

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

A. 510(k) Number:

k130080

B. Purpose for Submission:

New device

C. Measurand:

Creatine Kinase MB (CK-MB),
C-Reactive Protein (CRP)

D. Type of Test:

Quantitative, enzymatic
Quantitative, immunoturbidimetric

E. Applicant:

Medica Corporation

F. Proprietary and Established Names:

EasyRA Creatine Kinase-MB (CK-MB) Reagent
EasyRA C-Reactive Protein (CRP) Reagent,
EasyCAL C-Reactive Protein (CRP) Calibrator Kit
EasyQC C-Reactive Protein (CRP) Quality Control Material

G. Regulatory Information:

1. Regulation section:

Product Code	Classification	Regulation Section	Panel
JHY	Class II	21 CFR§862.1215 Creatine phosphokinase/creatine kinase or isozymes test system	Clinical Chemistry (75)
DCK	Class II	21 CFR§866.5270 C-reactive protein, Antigen, Antiserum and Control	Immunology (82)
JIT	Class II	21 CFR§862.1150 Calibrator	Clinical Chemistry (75)

JJX	Class I, reserved	21 CFR§862.1660 Quality control material	Clinical Chemistry (75)
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H. Intended Use:

1. Intended use(s):

See indications for use below.

2. Indication(s) for use:

EasyRA Creatine Kinase-MB (CK-MB) Reagent

The EasyRA Creatine Kinase-MB (CK-MB) Reagent is intended for the quantitative determination of CK-MB activity in human serum and plasma, using the MEDICA "EasyRA Chemistry Analyzer" in clinical laboratories.

Measurements of CK-MB activity are used in the diagnosis and treatment of myocardial infarction and muscle diseases such as progressive, Duchenne-type muscular dystrophy.

EasyRA C-Reactive Protein (CRP) Reagent,

Medica's EasyRA C-Reactive Protein (CRP) Reagent is intended for use in the quantitative in-vitro diagnostic determination of C-reactive protein in human serum or plasma using the EasyRA clinical chemistry analyzer. Measurements of C-reactive protein aids in evaluation of the amount of injury to body tissues.

EasyCal C-Reactive Protein (CRP) Calibrator Kit

The EasyCAL C-Reactive Protein Calibrator (CRP) Kit is used for calibrating the CRP assay on the EasyRA clinical chemistry analyzer when used in conjunction with EasyRA CRP Reagent. The CRP calibrators are used to establish points of reference that are used in the determination of values in the measurement of CRP in human serum and plasma.

EasyQC C-Reactive Protein (CRP) Quality Control Material

The EasyQC C-Reactive Protein (CRP) Quality Control Materials are intended for use as quality control material for the CRP turbidimetric assay, using the EasyRA CRP Reagent and calibrator kit on the EasyRA clinical chemistry analyzer.

3. Special conditions for use statement(s):

For *in vitro* diagnostic use only

For prescription use only.

4. Special instrument requirements:

EasyRA Chemistry Analyzer

I. Device Description:

The EasyRA Creatine Kinase-MB (CK-MB) Reagent kit is ready to use in vitro diagnostic reagent supplied in wedges. Each wedge contains the following two reagent bottles:

CK-MB Buffer Reagent (R1):

Imidazole Buffer (pH 6.3) 50 mmol/L, Glucose 20 mmol/L, N-Acetyl-L-Cysteine 20 mmol/L, Magnesium Acetate 10.0 mmol/L, NADP 2.0 mmol/L, EDTA 2.0 mmol/L, Hexokinase 5.0 kU/L, LDH 1.5 kU/L, Sodium Azide < 0.1%, Monoclonal antibodies (mouse) against human CK-M, inhibiting capacity.

CK-MB Substrate Reagent (R2):

Imidazole Buffer (pH 9.0) 50 mmol/L; Creatine phosphate 150 mmol/L; ADP 10 mmol/L; AMP 20 mmol/L; Diadenosine pentaphosphate 50 µmol/L, Glucose-6-PDH 20 kU/L, Sodium Azide < 0.1%

EasyRA C-Reactive Protein (CRP) Reagent kit is ready to use in vitro diagnostic reagent supplied in wedges. Each wedge contains the following two reagent bottles:

CRP Reagent (R1):

