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Original Article

# Beverage-induced enhanced bioavailability of carbamazepine and its consequent effect on antiepileptic activity and toxicity

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ARTICLE INFO

*Article history:*
Received 14 April 2014
Received in revised form
14 July 2014
Accepted 16 July 2014
Available online 3 December 2014

*Keywords:*
bioavailability
black tea
carbamazepine
grapefruit juice
HPLC

ABSTRACT

The present study was undertaken to investigate the food–drug interaction of carbamazepine (CBZ). Common fruit juices [grapefruit juice (GFJ), lime juice (LJ)], known to inhibit the enzyme cytochrome P450 3A4 (CYP3A4), and some widely consumed beverages [milk (M), black tea (BT)] were involved in this study in the presence of CBZ, as might happen during clinical therapy. The effects of the beverages on the pharmacokinetics and drug-induced toxicity of CBZ was observed after concomitant administration for a period of 28 days. Accordingly, the influence of altered bioavailability of CBZ on its antiepileptic activity was investigated. A significant shift in the $C_{max}$ as well as $T_{max}$ of CBZ was observed in the presence of LJ and GFJ. This increase in bioavailability significantly enhanced hepatotoxicity and delayed the onset of tremor and piloerection against pentylene tetrazole (PTZ)-induced seizure in experimental animals. However, increased toxicity of CBZ was found to be absent with BT. Thus, from our observation, LJ or GFJ in the presence of CBZ significantly increased the bioavailability of CBZ, which might lead to increased toxicity and antiepileptic activity of the drug.



## 1. Introduction

Antipsychotics and antidepressants are clinically important drugs often used for chronic treatment thereby rendering adverse effects and aspects of drug–drug or food–drug interactions of these drugs to serious clinical vigilance [1]. Carbamazepine (CBZ), a drug with a comparatively narrow therapeutic index, is a concern among health care professionals because it may result in clinically significant drug interactions. Therefore, bioavailability studies in the presence of other drugs and food are becoming increasingly necessary to avoid toxic effects [2] or clinical failure. It has been observed that inhibition of cytochrome P450 (CYP) enzymes is the most common mechanism that produces serious and potentially life-threatening drug interactions [3]. CBZ undergoes

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http://dx.doi.org/10.1016/j.jfda.2014.07.012

biotransformation by CYP3A4 into carbamazepine-10,11-epoxide [4] and as a consequence of CYP inhibition or induction, plasma concentrations of CBZ may reach toxic or subtherapeutic levels.

Similar studies are available on incidents of increased bioavailability and enhanced pharmacodynamic effects following concomitant administration of citrus juice and calcium channel blockers such as felodipine and nifedipine [5].

In the present study, we investigated the *in vivo* pharmacokinetic interaction and pharmacodynamic effects of CBZ with commonly consumed beverages including lime juice (*Citrus aurantifolia*; LJ), grapefruit juice (*Citrus paradise*; GFJ), milk (M), and black tea (*Camellia sinensis*; BT) in experimental animals.

## 2. Methods

### 2.1. Chemicals and reagents

A CBZ suspension (Tegretol; Novartis Pharmaceuticals, Hyderabad, India), acetonitrile [high performance liquid chromatography (HPLC) grade; Sigma Aldrich, Maharashtra, India], potassium dihydrogen phosphate (Merck Pvt. Ltd, Mumbai, India), ortho-phosphoric acid (Merck Pvt. Ltd, Mumbai, India), methanol (HPLC grade, Sigma Aldrich, Maharashtra, India), and HPLC grade water (resistivity of 18.2 MU cm) generated from a Milli Q water purification system (Elix; Milli Q A10 Academic, Molsheim, France) were used throughout the analysis. Biochemical kits and pentylene tetrazole (PTZ) were obtained from Merck Pvt. Ltd. (Mumbai, India) Crude CBZ and valdecoxib were obtained as gifts from Cosmas Pharmacls (Ludhiana, Punjab, India).

### 2.2. Animal husbandry and maintenance

Healthy adult male Wistar rats weighing 170–180 g and male Swiss albino mice of 25–30 g, procured from TAAB Biostudy Services, 67/1B, Ibrahimpur Road, Kolkata 700032, India, were used for the study. The animals were grouped and housed in wire cages with not more than six animals per cage in a controlled environment (12 h light–dark cycle, temperature of 25 ± 2°C and 50 ± 20% relative humidity). During the period of study, the animals had free access to standard dry pellet diet (Nutrilab Rodent; Provimi) and water *ad libitum*. The study was conducted in accordance with the Institutional Ethical Committee (constituted under the Guidelines Committee for the Purpose of Control and Supervision of Experiments on Animals, Reg. No. 367).

### 2.3. Preparation of beverages

LJ (*Citrus aurantifolia*), GFJ (*Citrus paradise*), and M were obtained from local commercial sources. Juice was obtained by squeezing the edible portion of the fruits and then filtered. BT (*Camellia sinensis*) extract was prepared by soaking 2 g of black tea leaves (Lipton; Hindustan Unilever Ltd.) in 10 mL of boiling water followed by filtration [6]. Beverages were administered at a dose of 10 mL/kg.

