Anti-hu tTG IgA		
		Positive	Negative	Total
EL-tTG™ IgA	Positive	40	2	42
	Negative	0	64	64
	Total	40	66	106

The comparative study on anti-tTG IgG had a Positive Percent Agreement of 84% (27/32) (95% CI: 67%-95%); 95% Negative Percent Agreement (62/65) (95% CI: 87%-99%); and 92% Overall agreement (89/97) (95% CI: 84%-

96%). Refer to table below:

		ImmuLisa™ Anti-hu tTG IgG			
		Positive	Equivocal*	Negative	Total
EL-tTG™ IgG	Positive	27	(4)	3	30
	Equivocal*	0	(1)	(3)	(4)
	Negative	5	(1)	62	67
	Total	32	(6)	65	97

*Equivocals were excluded from the calculation.

The comparative study on anti-Gliadin IgA had a 95% Positive Percent Agreement (21/22) (95% CI: 77%-100%); 96% Negative Percent Agreement (78/81) (95% CI: 90%-99%); and 96% Overall Agreement (99/103) (95% CI: 90%-99%). Refer to table below:

		ImmuLisa™ Anti-Gliadin IgA		
		Positive	Negative	Total
EL-Glia™ IgA	Positive	21	3	24
	Equivocal*	(2)	(1)	(3)
	Negative	1	78	79
	Total	22	81	103

*Equivocals were excluded from the calculation.

The comparative study on anti-Gliadin IgG had a 81% Positive Percent Agreement (29/36) (95% CI: 64%-92%); 92% Negative Percent Agreement (58/63) (95% CI: 82%-97%); and 88% Overall Agreement (87/99) (95% CI: 80%-94%). Refer to table below:

		ImmuLisa™ Anti-Gliadin IgA		
		Positive	Negative	Total
EL-Glia™ IgG	Positive	29	5	34
	Equivocal*	(6)	(1)	(7)
	Negative	7	58	65
	Total	36	63	99

*Equivocals were excluded from the calculation.

b. Matrix comparison:

Both assays use serum as the sample matrix.

3. Clinical studies:

a. Clinical Sensitivity and specificity

Samples used in the studies for determination of clinical sensitivity and specificity consisted of 44 samples from Celiac disease patients, 40 IBD, 16 thyroid disease patients and 100 blood bank donors. Samples from IBD, thyroid disease and the blood donors were combined as the control group. For anti-tTG IgA, clinical sensitivity was 95% (42/44) and clinical specificity was 98% (153/156) (refer to table below):

		Diagnosis		
		Celiac disease* (n=44)	Control group (n=156)	Total (n=200)
EL-tTG™ IgA	Positive	42	3	45
	Equivocal	0	0	0
	Negative	2**	153	155
	Total	44	156	200

* The group includes patients on gluten-containing and gluten-restricted diet, as well.

** IgA deficient patients

For anti-tTG IgG, clinical sensitivity was 48% (21/44) and clinical specificity was 97% (152/156) (refer to table below):

		Diagnosis		
		Celiac disease* (n=44)	Control group (n=156)	Total (n=200)
EL-tTG™ IgG	Positive	21	3	24
	Equivocal	4	1	5
	Negative	19	152	171
	Total	44	156	200

* The group includes patients on gluten-containing and gluten-restricted diet, as well.

For anti-Gliadin IgA, clinical sensitivity was 55% (24/44) and clinical specificity was 95% (148/156) (see table below):

		Diagnosis		
		Celiac disease* (n=44)	Control group (n=156)	Total (n=200)
EL-Glia™ IgA	Positive	24	4	28
	Equivocal	3	4	7
	Negative	17**	148	165
	Total	44	156	200

* The group includes patients on gluten-containing and gluten-restricted diet, as well.

**Two patients in this group are IgA deficient

For anti-Gliadin IgG, clinical sensitivity was 55% (24/44) and clinical specificity was 94% (146/156) (see table below):

		Diagnosis		
		Celiac disease* (n=44)	Control group (n=156)	Total (n=200)
EL-Glia™ IgG	Positive	24	6	30
	Equivocal	5	4	9
	Negative	15	146	161
	Total	44	156	200

* The group includes patients on gluten-containing and gluten-restricted diet, as well

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

Not applicable.

4. Clinical cut-off:

Same as assay cut-off.

5. Expected values/Reference range:

Expected values in the normal population should be negative.

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.