



Paracetamol has analgesic and antipyretic properties. It is given by oral or as per rectal for mild to moderate pain and pyrexia and indeed is much safer than aspirin.<sup>4</sup> The severe and fatal liver damage following overdose of paracetamol was first reported from Scotland.<sup>5-6</sup> Similar reports came from other countries.<sup>7-9</sup> Nausea, vomiting and abdominal pain usually occur within a few hour of ingestion of a hepatotoxic dose of paracetamol followed by hepatic tenderness. Mild jaundice may become apparent followed by rapid and complete recovery.<sup>10</sup> The onset of back pain is due to development of oliguric renal failure.<sup>11-15</sup> Other complications observed are metabolic acidosis, pancreatitis and cardiac toxicity.<sup>16-18</sup>

Paracetamol is metabolized in liver and excreted through kidney mainly as glucuronide and sulphate conjugates. The metabolism of other drugs is also impaired in patients with liver damage following paracetamol overdose.

The biochemical mechanism of paracetamol hepatotoxicity was first described by Mitchell and his colleagues.<sup>19-21</sup>

Maximum single therapeutic doses of paracetamol do not produce liver toxicity in man and, although the threshold dose has been estimated as 10 to 15 g. Unfortunately, no reliance can be placed on the dose claimed to have been taken by the patient and unknown amount may be lost by gastric lavage and vomiting. However, the amount absorbed can be estimated as the product of plasma paracetamol concentration three hour after ingestion and apparent volume of distribution ( $0.8 \text{ L Kg}^{-1}$ ). On this basis the threshold dose in man appears to be  $250 \text{ mg Kg}^{-1}$  body weight. All patients who absorb more than  $250 \text{ mg Kg}^{-1}$  body weight had severe liver damage.<sup>22</sup>

Paracetamol is often recommended instead of aspirin for alcoholic patients who have high incidence of G.I. bleeding. However chronic ethanol consumption induces hepatic cytochrome p-450.<sup>23</sup> and depletes hepatic glutathione and rises the possibility of a potential toxic interaction between paracetamol and ethanol. This statement was also confirmed by the work of Peterson F.J.<sup>24</sup>

Several sulphhydryl compounds are used as antidotes which protect the effect of paracetamol metabolite that causes depletion of hepatic glutathione. Activated charcoal may

adsorb antidote and reduces its bioavailability in treatment of paracetamol poisoning. The present study was performed using sulphhydryl compound (methionine) to find out any effect of charcoal on its bioavailability. For the treatment of over dose of paracetamol activated charcoal may be used to reduce the bioavailability of paracetamol due to its adsorption on activated charcoal.

## MATERIALS AND METHODS

The following materials which were of analytical grade were used, paracetamol, sodium hydroxide, monobasic potassium phosphate, distilled water and Carbomix (activated charcoal). Whatman filter papers and paracetamol tablets 500 mg, B.P, (Rickett & Colman) were also used. pH meter, UV/ visible spectrophotometer (Model 2000), weighing balance (Petit Balance Ohyo Balance Corporation, Japan) were the equipment used.

### Preparation of sodium hydroxide solution

Sodium hydroxide solution 0.2 M was prepared by dissolving 8 gm of sodium hydroxide per liter of distilled water.

### Vehicle preparation for adsorption studies

Simulated intestinal fluid (SIF) was prepared in accordance with the method described in the USP<sup>25</sup> except the enzyme pancreatin was omitted in this study.