Glycine Buffer Reagent (170 mM)
Sodium Chloride (100 mM)
Sodium EDTA disodium SALT dihydrate (50 mM)
Bovine serum albumin (1 %)
Sodium Azide < 0.1%

CRP Reagent (R2):

Latex particles coated with antibody to CRP
Sodium Azide < 0.1%

EasyCAL C-Reactive Protein (CRP) Calibrator Kit contains 6 vials each containing 1 ml of liquid stable calibrator based on human source material with different levels of CRP (0.0 mg/L, 2.5 mg/L, 10 mg/L, 20 mg/L, 80 mg/L, and 160 mg/L) for the EasyRA CRP assay. The six pre-assayed levels are used to establish the calibration curve using a spline curve fitting routine that is a prerequisite in determining the CRP concentration in unknown samples.

EasyQC C-Reactive Protein (CRP) Quality Control Material is a human based control consisting of three levels each containing different concentrations of CRP. The concentration of each level is assigned with every new lot of material. The EasyQC material is used to establish the accuracy and precision of the CRP assay on the EasyRA analyzer.

All human source materials were tested by FDA approved methods and found to be negative for the presence of HBsAg and antibody to HIV1/HIV2, and HCV.

J. Substantial Equivalence Information:

1. Predicate device name(s):
Creatine Kinase MB determination (UV)
Randox C-reactive Protein
Randox C-Reactive Protein Calibrator
Randox Liquid CRP Controls
2. Predicate 510(k) number(s):
k942958
k001009
k022682
k040351
3. Comparison with predicate:

CK-MB:

Similarities and Differences		
Item	Candidate Device The EasyRA Creatine Kinase-MB Reagent	Predicate Device Creatine Kinase MB Determination (UV) (k942958)
Intended Use/Indications for Use	For the in vitro quantitative determination of CK-MB in human serum and plasma on automated chemistry analyzers.	Same
Specimen	Serum and plasma	Same
Methodology	Enzymatic	Same
Wavelength	340 nm	Same
Antibody	Monoclonal mouse antibody against human CK-MB	Same
Calibration	The enzymatic method does not use a calibrator	Same
Storage	2-8°C	Same
Measuring range	5-100 U/L	Up to 175 U/L

CRP:

Similarities and Differences		
Item	Candidate Device <i>EasyRA C-Reactive Protein (CRP) Reagent</i>	Predicate Device Randox C-reactive Protein (k001009)
Intended Use/Indications for Use	For the <i>in vitro</i> quantitative determination of C-reactive protein (CRP) in human serum and plasma on automated chemistry analyzers.	Same
Specimen	Serum and plasma	Same
Storage	2-8°C	Same
Measuring range	0.5-160 mg/L	0.3-161 mg/L
Wavelength	600 nm	570 nm

Similarities and Differences		
Item	Candidate Device <i>EasyRA C-Reactive Protein (CRP) Calibrator Kit</i>	Predicate Device Randox CRP Calibrator (k022682)
Intended Use	For use in the calibration of the C-Reactive Protein (CRP) assay	Same
Levels	6 levels (1 ml each)	Same
Open Vial Stability	3 months at 2-8°C.	Same

Similarities and Differences		
Item	Candidate Device <i>EasyRA, C-Reactive Protein (CRP) Quality Control Material</i>	Predicate Device Randox C-reactive Protein QC Material (k040351)
Intended Use	For use as quality control for the C-Reactive Protein (CRP) assay	Same

Levels	3 levels (1 ml each)	Same
Open Vial Stability	3 months at 2-8°C.	Same

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

- CLSI EP5-A2: *Evaluation of Precision Performance of Clinical Chemistry Devices*
- CLSI EP9-A2: *Method Comparison and Bias Estimation Using Patient Samples*
- CLSI EP6-A: *Evaluation of the Linearity of Quantitative Analytical Methods: A Statistical Approach*
- CLSI EP17-A: *Protocols for Determination of Limits of Detection and Limits of Quantitation*
- CLSI EP7-A2: *Interference Testing in Clinical Chemistry*

L. Test Principle:

The EasyRA Creatine Kinase-MB (CK-MB) Reagent is provided in a ready-to-use plastic wedge. An antibody inhibits M subunits of CK-MM and CK-MB. The activity of the non-inhibited CK-B is then determined. The CK-B catalyzes the reversible phosphorylation of ADP, in the presence of creatine phosphate, to form ATP and creatine. Hexokinase catalyzes the phosphorylation of glucose by the ATP formed, to produce ADP and glucose-6-phosphate (G₆P). The G₆P is oxidized to 6-phosphogluconate with the concomitant production of NADH. The rate of NADH formation, measured at 340nm, is directly proportional to CK-B activity, which is half the CK-MB activity.