### 2.4. High performance liquid chromatography conditions

The HPLC system consisted of an LC-20A Dvp pump (Shimadzu, Kyoto, Japan), a Shimadzu UV absorbance detector, a Hamilton syringe, and a Shimadzu SDP-20Avp system controller. The system was equipped with a Luna 5μ C18 column (250 mm × 4.6 mm, 5 μm; Phenomenex, Torrance, CA, USA), preceded by a precolumn. The isocratic mobile phase comprised a mixture of 100 mM potassium dihydrogen orthophosphate ($KH_2PO_4$, pH 3.2) and acetonitrile in a ratio of 60:40 v/v was delivered at a flow rate of 0.8 mL/min at 25°C. The UV detector was set at 245 nm and the volume of injection was 20 μL. The column was equilibrated for at least 20 minutes with the mobile phase flowing through the system prior to the injection of the drug standards. The run time was set at 15 minutes with the system operating at air-conditioned temperature (20°C).

### 2.5. Grouping and dosing of animals for pharmacokinetic study

The maximum tolerated dose of CBZ in humans was extrapolated to a rat dose [7]. Rats were randomly grouped into nine ($n = 9$). CBZ at a dose of 105.70 mg/kg was administered orally (equivalent to 17.14 mg/kg in humans). Group I received CBZ and water (W) whereas Groups II, IV, VI, and VIII received CBZ 30 minutes after a dose of LJ, GFJ, M, and BT, respectively. Groups III, V, VII, and IX received LJ, GFJ, M, and BT, respectively.

### 2.6. Serum biochemistry

Serum samples collected on Day 0, Day 2, Day 15, and Day 29 were analyzed for serum glutamic pyruvic transaminase (SGPT), serum glutamic oxaloacetic transaminase (SGOT), alkaline phosphate (ALP), total protein (TP), albumin (ALB), albumin/globulin ratio (A/G), blood urea nitrogen (BUN), and creatinine (CR) using a Microlab-300 auto-analyzer.

### 2.7. Histopathological study

On Day 29, the animals were sacrificed by cervical decapitation under anesthesia. The liver and kidney tissues were fixed in neutral phosphate-buffered 10% formalin solution and kept in a fridge until the tissues were prepared for histological examination. The tissues were processed, embedded in paraffin, and sectioned at 3–5 μm. The sections were examined after staining with hematoxylin and eosin (H&E).

### 2.8. Pharmacokinetic experiment

On Day 1 and Day 28, each animal of Groups I, II, IV, VI, and VIII was anesthetized and blood was collected from the retro-orbital plexus or tail vein into tubes containing EDTA at time points of 0 hours, 0.25 hours, 0.50 hours, 1 hour, 3 hours, 6 hours, and 8 hours. The samples were immediately centrifuged at 1150*g* at 15°C for 5 minutes and the plasma was separated. The plasma sample collected was stored at −20°C until analysis.

The standard stock solution of analyte was prepared by dissolving the accurately weighed CBZ in dimethyl sulfoxide (DMSO) to get a final concentration of 2 mg/mL. The working stock solutions for the calibration curve were 4.88 μg/mL, 9.77 μg/mL, 19.53 μg/mL, 39.06 μg/mL, 78.125 μg/mL, 156.25 μg/mL, 312.5 μg/mL, 625 μg/mL, 1250 μg/mL, and 2500 μg/mL in methanol. A 10-point standard curve was prepared by spiking 2 μL of the working stock to 98 μL of blank plasma to get final concentrations of 0.1 μg/mL, 0.2 μg/mL, 0.4 μg/mL, 0.8 μg/mL, 1.56 μg/mL, 3.125 μg/mL, 6.25 μg/mL, 12.5 μg/mL, 25 μg/mL, and 50 μg/mL, respectively. The internal standard (Valdecoxib, 20 μg/mL) solution was made in the 100% acetonitrile that had been used to precipitate the protein samples. All stocks were stored in polypropylene tube vials at 2–8°C. The plasma sample aliquot was taken in a microcentrifuge tube and then direct precipitation of the matrix was performed by adding ice-cold acetonitrile (3 × the sample solution) containing the internal solution. The mixture was vortexed in a cyclomixer for 5 minutes and centrifuged at 1500*g* for 10 minutes at 15°C; 120 μL of the clear supernatant was collected and 20 μL of it was used for HPLC analysis.

