Kinetic Studies of the Interaction Between Isoniazid and Reducing Sugars

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Abstract ☐ The interaction between isoniazid and reducing sugars is acid-catalyzed and reversible. Kinetic studies of hydrazone formation from isoniazid and glucose, lactose, maltose, and galactose have been carried out in simulated gastric juice at 37°C. The forward reaction was found to follow second-order kinetics, while the reverse reaction, the hydrolysis of the sugar isonicotinoyl hydrazone, is pseudo-first-order. The effects of the concentration of reactants, pH, and temperature on the rate have been studied, and the rate constants and the energy of activation were determined.

Isoniazid is incompatible with aldehydes and ketones.[1] It is also known to react with reducing sugars to form hydrazones.[2] Earlier, the solid-state interaction of isoniazid with lactose had been reported.[3] The interaction between isoniazid and sugar adjuvants in liquid and solid dosage forms has been studied.[4-7] Isoniazid has been found to condense with glucose in a commercial isoniazid syrup to the extent of 60–70%.[4] The presence of lactose isonicotinoyl hydrazone has been estimated in lactose-containing isoniazid tablets by many workers.[5-7] Samples of isoniazid tablets have been found to contain lactose isonicotinoyl hydrazone in amounts of 0.3–14%.[5,6] This reaction of isoniazid with lactose is more significant in tropical climates[3] where the hydrazone content in isoniazid tablets has been found to be as high as 22%.[7]

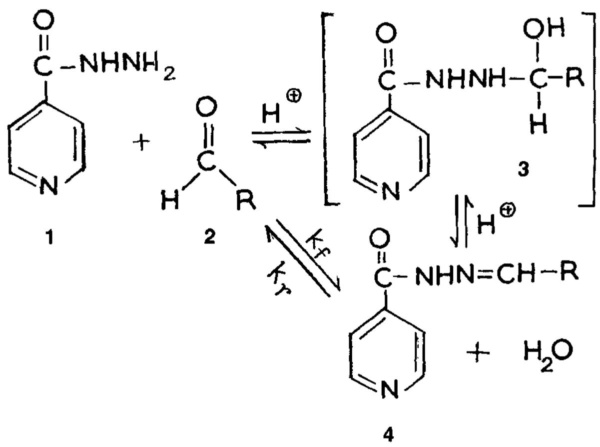
In an acidic medium, sugar isonicotinoyl hydrazones are known to hydrolyze to free isoniazid. However, an in vivo study in humans has revealed that the hydrolysis of glucose isonicotinoyl hydrazone is suppressed by the presence of glucose.[8] Furthermore, sugar isonicotinoyl hydrazones are known to be poorly absorbed from the GI tract.[4,8]

An earlier in vitro study indicated an interaction between isoniazid and reducing sugars present in such food materials as milk and fruit juices.[9] It was therefore of interest to investigate the factors affecting the formation of sugar isonicotinoyl hydrazones under physiological conditions and to study the possible impact of these substances on the bioavailability of free drug.

The present study reports the kinetics of the reaction of isoniazid with the sugars glucose, lactose, galactose, and maltose, in simulated gastric juice (pH 1.8), at 37°C. The effects of pH, concentration of reactants, and temperature on hydrazone formation have been studied. The rate constants of formation and hydrolysis of hydrazones have been determined.

Experimental Section

Materials—Isoniazid (B.P.) recrystallized from ethanol (mp 170–171°C), glucose, lactose, galactose, and pepsin from BDH, India; fructose from E. Merck; and maltose from Biddle Sawyer and Co., Holland, were employed in this study. All other reagents were of analytical grade. Glucose isonicotinoyl hydrazone[2] and lactose isonicotinoyl hydrazone[10] were prepared as described earlier and their purity was confirmed by TLC.[3,11]



Buffer Solutions—The buffer solutions used in the kinetic studies were HCl:NaCl (pH 1.2 and 1.8), citrate buffer (pH 2.6, 3.1, 3.5, 3.9, 5.1, and 6), and phosphate buffer (pH 7 and 8). The buffers were 0.1 M with respect to HCl, citrate, and phosphate ions (except when a buffer effect was investigated), and they were adjusted to an ionic strength of 0.5 with NaCl. The pH of each solution was measured using a Systronic's pH meter[4] equipped with a combined glass-calomel electrode.

Simulated gastric juice, modified from USP,[12] was prepared by dissolving NaCl (2 g), pepsin (3.2 g), and HCl (2 mL) in sufficient water to make 1000 mL. The solution had a pH of 1.8 ± 0.05. Preliminary studies indicated that the pepsin present in the simulated gastric juice had no appreciable effect on the rate constants.

