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

I Background Information:

A 510(k) Number

K221813

B Applicant

Nova Biomedical Corporation

C Proprietary and Established Names

Nova Allegro UACR Assay, Nova Allegro Analyzer

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
CGX	Class II	21 CFR 862.1225 - Creatinine Test System	CH - Clinical Chemistry
JIQ	Class I	21 CFR 862.1645 - Urinary protein or albumin (nonquantitative) test system	CH - Clinical Chemistry
JQT	Class I	21 CFR 862.2400 - Densitometer/ scanner (integrating, reflectance, TLC, or radiochromatogram) for clinical use	CH - Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

New Device

B Measurand:

Urine Albumin and Creatinine

C Type of Test:

Immunoturbidimetric (Albumin)
Colorometric (Creatinine)

III Intended Use/Indications for Use:

A Intended Use(s):

The Nova Allegro urine albumin creatinine ratio (UACR) Assay is intended for the quantitative determination of albumin, creatinine, and the albumin/creatinine ratio (UACR) in human urine. The measurement of urine albumin, creatinine, and albumin/creatinine ratio aids in the early diagnosis of nephropathy.

The Nova Allegro Analyzer is intended for in vitro diagnostic use in clinical laboratory and near-patient testing (point-of-care) settings for the quantitative determination of Nova Allegro Assays using Nova Allegro Test Cartridges.

B Indication(s) for Use:

See Intended Use above.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

The UACR Assay is used with the Nova Allegro Analyzer.

IV Device/System Characteristics:

A Device Description:

Nova Allegro UACR Assay

The Allegro UACR Assay measures Albumin and Creatinine in human urine. The UACR is calculated by the Allegro Analyzer. The Allegro UACR Test Cartridge contains all of the reagents for measuring albumin and creatinine.

Cartridge Description:
The main components of the Test Cartridge are the Capillary, the reaction chamber, and the barcode label.

The Allegro UACR test cartridge contains 3 reagent solutions as described in the following table:

Sample Diluent	0.2% w/v 3,5-dinitrobenzoic acid, 4% polyethylene glycol in 100 mM tris buffer, pH 7.5, 150 mM NaCl, with 2.5% other ingredients (stabilizers, preservatives) (350 µL per cartridge)
Albumin Reagent	10 – 15 mg/mL delipidated goat IgG fraction containing anti-human albumin in 50 mM Tris, 150 mM NaCl with 2.2% other ingredients (stabilizers, preservatives) (105 µL per cartridge)
Creatinine Reagent	15% potassium hydroxide, 0.5M dipotassium hydrogen phosphate, with 2% other ingredients (stabilizers, preservatives). (120 µL per cartridge).

Nova Allegro Analyzer
The Nova Allegro Analyzer is a compact, point-of-care analyzer. All tests are measured with disposable, ready-to-use cartridges. The analyzer supports multiple wavelengths that are used to measure the assay of interest. The analyzer consists of the following key systems/components that the user interacts with:

- Two analytical bays where the single use test cartridges are analyzed
- Color Touchscreen Display
- Barcode Scanner
- Printer
- Data Export Options
- Ethernet Connection
- USB Port

B Principle of Operation:

The Allegro UACR Test Cartridge contains all reagents required for measuring albumin and creatinine. When a human urine sample is introduced into a Test Cartridge by a capillary, albumin present in a urine sample reacts with antibody specific against human albumin and antibody-albumin complex is formed resulting in turbidity increase measured by absorbance at

405 nm. The amount of complex is in direct proportion to the amount of albumin in the sample. The albumin is then quantified using a calibration curve.

The Benedict/Behre chemistry is the basis for the creatinine assay. 3,5-dinitrobenzoic acid at high pH reacts with creatinine to form a colored complex at 530 nm. The colored complex is in direct proportion to the amount of creatinine in the sample which is determined from a stored calibration curve. The albumin to creatinine ratio, UACR, is then calculated and displayed on the Nova Allegro Analyzer.

C Instrument Description Information:

1. Instrument Name:

   Nova Allegro Analyzer

2. Specimen Identification:

   Specimen identification can be done by scanning a patient identification bar code or by manual entry.

