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Inhibitory Effect of Various Iron Salts on the Absorption of Tetracycline in Man

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Summary. The inhibitory effect of various iron salts, all containing 40 mg elemental iron, on the absorption of tetracycline (500 mg) administered simultaneously has been compared in a double-blind cross-over study in 6 healthy human volunteers. On the basis of changes in peak serum tetracycline concentration, area under individual serum tetracycline concentration-time curves and urinary excretion of tetracycline, the following order of inhibition of tetracycline absorption was found: ferrous sulphate > ferrous fumarate, ferrous succinate, ferrous gluconate > ferrous tartrate > ferric sodium edetate. Thus, in addition to different pharmaceutical properties of iron tablets or capsules, the type of iron salt used may significantly influence the absorption of simultaneously ingested tetracycline.

Key words: Tetracycline absorption, iron salts, drug absorption, man.

Intestinal absorption of most tetracycline derivatives is rapid but incomplete, and it may be further reduced by simultaneous ingestion of antacids or milk (Waisbren and Hueckel, 1950; Price *et al.*, 1957; Barr *et al.*, 1971). It has recently been shown that ferrous sulphate taken at the same time as tetracycline derivatives seriously impairs absorption of the latter in man (Neuvonen *et al.*, 1970). The interaction, which is probably due to the ability of tetracyclines to form chelates with bivalent and trivalent cations, appears to take place in the upper part of the gastrointestinal tract. The present study was intended to demonstrate any differences between various iron salts in their ability to affect the absorption of tetracycline.

Material and Methods

A double-blind cross-over study was performed in six healthy pharmacists (5 women, 1 man), aged 20—30 y, body weight 48—85 kg, who volunteered for the experiment.

The volunteers were given written instructions forbidding the use of any drug one day before and on the day of the experiment. After an overnight fast, a cup of coffee with toast (but no milk or other dairy products) was allowed in the morning before 7 a.m. The drugs were given simultaneously at 8 a.m. with a glass of water. A light meal, again without any dairy products, was taken after a venepuncture at 10.30 a.m.

Each drug combination studied consisted of 500 mg tetracycline hydrochloride and 40 mg of elemental iron as various salts, or a placebo instead of the iron. The tetracycline was given as two gelatine capsules, each of 250 mg (caps. Medicyclin, Medica Ltd., Helsinki). All iron salts and the placebo were packed in identical gelatine capsules; the following salts were employed, in doses equivalent to 40 mg Fe^{++} or Fe^{+++}; ferrous fumarate, ferrous gluconate, ferrous succinate, ferrous sulphate, ferrous tartrate and ferric sodium edetate. Each combination of tetracycline and iron or placebo capsules was packed individually in closed envelopes. The envelopes were coded and key was read only after all the analyses had been performed and the statistical assessment completed. The drug combinations were given in an arbitrary sequence. The interval between two successive tetracycline-iron combinations was at least one week, i.e. more than 15 times the biological half-life of tetracycline. Blood samples were taken 0, 1, 2.5, 4.5, 8 and 24 h after dosing, and a 24 h urine was collected. Serum and urine samples were stored at −18° C until analyzed by the fluorometric method of Kohn (1961), using an Hitachi-Perkin-Elmer MPF-3 fluorescence spectrophotometer. If serum samples of 1.0 ml were employed, the sensitivity of this method was at least 0.1 μg. The relative areas under individual 24 h serum tetracycline concentration curves were calculated for each experiment.

Arithmetic means with standard errors (S.E.M.) were calculated for all groups. The statistical significance of the difference between the placebo and each of the iron groups was calculated by use of the t-test for paired values. To evaluate the difference between individual iron salts, the Friedman two-way analysis of variance and the Wilcoxon matched-pairs signed-ranks test were used (Siegel, 1956).

Results

After ingestion of a single dose of tetracycline and placebo, good absorption of tetracycline was shown by its high serum level and high urinary excretion. Administration of any of the iron salts resulted in significant lowering of peak serum tetracycline concentrations with concomitant reduction of the areas under the serum curves and diminution in the amount excreted in the urine (p for individual results ranged from 0.05 to 0.001).

There was a highly significant difference ($p<0.001$) between the various iron salts in their ability to interfere with the absorption of tetracycline (Figs. 1a, b, 2 and 3).

Ferrous sulphate caused a reduction of 80–90% in serum antibiotic concentration and area under the serum curves, as compared with the values found at similar times in the group given tetracycline + placebo ($p<0.001$). The urinary excretion of tetracycline was diminished to a similar extent ($p<0.001$).