Analytical Procedures—In the reaction mixtures of isoniazid with sugars, the unreacted isoniazid was determined spectrophotometrically, using 2,3-dichloro-1,4-naphthoquinone in the presence of ammonia:ammonium chloride buffer (pH 10.8).[13]

Preliminary Study—*Effect of pH on the Formation and Hydrolysis of Hydrazones*—In a series of 50-mL volumetric flasks, isoniazid (0.5 mmol) and the sugar (2.5 mmol) were dissolved and diluted to mark with buffer solution (pH 1.2–8.0), previously equilibrated at 37°C. The mixtures were placed in a constant-temperature water bath (Townson, Mercer Ltd., England) at 37 ± 0.1°C. At 1 h, 2 mL of sample solution was withdrawn and diluted to 50 mL with chilled water (3°C). A 4-mL aliquot of this solution was immediately transferred by pipet into a 25-mL volumetric flask. Ammonia:ammonium chloride buffer (1 M, pH 10.8; 1 mL) and 2,3-dichloro-1,4-naphthoquinone solution (0.03%, w/v in ethanol; 6 mL) were added, and the volume was adjusted with water. This mixture was allowed to stand at 0°C for 40 min. The absorbance was measured on a Beckman Model 25 spectrophotometer with two matched cells and a 1-cm light path at 610 nm, using a blank

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prepared by mixing appropriately diluted buffer solutions (4 mL), ammonia:ammonium chloride buffer (1 mL), and the reagent solution (6 mL) in a 25-mL volumetric flask, and adjusting the volume.

The amount of isoniazid was determined by comparing the absorbance of the sample solution with that of a standard, run simultaneously without adding the sugar. The decrease in the initial concentration of isoniazid was considered equivalent to the amount of hydrazone formed. The liberated isoniazid during the hydrolysis of sugar isonicotinoyl hydrazone (0.01 M solution) in buffers (pH 1.2–8.0) was determined as above.

Effect of the Concentration of Sugar on Formation and Hydrolysis of Hydrazone in Buffer pH 1.8—For the study of the hydrazone formation, isoniazid (0.5 mmol) and glucose or lactose (0.5–4.5 mmol) were transferred to 50-mL volumetric flasks. Similarly, for the study of the hydrolysis of the hydrazones, glucose isonicotinoyl hydrazone or lactose isonicotinoyl hydrazone (0.5 mmol) and glucose or lactose (0.5–4.5 mmol), respectively, were transferred to 50-mL volumetric flasks. The contents were dissolved in and diluted with buffer (pH 1.8). The mixture was maintained for 1 h at 37°C, and then a 2-mL aliquot was withdrawn and assayed for isoniazid as described above.

Kinetic Study—*Effect of the Concentration of Reactants on the Rate Constants*—Stock solutions of isoniazid and the sugars were prepared in simulated gastric juice and equilibrated at 37°C. The kinetic experiments were carried out by mixing the requisite volumes of both solutions and diluting with simulated gastric juice to obtain the initial concentrations of isoniazid (0.8×10^{-2} M to 1.2×10^{-2} M) and sugars (2.0×10^{-2} M to 15.0×10^{-2} M). The mixture was placed in a constant-temperature water bath at 37°C. Samples were withdrawn at 15-min intervals during 3 h and assayed for isoniazid, as described above. Similarly, the kinetics of hydrolysis of glucose or lactose isonicotinyl hydrazone (2×10^{-4} M to 1.2×10^{-2} M solution) in simulated gastric juice was studied at 37°C.

Effect of Temperature on the Rate of Reaction—The rate of reaction of isoniazid (0.5 mmol) with glucose or lactose (2.5 mmol) in simulated gastric juice (50 mL) was determined at different temperatures.

Effect of Buffer Concentration on the Rate of Reaction—The catalytic effect of the buffer concentration on the rate of reaction of isoniazid (0.5 mmol) with glucose (2.5 mmol) was determined at constant temperature (37°C), ionic strength (μ = 0.5) and two pH values (2.6 and 3.1), with only the buffer concentration varying (0.05–0.2 M).

Effect of pH on the Rate of Reaction—The pH dependency on the rate of hydrazone formation of isoniazid (0.5 mmol) and glucose or lactose (2.5 mmol) was studied in 0.1 M buffers (pH 1.2–5.0; μ = 0.5) and at 37°C.

Results and Discussion

Preliminary experiments to study the effect of pH on hydrazone formation were carried out by allowing isoniazid and the sugars to react in different buffer solutions. Plot of percentage hydrazone formed versus pH (Fig. 1) shows that the reaction of isoniazid with sugars takes place only in an acidic medium (pH 1.0–6.0). The maximum amount of hydrazone is formed at pH 3.1. It is noteworthy that under experimental conditions, the formation of fructose isonicotinoyl hydrazone is negligible in the entire pH range.

It is evident from Fig. 2 that the hydrolysis of glucose and lactose isonicotinoyl hydrazones occurs in an acidic medium (pH 1.0–6.0). The hydrolysis of hydrazone decreases with the decrease in hydrogen-ion concentration. The results conform with the observations of the earlier workers.[8] As observed in Fig. 3, the hydrazone formation of isoniazid with glucose and lactose increases proportionately with the increase in the concentration of these sugars, whereas, as indicated in Fig. 4, the hydrolysis of glucose isonicotinoyl hydrazone and lactose isonicotinoyl hydrazone decreases with the increase in the concentration of sugars.

