1. INTENDED USE
The kit has been designed for the quantitative determination of procalcitonin (PCT) in human serum, plasma and whole blood. The method can be used for samples over the range of 0-100 ng/ml. The test has to be performed on the Maglumi fully auto analyzer (including Maglumi 1000, Maglumi 2000, Maglumi 2000 plus).

2. SUMMARY AND EXPLANATION OF THE TEST
Procalcitonin (PCT) is the hormonally inactive propodeide of calcitonin with a molecular weight of 12.6 kDa. Because PCT is decomposed by proteolysis in individuals with a normal metabolism, PCT normally exists at undetectable levels (< 0.1 ng/ml) in healthy subjects. In severe infections due to bacteria, fungi and parasites as well as in sepsis, PCT serum titres may increase to over 500 ng/ml. Mononuclear blood cells are, among others, currently regarded as the site of procalcitonin synthesis under the conditions of a systemic inflammatory response (1)

Clinical evaluations in various specialised fields of medicine (2-5) have shown that PCT is an excellent parameter - for early diagnosis of generalised bacterial and mycotic infection and sepsis - to assess the degree of severity and prognosticate the outcome of systemic infection, sepsis, and multiple organ failure - for monitoring high-risk patients for the development of infections, e.g. after surgery or organ transplantation, under immunosuppression, or in patients with multiple trauma - for differential diagnosis between systemic infection and acute inflammatory disease - for differential diagnosis between bacterial and viral infection.

3. PRINCIPLE OF THE TEST
Sandwich immunoluminometric assay;
Use an anti-PCT monoclonal antibody to label ABEI, and use another monoclonal antibody to label FITC. Sample, Calibrator or Control, with ABEI Label, FITC Label and magnetic microbeads coated with anti-FITC are mixed thoroughly and incubated at 37°C for 1 time. Subsequently, the starter reagents are added and a flash chemiluminescent reaction is initiated. The light signal is measured by a photomultiplier as RLU within 3 seconds and is proportional to the concentration of PCT present in controls or samples.

4. KIT COMPONENTS

4.1 Material supplies

<table>
<thead>
<tr>
<th>Reagent Integral for 100 determinations</th>
<th>2.5ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nano magnetic microbeads: TRIS buffer, 1.2%(W/V), 0.2%NaNO₃, coated with sheep anti-FITC polyclonal antibody.</td>
<td></td>
</tr>
<tr>
<td>Calibrator: low</td>
<td>2.5ml</td>
</tr>
<tr>
<td>Calibrator: high</td>
<td>2.5ml</td>
</tr>
<tr>
<td>ABEI Label: anti-PCT monoclonal antibody labeled ABEI, contains BSA, 0.2%NaNO₃,</td>
<td>10.5ml</td>
</tr>
<tr>
<td>FITC Label: anti-PCT monoclonal antibody labeled FITC, contains BSA, 0.2%NaNO₃,</td>
<td>10.5ml</td>
</tr>
<tr>
<td>Diluent</td>
<td>25ml</td>
</tr>
</tbody>
</table>

All reagents are provided ready-to-use.

*Please prepare 0.9% sodium chloride solution in case of insufficient diluents.

4.2 Preparation of the Reagent Integral
Before the sealing is removed, gentle and careful horizontal shaking of the Reagent Integral is essential (avoid foam formation!). Remove the sealing and turn the small wheel of the magnetic microbeads compartment to and fro, until the colour of the suspension has changed into brown. Place the integral into the reagent area and let it stand there for 30 mins. During this time, the magnetic microbeads are automatically agitated and completely
Do not interchange nano magnetic microbeads from different reagents.  

4.3 Storage of the Reagents Integral
- Seal, Stored at 2-8°C until the expiry date.
- Opened, Stable for 4 weeks. After this period, it is still possible to keep on using the Reagent Integral provided that the controls are found within the expected ranges.
- Keep upright for storage.
- Keep away from direct sunlight.

5. Origin of Calibrators
Calibrators in the Reagent Kit are from RANDOX. Biological root: synthetic materials, processed by SDS PAGE purification, with a purity ≥95%. No HBsAg, anti-HCV, and anti-HIV is found.

6. Calibration
6.1 2 point recalibration
Via the measurement of calibrators, the predefined master curve is adjusted (recalibrated) to a new, instrument-specific measurement level with each calibration.

6.2 Frequency of Recalibration
- After each exchange of lot (Reagent Integral or Starter Reagents).
- After 4 weeks and/or each time a new Integral is used (recommendation).
- After each servicing, of the Maglumi Fully Auto analyzer.
- If controls are beyond the expected range.

7. Sample Collection, Material and Storage

7.1 Serum
- Elbow vein blood 5ml in the tube, centrifugation at room temperature, serum was separated and stored at 2°C-8°C.
- Serum samples were stable for 12 hours at 2-8°C. If preserved more than 12 hours, please packed, -20°C can be stored for 30 days,
- to avoid repeated freezing and thawing.

7.2 Plasma
- Elbow vein blood 5ml in the tube, then add EDTA anticoagulant(50ul 0.3M EDTA per 5ml blood), centrifuged and separated plasma, stored at 2-8°C.
- Plasma samples were stable for 12 hours at 2-8°C. If preserved more than 12 hours, please packed, -20°C can be stored for 30 days,
- to avoid repeated freezing and thawing.

7.3 Whole blood
- Elbow vein blood 5ml in the tube, then add EDTA anticoagulant, (50ul 0.3M EDTA per 5ml blood),
- (Note: recommended EDTA as an anticoagulant, can not use heparin as an anticoagulant).
- Plasma was stable at 2-8°C for 24 hours. If preserved more than 12 hours, please packed, -20°C can be stored for 30 days,
- to avoid repeated freezing and thawing.
- Seal the nozzle, and upside down several times stored in room temperature. If preserved for long time, please packed, -20°C can be stored for 30 days.
- to avoid repeated freezing and thawing.