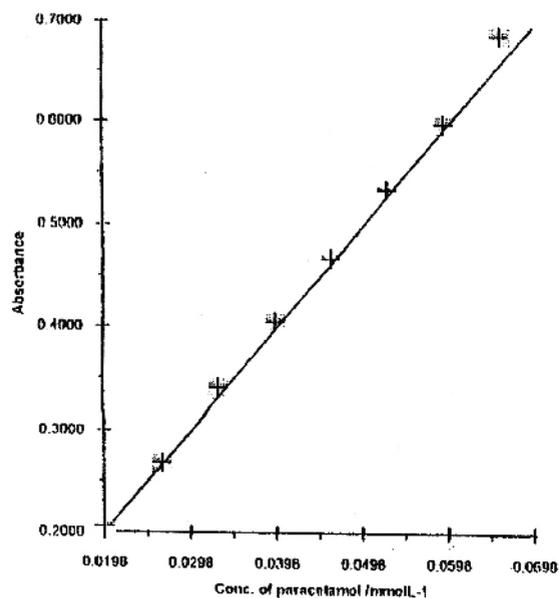
Monobasic potassium phosphate 34.0022 g was taken in 5 liter volumetric flask and mixed with 1.250 ml of water, 950 ml of 0.2 M sodium hydroxide and 2 liter of water were added and the resulting solution was adjusted to  $\text{pH } 7.5 \pm 0.1$  with 0.2 M NaOH. The volume was made up to 5 liter with distilled water.

### Preparation of calibration graph

A graph of known concentrations verses absorbance was plotted and then concentration of the unknown solution was found by using the calibration graph of absorbance and concentration at 257 nm.

Paracetamol 0.1 g of powder was taken in 100 ml volumetric flask. It was first dissolved with SIF to make the volume. Ten ml of this solution was transferred to 100 ml volumetric flask and diluted to 100 ml with SIF. Similarly further dilutions were made. The data of

calibration graph is mentioned in Table 1 and Fig 1.



**Fig. 1:** Calibration curve of paracetamol in SIF (pH 7.5).

#### Determination of Equilibrium Contact Time

Equivalent to 1.512 g of paracetamol was transferred into 500 ml volumetric flask and dissolved in SIF and made up the volume with SIF. Aliquots of 25 ml of paracetamol solution (0.5mmol of paracetamol) were pipetted into six stoppered tube containing 0.3024 g of activated charcoal (charcoal to drug ratio 4:1). A sample was removed at 5, 10, 15, 30, 45 and 60 minutes intervals. The samples were filtered and analyzed at 257 nm using SIF as blank.

#### Paracetamol adsorption on different concentrations of activated charcoal at pH 7.5

Aliquots of 25 ml of paracetamol solution were placed in glass tubes containing 0.30, 0.38, 0.45, 0.60 and 7.5 g of activated charcoal. The mixtures were filtered and absorbance was measured at 257 nm using SIF with charcoal.

#### Determination of adsorption of Paracetamol tablets on different concentrations of activated Charcoal at pH 7.5

- Each tablet contains paracetamol = 500 mg
- Weight of 20 paracetamol tablets = 11.933 g
- Weight of one paracetamol tablet = 0.5966 g

20 tablets were weighed and powdered and 0.1779 g powder of paracetamol (equivalent to 0.150 g) was taken in 200 ml volumetric flask. 50 ml of 0.1 M NaOH was transferred to the flask and diluted with 100 ml of water and volume was made to 200 ml. 10 ml of solution was diluted to 100 ml. From this 10 ml solution was further diluted to 100 ml with 0.1 M NaOH. The absorbance was measured at 257 nm.

### RESULTS AND DISCUSSION

The results of the studies on adsorption of Paracetamol powder on activated charcoal were presented in Table 2. It is clear from the table that adsorption of paracetamol was 95.6% within 30 minutes. Adsorption was completed within 45 minutes.

**Table 1:** Standard curve for determination of paracetamol.

S.S ml	F.V MI	Conc./m MI <sup>-1</sup>	ABSORBANCE				
			1	2	3	4	Mean
10	100	0.661	0.669	0.689	0.689	0.686	0.686
9	100	0.0595	0.599	0.606	0.588	0.600	0.598
8	100	0.0529	0.540	0.532	0.525	0.534	0.533
7	100	0.0463	0.467	0.473	0.461	0.467	0.467
6	100	0.0397	0.401	0.409	0.403	0.403	0.404
5	100	0.0331	0.333	0.343	0.347	0.341	0.341
4	100	.0265	0.265	0.270	0.269	0.269	0.268
3	100	0.198	0.203	0.210	0.205	0.205	0.206