3. Specimen Sampling and Handling:

   A single-use, disposable capillary sample collection device is used to obtain and load the sample. The capillary sample collection device containing the sample is loaded onto the test cartridge, then the test cartridge is placed into the sample bay within 1 minute of sample collection and the start icon is pressed to start analysis.

4. Calibration:

   The reagent test cartridge includes a scannable 2D barcode containing the stored calibration curve.

5. Quality Control:

   External Quality Control (QC) material is used to monitor the performance of the analyzer and the test cartridges used with the analyzer and to verify the analyzer is performing to specifications. QC recommendations are found in the labeling.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Afinion ACR and Afinion ACR Control, Afinion 2 analyzer

B Predicate 510(k) Number(s):
K072409, K171650

C Comparison with Predicate(s):

Device & Predicate Device(s):	K221813	Predicate
Device Trade Name	Nova Allegro UACR Assay	Afinion ACR and Afinion ACR Control
General Device Characteristic Similarities		
Intended Use/Indications For Use	For the quantitative determination of albumin, creatinine, and the albumin/creatinine ratio (UACR) in human urine. The measurement of urine albumin, creatinine, and albumin/creatinine ratio aids in the early diagnosis of nephropathy.	Same
Matrix	Urine	Same
Intended Users	Professional	Same
General Device Characteristic Differences		
Measurement Range - Albumin	5-300 mg/L	5- 200 mg/L
Measurement Range - Creatinine	15-500 mg/dL	16.4 – 339.9 mg/dL
Measurement Range - ACR	1-2000 mg/g	1 – 1250 mg/g
Test Principle – Albumin	Immunoturbidimetric measurement	Immunometric membrane flow-through principle
Test Principle – Creatinine	Benedict/Behre (non enzymatic) alkaline colorimetric	Enzyme Colorimetric
Sample Volume	25 µL	3.5 µL

Device & Predicate Device(s):	K221813	K171650
Device Trade Name	Nova Allegro Analyzer	Afinion 2 analyzer
General Device Characteristic Similarities		

Intended Use/Indications For Use	For in vitro diagnostic use for the quantitative determination of assays using test cartridges.	Same
General Device Characteristic Differences		
Cartridge Interface	Physical processing of cartridge (locking, docking, foil penetration, mixing assay)	Physical processing of cartridge (locking, docking, splitting, foil penetration, merging)
Peripheral units	Barcode reader and printer integrated into analyzer	A barcode reader and printer can be connected

VI Standards/Guidance Documents Referenced:

- CLSI Guideline "Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guidelines – Third Edition", CLSI EP5-A3
- CLSI Guideline, "Evaluation of the Linearity of Quantitative Measurement Procedures", EP06-Ed2
- Interference Testing in Clinical Chemistry; Approved Guideline -Third Edition: CLSI EP07-A3
- CLSI "Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition", CLSI EP09c

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Measures of Imprecision for the Nova Allegro UACR Assay were assessed using methods described in CLSI "Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guidelines – Second Edition", CLSI EP5-A3 as guidance.

A total of 8 Nova Biomedical Allegro Analyzers (2 per site) and 3 lots of Allegro UACR Test Cartridges were used in the study.

Precision studies were conducted at four different sites with 12 operators. At each of the 4 sites, 3 operators ran 2 levels of quality control material (UACR Control Solution Level 1 and UACR Control Solution Level 2) and 3 levels of urine specimens four times a day, twice in the morning and twice in the afternoon, for 20 days to obtain 80 data points per level.

Repeatability

Urine Samples		Site 1		Site 2		Site 3		Site 4		Overall		
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	% CV
1	ALB	125	4.4	125	4.5	124	5.0	120	4.0	123	4.5	3.6%
	CRE	62	2.2	61	2.4	59	2.9	59	2.2	60	2.4	4.0%
	ACR	202	4.2	203	5.5	210	5.0	202	5.5	204	5.1	2.5%
2	ALB	134	5.1	131	5.9	131	4.8	130	5.1	131	5.3	4.0%
	CRE	460	13.2	453	15.6	440	12.3	454	14.0	452	13.8	3.1%
	ACR	29	0.6	29	0.8	30	0.8	29	0.7	29	0.7	2.5%
3	ALB	251	8.4	248	8.6	251	8.0	237	10.5	247	8.9	3.6%
	CRE	82	2.3	81	2.2	79	3.0	79	3.2	80	2.7	3.4%
	ACR	308	6.1	308	6.6	316	7.9	302	9.1	308	7.5	2.4%