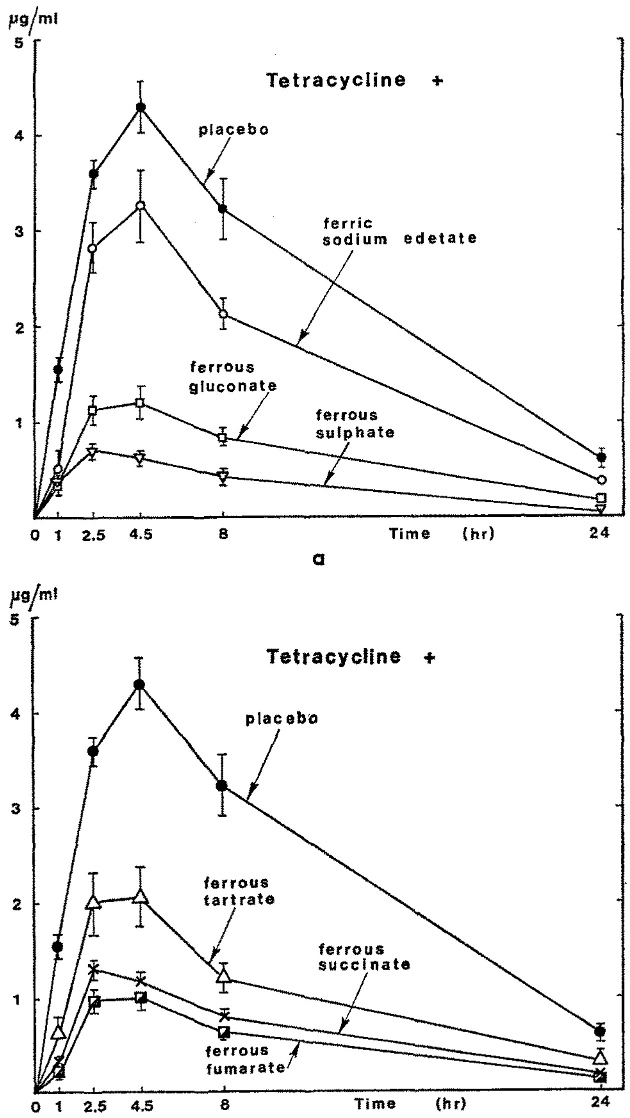

Fig. 1a and 1b. Effect of simultaneous ingestion of various iron salts (dose equivalent to 40 mg elemental iron) and tetracycline hydrochloride (500 mg) on serum concentrations of tetracycline. Means (± S.E.M.) of serum concentrations of tetracycline are shown (n = 6)

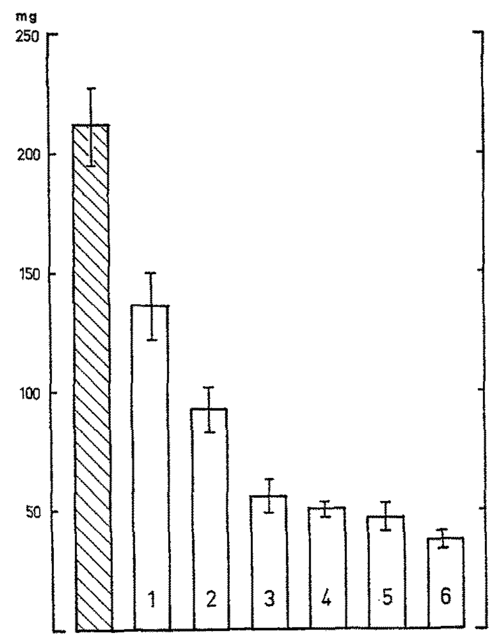

Fig. 2. Effect of simultaneous ingestion of various iron salts and tetracycline hydrochloride (500 mg) on 24 h urine excretion of tetracycline. Ferric sodium edetate (1), ferrous tartrate (2), ferrous succinate (3), ferrous gluconate (4), ferrous fumarate (5) or ferrous sulphate (6) were administered in doses corresponding to 40 mg elemental iron, or a placebo (hatched bar). Excretion of tetracycline was measured during the following 24 h. (Means ± S.E.M.; n = 6)

Ferrous fumarate, ferrous succinate and ferrous gluconate taken simultaneously with tetracycline reduced serum tetracycline concentrations, the areas under serum tetracycline curves and the urinary excretion of tetracycline by 70–80% from the control values. This reduction was significantly less than that caused by ferrous sulphate ($p<0.05$).

When *ferrous tartrate* was administered at the same time as tetracycline, the serum tetracycline curves and the urinary excretion of tetracycline were about 50% of the control values ($p<0.001$). Inhibition of the absorption of tetracycline by ferrous tartrate was less profound than that caused by ferrous sulphate, ferrous fumarate, ferrous succinate or ferrous gluconate ($p < 0.05$).

Ferric sodium edetate diminished the absorption of tetracycline by 30% ($p < 0.05$), a significantly smaller effect than that of any other iron salt.