### 2.9. Pharmacodynamic analysis/PTZ-induced convulsion

Male Swiss albino mice were grouped into six ($n = 9$). Group I received PTZ at a dose of 60 mg/kg intraperitoneally (i.p.) [8] and Group II received CBZ and PTZ. Groups III, IV, V, and VI were administered CBZ and PTZ along with LJ, GFJ, M, and BT, respectively. Group II received PTZ 30 minutes after administration of CBZ, and Groups III, IV, V, and VI received PTZ 1.5 hours after CBZ. This was in accordance with the $T_{max}$ obtained from the plasma concentration time profile of rats. The dose of CBZ was 52.86 mg/kg per os (orally, p.o.; equivalent to 4.29 mg/kg in humans). The onset of tremor, piloerection, and convulsions, the number of convulsions, the recovery from the convulsive phase, and the subsequent mortality were recorded for experimental animals as well as the control group.

### 2.10. Data analysis

Results were expressed as mean ± standard error of the mean (SEM; $n = 9$). Statistical analyses were performed with one-way analysis of variance followed by the post-hoc Dunnett's test; $p < 0.05$ was considered to be statistically significant. To determine the toxicity and pharmacokinetic parameters of CBZ, the concentration–time data were analyzed by the noncompartmental analysis method. The area under the plasma concentration–time curve (AUC) from 0 hours to 8 hours after administration was calculated by the linear trapezoidal method. Mean plasma concentration–time profiles of CBZ per dose group were obtained by averaging the individual concentrations per sampling time point and by plotting the mean concentrations versus the sampling time point. The $C_{max}$ was defined as the highest observed concentration of CBZ in plasma. The $T_{max}$ was defined as the time to reach the $C_{max}$. Half-lives were determined from rate constants using the relationship: $t_{1/2} = \ln 2/k$.

## 3. Results

### 3.1. Serum biochemistry

SGOT, SGPT, CR, and BUN were significantly increased whereas TP was significantly decreased in rats belonging to Groups II and IV. There was a significant elevation in SGPT and CR in rats belonging to Group VI. However, it was observed that significant changes in serum biochemistry parameters were higher in Groups II and IV in comparison to Group VI. Thus, LJ and GFJ caused a greater change in parameters than M. No significant changes were observed in other groups. The data for Day 0 and Day 29 are represented in Table 1.

### 3.2. Morphologic pathology

Photomicrographs of the liver (Fig. 1) and kidney (Fig. 2) samples showed normal architecture in Group I (CBZ + W) rats. Significant toxic changes were observed in liver and kidney of rats treated with CBZ along with LJ, GFJ, and M. In Group II (CBZ + LJ), histological study of the liver revealed fatty changes, pyknotic nucleus, nuclear vacuolation, central vein congestion, peri-biliary infiltration, and widening of the sinusoidal space. Histological study of the kidney revealed pyknotic necrosis of the nucleus, necrosis of the tubular epithelium, and necrosis of the epithelial cell lining of the glomerulus. In Group IV (CBZ + GFJ), histopathology of the liver showed hepatocyte and hepatic plate disruption, hemorrhage from the central vein, necrotic and degenerative changes, focal fibrosis, and fatty changes in the peri-biliary area. Histological study of the kidney expressed focal necrosed glomeruli, hemorrhage, focal epithelial swelling, Hyalinization of the focal tubule, degenerative changes of the epithelium in the distal convoluted tubule, and swelling. In Group VI (CBZ + M), liver tissues showed biliary hyperplasia, eosinophilia, necrotic changes, granular cytoplasm, cell disruption, and fatty changes whereas kidney tissues revealed hemorrhage, protein cast, coagulative necrosis, and severe hemorrhage. There were no significant histopathological changes in the liver and kidney tissues in Group VIII (CBZ + BT).

### 3.3. Pharmacokinetic analysis

Significant increases ($p < 0.05$) in the $T_{max}$ and $AUC_{0-8}$ of CBZ in rats belonging to Groups II, IV, VI, and VIII were observed in comparison to rats belonging to Group I, both on Day 1 and Day 28 (Table 2), whereas the $C_{max}$ values of Groups II and IV were significantly increased on Day 1. Therefore, beverages increased the bioavailability of CBZ (AUC increased) when both were administered p.o. concomitantly. However, when we compared the $C_{max}$, $T_{max}$, and $AUC_{0-8}$ of Day 1 with the corresponding $C_{max}$, $T_{max}$, and $AUC_{0-8}$ of Day 28 of the same group, we observed that there was a decrease in $C_{max}$ and $AUC_{0-8}$ and an increase in $T_{max}$. The plasma concentration time profiles for Day 1 and Day 28 are represented in Figs. 3 and 4, respectively.