As the aim of the present work was to investigate the interaction of isoniazid with sugars under physiological conditions, the detailed kinetics of hydrazone formation and hydrolysis were studied in simulated gastric juice (pH 1.8) at 37°C.[8] The initial concentrations of the reactants for the kinetic study of hydrazone formation were selected on the basis of the normal oral dose of isoniazid and the availability of sugars from food.

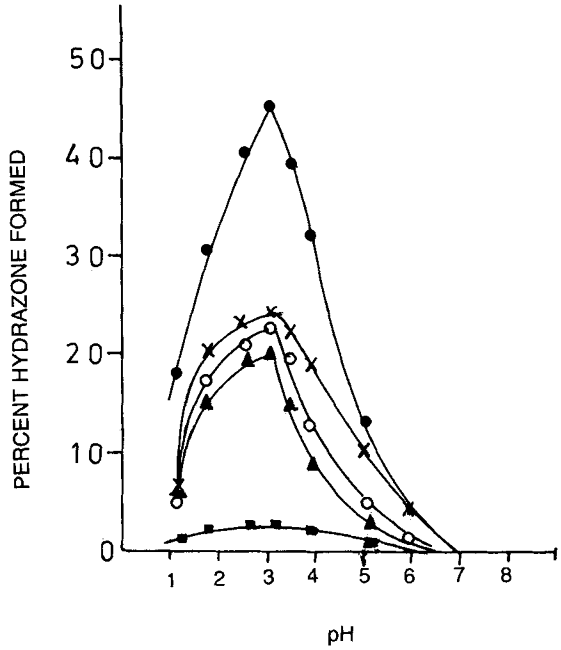

Figure 1—*Hydrazone formation of isoniazid with sugar in buffers (0.1 M; μ = 0.5) at various pH; [Isoniazid] = 1×10^{-2} M; [Sugar] = 5×10^{-2} M; temp. = 37°C; time = 1 h. Key: (●) galactose; (×) lactose; (○) glucose; (▲) maltose; (■) fructose.*

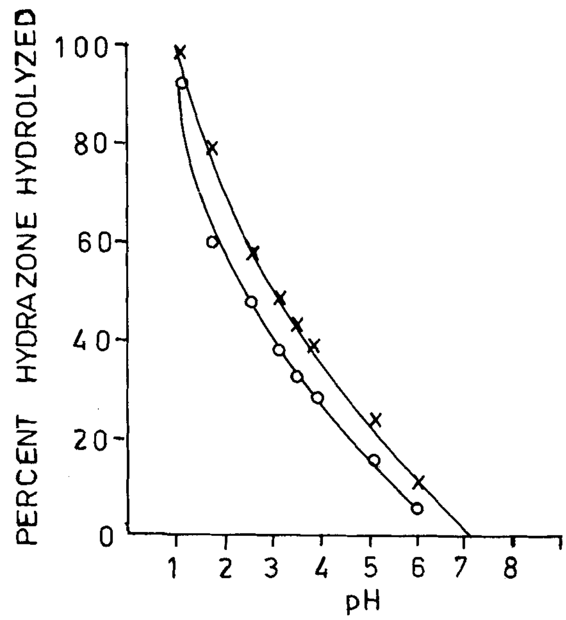

Figure 2—*Hydrolysis of sugar isonicotinoyl hydrazone in buffers (0.1 M; μ = 0.5) at various pH; temp. = 37°C; time = 1 h. Key: (○) glucose isonicotinoyl hydrazone (1×10^{-2} M); (×) lactose isonicotinoyl hydrazone (1×10^{-2} M).*

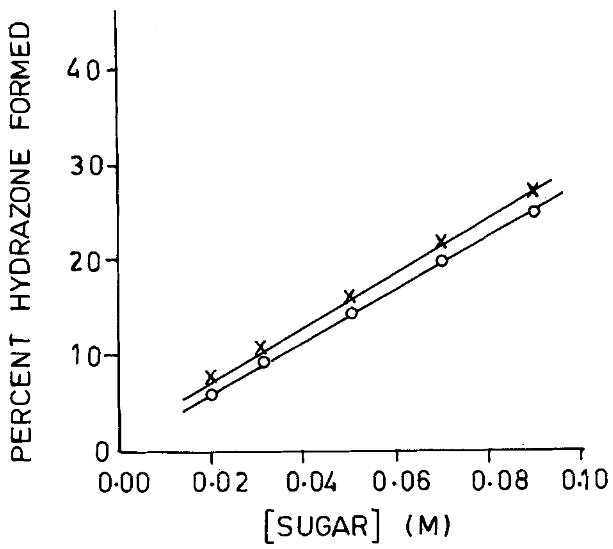

Figure 3—*Hydrazone formation of isoniazid (1 × 10^{-2} M) with different concentrations of sugar in buffer pH 1.8 at 1 h and 37°C. Key:* (○) *glucose;* (×) *lactose.*

