INTENDED USE
Reagent for the quantitative determination of calcium in human serum, plasma, or urine with MEDILYZER® series automated analyzers and other types of discrete analyzers mentioned in the special leaflet accompanying the insert. For in vitro diagnostic use only.

CLINICAL SIGNIFICANCE
Calcium concentration in serum increases in cases of primary and tertiary hyperparathyroidism, in hyperthyroidism, in conditions with bone involvement (in particular metastatic bone tumours, multiple myeloma, lymphomas, leukemia), excessive calcium intake, excessive vitamin D levels, Paget's disease, acromegaly, sarcoidosis, milk-alkali syndrome, liver or kidney disease, idiopathic hypercalcemia during infancy, drug induced hypercalcemia, dehydration. Lower calcium levels can be observed in hypothyroidism, pseudo-pseudohypoparathyroidism, vitamin D deficiency, chronic renal failure, liver disease causing decreased albumin production, low serum magnesium, hyperphosphatemia, acute pancreatitis, osteomalacia, prolonged anticoagulant therapy, inadequate nutrition.

METHOD PRINCIPLE
The calcium ions react with the Arsenazo III dye in acidic pH, to form a violet complex. The absorption at 650 nm is proportional to the calcium concentration in the sample. End point reaction.

METHOD LIMITATIONS
Refer to the book “Effects of Preanalytical Variables on Clinical Laboratory Tests” for possible interference of other pharmaceutical agents in this particular test. Interference of other agents is described in the “Clinical Guide to Laboratory Tests”.

REAGENT COMPOSITION
MES buffer (pH 6.5): 100 mMol
Arsenazo III: 1.5 mMol
Non reacting ingredients, preservative

WARNINGS - PRECAUTIONS
- This reagent is designed for in vitro diagnostic use. In vitro diagnostic reagents can be hazardous. They should be handled according to good laboratory techniques. Avoid inhalation and contact with eyes and skin.
- Samples should be considered as potentially infectious. Handle with special caution.
- The reagent contains Arsenazo III. Avoid swallowing or contact with skin and mucous membranes.
- Dispose all waste according to national laws.
- MSDS is available by MEDICOM upon request.

REAGENT PREPARATION
Reagent ready-to-use. The reagent bar code for automatic recognition by MEDILYZER®series analyzers.

REAGENT DETERIORATION
The reagents should not be used:
- When they do not exhibit the specified linearity or control values lies outside the acceptable range after recalibration.
- When they appear cloudy or decolorized.
- After prolonged exposure to sunlight or high temperature.

SHELF LIFE
Unopened, the reagent is stable at 2 – 8°C up to the expiry date stated on the label. After opening it remains stable for 2 months when stored in the reagent sampler of the MEDILYZER® or the MEDILYZER® BT analyzer.

SAMPLE
Fresh non hemolyzed serum or heparinized plasma from overnight fasting patient. Do not use citrate, oxalate, EDTA or any anticoagulant as they form complexes with Ca2+ ions. Serum is stable for at least 4 days at 4°C. Plastic or glass testing tubes may absorb calcium during storage. Precipitation of calcium with fibrinogen or lipids in heparinized plasma during storage or refrigeration has also been reported. Avoided repeated freezing and thawing of samples.

The right before sample collection, the patient must not eat, as calcium levels depend on patients' digestive state. Calcium excretion fluctuates during the day, declining after night. Obtain sample with minimal venous occlusion, avoiding patient exercise or after resting for circulation >1min, since the pressure from the tourniquet may lead to increased total calcium concentration due to water diffusion from the vein pores. Upright position causes a 0.2 – 0.8 mg/dl, increase in calcium levels.

Collect 24-hr urine sample in a bottle containing 10 ml HCl or 6M acetyl after collection to pH <2 and wait for 1 hour before analysis to allow any calcium salts to dissolve. Urine samples should be diluted in a 1.5 ratio with deionized water. Calcium concentration should be corrected for the added volume of HCl.

CALIBRATION
MEDICOM provides MEDIS-CAL (1578-0918) for serum calibration and MEDI-CAL U (1579-0185) for urine calibration. Calibrate the assay every 1 month when used on MEDILYZER® or on MEDILYZER® BT. Recalibration should also be repeated a major maintenance is performed on the analyzer or a critical part is replaced or when a significant shift in control values occurs.

QUALITY CONTROL
MEDICOM provides the MEDICOM Clinical Chemistry Control Level 1 & 2 (1578-0901-12 & 1578-0902-12) respectively and the MEDITROL-U 1/2 (1579-081I) or the BIORAD Lynchiqckette Quantitative Urine Control Normal (Cat. No: 397, MEDICOM code: 1778-0185), Abnormal (Cat. No: 398. MEDICOM code: 1778-0182). Control recovery should lie within the acceptable range. Results outside the acceptable range even after recalibration could be due to reagent deterioration, unstable storage conditions or control deterioration, instrument malfunction, or error during test procedure.

MATERIALS REQUIRED BUT NOT SUPPLIED WITH THE KIT
Calibrator control materials
MEDILYZER® FT or other automated biochemistry analyzer
Common laboratory equipment.

CALCULATIONS
The analyzer automatically computes the urea concentration of each sample. Calculate the urine calcium result by multiplying the test result by the dilution factor of the sample.

REFERENCE INTERVALS
Serum, plasma: 8.1 – 10.4 mg/dl
Urine: Up to 40 mg/dl
In low calcium diet the reference interval for urine is 50 – 150 mg/24h.
In calcium free diet the reference interval for urine is 5 – 40 mg/24h.
Expected values may vary with age, sex, sample type, diet and geographical location. Each laboratory should determine its own expected values as dictated by good laboratory practices.

WASTE DISPOSAL
The reagent contains Arsenazo III. Flush waste pipes with water after the disposal of undiluted reagent in the drain.

SPECIFIC PERFORMANCE CHARACTERISTICS
The following values are representative of the reagent performance on MEDILYZER® series analyzers. The performance has been evaluated on other types of analyzers, covering all requirements of the 98/79 IVD Directive. A list of analyzers with the corresponding performance characteristics is available in the special leaflet accompanying the insert. The results taken in your laboratory may differ from these values.

**SERUM**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MEDILYZER® BT</th>
<th>MEDILYZER® F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Linearity</td>
<td>Serum: Up to 20 mg/dl</td>
<td>Serum: Up to 20 mg/dl</td>
</tr>
<tr>
<td>Lowest detection limit</td>
<td>Serum: 0 mg/dl</td>
<td>Serum: 0.31 mg/dl</td>
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</tbody>
</table>

**Urine**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MEDILYZER® BT</th>
<th>MEDILYZER® F</th>
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</thead>
<tbody>
<tr>
<td>Linearity</td>
<td>Lactate: up to 5 mmol/l</td>
<td>Lactate: up to 5 mmol/l</td>
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<tr>
<td>Lowest detection limit</td>
<td>Lactate: 0.8 mg/dl</td>
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<tr>
<td>Lowest detection limit</td>
<td>Non reacting ingredients, preservative</td>
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**INTERFERENCES**

- Insignificant
- Significant
- Critical

**SYMBOLES**

<table>
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<tr>
<td>Ca++</td>
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<tr>
<td>Ni</td>
<td>Nickel</td>
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**BIBLIOGRAPHY**