**Table 1 – Biochemical analysis of blood serum after administration of CBZ with W, LJ, GFJ, M, or BT.**

| Parameter | Day 0 | | | | | Day 29 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | CBZ + W | CBZ + LJ | CBZ + GFJ | CBZ + M | CBZ + BT | CBZ + W | CBZ + LJ | CBZ + GFJ | CBZ + M | CBZ + BT |
| SGOT | 130.33 ± 17.01 | 131.25 ± 17.61 | 131.11 ± 17.65 | 136.67 ± 18.40 | 132.44 ± 18.49 | 141.11 ± 18.41 | 286.52 ± 40.90* | 270.00 ± 38.45* | 156.89 ± 23.4 | 129.44 ± 15.95 |
| SGPT | 51.89 ± 3.91 | 55.89 ± 7.25 | 45.22 ± 5.82 | 42.78 ± 5.58 | 49.89 ± 3.74 | 56.56 ± 4.65 | 138.52 ± 18.80* | 83.67 ± 9.37* | 72.11 ± 10.78* | 49.22 ± 4.26 |
| ALP | 31.33 ± 3.83 | 30.78 ± 3.94 | 29.44 ± 3.42 | 28.22 ± 2.98 | 33.89 ± 5.48 | 33.33 ± 4.06 | 45.78 ± 7.81 | 41.44 ± 5.98 | 36.56 ± 4.86 | 32.89 ± 5.04 |
| TP | 7.42 ± 1.05 | 7.40 ± 0.72 | 7.51 ± 0.93 | 8.00 ± 0.83 | 7.54 ± 0.79 | 6.84 ± 0.84 | 4.74 ± 0.43* | 4.71 ± 0.63* | 6.67 ± 0.91 | 7.88 ± 1.38 |
| ALB | 3.84 ± 0.46 | 3.90 ± 0.52 | 3.42 ± 0.47 | 3.73 ± 0.42 | 3.73 ± 0.48 | 3.52 ± 0.39 | 2.97 ± 0.24 | 2.72 ± 0.40 | 3.02 ± 0.22 | 3.90 ± 0.64 |
| A/G | 1.45 ± 0.26 | 1.53 ± 0.18 | 1.43 ± 0.27 | 1.34 ± 0.19 | 1.48 ± 0.26 | 1.37 ± 0.28 | 2.05 ± 0.27 | 1.67 ± 0.29 | 1.56 ± 0.15 | 1.55 ± 0.25 |
| CR | 0.35 ± 0.04 | 0.34 ± 0.05 | 0.36 ± 0.04 | 0.31 ± 0.04 | 0.30 ± 0.04 | 0.38 ± 0.06 | 2.58 ± 0.43* | 2.57 ± 0.04* | 2.10 ± 0.28* | 0.31 ± 0.05 |
| BUN | 15.48 ± 2.54 | 15.96 ± 1.59 | 17.62 ± 2.56 | 13.36 ± 1.34 | 17.62 ± 1.85 | 16.39 ± 1.98 | 24.64 ± 2.94* | 25.86 ± 3.05* | 18.66 ± 2.29 | 18.54 ± 1.98 |

Data are presented as mean ± SEM ($n = 9$).
*$p < 0.05$ versus CBZ + W group.
A/G = albumin/globulin ratio; ALB = albumin; ALP = alkaline phosphate; BT = black tea; BUN = blood urea nitrogen; CBZ = carbamazepine; CR = creatinine; GFJ = grapefruit juice; LJ = lime juice; M = milk; SEM = standard error of the mean; SGOT = serum glutamic oxaloacetic transaminase; SGPT = serum glutamic pyruvic transaminase; TP = total protein; W = water.

### 3.4. *PTZ-induced convulsions in mice*

The mice of Groups III–VI showed significant delayed onset of tremor and piloerection, and episodes of convulsion were absent when compared with Group I. The disappearance of signs and symptoms of convulsion were also significantly faster in Groups III–VI than in Group I. No mortality was observed in Groups III–VI. The anticonvulsive effect against PTZ-induced convulsion was found to be absent in Group II (CBZ + PTZ). However, the increased bioavailability of CBZ in Groups III–VI probably resulted in the anticonvulsive effect of CBZ against PTZ-induced convulsion at the same dose. Data relating to the effect of the beverages on the anticonvulsive effect of CBZ are represented in Table 3.

## 4. Discussion

CYP enzymes are important for the metabolism of drugs and xenobiotics. This is particularly important for drugs with a narrow therapeutic index that are generally advocated for chronic treatment where the change in bioavailability may eventually affect efficacy and toxicity [9–11]. It has also been documented that such drugs often present interaction with some commonly consumed foodstuffs having relevance to involvement of similar CYP enzyme isoforms [12]. CBZ is one such drug; it is used for epilepsy on a long-term basis with reported clinically relevant liver toxicity [13]. During long-term drug therapy, restrictions regarding food are not convenient as far as patient compliance is concerned. Accordingly, this study concerning the interaction of CBZ with common beverages (LJ, GFJ, BT, and M) was undertaken. CBZ is a known substrate of CYP3A4 [14] whereas the beverages LJ and GFJ are known inhibitors of the same isoform [15,16]. In our study, beverages were co-administered on a chronic basis along with CBZ and some general indicators of toxic manifestations relevant to CBZ were monitored in experimental animals. It was found that LG and GFJ in combination with CBZ resulted in significant toxicity with particular reference to reduction of body weight, food and water intake, and increases in hepatic markers namely SGPT, SGOT, TP, CR, and BUN. Toxicity was also confirmed through histopathological studies of kidney and liver samples in the affected groups of animals. Histological study of the liver revealed fatty changes, pyknotic nucleus, nuclear vacuolation, central vein congestion, peri-biliary infiltration, and widening of the sinusoidal space while that of the kidney revealed focal necrosed glomeruli, hemorrhage, focal epithelial swelling, hyelinization of the focal tubule, degenerative changes of the epithelium in the distal convoluted tubule, and swelling. The observed toxicity complies with the reported toxic manifestation of CBZ that might have occurred due to the reduced activity of CYP3A4, eventually leading to increased bioavailability of CBZ in the presence of LJ or GFJ. [It has been reported that pomegranate juice increases the bioavailability of CBZ in rats [12].]