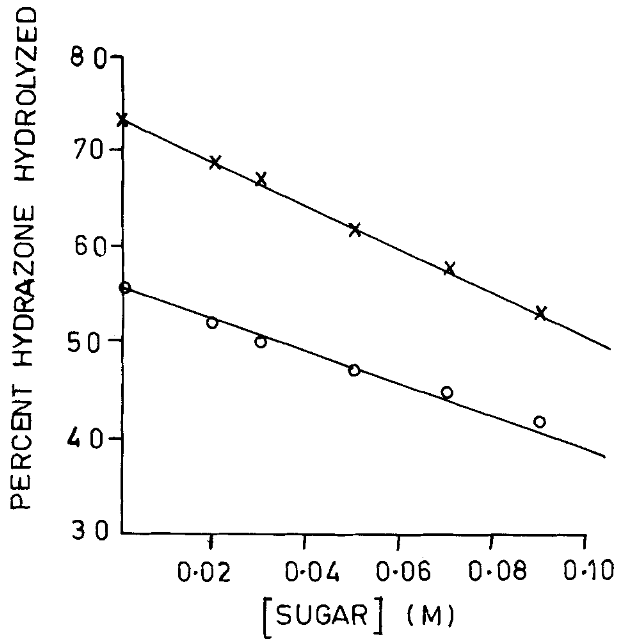

Figure 4—*Hydrolysis of sugar isonicotinoyl hydrazone (1 × 10^{-2} M) in presence of different concentrations of sugar in buffer pH 1.8 at 1 h and 37°C. Key:* (○) *glucose isonicotinoyl hydrazone + glucose;* (×) *lactose isonicotinoyl hydrazone + lactose.*

The rate constants of the kinetic runs were computed from the slopes of the linear regression lines fitted to the following equations.[14] Formation of hydrazone:

$$\text{Isoniazid} + \text{Sugar} \underset{k_h}{\overset{k_f}{\rightleftarrows}} \text{Hydrazone} + H_2O \tag{1}$$

Pseudo-first-order reversible reaction:[14]
when $b \gg a$,

$$k_1 = \frac{x_e}{at} \cdot \ln \frac{x_e}{x_e - x} \tag{2}$$

where a is the initial concentration of isoniazid, b is the initial concentration of sugar, x is the amount of hydrazone formed at time t, x_e is the amount of hydrazone formed at equilibrium (24 h), and k_1 is the pseudo-first-order rate constant of formation.

For the second and pseudo-first-order reversible reaction when $a \neq b$, assuming no hydrazone is present initially, the rate of reaction of isoniazid with sugar at any instant is:

$$\frac{dx}{dt} = k_f(a - x)(b - x) - k_h x \tag{3}$$

where $a - x$ and $b - x$ are concentrations of isoniazid and sugar, respectively, at time t, k_f is the second-order rate constant of hydrazone formation, and k_h is the pseudo-first-order rate constant of hydrolysis of hydrazone.

At equilibrium, the rates of forward and reverse reactions are equal, so that:

$$k_f(a - x_e)(b - x_e) = k_h x_e \tag{4}$$

where x_e is the amount of hydrazone formed at equilibrium. Therefore:

$$\frac{k_f}{k_h} = \frac{x_e}{(a - x_e)(b - x_e)} = K \tag{5}$$

where K is the equilibrium constant of the reaction. Substituting the value of k_h obtained from eq. 4 in eq. 3 and integrating by partial fractions:

$$k_f = \frac{x_e}{t(ab - x_e^2)} \cdot \ln \frac{x_e(ab - x \cdot x_e)}{ab(x_e - x)} \tag{6}$$

Hydrolysis of hydrazone:

$$\text{Hydrazone} \underset{k_f'}{\overset{k_h'}{\rightleftarrows}} \text{Isoniazid} + \text{Sugar} \tag{7}$$

For the pseudo-first- and second-order reversible reactions,[14] at equilibrium:

$$\frac{k_h'}{k_f'} = \frac{x_e^2}{(a - x_e)} = K' \tag{8}$$

and:

$$k_h' = \frac{x_e}{t(2a - x_e)} \cdot \ln \frac{ax_e + x(a - x_e)}{a(x_e - x)} \tag{9}$$

Table I—Rate Constants for Hydrazone Formation Between Isoniazid and Excess Glucose and Lactose in Simulated Gastric Juice[a]

[Isoniazid] × 10^{-2} M	[Sugar] × 10^{-2} M	$10^2 k_1$, min^{-1}	$10^2 k_f$, $M^{-1} \cdot \text{min}^{-1}$ [b]	r[c]
		Glucose		
0.8	15	1.2430	—	0.998
1.0	15	1.2680	—	0.999
1.2	15	1.2470	—	0.997
1.0	5	0.4121	8.242	0.999
1.0	7	0.5795	8.279	0.999
1.0	9	0.7398	8.220	0.998
1.0	15	1.2680	8.453	0.999
		Lactose		
0.8	15	1.5070	—	0.999
1.0	15	1.5095	—	0.999
1.2	15	1.5230	—	0.999
1.0	5	0.5092	10.184	0.999
1.0	7	0.7240	10.343	0.998
1.0	9	0.9134	10.149	0.999
1.0	15	1.5095	10.063	0.998

[a] At pH 1.8 at 37°C. [b] $k_f = k_1/[\text{Sugar}]$. [c] Correlation coefficient for the linear regression line, data fitted to eq. 2.

where a is the initial concentration of hydrazone; x is the amount of isoniazid liberated at time t, which is equivalent to the amount of hydrazone hydrolyzed; x_e is the amount of hydrazone hydrolyzed at equilibrium; k_h' is the pseudo-first-order rate constant of hydrolysis; k_f' is the second-order rate constant of hydrazone formation; and K' is the equilibrium constant of the reaction.