S.S = Standard solution F.V = Full Volume

**Table 2:** Amount of paracetamol adsorbed at different time intervals.

T/Min	Absorbance				C/X10 <sup>2</sup> mmol/L	Dilution Factor	Ceq/ mmol/L	Ceq/ mmol/25ml	X/ mmol	% Adsorbed
	1	2	3	Mean						
5	0.309	0.309	0.311	0.310	30.420	1 in 100	3.0420	0.0760	0.4240	48.8
10	0.195	0.193	0.197	0.196	1.9219	1 in 100	1.9219	0.0480	0.4520	90.4
15	0.300	0.304	0.301	0.302	2.9635	1 in 50	1.4818	0.0370	0.4630	92.6
30	0.178	0.176	0.130	0.178	1.7450	1 in 50	0.8725	0.0218	0.4782	95.6
45	0.130	0.129	0.177	0.130	1.2733	1 in 50	0.6366	0.0159	0.4841	96.8
60	0.643	0.646	0.646	0.646	6.3492	1 in 10	0.6349	0.0159	0.4841	96.8

T = Time

C = Concentration

Ceq = Equilibrium concentration

X = Amount of paracetamol adsorbed

The findings of the investigations on adsorption of paracetamol on activated charcoal with various charcoal to drug proportions are given in Table 3. The adsorption was almost complete at 5:1 charcoal drug ratio and is 98.86%. There is increase in adsorption with the increase in concentration of charcoal, but the percentage adsorption was negligible as compared to increase in concentration of charcoal. The data for the adsorption of paracetamol for tablets is given in Table 4. The decrease in adsorption may be due to adsorption of the drug to tablet inactive ingredients. The blank was only SIF. The ideal blank should contain all ingredients which are present in tablets except paracetamol.

The effect of alcohol (6% v/v) on paracetamol adsorption is shown in Table 5. It was found that alcohol has no effect on the adsorption of paracetamol on activated charcoal. Beside other physiological properties molecular weight of alcohol and paracetamol (46 and 151.2). The molecular size of alcohol is one third the size of paracetamol, therefore alcohol is very less likely to have any effect on the adsorption of paracetamol onto activated charcoal. Patients with history of alcoholism may have increased susceptibility to paracetamol induced toxicity. Paracetamol is metabolized predominately in the liver and is excreted mainly as glucuronide and sulphate conjugates which are very polar and are ionized at pH of blood and urine and are excreted easily. The combination of paracetamol with glucuronic acid occurs readily with compounds having reactive proton, usually attached to hetro atom (hydroxyl, carboxylic, amino and sulfhydryl).

The adsorption results of paracetamol in the presence of 0.01 M methionine using SIF of pH 7.5 and using charcoal to drug ratio between 4:1 and 10:1 are given in Table 6. There is reduction of paracetamol adsorption in presence of methionine. Beside other physiochemical properties there is similarity in size of Paracetamol and methionine which play role in competing for adsorption sites on activated charcoal.

Since the adsorption of paracetamol on activated charcoal is only reduced by 8% in the presence of methionine, charcoal can be used in removing paracetamol molecules, while at the same time only slightly reducing the bioavailability of methionine. Hepatotoxicity is related to the conversion of a small fraction of the dose to a highly reactive alkylating metabolite which is normally inactivated by preferential conjugation with glutathione and eventually excreted as cystine and mercapturic acid conjugates. Liver damage is associated with glutathione depletion, and this compound would seem to be an ideal antidote. Unfortunately, glutathione itself does not enter cells readily and it is ineffective in preventing experimental paracetamol hepatotoxicity. In contrast, glutathione precursors and related compounds such as cysteine, N-acetylcysteine, cysteamine and methionine have been studied clinically in paracetamol poisoning. They prevent covalent binding and hepatic necrosis in animals and have been remarkably successful in preventing liver damage following paracetamol overdose in man.