Accordingly, an attempt was made to justify the above apprehension where the bioavailability of CBZ in the presence of beverages, namely LJ, GFJ, M, and BT, was quantitatively analyzed using HPLC. Our observation indicated a significant shift in the $C_{max}$ as well as $T_{max}$ of CBZ in the presence of LJ or

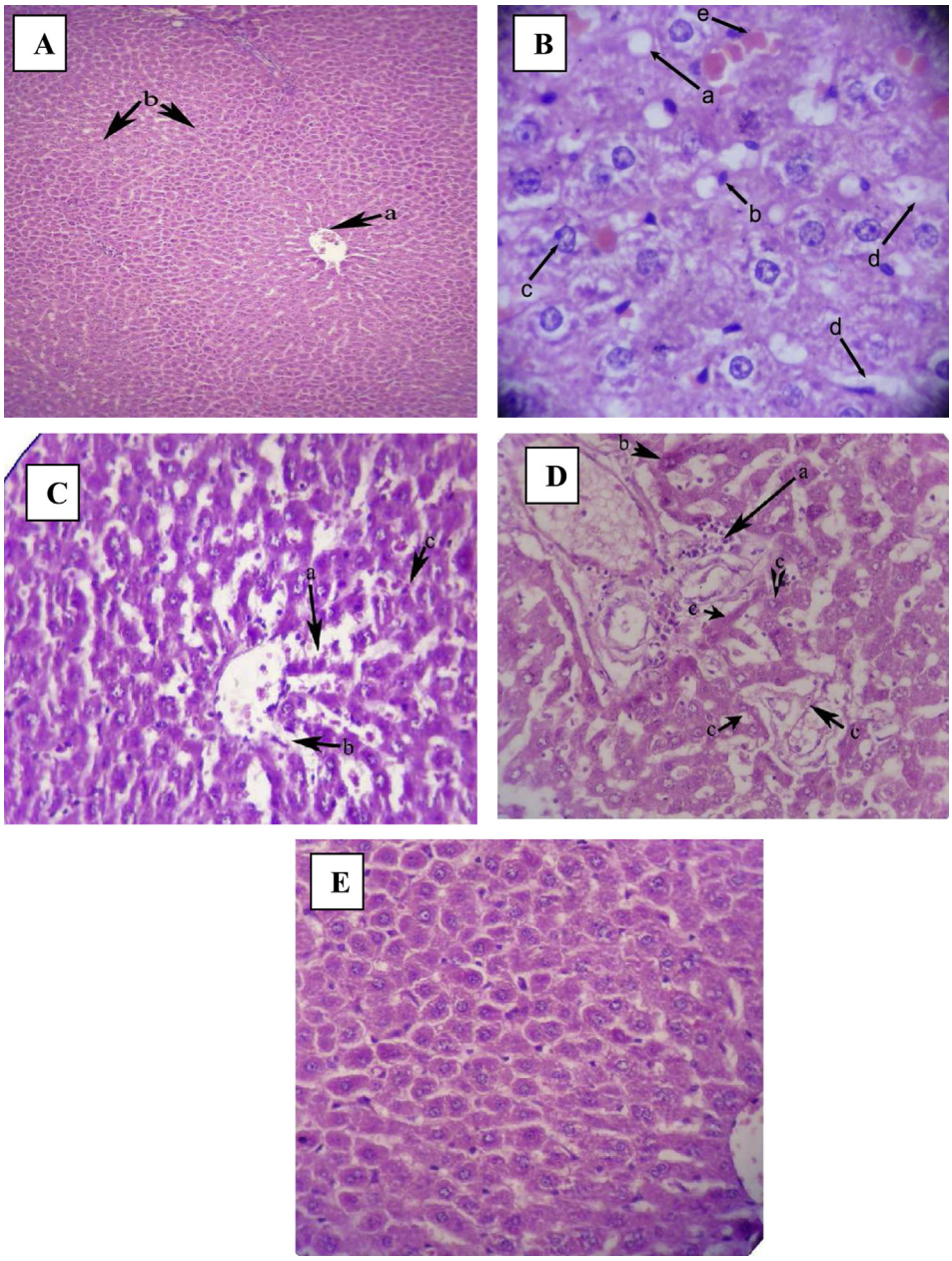


**Fig. 1 – Representative histopathological picture of the male rat liver (H&E, 10×). (A) CBZ and W: (a) central vein and (b) normal cells. (B) CBZ and LJ: (a) fatty changes; (b) pyknotic nucleus; (c) nuclear vacuolation; (d) widening of the sinusoidal space; and (e) central vein congestion. (C) CBZ and GFJ: (a) hepatocyte and hepatic plate disruption; (b) hemorrhage from the central vein; and (c) necrotic and degenerative changes. (D) CBZ and M: (a) biliary hyperplasia; (b) increased eosinophilia; and (c) necrotic changes. Fatty changes are present in the entire area. (E) CBZ and BT leaf extract. BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; H&E = hematoxylin and eosin; LJ = lime juice; M = milk; W = water.**

GFJ, although the maximum shift was found with LJ (at least in our experimental conditions).

Attempts were also made to investigate the anticonvulsant effects owing to the observed increase in bioavailability of CBZ in the presence of LJ or GFJ. Accordingly, the antiepileptic activity of the said drug against PTZ-induced seizure in experimental animals was measured. It was observed that in the presence of LJ or GFJ the onset of tremor and piloerection was significantly delayed, the disappearance of signs and symptoms of seizures was significantly shortened, and episodes of seizures and even mortality were found to be completely absent as compared to the control group of animals.

In this context, it is interesting to consider that similar effects on CBZ were also observed with BT or M, although these beverages are not reported to be significantly potent CYP3A4 inhibitors; however, a few reports are available in support of