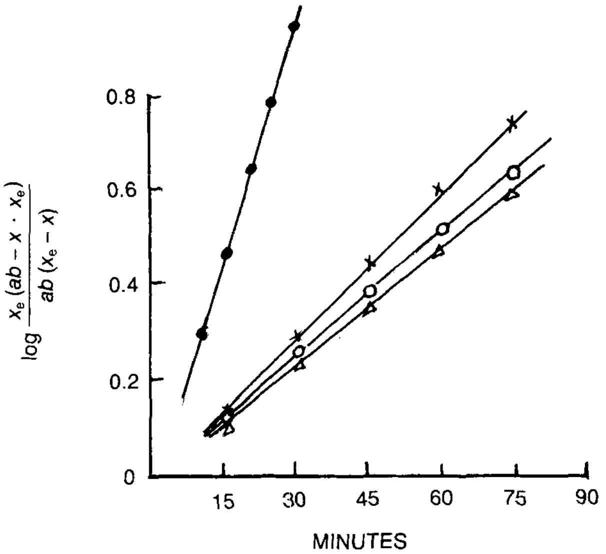

Figure 5—*Plot according to eq. 6 for hydrazone formation of isoniazid with sugar in simulated gastric juice at 37°C; [Isoniazid] = 1×10^{-2} M; [Sugar] = 5×10^{-2} M. Key: (●) galactose; (×) lactose; (○) glucose; (▲) maltose.*

The kinetic data obtained on treating isoniazid with a large excess of the sugars when fitted to eq. 2 gave straight-line plots ($r = 0.999$). The values of pseudo-first-order rate constant of formation (k_1), calculated from the slopes of the lines, were constant for different initial concentrations of isoniazid (Table I). The increase in the concentration of sugar, at the same initial concentration of isoniazid, resulted in the increase in the value of k_1. The values of second-order rate constants of formation ($k_f = k_1/[\text{sugar}]$) were constant for different initial concentrations of sugar (Table I). Therefore, the results indicate that the reaction of isoniazid with sugar is first-order dependent on the concentrations of both the reactants.

Further, the second-order formation constant (k_f) was also evaluated from the slopes of the linear plots of eq. 6 (Fig. 5). The values of k_f remained constant for the different initial concentrations of both the reactants (Table II). The equilibrium constant (K) and the hydrolysis constant (k_h) were determined using eq. 5.

The value of the pseudo-first-order rate constant for hydrolysis of the hydrazones (k_h'), calculated from the slopes of the linear plots of eq. 9 (Fig. 6), remained constant for the various initial concentrations of hydrazones (Table III). The equilibrium constant (K') and the formation constant (k_f') were computed from eq. 8 (Table III). To further confirm the kinetics of hydrolysis, k_h was also determined at low concentrations of hydrazone (2×10^{-4} and 8×10^{-4} M), presuming that the reaction will follow the simple first-order kinetics since the

Table II—Rate Constants for Reactions of Isoniazid with Glucose, Lactose, Maltose, and Galactose in Simulated Gastric Juice[a]

[Isoniazid] $\times 10^{-2}$ M	[Sugar] $\times 10^{-2}$ M	$10^2 k_f$, $M^{-1} \cdot min^{-1}$	K, M^{-1}	$10^2 k_h$, min^{-1} [b]	r [c]
		Glucose			
0.8	5	8.401	5.441	1.544	0.999
1.0	5	8.347	5.579	1.496	0.999
1.2	5	8.362	5.692	1.469	0.998
1.0	2	8.268	5.508	1.501	0.999
1.0	3	8.188	5.701	1.436	0.999
1.0	7	8.310	5.731	1.450	0.999
1.0	9	8.244	5.551	1.485	0.999
Mean	—	8.303	5.584	1.487	—
± SD	—	± 0.074	± 0.110	± 0.036	—
		Lactose			
0.8	5	10.115	5.975	1.693	0.999
1.0	5	10.217	5.899	1.732	0.999
1.2	5	10.230	5.989	1.708	0.999
1.0	2	10.314	5.917	1.743	0.998
1.0	3	10.368	6.000	1.728	0.999
1.0	7	10.408	5.920	1.758	0.999
1.0	9	10.194	5.951	1.713	0.999
Mean	—	10.264	5.950	1.725	—
± SD	—	± 0.104	± 0.039	± 0.022	—
		Maltose			
0.8	5	7.513	5.280	1.423	0.999
1.0	5	7.503	5.310	1.413	0.999
1.2	5	7.519	5.333	1.410	0.999
1.0	2	7.515	5.322	1.412	0.999
1.0	3	7.502	5.302	1.415	0.999
1.0	7	7.575	5.331	1.421	0.999
Mean	—	7.521	5.313	1.416	—
± SD	—	± 0.027	± 0.020	± 0.005	—
		Galactose			
1.0	5	45.261	9.292	4.871	0.999
1.2	5	44.950	9.390	4.787	0.999
1.0	2	45.789	9.333	4.906	0.998
1.0	3	45.598	9.260	4.924	0.999
Mean	—	45.399	9.319	4.872	—
± SD	—	± 0.371	± 0.056	± 0.061	—