Between Run/Operator

Urine Samples		Site 1		Site 2		Site 3		Site 4		Overall		
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	% CV
1	ALB	125	2.8	125	3.9	124	2.1	120	3.7	123	3.2	2.6%
	CRE	62	1.3	61	1.8	59	0.8	59	1.4	60	1.4	2.3%
	ACR	202	1.4	203	0.0	210	2.6	202	3.8	204	2.4	1.2%
2	ALB	134	0.9	131	0.0	131	0.0	130	0.0	131	0.5	0.3%
	CRE	460	0.0	453	2.8	440	3.9	454	0.0	452	2.4	0.5%
	ACR	29	0.2	29	0.0	30	0.0	29	0.3	29	0.2	0.6%
3	ALB	251	6.0	248	3.0	251	3.2	237	0.0	247	3.7	1.5%
	CRE	82	1.5	81	2.2	79	2.9	79	1.8	80	2.2	2.7%
	ACR	308	0.0	308	5.9	316	9.7	302	0.0	308	5.7	1.8%

Between Day/Lot

Urine Samples		Site 1		Site 2		Site 3		Site 4		Overall		
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	% CV
1	ALB	125	1.4	125	1.7	124	1.4	120	1.8	123	1.6	1.3%
	CRE	62	0.7	61	1.0	59	0.0	59	0.0	60	0.6	1.0%
	ACR	202	0.0	203	0.0	210	1.9	202	1.0	204	1.1	0.5%
2	ALB	134	0.0	131	2.1	131	2.7	130	0.0	131	1.7	1.3%
	CRE	460	1.8	454	3.9	440	0.9	454	0.0	452	2.2	0.5%
	ACR	29	0.0	29	0.2	30	0.5	29	0.2	29	0.3	1.1%
3	ALB	251	0.0	248	1.5	251	0.0	237	1.9	247	1.2	0.5%
	CRE	82	0.0	81	0.0	79	0.0	79	0.7	80	0.4	0.5%
	ACR	308	0.0	308	1.0	316	1.0	302	0.0	308	0.7	0.2%

Within Laboratory

Urine Samples		Site 1		Site 2		Site 3		Site 4		Overall		
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	% CV
1	ALB	125	5.6	125	5.7	124	5.5	120	5.8	123	5.6	4.6%
	CRE	62	2.9	61	2.8	59	2.9	59	2.9	60	2.9	4.8%
	ACR	202	4.5	203	5.5	210	7.0	202	6.7	204	6.0	2.9%
2	ALB	134	5.0	131	6.3	131	5.4	130	5.0	131	5.4	4.1%
	CRE	460	13.6	453	18.3	440	13.6	454	14.7	452	15.2	3.4%
	ACR	29	0.7	29	0.9	30	1.1	29	0.7	29	0.9	3.0%
3	ALB	251	11.2	248	9.2	251	9.9	237	11.2	247	10.4	4.2%
	CRE	82	3.1	81	3.0	79	3.9	79	4.1	80	3.6	4.5%
	ACR	308	7.0	308	8.4	316	14.1	302	8.7	308	9.9	3.2%

Between-Site

Control Samples		Cartridge Lot 1		Cartridge Lot 2		Cartridge Lot 3		Overall		
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	% CV
L1	ALB	47	0.4	47	0.5	46	0.4	47	0.5	1.0%
	CRE	85	0.0	85	1.4	85	0.4	85	0.8	1.0%
	ACR	55	0.7	55	0.5	55	0.7	55	0.6	1.2%
L2	ALB	153	0.0	153	3.7	151	1.9	152	2.4	1.6%
	CRE	216	1.9	216	2.40	215	1.6	216	2.0	0.9%
	ACR	71	0.5	71	0.9	70	1.0	71	0.8	1.2%

2. Linearity:

A study was performed to validate the Nova Allegro UACR Assay linearity and reportable ranges described below:

	Range Evaluated	Levels	Slope	Intercept	r	Reportable Range
Albumin	0 – 362 mg/L	9	0.994	2.52	0.997	5 – 300 mg/L
Creatinine	0 – 507 mg/dL	9	0.997	4.73	0.999	15 – 500 mg/dL

If the albumin or creatinine value is outside the reportable range, no ACR test result will be reported.