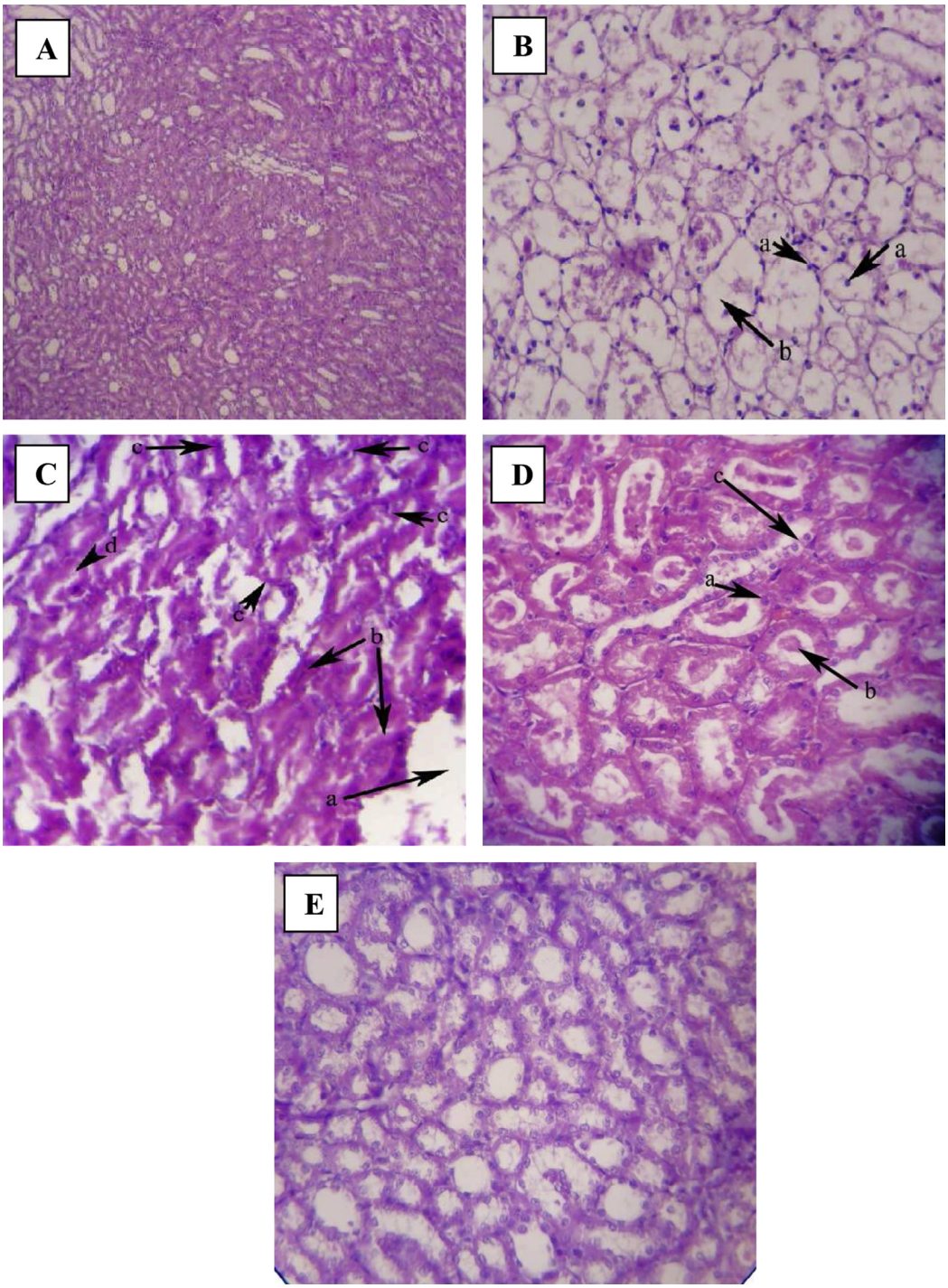


**Fig. 2 – Representative histopathological picture of the male rat kidney. (A) CBZ and W (H&E, 10×). (B) CBZ and LJ (H&E 40×): (a) pyknotic nucleus of the necrotic cell and (b) necrosis of the tubular epithelium. (C) CBZ and GFJ (H&E 40×): (a) focal epithelial swelling; (b) hyelinization of the focal tubule; (c) degenerative changes of the epithelium in the distal convoluted tubule; and (d) swelling. (D) CBZ and M (H&E 40×): (a) hemorrhage; (b) protein cast; and (c) coagulative necrosis. (E) CBZ and BT leaf extract (H&E 40×). BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; H&E = hematoxylin and eosin; LJ = lime juice; M = milk; W = water.**

this theory [17,18]. It is important to mention that the solubility of CBZ is known to increase with an increase in pH, which might be a possible reason for enhanced bioavailability of CBZ when concomitantly administered with M [19,20]. Interestingly enough, the increased bioavailability-related toxicity of CBZ was found to be absent with BT, which might have some relation to the *in vivo* antioxidant property of BT [21,22] while similar protection was found to be absent in M. Therefore,

**Table 2 – Pharmacokinetic parameters of CBZ after administration of CBZ with W, LJ, GFJ, M, or BT on Day 1 and Day 28.**

| | | Pharmacokinetic parameters of CBZ | | | | |
|---|---|---|---|---|---|---|
| | | Group I | Group II | Group IV | Group VI | Group VIII |
| | | CBZ + W | CBZ + LJ | CBZ + GFJ | CBZ + M | CBZ + BT |