The EasyRA C-Reactive Protein (CRP) Reagent is a turbidimetric immunoassay using two reagents sequentially. The serum or plasma is first reacted to a buffer (Reagent 1) and then with Latex particles coated with antibody to CRP (Reagent 2). The formation of the antibody-antigen complex during the reaction results in an increase in turbidity, which is measured as the amount of light absorbed at 600nm. The increase in absorbance is related to the concentration of the CRP in serum or plasma sample based on a pre-established calibration curve. The CRP concentration of the sample can be determined using a spline curve fitting routine.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

The sponsor carried out precision studies for CK-MB and CRP assays according to the recommendations in the CLSI EP5-A2 guideline. Duplicate measurements of each of two levels of serum QC material were tested on three instruments, twice a day for 20 days (n=80 per instrument) by one operator

using one reagent lot. Both within-run and total precision for CK-MB and CRP were determined from these data and results are presented in the tables below.

CK-MB QC Level (U/L)	Within run imprecision		Total Imprecision	
	SD (U/L)	CV (%)	SD (U/L)	CV (%)
14	0.56	3.94	0.79	5.54
30	0.72	2.41	0.88	2.96
94	0.85	0.90	1.72	1.83

CRP QC Level (mg/L)	Within run imprecision		Total Imprecision	
	SD (mg/L)	CV (%)	SD (mg/L)	CV (%)
2.7	0.06	2.33	0.09	3.32
22.8	0.34	1.50	0.65	2.85
135.0	1.82	1.35	3.29	2.44

b. Linearity/assay reportable range:

CK-MB assay:

The sponsor conducted a linearity study based on the recommendations in CLSI EP6-P2. A concentrated CK-MB stock solution was used to spike a serum sample to a CK-MB concentration just above 100U/L, the upper reportable range. This solution was volumetrically serially diluted with saline solution to make six more solutions of known CK-MB concentrations. The total of seven solutions with values ranging from 5 to 100.5 U/L were assayed in duplicate and the mean values were obtained and compared to the expected values to determine percent recovery. The results met the sponsors' acceptance recovery criteria of $\leq 10\%$.

The EasyRA CK-MB Reagent is linear from 5 U/L to 100 U/L with slope= 1.009 ± 0.0159 , Intercept = -0.2291 ± 0.9533 and $r^2 = 0.9994$

An extended linearity study was carried out to support dilution extension of the measuring range to up to 500 U/L. A pooled serum sample was spiked with different amounts of CK-MB to make 5 samples ranging from 128-444 U/L. The samples were then split to be diluted 1:4 manually (volumetrically) and by the instrument re-run function (1/5 volume - sample and 4/5 volume - diluent). The results of the study showed that the sponsor validated their dilution scheme and their claimed 100-500 U/L extended measuring range with 99.2% average CK-MB recovery.

CRP assay:

The sponsor conducted a linearity study based on CLSI EP6-P2. A concentrated CRP stock solution was used to spike a pooled human serum sample to a CRP

concentration just above 160 mg/L, the upper reportable range. The high pool solution was serially diluted with pooled human serum which was also used as the low CRP level in the study was used to volumetrically to make seven more CRP concentrations with values ranging from 0.27 to 176.77. The total of nine CRP concentrations covered the sponsor's claimed linear range of 0.5 to 160 mg/L. Each dilution sample was assayed in duplicate and the mean values were obtained and compared to the expected values to determine percent recovery. The results met the sponsor's acceptance recovery criteria of $\leq 10\%$.

The EasyRA CRP Reagent is linear from 0.5 mg/L to 160 mg/L with slope = 1.0086 ± 0.0101 , Intercept = 0.0526 ± 0.9216 and $r^2 = 0.9984$

An extended linearity study was carried out to support dilution extension of the measuring range to up to 480 U/L. A pooled serum sample was spiked with different amounts of CRP to make 5 samples ranging from 160-480 U/L. The samples were then split to be diluted 1:2 manually (volumetrically) and by the instrument re-run function (1/3 volume - sample and 2/3 volume - diluent). The results of the study showed that the sponsor validated their dilution scheme and their claimed 160-480U/L extended measuring range with 101.4% average CRP recovery.