Hook Effect Study
A study was performed to validate and determine at what concentration level the Nova Allegro UACR Assay exhibits a characteristic "Hook Effect" when measuring albumin. No "Hook effect" was observed with albumin up to 50,000 mg/L.

3. Analytical Specificity/Interference:

A study was performed to test for potentially interfering substances with the Nova Allegro

UACR Assay according to Interference Testing in Clinical Chemistry; Approved Guideline - Third Edition: CLSI EP07-A3.

Ten (10) replicate measurements were made on both prepared specimens containing each potential interfering substance and control specimens containing no interfering substance. The mean albumin and creatinine concentration of the ten (10) replicates for both the test and control pools for each of the interfering substances, and percent difference between the mean test value and the mean control value were calculated. Acceptance criteria for interfering substance is ≤10%.

Albumin concentrations were targeted to approximately 20 mg/L and 220 mg/L and creatinine, targeted to approximately 56 mg/dL and 450 mg/dL with the interferents as described in the table below.

Substances	**Highest tested concentration without significant interference**
Acetaminophen	20 mg/dL
Acetaminophen – glucuronide	1050 mg/dL
Acetone	80 mg/dL
Albumin	1000 mg/dL
Ammonium Chloride	100 mg/dL
β-Hydroxybutyric Acid	590 mg/dL
β-Microglobulin	2 mg/dL
Blood	50 Ery/µL
Calcium Chloride	180 mg/dL
Citric Acid	75 mg/dL
Creatine	1000 mg/dL
Creatinine	620 mg/dL
Digoxin	0.03 mg/dL
Ethyl Acetoacetate	84 mg/dL
Fructose	100 mg/dL
Galactose	80 mg/dL
Glucose	4500 mg/dL
Glybenclamide	1.48 mg/dL
Glyburide	1.48 mg/dL
Glycine	450 mg/dL
Hemoglobin	10 mg/dL
Ibuprofen	200 mg/dL
IgA	25 mg/dL
IgG	2000 mg/dL
Insulin	500 µU/mL
Lactose	10 mg/dL
Lithium Acetoacetate	250 mg/dL
Metformin	400 mg/dL

Myoglobin	2 mg/dL
Sodium Acetate	2.25 mg/dL
Sodium Bicarbonate	1500 mg/dL
Sodium Nitrate	10 mg/dL
Sodium Nitrite	10 mg/dL
Sodium Phosphate	500 mg/dL
Theophylline	100 mg/dL
Transferin	200 mg/dL
Trichlormethiazide	2 mg/dL
Urea	3000 mg/dL
Uric acid	150 mg/dL
Urobilinogen	20 mg/dL

Significant interference (>10%) was observed at the following concentration levels.

Interfering Substances	**Targeted Tested Concentration**	**Highest tested concentration that does not cause interference**	
		Albumin	**Creatinine**
Oxalic Acid	70 mg/dL	40 mg/dL	70 mg/dL
Conjugated Bilirubin	20 mg/dL	4.2 mg/dL	20 mg/dL
Riboflavin – Vitamin B2	10 mg/dL	2.5 mg/dL	10 mg/dL
Sodium chloride	5500 mg/dL	2000 mg/dL	5500 mg/dL
Potassium chloride	1500 mg/dL	1200 mg/dL	1500 mg/dL
Leukocytes	2500/µL	2500/µL	1250/µL
Ascorbic acid	300 mg/dL	300 mg/dL	100 mg/dL

Effects of urine pH: Five (5) fresh urine specimens were pooled and divided into two sample pools. Both samples were then divided into 7 aliquots and the pH of the aliquots were adjusted to have pH values of 4.0, 4.3, 4.5, 5.0, 6.0, 7.0, 8.0, 9.0, and 10.0. The results demonstrated that changes in pH ranging from 4.3 to 9.0 do not interfere with the results of albumin and creatinine.

