

# CURRENT

Category : Patient Care  
Title : Procedure for estimation of Paracetamol in human serum by Spectrophotometer

SOP No. and Version: TDM 14/01  
Date first effective: 1<sup>st</sup> January 2025 Review date: 31<sup>st</sup> December 2025  
Department of Clinical Pharmacology, 1st Floor, New MS Building,  
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### 1. Purpose:

This SOP describes the technique for qualitative and quantitative estimation of Paracetamol in human Serum by Spectrophotometer

### 2. Scope:

This SOP is limited to estimation of Paracetamol in  $\mu\text{g/mL}$  in human Serum by Spectrophotometer

### 3. Responsibilities:

The Head of the department is responsible for the medical care and welfare of all patients pertaining to TDM of Paracetamol. The task of performing estimation of Spectrophotometer will be delegated to trained personnel who will perform this function.

### 4. Applicable rules, regulations and guidelines

- ICMR Good Clinical Laboratory Practices Guidelines 2008 (<http://icmr.nic.in/guidelines/GCLP.pdf>)

### 5. Reference to other applicable SOPs

- SOP No.24/02: Biomedical waste management.
- SOP No. TDM01/02: Collection and separation blood plasma/serum for TDM

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## 6. Detailed instructions

### 1. Preparation of standard and calibrator

#### a. Preparation of Stock Standard (Paracetamol acid 1mg/mL) :(0.010g)

10 mg of pure powder of Paracetamol + 10 mL of methanol

#### b. Preparation of Reagent & Chemical Solvent

a. Trichloroacetic acid (TCA) 24g in 100mL Distilled water

b. Sodium Hydroxide (NaOH) 4g in 100 mL Distilled water

### Preparation of Serum standards:

Plasma Standard	25µg/mL	50µg/mL	100µg/mL
Blank Serum	1000µL	1000µL	1800µL
Paracetamol acid 1mg/mL			200µl mix well and transfer to
100µg/mL(stock)	1000µL	1000µL	

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## 2. Extraction procedure

1000 $\mu$ L (1mL) of Serum (serum/standards/quality control/patient sample) in 10mL centrifuge tube



Add 2000 $\mu$ L (2mL) of Trichloroacetic acid (TCA) Vortex mix thoroughly for 1 min.



Centrifuge at 3500 rpm for 10mins



Transfer the supernatant in another test tube



Again, add 1000 $\mu$ L (1mL) of TCA to the Supernatant



Vortex and Centrifuge at 3500rpm for 10 min



Separate 2mL of supernatant and add 400 $\mu$ L of NaoH to it



Vortex and take OD on spectrophotometer at 290nm

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### 3. Spectrophotometer Conditions

- a. Wavelength: 290 nm

### 4. Abbreviations:

- a. SPECTO= Spectrophotometer