[a] At pH 1.8 at 37°C. [b] $k_h = k_f/K$. [c] Correlation coefficient for the linear regression line, data fitted to eq. 6.

back-reaction (formation of hydrazone) would be negligible in the presence of small amounts of sugar and isoniazid produced. The k_h' values at the lower concentration of hydrazone were found to agree closely with the k_h' values calculated at the higher concentration when the reversible reaction becomes significant (Table III). Thus, it may be concluded that the formation of hydrazone is a second-order reaction, while its hydrolysis follows pseudo-first-order kinetics.

The formation constants of sugar isonicotinoyl hydrazones at different temperatures were evaluated from the slopes of the linear plots of eq. 6. The rate of formation increased with the increase in temperature (Table IV). The energy of activation for the reaction of isoniazid with glucose and lactose was determined from the slopes of the Arrhenius plots (Fig. 7).

In Fig. 8, the formation rate constants (k_f) of the reaction of isoniazid with glucose are plotted against the buffer concentration at constant pH 2.6 and 3.1. The rate constant increases with the increase in molarity of the buffers. Figure 9 indicates the pH–rate profiles of the reaction of isoniazid with glucose and lactose at pH 1.0–5.0, exhibiting a maximum formation rate at pH 3.1. Beyond pH 5.0, the reactions were too slow to be determined kinetically.

As shown in Scheme I, isoniazid (**1**) reacts with sugar **2** to form hydrazone **4**, probably through the intermediate **3**. The initial step in the mechanism of the reaction is the nucleophilic attack of hydrazide **1** nitrogen on the carbonyl carbon of the sugar moiety **2** to form the addition product (**3**) which undergoes an acid-catalyzed dehydration to form hydrazone **4**, the dehydration being the rate-determining step. In this reaction, it is necessary to control the hydrogen-ion concentration of the medium to prevent the protonation of the hydrazide base. A decrease in pH value from 5.0 to 3.1 increases the reaction rate (Fig. 9) because it increases the rate of dehydration. However, with further increase in the acidity, although the dehydration rate increases, the protonation of the hydrazide nitrogen slows down the formation of an addition product to such an extent that this now becomes the rate-determining step in the reaction. As a result, a further decrease in pH value decreases the overall reaction rate. Such pH dependency of the reaction kinetics indicates that the reaction of isoniazid with sugar is a general acid-catalyzed reaction. The effect of buffer concentration on the rate constant (Fig. 8) supports this observation.

The reverse reaction, the hydrolysis of hydrazone, also occurs in the acidic region (Fig. 2). The water molecule attacks hydrazone **4** (Scheme I) at the electrophilic carbon of C=N, resulting in the formation of an intermediate **3**. In the acidic region, the rate-determining step is the cleavage of **3** to **1**. A similar reaction mechanism has been proposed for the acid-catalyzed

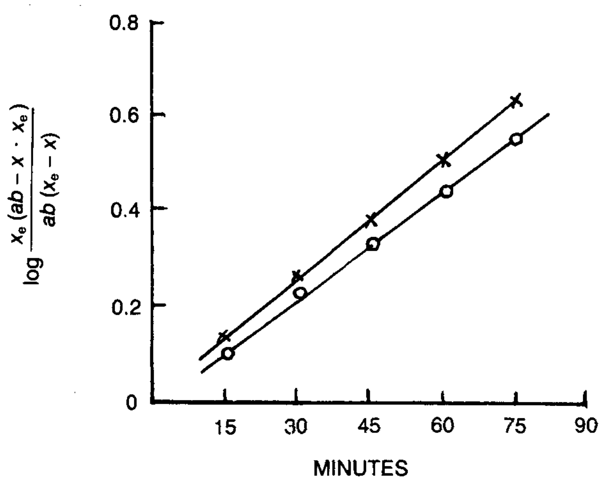

Figure 6—*Plot according to eq. 9 for hydrolysis of hydrazone in simulated gastric juice at 37°C; Key:* (O) *glucose isonicotinoyl hydrazone (1 × 10⁻² M);* (×) *lactose isonicotinoyl hydrazone (1 × 10⁻² M).*

Table IV—Rate Contants for Hydrazone Formation Between Isoniazid and Glucose and Lactose in Simulated Gastric Juice at Different Temperatures[a]

