



## METHODS

### 1. Animal

The study was carried out on young male albino rats weighing between 40–60 g and albino rats weighing between 180–220 g of Wistar strain. They were housed under standard laboratory conditions with controlled temperature ( $26^{\circ} \pm 2^{\circ}\text{C}$ ) and lighting (lights on 7 a.m. to 6 p.m.) and fed *ad libitum* with rat diet in pellet form with free access to water.

### 2. Exposure to heat stress

The animals were divided into two groups – control and experimental. The animals of the experimental group were exposed to acute heat stress at  $38^{\circ}\text{C}$  for 2 hours beginning at about 9–30 a.m. At the end of the exposure to high ambient temperature, the experimental animals were allowed to rest for 20 to 30 minutes at the control room temperature ( $26^{\circ}\text{C}$ ). Thereafter the animals were used for motility studies.

The animals were exposed to high ambient temperature in a chamber measuring 60 cm × 40 cm × 40 cm with provision for an inlet and an outlet for the atmospheric air. The animals were confined on a wire bottom platform in a cage approximately 30 cm from the floor of the chamber. The temperature of the chamber was maintained within an error of  $\pm 1^{\circ}\text{C}$ .

### 3. Measurement of gastrointestinal motility

After overnight starvation the animal was given just sufficient ether so that it submitted passively to intubation with a

human infant feeding tube and 1 ml of barium sulphate suspension in isotonic saline (0.9% w/v) containing 100% w/v barium sulphate was introduced into stomach by a syringe in case of a young rat (3 ml of barium sulphate was introduced in case of an adult rat). After an interval of 15 minutes the animal was killed by cervical dislocation; the abdomen was opened by a midline incision and ligatures were rapidly applied at the gastro-oesophageal junction, ileocaecal junction and at the barium column which could be seen through the thin wall of the intestine. The stomach and the small intestine were removed from the abdomen. The stomach was cut open and washed for its luminal contents into a beaker with normal saline. The stomach washing was centrifuged at 3000 rpm for 5 minutes in a clinical centrifuge. The sediments were dried in a hot-air oven ( $100 - 110^{\circ}\text{C}$ ) to a constant weight. The gastric emptying and the intestinal transit were calculated on the basis of methods described earlier (5). Gastric emptying was expressed as the percent release of barium sulphate as follows:

$$\text{Gastric emptying} = \frac{A - B}{A} \times 100, \text{ where}$$

A = barium sulphate introduced into the stomach

B = barium sulphate from the stomach.

Intestinal transit was expressed as the percentage of the total length of the small intestine traversed by barium sulphate in the study period of 15 minutes. Total length of the intestine was measured by the method of Barry *et al* (6).

#### 4. Absorption studies

After anaesthetising the rats with sodium pentothal (40 mg/kg body weight I.P) absorptive studies for D-glucose and L-proline were undertaken by the in vivo loop technique from proximal jejunum and terminal ileum following the procedure of Crampton *et al* (7) with some modifications. D-glucose and L-proline were estimated by the methods described earlier (8, 9, 10). The results were expressed in terms of micromoles per centimeter length per hour.

#### 5. Injection of drugs

The drugs atropine sulphate (obtained from Bengal Immunity, India) and cyproheptadine hydrochloride (obtained from Sigma Chemical, USA) were injected via the intraperitoneal route at the dose of 5 mg/kg and 20 mg/kg respectively.

#### 6. Statistical analysis

It was done by analysis of variance and students' 't' test.

## RESULTS

Table I shows the effect of heat stress on gastric emptying and intestinal transit in adult rats. Compared to the controls there was no significant change in the gastric emptying and intestinal transit in heat exposed adult rats. Table II shows the effect of heat stress on gastric emptying and intestinal transit in young rats. Compared to the controls, gastric emptying and intestinal transit were significantly increased in heat exposed young rats. But in cyproheptadine hydrochloride pretreated heat exposed young rats there was no significant change in gastrointestinal motility compared to the controls. Table III shows the effect of heat stress on intestinal absorption of D-glucose and L-proline in control and young rats; compared with the controls, absorption of D-glucose and L-proline was significantly reduced from both jejunum and ileum in heat exposed young rats.