To test for High Dose "Hook Effect", a CRP concentrate solution (2800mg/L) was combined with pooled serum into 11 different concentration samples to form a series of CRP spiked samples. The CRP testing was performed in triplicate on a single EasyRA Analyzer. The sponsor's study demonstrated that no Hook Effect was observed up to 2800 mg/L.

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

CK-MB:

The sponsor's protocols and acceptance criteria were reviewed and found acceptable and support the following claims: On-board stability for the reagent is 31 days at 2-8°C.

CRP:

The CRP calibrators are traceable to European Reference Material ERM-DA472/IFCC

The sponsor conducted a real time stability study to validate the claimed shelf-life of the reagent, controls and calibrators. The study results met the sponsor's predetermined acceptance criteria of $\pm 10\%$ and confirmed claimed stability of the reagent, controls and calibrators. The sponsor conducted on board stability studies on the MEDICA Easy RA Clinical Chemistry Analyzer. The sponsor's protocols and acceptance criteria were reviewed and found acceptable and support the following claims:

On-board stability for the reagent is 61 days at 2-8°C. Open calibrators are

stable for 3 months at 2-8°C. The components of the QC material are stable for 3 months after opening at 2-8°C.

Quality Control material is obtained by the sponsor from a commercial vendor. Each new lot of the QC material is tested in triplicate on the EasyRA for each QC level using a released reagent and calibrator lot. The acceptance criteria for QC level 1 are $\pm 20\%$, and $\pm 10\%$ for QC levels 2 and 3 to transfer the values to the EasyRA.

d. *Detection limit:*

CK-MB assay:

The sponsor conducted limit of detection studies following the recommendations in CLSI EP-17A.

LoB: To determine the limit of the blank (LOB), deionized water was analyzed 20 times on three EasyRA analyzers and the results were rank ordered to determine the LoB. The LoB was calculated to be 1.7 U/L.

LoD: A serum sample was prepared with a CK-MB value above the LoB (1.7U/L) and analyzed 20 times on three EasyRA analyzers to determine the typical precision of the low concentration sample. The average standard deviation of the low concentration sample was subsequently used, in conjunction with the previously calculated LoB to determine the LoD.

The LoD is equal to the LoB plus 1.653 times the average standard deviation of the Low Concentration Sample ($\text{LoD} = 1.7 + 1.653 \times 0.74$).

The LoD is calculated to be 2.9 U/L.

LoQ: The LoQ was determined to be the lowest concentration sample that had a %CV of $\leq 20\%$. The studies yielded the LoQ of 4.98 U/L. The sponsor's claimed measuring range is 5-100U/L.

CRP assay:

The sponsor conducted limit of detection studies following the recommendations in CLSI EP-17A.

LoB: To determine the limit of the blank (LOB), deionized water was analyzed 20 times on three EasyRA analyzers and the results were rank ordered to determine the LOB. The LOB was calculated to be 0.04 mg/L.

LoD: The LoD was determined per CLSI EP-17A. A serum sample was prepared with a CRP value above the LoB (1.7U/L) and analyzed 20 times on three EasyRA analyzers to determine the typical precision of the low concentration sample. The average standard deviation of the

low concentration sample was subsequently used, in conjunction with the previously calculated LoB to determine the LoD.

The LoD is equal to the LoB plus 1.653 times the average standard deviation of the Low Concentration Sample ($LoD = 0.04 + 1.653 \times 0.072$).

The LOD is calculated to be 0.16 mg/L.

LoQ: The LoQ was determined to be the lowest concentration sample that had a %CV of $\leq 10\%$. The studies yielded the LoQ of 0.5 mg/L. The sponsor's claimed measuring range is 0.5-160mg/L.

e. *Analytical specificity:*

The sponsor evaluated common interfering substances on the EasyRA Clinical Chemistry Analyzer according to the recommendations in CLSI EP7-A2 for their candidate CK-MB and CRP assays. Three different CK-MB (35, 90, and 240 U/L) and CRP (3, 7, and 40 mg/L) levels were spiked with various concentrations of potentially interfering compounds and triplicate measurements were compared to the samples without added interferent. The samples were tested using one instrument and one reagent lot. The following substances at the concentrations tested demonstrated less than 10% interference when comparing the spiked sample to the control sample. The sponsor claims that the following substances (except hemoglobin for CK-MB) did not interfere with the performance of the device. The labeling includes the limitation that hemoglobin interferes with the CK-MB assay and hemolyzed samples should not be used.