Temp., °C	Glucose 10^2k_f, $M^{-1}\cdot min^{-1}$	Glucose r^b	Lactose 10^2k_f, $M^{-1}\cdot min^{-1}$	Lactose r^b
27	3.931	0.998	5.010	0.999
32	5.737	0.997	6.772	0.997
37	8.347	0.999	10.217	0.999
42	12.370	0.998	15.607	0.999
47	17.398	0.999	20.174	0.997
Slope, degree min^{-1c}		−3124.4		−3022.5
log A, min^{-1c}		9.0006		8.7579
E act, kcal/mol[c]		14.30		13.83
r^d		0.999		0.997

[a] At pH 1.8. Concentration of isoniazid = 1.0 × 10^{-2} M, glucose = 5.0 × 10^{-2} M, and lactose = 5.0 × 10^{-2} M. [b] Correlation coefficient for linear regression line, data fitted to eq. 6. [c] The linear regression parameters from Arrhenius plots, A is frequency factor. [d] Correlation coefficient for the linear regression line, data fitted to Arrhenius equation.

Table III—Rate Constants for Hydrolysis of Glucose and Lactose Isonicotinoyl Hydrazones in Simulated Gastric Juice[a]

[Hydrazone] × 10^{-2} M	$10^2k_h'$, min^{-1}	K', M	$10^2k_f'$, $M^{-1}\cdot min^{-1b}$	$1/K' = K$, M^{-1}	r^c
		Lactose Isonicotinoyl Hydrazone			
0.8	1.738	0.1675	10.376	5.970	0.999
1.0	1.717	0.1672	10.269	5.981	0.999
1.2	1.730	0.1687	10.255	5.927	0.999
Mean	1.728	0.1678	10.300	5.959	—
± SD	± 0.011	± 0.0008	± 0.066	± 0.028	—
		Glucose Isonicotinoyl Hydrazone			
0.8	1.478	0.1843	8.017	5.426	0.998
1.0	1.481	0.1846	8.025	5.417	0.996
1.2	1.526	0.1855	8.226	5.391	0.997
Mean	1.495	0.1848	8.089	5.411	—
± SD	± 0.027	± 0.001	± 0.118	± 0.018	—
0.02	1.345	—	—	—	0.998[d]
0.04	1.503	—	—	—	0.999
0.08	1.545	—	—	—	0.999
Mean	1.464	—	—	—	—
± SD	± 0.105	—	—	—	—

[a] At pH 1.8 at 37°C. [b] $k_f' = k_h'/K'$. [c] Correlation coefficient for the linear regression line, data fitted to eq. 9. [d] Correlation coefficient for the linear regression line, data fitted to simple first-order equation (ref. 14) at low concentration of hydrazone.

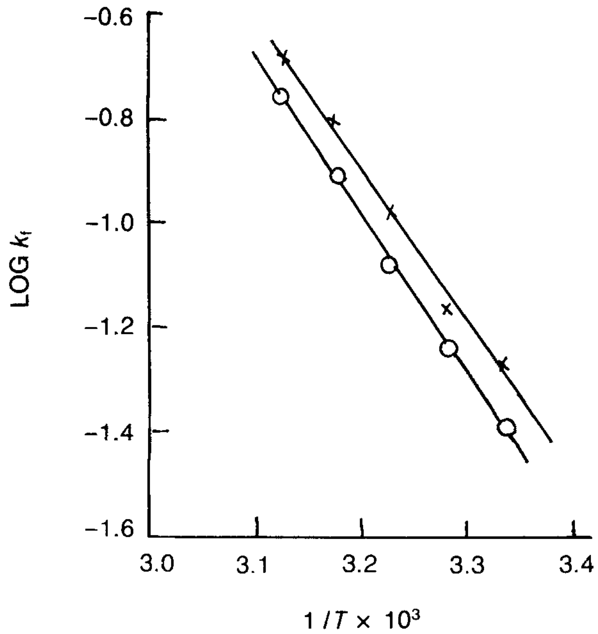

Figure 7—*Arrhenius plot for data shown in Table IV; Key: (×) lactose; (○) glucose.*

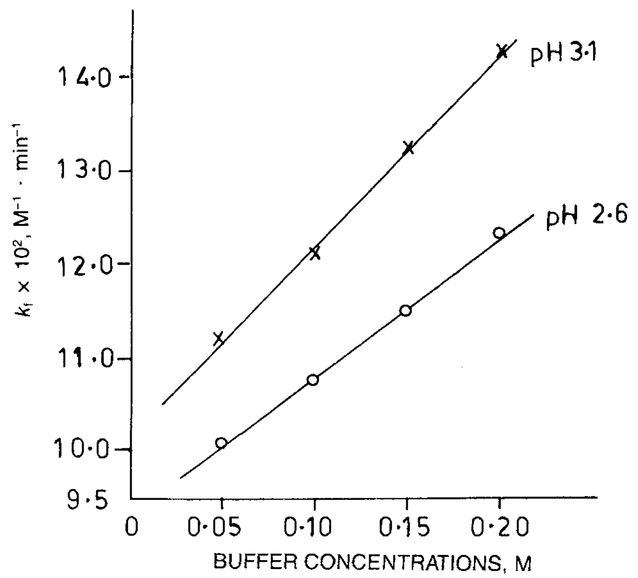

Figure 8—*Plot of k_f versus buffer concentration (μ = 0.5) for the reaction of isoniazid (1 × 10^{-2} M) and glucose (5 × 10^{-2} M) at 37°C; citrate buffer pH 2.6 (○) and pH 3.1 (×).*

reversible reaction of aldehydes and ketones with semicarbazides, hydroxylamines, and amines.[15-17]