TABLE I: Gastric emptying in 15 min-percent release and intestinal transit in 15 min-percent travelled in control and heat exposed adult rats.

Parameter	(Mean $\pm$ SEM for 6 animals)		Level of significance
	Control	Adult rats exposed to heat for 2 hrs.	
Gastric emptying (% release)	76.6 $\pm$ 1.8	76.4 $\pm$ 1.9	NS
Intestinal transit (% travelled)	66.5 $\pm$ 2.2	68.4 $\pm$ 2.3	NS

TABLE II: Gastric emptying in 15 min-percent release and intestinal transit in 15 min-percent travelled in control and heat exposed young rats.

Parameter	(Mean $\pm$ SEM for 6 animals)			
	Control	Young rats exposed to heat for 2 hrs.	Heat exposed young rats pretreated with atropine sulphate	Heat exposed young rats pretreated with cyproheptadine hydrochloride
Gastric emptying (% release)	75.5 $\pm$ 1.7	86.7 $\pm$ 2.2*	87.1 $\pm$ 1.8	74.6 $\pm$ 1.6
Intestinal transit (% travelled)	68.3 $\pm$ 2.1	79.3 $\pm$ 1.8*	79.1 $\pm$ 2.1	66.4 $\pm$ 1.9

\*In comparison with control P&lt;0.05

TABLE III: *In vivo* intestinal absorption of D-glucose and L-proline in control and heat exposed young rats.

Group	(Mean $\pm$ SEM for 6 animals)			
	D-glucose		L-proline	
	Jejunum	Ileum	Jejunum	Ileum
Control	34.84 $\pm$ 1.16	18.58 $\pm$ 0.86	42.52 $\pm$ 1.22	24.38 $\pm$ 1.42
Heat stress	20.62 $\pm$ 1.02*	10.16 $\pm$ 0.92*	20.64 $\pm$ 1.36*	12.44 $\pm$ 1.36*

\*In comparison with control P&lt;0.05

## DISCUSSION

These results indicate that acute heat stress increases gastrointestinal motility in young rats, while decreasing the absorption of nutrients from the intestinal tract. In a recent study (4), it was shown that heat stress altered various morphometric dimensions; there was significant reduction in dry weight, villus height, villus surface area while the absorptive study showed significant decrease in absorption as compared to the control. Our results correlate well with this study and it may be suggested that intestinal absorption is decreased following increased gastrointestinal motility allowing the nutrients a shorter period of stay in the gut lumen. There are evidences that the decreased intestinal absorption could also be hormonally mediated (11); however the structural change as shown by Sengupta

and Sharma (4) appears not to be so (12). It is unlikely that the structural changes as shown by Sengupta and Sharma (4) would be responsible for hypermotility. There are evidences that the structural changes have no bearing to alteration in the motor activity of the gut (13, 14). The hypermotility of gastrointestinal tract is also not due to bacterial toxins, as the rats were exposed to heat for a short period, when there is little chance of the pathogens in the gut lumen to grow and flourish more to produce increased motility. It is known that intestinal motility serves not only the purpose of mixing the luminal contents and propelling them in the aboral direction but also as an important factor in maintaining the relative sterility of the gut (15). Hypermotility following acute heat stress as seen in this study may thus be considered as a cause of relative sterility of the gut as seen in some children suffering from

summer diarrhoea (3). Atropine sulphate, cholinergic receptor blocker could not alter this change indicating non-involvement of the cholinergic mechanism in the increase of gastrointestinal motility. Rather this increase in gastrointestinal motility could be mediated by serotonergic mechanism as this increase was not found in serotonergic receptor blocker cyproheptadine hydrochloride pretreated rats. Serotonin is present in and secreted by endocrine enterochromaffin cells as well as nerve endings, so in the gastrointestinal tract it is both a hormone and a neurotransmitter (16). At the present juncture it is however not possible to explain how exposure of heat increases gastrointestinal motility through

serotonergic mechanism. It may be concluded that increased gastrointestinal motility in young growing rats could have a bearing on the etiopathology of diarrhoea seen in children during summer season in tropical countries and cyproheptadine hydrochloride may be used in the treatment of summer diarrhoea in children particularly when no specific etiology is found.

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