Effects of urine specific gravity:
Fresh urine samples with low specific gravity (≤1.005) were pooled and divided into six (6) aliquots. Five (5) aliquots were adjusted to specific gravity of 1.01, 1.02, 1.03, 1.04, and 1.05 and spiked with a combination of albumin and creatinine. The results demonstrated that changes in specific gravity ranging from 1.003 to 1.048 do not interfere with the results of albumin and creatinine.

4. Assay Reportable Range:

   Albumin: 5 – 300 mg/L
   Creatinine: 15 – 500 mg/dL

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

   Albumin calibration standards are traceable to BCR/CRM 470 IFCC reference materials and creatinine calibration standards are traceable to NIST SRM 914.

6. Detection Limit:

   Studies for detection limit were conducted following CLSI EP 17-A2.

   Three (3) Nova Allegro Analyzers and three (3) lots of Nova Allegro UACR Test Cartridges were used in the study. Sixty (60) replicate measurements were tested to determine the limit of blank (LOB) and the limit of detection (LOD) and thirty six (36) replicate measurements were tested to determine the limit of quantitation (LOQ). Nova Allegro Urine Calibrator base matrix was used for determining the LOB. Urine samples spiked with albumin and creatinine were used for determining the LOD and LOQ. The total error goal for LOQ were defined as ≤2 mg/L for albumin and ≤3 mg/dL for creatinine.

Analyte	LOB	LOD	LOQ
Albumin	1.9 mg/L	2.1 mg/L	3.5 mg/L
Creatinine	1.4 mg/dL	2.6 mg/dL	4.4 mg/dL

7. Assay Cut-Off:

   Not applicable

8. Accuracy (Instrument):

   See Section VII.B.1 Method Comparison with Predicate Device.

9. Carry over:

   The Nova Allegro UACR Test Cartridges are fully self-contained test cartridges. The sponsor describes that no contact is made between the system instrumentation and the reagents or test sample.

B Comparison Studies:

1. Method Comparison with Predicate Device:

   Point-of-Care (POC) Method Comparison studies on fresh urine specimens were conducted within four (4) different POC sites using methods described in CLSI "Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition", CLSI EP09c.

   A total of eight (8) Nova Allegro Analyzers (2 per site) and three (3) lots of Allegro UACR Test Cartridges were used in the study. At each site, a minimum of 3 operators conducted the

testing over 20 days using freshly collected urine specimens. A small percentage of test specimens were altered to cover the analytical measurement range of the Nova Allegro UACR Assay for albumin and creatinine.

Specimens run on the Nova Allegro Analyzers using the Nova Allegro UACR Assay were compared to the Siemens Dimension EXL 200 Integrated Chemistry System utilizing Dimension® Flex® reagent cartridge MALB, Dimension calibrator cartridge MALB CAL, Dimension Flex Reagent Cartridge CRE2, and Dimension CHEM 1 CAL as the comparator method. Least Squares Linear Regression Analyses were performed with the following results for all 4 sites combined:

Analyte	Total No. of Samples	Sample Range	No. of Samples within AMR	Slope	Intercept
Albumin (mg/L)	659	5 – 300	535	0.98	0.86
Creatinine (mg/dL)	659	16 – 498	653	0.95	3.78
UACR (mg/g)	659	1 – 1592	531	0.99	1.10

2. Matrix Comparison:

   Not Applicable

C Clinical Studies:

1. Clinical Sensitivity:

   Not applicable

2. Clinical Specificity:

   Not applicable

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

   Not applicable.

D Clinical Cut-Off:

Not Applicable

E Expected Values/Reference Range:

	Albumin (Timed Collection)	Creatinine	UACR (Spot Collection)
Normal	< 20 µg/min	34 – 147 mg/dL	< 30 mg/g
Microalbuminuria	20 – 200 µg/min		30 – 300 mg/g
Clinical Albuminuria	>200 µg/min		> 300 mg/g

F Other Supportive Instrument Performance Characteristics Data:

Electrical safety and electromagnetic compatibility (EMC) testing had been performed and the system was found to be compliant.

Software and cybersecurity documentation was reviewed and found to be acceptable.

The Nova Allegro UACR Assay on the Nova Allegro Analyzer is not impacted by altitude up to 12,000 feet/3650 meters.

VIII Proposed Labeling:

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

IX Conclusion:

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