| $C_{max}$ (μg/mL) | Day 1 | 26.40 ± 2.54 | 32.26 ± 2.84* | 30.98 ± 2.49* | 28.97 ± 3.16 | 28.13 ± 3.44 |
| | Day 28 | 21.88 ± 2.59 | 18.47 ± 3.15 | 17.01 ± 2.46 | 17.49 ± 1.88 | 17.05 ± 1.67 |
| $T_{max}$ (h) | Day 1 | 0.5 ± 0.00 | 1.5 ± 0.00 | 1.5 ± 0.00 | 1.5 ± 0.00 | 1.5 ± 0.00 |
| | Day 28 | 0.25 ± 0.00 | 3 ± 0.00 | 3 ± 0.00 | 3 ± 0.00 | 3 ± 0.00 |
| $AUC_{0-8}$ | Day 1 | 147.79 ± 4.69 | 185.50 ± 5.31* | 177.16 ± 3.88* | 173.2 ± 4.62* | 168.75 ± 5.02* |
| | Day 28 | 72.35 ± 4.36 | 96.83 ± 5.16* | 92.84 ± 4.36* | 87.28 ± 3.84* | 90.56 ± 4.44* |

Data are presented as mean ± SEM ($n = 9$).
*$p < 0.05$ versus CBZ + W group.
AUC = area under the plasma concentration–time curve; BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; LJ = lime juice; M = milk; SEM = standard error of the mean; W = water.

among the various combinations of beverages and CBZ used in this study, BT appears to be comparatively safe.

Thus, from our observation, the presence of LJ or GFJ along with CBZ leads to increased bioavailability of CBZ. This in turn may increase the probability of liver and kidney toxicity of the drug in a dose that is otherwise expected to be safe.

In summary, this study demonstrates that concomitant administration of beverages such as GFJ, LJ, M, and BT along with a narrow therapeutic index drug like CBZ may result in an increased bioavailability where subsequent monitoring of toxicity to the liver and kidney will be of clinical importance.

