MATERIALS

The following materials were used as procured from their manufacturer: metephosphoric acid, sodium hydroxide, acetic acid (Fyrite Chemical Co., St. Louis, Mo), sodium nitroprusside (Roche Prod., Velwex Garden, U.S.A.), INH powder, and tablets (Ciba-Geigy, Zurich, Switzerland) and CP tablets (Ciprobloc, Indonesia Pharm. Co., Jakarta).

STUDY DESIGN

The study was in two phases and the two phases were separated by a 1-week drug washout. Six healthy female volunteers of age in the range of 15 to 22 years and weights in the range of 45 to 72 kg participated in the study. The volunteers were not taking any prescribed or over-the-counter medications. They were non-smokers, had no history of tuberculosis or leprosy, and had not consumed INH or CP within 2 months before the commencement of the study. The study design met the requirements of good clinical practice.

FIRST PHASE

During the first phase, each subject received 300 mg of INH in tablet form, with 400 ml of spring water (Evian pure water, NLC, Lagos, Nigeria) after an overnight fast. Food was restricted for 4 hours after the drug ingestion period. A 5-ml quantity of blood was collected at 0, 1, 2, 3, 4, 5, 6, 7, and 12 hours and at the 24th hour in 5 ml of spring water. The blood samples were preserved in ethylenediaminetetraacetic acid bottles and centrifuged 1 hour after collection at 3000 rpm for 5 minutes. A 2 ml quantity of plasma from each sample was harvested into a plastic test tube. The plasma samples were kept frozen at -20°C until analysis.

SECOND PHASE

In the second phase, each subject received, in addition to 300 mg of INH, a 500-mg tablet of CP (Ciprobloc) concurrently after an overnight fast. The same protocol as in the first phase was followed.

SAMPLE ANALYSIS

Plasma INH concentration was analyzed using the method of indifference and linearity. In each case, a 2-ml plasma sample was added to a test tube followed by the addition of 4 ml of purified water. This was vortex mixed. A 2-ml volume of 20% metephosphoric acid was added and the mixture was vortex mixed. This was followed by centrifugation at 3000 rpm for 5 minutes, A 2-ml supernatant was pipetted into a 10-ml glass cuvette. A 2-ml volume of 2 M acetic acid solution was added to the cuvette followed by 2 ml of a chromogenic reagent (equal volume of 2% sodium nitroprusside and 4 M sodium hydroxide freshly prepared). After standing for 2 minutes, the absorbance of the sample was read at 440 nm in a Spectronic 20 colorimeter. Sample absorbances were converted to concentration from Beer's calibration curve previously determined.

ANALYSIS OF DATA

To analyze the data, it was assumed that INH absorption and elimination followed first-order kinetics at all concentrations encountered. The differences between sample means were analyzed using Student t-tests for paired data with statistical significance defined as P = 0.05. For each subject, a plasma concentration versus curve (AUCt0-t) was calculated using trapezoidal rule and the elimination rate constant (K) was eliminated from a graphical representation of equation using the line of best fit of four points in the elimination phase.

Cp = Cpo e^-kt

Where Cp and Cpo are plasma concentration of INH at time t and t = 0. INH half-life (t1/2) was obtained from equation 2.

1 0.693
12 = K

Peak plasma concentration (Cmax) and time for peak plasma concentration (tmax) were estimated from the plasma level-time curves.

RESULTS

Some important pharmacokinetic parameters of INH in the absence and in the presence of the coadministered CP are shown in Table 1. There was an increase of about 15% in the extent of the absorption of INH (AUCt0-t) in the presence of CP. There was, however, a slight reduction in the peak plasma concentration (Cmax) concentration of INH and a 1-hour shift in the time to attain peak plasma concentration (tmax) (Fig. 1). The
LEONARDO AND FLUIDOGENICINES

Table 1. Important pharmacokinetic parameters of INH in the healthy female volunteers.

<table>
<thead>
<tr>
<th>Pharmacokinetic parameters</th>
<th>INH</th>
<th>INH + CP</th>
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<tbody>
<tr>
<td>AUC (µg/mL)</td>
<td>60.55 ± 33.69</td>
<td>60.64 ± 22.48</td>
</tr>
<tr>
<td>t1/2 (h)</td>
<td>6.127 ± 0.020</td>
<td>6.115 ± 0.006</td>
</tr>
<tr>
<td>tmax (h)</td>
<td>5.44 ± 0.660</td>
<td>6.92 ± 0.222</td>
</tr>
<tr>
<td>Cmax (µg/mL)</td>
<td>10.55 ± 0.111</td>
<td>9.20 ± 0.508</td>
</tr>
<tr>
<td>C (µg/mL)</td>
<td>3.0</td>
<td>4.0</td>
</tr>
</tbody>
</table>

![Graph showing plasma level-time curves of leonardo (INH) and drug absorption and excretion in the presence of CP.](image)