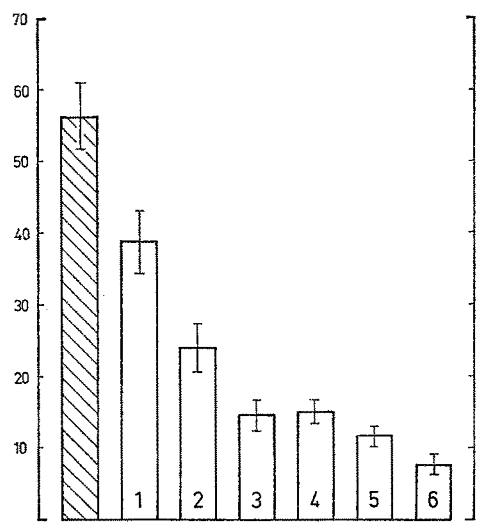

Fig. 3. Effect of simultaneous oral administration of various iron salts (dose corresponding to 40 mg elemental iron; for symbols see legend to Fig. 2) or placebo (hatched bar) and tetracycline hydrochloride (500 mg) on the areas under individual serum concentration-time curves (mean $\pm$ S.E.M; n = 6)

Discussion

It has been recently shown that ferrous sulphate inhibits the absorption of various tetracycline derivatives (Neuvonen *et al.*, 1970). The time interval between the ingestion of the iron salt and tetracycline, as well as the pharmaceutical properties of both preparations, have important influences on this interaction (Neuvonen *et al.*, 1971; Gothoni *et al.*, 1972; Mattila *et al.*, 1972). All iron salts in the present study were packed in rapidly dissoluble gelatine capsules and the inhibitory effect of ferrous sulphate, taken as the reference compound, was greater than that found previously after administration of conventional sugar-coated or slow release tablets (Neuvonen *et al.*, 1970; Mattila *et al.*, 1972).

Gastrointestinal absorption of various iron salts differs to some degree, although most of their iron content remains unabsorbed in the intestinal lumen, at least in non-anaemic persons (Brise and Hallberg, 1962). It seems likely that the amount of iron remaining unabsorbed in the gut lumen could not account for the considerable quantitative differences observed in the interaction between tetracycline and various iron compounds. In addition, the presence of iron in the ferric form, in ferric sodium edetate, cannot explain the lesser ability of this particular compound to interact with the absorption of tetracycline, because Fe^{+++} *in vitro* forms even more stable complexes with it than does Fe^{++} (Albert and Rees, 1956).

Ferrous sulphate dissolves in water more rapidly than organic iron compounds. Furthermore, in some iron compounds, especially in ferrous tartrate and ferric sodium edetate, the iron exists as a complex form. The ability of various iron compounds to liberate ferrous or ferric ions in the upper part of the gastrointestinal tract, before tetracycline is absorbed, would seem to be essential for the interaction of iron and tetracycline. If the stability of the iron complex were high, e.g. in ferric sodium edetate, the degree of inhibition of tetracycline absorption is low. On the other hand, if the stability of the iron complex in the compound used were low in comparison to that of the iron-tetracycline complex, the inhibitory effect on tetracycline absorption was high. It is interesting that the order of activity of different iron salts in the inhibition of tetracycline absorption seen in the present study is the same as the order of the intestinal absorption of these iron compounds (Brise and Hallberg, 1962). Thus ferrous tartrate and ferric sodium edetate, from which the percentual absorption of iron is poorer than from other iron compounds used, had less action on the absorption of tetracycline than other iron salts.

It can be concluded that there are considerable quantitative differences between various iron compounds in their ability to interfere with the absorption of tetracycline swallowed at the same time. The differences appear to be related to the same physicochemical properties of various iron compounds that also affect their intestinal absorption.

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References

Albert, A., Rees, C. W.: Avidity of the tetracyclines for the cations of metals. Nature (Lond.) 177, 433—434 (1956)

Barr, W. H., Adir, J., Garrettson, L.: Decrease of tetracycline absorption in man by sodium bicarbonate. Clin. Pharmacol. Ther. 12, 779—784 (1971)

Brise, H., Hallberg, L.: Absorbability of different iron compounds. Acta med. scand., Suppl. 376 171, 23—37 (1962)

Gothoni, G., Neuvonen, P.J., Mattila, M., Hackman, R.: Iron-tetracycline interaction: effect of time interval between the drugs. Acta med. scand. 191, 409–411 (1972)

Kohn, K.W.: Determination of tetracyclines by extraction of fluorescent complexes. Application to biological materials. Analyt. Chem. 33, 862–866 (1961)

Mattila, M.J., Neuvonen, P.J., Gothoni, G., Hackman, C.R.: Interference of iron preparations and milk with the absorption of tetracyclines. In: Excerpta Medica Int. Congress Series no. 254: Toxicological problems of drug combinations, pp. 128–133 (1972)

Neuvonen, P.J., Gothoni, G., Hackman, R., af Björksten, K.: Interference of iron with the absorption of tetracyclines in man. Brit. med. J. 1970 4, 532–534

Neuvonen, P.J., Mattila, M., Gothoni, G., Hackman, R.: Interference of iron and milk with absorption of tetracycline. Scand. J. clin. Lab. Invest. 27, suppl. 116, 76 (1971)

Price, K.E., Zolli, Z., Jr., Atkinson, J.C., Luther, H.G.: Antibiotic inhibitors. Antibiot. and Chemother. 7, 672–688 (1957)

Siegel, S.: Nonparametric statistics. Tokyo: McGraw-Hill Book Company 1956

Waisbren, B.A., Hueckel, J.S.: Reduced absorption of aureomycin caused by aluminium hydroxide gel. Proc. Soc. exp. Biol. 73, 73–74 (1950)

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