8. WARNING AND PRECAUTIONS FOR USERS
- For use in IN-VITRO diagnostic procedures only.
- Do not interchange reagents from different lots. Do not use kit components beyond their labeled expiry date.
- All samples, biological reagents and materials used in the assay must be considered potentially able to transmit infectious agents. They should therefore be disposed of in accordance with the prevailing regulations and guidelines of the agencies holding jurisdiction over the laboratory, and the regulations of each country. Disposal materials must be incinerated; liquid waste must be decontaminated with sodium hypochlorite at a final concentration of 5% for at least half an hour. Any materials to be reused must be autoclaved using an overkill approach(USP 24:2000.p.2.143J). A minimum of one hour at 121°C is usually considered adequate, though the users must check the effectiveness of their decontamination cycle by initially validating it and routinely using biological indicators.
- The calibrators in this kit are prepared from bovine serum products. However, because no test method can offer complete assurance that HIV, Hepatitis B Virus or other infectious agents are absent, these reagents should be considered a potential biohazard and handled with the same precautions as applied to any serum or plasma specimen.

9. Test Procedure
To ensure proper test performance, strictly adhere to the operating instructions of the Maglumi Fully Auto Analyzer. Each test parameter is tedly measured 10 times of its measurement, calculating three batches of kit, repeated measured 10 times of Calibrator High, calculating three batches of kit for Calibrator High between the measured values of the coefficients of variation, the results should ≤10%.

<table>
<thead>
<tr>
<th></th>
<th>Sample, calibrator or controls</th>
<th>ABEI Label</th>
</tr>
</thead>
<tbody>
<tr>
<td>+80μl</td>
<td>FITC Label Nano magnetic microbeads</td>
<td></td>
</tr>
<tr>
<td>+20μl</td>
<td>Incubation Cycle washing</td>
<td></td>
</tr>
<tr>
<td>+80μl</td>
<td>Measurement</td>
<td></td>
</tr>
</tbody>
</table>

10. Quality Control
- Observe quality control guidelines for medical laboratories.
- Use suitable controls for in-house quality control.

11 Results
11.1 Calculation of Results
- The analyzer automatically calculates the PCT concentration in each sample by means of a calibration curve which is generated by a 2-point calibration master curve procedure. The results are expressed in ng/ml. For further information please refer to the Maglumi Fully Auto Operator’s Manual.
- Conversion factor: 1 IU/ml PCT = 1.21 ng/ml PCT.
- 1 ng/ml PCT = 0.83 IU/ml PCT.

11.2 Interpretation of Results
- Reference values

<table>
<thead>
<tr>
<th>Clinical(serum or plasma)</th>
<th>concentration[ng/ml]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>&lt; 0.5</td>
</tr>
<tr>
<td>Chronic non-specific inflammation and autoimmune diseases</td>
<td>&lt; 0.5</td>
</tr>
<tr>
<td>Viral infection</td>
<td>&lt; 0.5</td>
</tr>
<tr>
<td>Moderate local bacterial infection</td>
<td>&lt; 0.5</td>
</tr>
<tr>
<td>Systemic inflammatory response syndrome (SIRS), multiple trauma, burns</td>
<td>0.5-2.0</td>
</tr>
<tr>
<td>Serious bacterial infection, sepsis, multiple organ failure</td>
<td>[2.0(can reach 10(100)]</td>
</tr>
</tbody>
</table>

- Results may differ between laboratories due to variations in population and test method. Each laboratory should establish its own reference range.

12. Limitations of the procedure
12.1 Patients with malignancies may exhibit PCT values within the normal range. PCT concentrations may be elevated in case of liver cirrhosis, hepatitis or tyrosinaemia. Thus, PCT determination is more suitable for therapeutic monitoring and follow-up as well as for a comparison with histological results.
- PCT serum levels may only be interpreted in context with the clinical picture and other diagnostic procedures. The PCT assay should not be used as the only criterion for cancer screening.
12.2 HAMA
Patient samples containing human anti-mouse antibodies (HAMA) may give falsely elevated or decreased values. Although HAMA-neutralising agents are added, extremely high HAMA serum concentrations may occasionally influence results.
12.3 High-Dose Hook
No high-dose hook effect was seen for PCT concentrations up to 10,000 ng/ml.

13. Performance Characteristics
13.1 Accuracy
Consider calibrator high of known concentration as a sample, dilute it by 1:2 ratio with diluent, and measure its diluted concentration for 10 times. Then calculate the recovery of measured concentration and expected concentration. The recovery should be within 90% -110%.
13.2 Precision
Intra-assay coefficient of variation was evaluated on Calibrator High repeatedly measured 10 times in the same assay, calculating their coefficient of variation, the results should ≤10%.
Inter-assay coefficient of variation was evaluated on three batches of kit, repeatedly measured 10 times of Calibrator High, calculating three batches of kit for Calibrator High between the measured values of the coefficients of variation.
variation, the results should ≤15%.

13.3 Sensitivity
The sensitivity is defined as the concentration of PCT equivalent to the mean RLU of 20 replicates of the zero standard plus two standard deviations corresponding to the concentration from the standard curve. The sensitivity is typically less than 0.13 ng/ml.

13.4 Specificity
The specificity of the PCT assay system was assessed by measuring the apparent response of the assay to various potentially cross reactive analytes. (When CT= 20ng/ml, the detection result of PCT is less than 1ng/ml.)

13.5 Linearity
Conduct a logarithmic transform to the RLU value and concentration value of 6 standards. After a double logarithmic fitting, the absolute value of its linearity should exceed 0.9800.

14. References