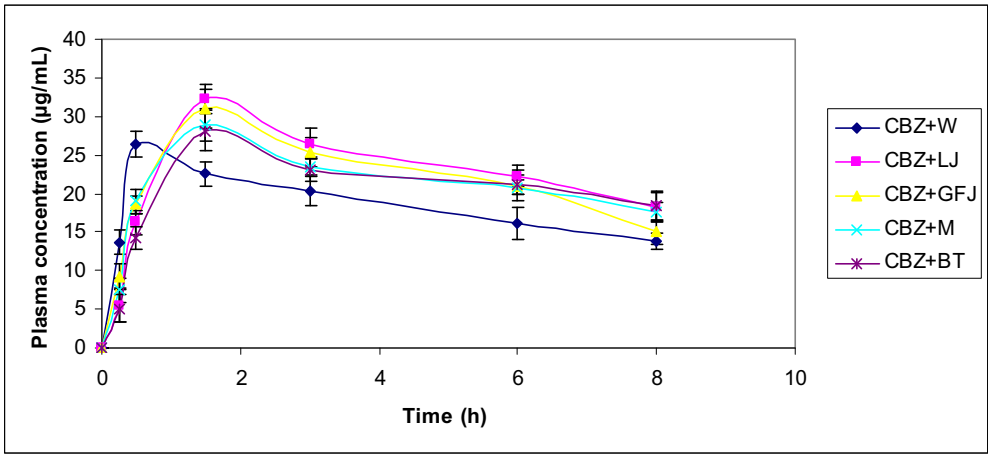


**Fig. 3 – Plasma concentration time profile of rats treated with CBZ and W, LJ, GFJ, M, or BT on Day 1. BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; LJ = lime juice; M = milk; W = water.**

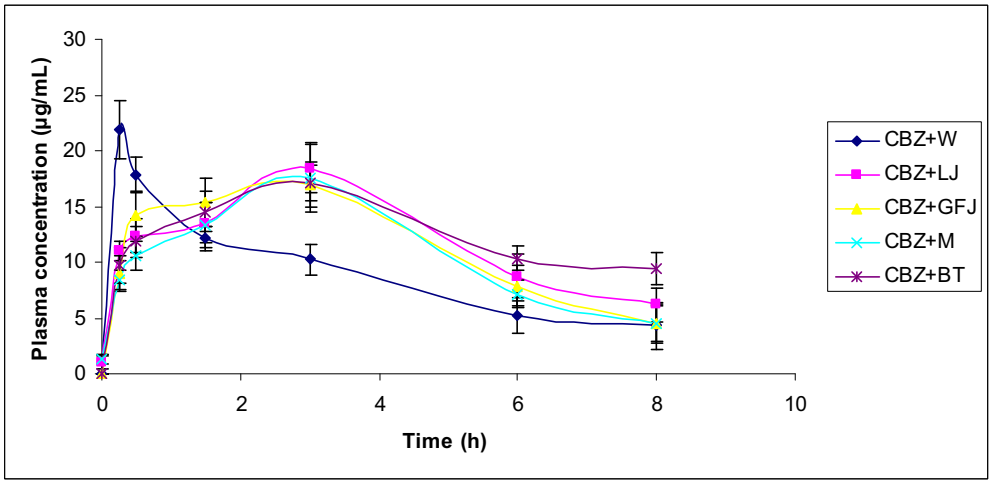


**Fig. 4 – Plasma concentration time profile of rats treated with CBZ and W, LJ, GFJ, M, or BT on Day 28. BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; LJ = lime juice; M = milk; W = water.**

**Table 3 – The effect of beverages on the anticonvulsive effect of CBZ against PTZ-induced convulsions.**

| Group | Onset of tremor (min) | Onset of piloerection (min) | Onset of convulsions (min) | Number of convulsions | Recovery from convulsive phase (min) | Mortality (%) |
|---|---|---|---|---|---|---|
| I (PTZ) | 0.83 ± 0.09 | 0.52 ± 0.08 | 12.667 ± 1.36 | 3.67 ± 0.38 | 73.25 ± 8.45 | 11.11 |
| II (CBZ + W + PTZ) | 1.40 ± 0.21 | 1.24 ± 0.16 | 11.56 ± 0.87 | 3.33 ± 0.33 | 68.63 ± 6.89 | 11.11 |
| III (CBZ + LJ + PTZ) | 6.90 ± 0.98* | 6.09 ± 0.86* | — | 0* | 38.56 ± 4.29* | 0 |
| IV (CBZ + GFJ + PTZ) | 2.94 ± 0.43* | 2.64 ± 0.29* | — | 0* | 45.00 ± 6.01* | 0 |
| V (CBZ + M + PTZ) | 2.77 ± 0.27* | 2.72 ± 0.38* | — | 0* | 43.67 ± 5.88* | 0 |
| VI (CBZ + BT + PTZ) | 2.60 ± 0.18* | 1.73 ± 0.22* | — | 0* | 48.44 ± 6.30* | 0 |

Data are presented as mean ± SEM ($n = 9$).
*$p < 0.05$ versus Group I.
BT = black tea; CBZ = carbamazepine; GFJ = grapefruit juice; LJ = lime juice; M = milk; PTZ = pentylene tetrazole; SEM = standard error of the mean; W = water.

## Conflicts of interest

The authors declare that there are no conflicts of interest.

## Acknowledgments

We would like to extend our sincere thanks to All India Council for Technical Education – Research Promotion Scheme (AICTE-RPS), the Government of India, University Grants Commission – Department of Special Assistance (UGC DSA), and University Grants Commission – Universities with Potential for Excellence Phase II (UGC UPE-II) for providing the financial support needed for this study.

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