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RUO Microplate/Manual Applications

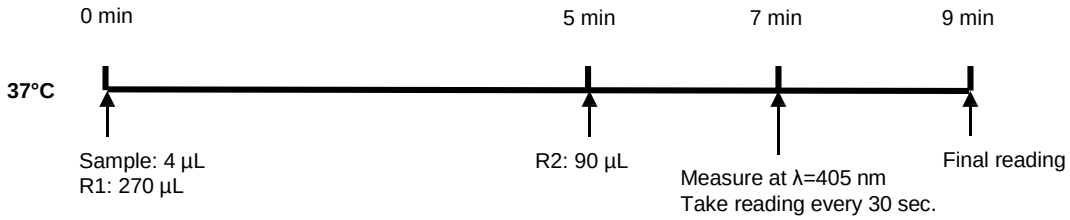
# Autokit Total Ketone Bodies Microtiter Procedure

For Research Use Only. Not for use in diagnostic procedures.
Cat. No. 415-73301 **Autokit Total Ketone Bodies R1 set**
Cat. No. 411-73401 **Autokit Total Ketone Bodies R2 set**
Cat. No. 412-73791 **Ketone Body Calibrator 300**
Cat. No. 418-73891 **Ketone Body Calibrator 40**

1. Prepare reagents R1 and R2 according to the package insert instructions.
2. Pipette 4µL of each of the following into assigned microplate well according to plate layout: saline (to be used as blank), each level of calibrator, and sample.*
3. Add 270µL of prepared R1 solution to each well.
4. Mix quickly and gently, and incubate for 5 minutes at 37°C.
5. Add 90µL of prepared R2 solution to each well.
6. Mix quickly and gently, and incubate at 37°C for 2 minutes.
7. Take initial absorbance readings at 405nm (T=0) after the 2 minute incubation (see step 6.) period. Continue to take readings every 30 seconds for 2 additional minutes (T=0.5min, T=1min, T=1.5 min, T=2min).
8. Determine the Δ OD/min by subtraction.
9. Calculate the Total Ketone Bodies concentration using the calibrator's concentration and delta absorbance/min values. See equation below:

$$\text{Sample conc. (µmol/L)} = \text{calibrator concentration (µmol/L)} \times \frac{\text{Sample}\Delta\text{OD/min}}{\text{Calibrator } \Delta \text{ OD/min}}$$

**Basic procedure outline:**




To increase the sensitivity of the method, use a sample volume of 17 µL