**Table 3:** Adsorption studies of paracetamol on activated charcoal.

m/g	Drug Ratio	Amount Used/g	ABSORBANCE				C/X 10 <sup>-2</sup> mmol/L	Dilution factor	Ceq/ mmol/L	Ceq/ x10 <sup>-2</sup> mmol/25ml	X /mmol	X/mg	X/m mmol/g	Ceq/X/m Per g per L	% Adsorbed
			1	2	3	Mean									
0.30	4:1	0.3001	0.304	0.317	0.291	0.302	2.9635	1 in 20	0.5927	1.4818	0.4852	73.362	1.6168	0.3666	97.06
0.38	5:1	0.3810	0.508	0.429	0.439	0.462	4.5358	1 in 5	0.2268	0.5670	0.4943	74.738	1.2974	0.1748	98.86
0.45	6:1	0.4488	0.783	0.733	0.785	0.775	7.6116	1 in 2	0.1522	0.3805	0.04962	75.025	1.1059	0.1377	99.24
0.60	8:1	0.6035	0.700	0.708	0.730	0.705	6.9237	None	0.0692	0.1730	0.4983	75.343	0.8257	0.0838	99.66
0.75	10:1	0.7530	0.505	0.575	0.564	0.541	5.3121	none	0.0531	0.1328	0.4987	75.403	0.6623	0.0802	99.74

M= amount of charcoal

C = Concentration obtained from calibration graph

Ceq = Equilibrium concentrations

**Table 4:** Adsorption studies of paracetamol tablets on activated charcoal.

m/g	Drug ratio	Amount used/g	Absorbance	C/ mmol/L	Dilution Factor	Ceq/ mmol/L	Ceq/ Mmol/L	Ceq/x 10 <sup>-2</sup> mmol/25ml	X/ mmol	X/mg	% adsorbed	X/m mmol/g	Ceq/ x/m/ g/L
0.30	4:1	0.3051	0.341	3.3467	1 in 50	1.6734	1.6734	4.1834	0.4581	69.26	91.62	1.5017	1.1140
0.38	5:1	0.3772	0.300	2.9439	1 in 50	1.4719	1.4719	3.6799	0.4632	70.04	92.64	1.2279	1.1986
0.45	6:1	0.4551	0.115	1.1259	1 in 25	0.2815	0.2815	0.7037	0.4930	74054	98.6	1.0832	0.2599
0.60	8:1	0.6401	0.259	2.5410	1 in 5	0.1270	0.1270	0.3176	0.4968	75.15	99.36	0.8224	0.1644
0.75	10:1	0.7552	0.190	1.8629	1 in 5	0.0931	0.0931	0.2329	0.4977	75.25	99.54.	0.6590	0.1413

M= amount of charcoal

C = Concentration obtained from calibration graph

Ceq = Equilibrium concentrations

**Table 5:** Adsorption studies of paracetamol in presence of alcohol 6% v/v with different drug to charcoal ratios.

Amount used/g	Absorbance	Dilution Factor	Ceq/ mmol/L	X/mmol	X/mg	% Adsorbed	X/m mmol/g	Ceq/x/m/g/L
0.3021	0.481	1 in 10	0.471	0.488	73.78	97.60	1.615	0.292
0.3778	0.424	1 in 5	0.208	0.495	74.84	99.99	1.310	0.178
0.4541	0.666	1 in 2	0.131	0.497	75.15	99.40	1.094	0.119
0.6051	0.681	None	0.067	0.498	75.30	99.60	0.823	0.081
0.7565	0.407	none	0.040	0.499	75.45	99.80	0.660	0.061

**Table 6:** Adsorption of paracetamol in the presence of methionine (0.1 M).

Amount used/g	Absorbance	Dilution factor	Ceq/ mmol/L	X/mm ol	X/mg	% Adsorbed	X/m mmol/g	Ceq/x/m/g/L
0.3034	0.239	1 in 25	0.583	0.485	73.33	97.00	1.599	0.365
0.3789	0.637	1 in 15	0.321	0.493	74.54	98.60	1.301	0.270
0.4558	0.146	1 in 10	0.142	0.496	75.00	99.20	1.090	0.130
0.6078	0.673	None	0.066	0.498	75.30	99.60	0.820	0.008
0.7581	0.404	none	0.040	0.499	75.45	99.80	0.658	0.006

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