The present study indicates that in the presence of excess sugars, the formation of sugar isonicotinoyl hydrazone will be favored over its hydrolysis in the acidic condition of the stomach (Figs. 3 and 4). Thus, the interaction of isoniazid with

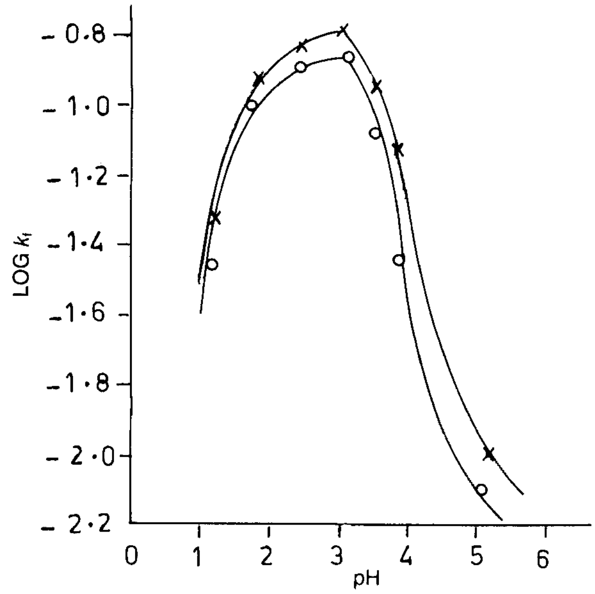

Figure 9—*Log k_f-pH profiles for the reaction of isoniazid (1 × 10^{-2} M) with glucose, 5 × 10^{-2} M (○) and lactose, 5 × 10^{-2} M (×). Buffer concentration 0.1 M; μ = 0.5; temp. = 37°C.*

sugars is likely to interfere with the bioavailability of isoniazid following its oral administration, as sugar isonicotinoyl hydrazones do not hydrolyze in alkaline media and they are poorly absorbed from the GI tract.

References and Notes

1. "The Extra Pharmacopoeia," 27th ed.; Pharmaceutical Press: London, 1977; p 1589.
2. Fox, H. H. *J. Org. Chem.* **1953,** *18,* 990.
3. Wu, W. H.; Chin, T. F.; Lach, J. L. *J. Pharm. Sci.* **1970,** *59,* 1234.
4. Rao, K. V. N.; Kailasam, S.; Menon, N. K.; Radhakrishna, S. *Indian J. Med. Res.* **1971,** *59,* 1343.
5. Bailey, L. C.; Abdou, H. *J. Pharm. Sci.* **1977,** *66,* 564.
6. Butterfield, A. G.; Lovering, E. G.; Sears, R. W. *J. Pharm. Sci.* **1980,** *69,* 222.
7. Devani, M. B.; Shishoo, C. J.; Patel, M. A.; Bhalara, D. D. *J. Pharm. Sci.* **1978,** *67,* 661.
8. Kakemi, K.; Arita, T.; Sezaki, H.; Takasugi, N. *Chem. Pharm. Bull.* **1965,** *13,* 551.
9. Devani, M. B.; Shishoo, C. J.; Doshi, K. J. *Indian J. Pharm. Sci.* **1979,** *41,* 261.
10. Yamamoto, R.; Tanaka, H. *Yakuzaigaku* **1957,** *17,* 219; *Chem. Abstr.* **1958,** *52,* 11849.
11. Pifferi, P. G. *Anal. Chem.* **1965,** *37,* 925.
12. "U.S. Pharmacopeia," 19th rev.; U.S. Pharmacopeial Convention, Inc.: Rockville, MD, 1980; p 765.
13. Devani, M. B.; Shishoo, C. J.; Doshi, K. J.; Bhalara, D. D. *Indian J. Pharm. Sci.* **1980,** *42,* 179.
14. Glasstone, S. "Text Book of Physical Chemistry," 2nd ed.; McMillan and Co. Ltd.: London, 1968; pp 1069–1071.
15. Conant, J. B.; Bartlett, P. D. *J. Am. Chem. Soc.* **1932,** *54,* 2881.
16. Jencks, W. P. *J. Am. Chem. Soc.* **1959,** *81,* 475.
17. Cordes, E. H.; Jencks, W. P. *J. Am. Chem. Soc.* **1962,** *84,* 4319.

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