**DISCUSSION.**

A survey of the above shows that information is sparse on the effect of pharmacokinetics and potential interaction in humans. Currently, isoniazid, another antituberculous drug, has been shown to increase the metabolism of CP in the rabbit. It is evident from our study that the presence of CP delayed the rate of absorption of INH, resulting in a reduction in the Cmax and a 1-hour increase in the t1/2. Because INH is not appreciably bound to plasma proteins, protein-binding interaction is ruled out. A form of pharmacokinetics interaction (absorption interaction) may have taken place between the two drugs. There are several mechanisms by which the absorption of a drug may be altered by another drug. These include decrease in gastrointestinal motility caused by morphine-like drugs and drugs with anticholinergic effects, such as the tricyclic antidepressants, and chelation of calcium, aluminium, magnesium, and iron salts by tetracyclines. In a recent study, CP was found to inhibit cholinergic neurotransmission, which resulted in part from stimulation of M3, K+ ATPase activity and the subsequent repolarization/hyperpolarization of postganglionic nerve fiber. It is possible that the effect of CP on INH absorption resulted from the drug's inhibiting action on gastric motility, which is capable of delaying gastric emptying.

The pHs of 1%, 5%, and 10% solution of INH in water are 5.5 to 6.5, 6 to 8, and 6 to 7.5, respectively. It has an amine group in its structure, which makes it slightly basic. Its absorption is expected to be maximally absorbed in the upper part of the small intestine. This further shows that the rate of gastric emptying affected its onset of absorption. Because INH is readily absorbed orally, delay in gastric emptying will affect only the rate and not the extent of its absorption, as the current study has shown. Peak plasma concentrations of INH of about 8 mg/mL appear in blood, 1 to 2 hours after a fasting dose of 300 mg by mouth. The current study shows peak plasma concentration of 10.50 µg/mL for INH + CP. Even though the difference between these two values is statistically significant (P = 0.02), the decrease in the peak plasma concentration in INH in the presence of CP may not be of clinical significance in terms of outcomes where the two drugs are combined in the treatment of tuberculosis. However, because INH is a known inhibitor of drug metabolism, its coadministration with CP in patients would be expected to produce greater variability than that which could be observed in patients not receiving potentially interacting drugs.

The presence of CP did not hasten the elimination of INH. This may well indicate that acetylation of INH was not accelerated in the presence of CP. There was a slight increase in the % of INH in the presence of CP. These data strengthen the earlier observation that the interaction between INH and CP in vivo may not result in recognizable clinical consequences. This
study supports the general consensus that CP is a relatively safe drug. None of the subjects complained of any unusual response after ingesting the two drugs concurrently. Results of the current study indicate that CP may be a suitable thioquinoxolone antibacterial agent to be combined with INH as the first-line agents in the treatment of tuberculosis because of its bactericidal activity against mycobacteria. Its capacity to act against intracellular pathogens, and its excellent tissue penetration without risk of toxicity, therapeutic failure, or any reaction of clinical consequence. The effect of CP on the urinary and salivary excretions of INH is currently being assessed in the same female volunteers, and this will constitute a separate report. This was intended to validate the results obtained from the plasma samples and to further prove the safety in coadministering the two drugs to patients. Our findings, however, may require knowledge of the effect of INH on CP pharmacokinetics before a firm conclusion can be drawn. This is also being investigated.

REFERENCES


Some Plasma Pharmacokinetic Parameters of Isoniazid in the Presence of a Fluoroquinolone Antibacterial Agent

Sobusua L. Olufolalu,1 Chioma E. Ohudo,1 Orieh E. Osisakwe,2 Nnidihamaka A. Iondu,2 Ogenmoji J. Monjor,2 Steve O. Maduka,3 Chikere A. Anu fun,2 and Patrick U. Agbasi2

The effects of ciprofloxacin (CP), a fluoroquinolone antibacterial agent, on the extent of absorption of Isoniazid (INH) and on some of its pharmacokinetic parameters were investigated in six healthy female volunteers between the ages of 22 and 32 years. The presence of CP led to increase in the amount of INH and to a slight reduction in its peak plasma concentration (Cmax). There was a

1-hour increase in the time to attain Cmax (Tmax) of INH, indicating absorption interaction between the two drugs. This absorption interaction was related to inhibition of cholinergic neurotransmission caused by CP, which is capable of inhibiting gastric motility, leading to a delay in gastric emptying. The rate of elimination (β) and plasma half-life (t1/2) of INH were not significantly affected (P > 0.05). The extent of absorption interaction that may have occurred (based on values of 2-hour values for area under the concentration curve, Cmax, Tmax, β, and t1/2) was considered to be of no therapeutic consequence and the coadministration of the two drugs may be recommended in clinics of practice.

Keywords: fluoroquinolone, isoniazid, ciprofloxacin, plasma level-time curve, pharmacokinetic parameters.