CK-MB Interference:	Concentration
Hemoglobin	Hemoglobin interferes, even in minimal concentrations. Do not use hemolyzed samples.
Unconjugated Bilirubin	28 mg/dL
Conjugated Bilirubin	25 mg/dL
Triglyceride (Intralipid)	775 mg/dL
Ascorbic acid	30 mg/dL

CRP Interference:	Concentration
Hemoglobin	1000 mg/dL
Unconjugated Bilirubin	30.1 mg/dL
Conjugated Bilirubin	27 mg/dL
Triglyceride (Intralipid)	1750 mg/dL
Ascorbic acid	500 mg/dL
Rheumatoid Factor	1024 mg/dL

- f. *Assay cut-off:*
Not applicable

2. Comparison studies:

a. *Method comparison with predicate device:*

CK-MB assay:

Forty one serum samples were tested using Medica's CK-MB Reagent on the Medica EasyRA Analyzer (candidate method) and compared to the Stanbio CK-MB Reagent on the Stanbio Sirrus analyzer (predicate method) following CLSI Guideline EP9-A. The slope, intercept and correlation coefficient were calculated by linear regression. The samples ranged from 6 to 96 U/L. The resulting equation was $y=0.9869x + 0.4702$ with a correlation coefficient of 0.9961.

CRP assay:

Sixty seven serum samples were tested using Medica's CRP Reagent on the Medica EasyRA Analyzer (candidate method) and compared to the performance using Randox Laboratories CRP Reagent on the Hitachi 911 Analyzer (predicate method) following the recommendations in CLSI EP9-A. The slope, intercept and correlation coefficient were calculated by linear regression. The samples ranged from 0.90 to 153.2 mg/L. The resulting equation was $y=-1.0001x - 0.0175$ with a correlation coefficient of 0.9988.

b. *Matrix comparison:*

CK-MB assay:

Matrix effects were evaluated using fifty seven matched serum and lithium heparin plasma samples. The samples were tested using the Medica CK-MB Reagent on the Medica EasyRA Chemistry Analyzer. The serum results were used as reference to which the lithium heparin plasma results were compared. The slope, intercept and correlation coefficient were calculated by linear regression. The samples ranged from 6 to 93 U/L. The resulting equation was $y=0.9797x + 1.4523$ with a correlation coefficient of 0.9945

CRP assay:

The matrix effects was evaluated using forty five matched serum and lithium heparin plasma samples. The samples were tested using Medica CK-MB Reagent on the Medica EasyRA Chemistry Analyzer. The serum results were used as reference to which the plasma results were compared. The slope, intercept and correlation coefficient were calculated by linear regression. The samples ranged from 0.51 to 151 mg/L. The resulting equation was $y=1.0021x - 0.0067$ with a correlation coefficient of 0.9999.

3. Clinical studies:

- a. *Clinical Sensitivity:*
Not applicable.
- b. *Clinical specificity:*
Not applicable.
- 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:

CK-MB:

The reference range for CK-MB activity on the EasyRA Clinical Chemistry Analyzer is below 24 U/L. This range has been validated by the sponsor according to CLSI C28-A3c.^{1, 2,3}

CRP:

CRP concentration from a healthy individual should be < 5 mg/L.⁴ The sponsor states that each laboratory should establish its own expected values using this kit.

- 1. Klein G, Berger A, Bertholf R et al. Abstract: Multicenter Evaluation of Liquid Reagents for CK, CK-MB and LDH with Determination of Reference Intervals on Hitachi Systems. Clin Chem 2001; 47:Suppl. A30.
- 2. Thomas L, Müller M, Schumann G, Weidemann G et al. Consensus of DGKL and VDGH for interim reference intervals on enzymes in serum. J Lab Med 2005; 29:301-308.
- 3. D'Souza, J.P., Sine HE, Horvitz RA, et al., The significance of the MB isozymes in patients with acute cardiovascular disease with abnormal or borderline total CPK activity. Clin Biochem 1978; 11: 204-209.
- 4. Tietz Clinical Guide to Laboratory Tests, 4th Edition, 480-483 (2006).

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.