

FIG. 4. Whole blood levels of thiamin in subjects on long-term HTPN in the course of daily administration of adult AMA-FDA thiamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

cluded with both formulations. At each sampling on MVI-12 the means for all four of these vitamins were well within normal limits. It is apparent that 50 µg of B₁₂ given once weekly in the original formulation was adequate as was 5 µg given daily in MVI-12. There were some values for pantothenate and biotin which were consistently above the upper limit of normal. These were associated with patients with renal dysfunction.

Folate (Fig. 11)

The average daily amount of folate provided in both formulations was close to 400 µg. However, in contrast to its daily provision of 400 µg with the AMA-FDA formula, the previous formulation provided 1.5 mg of folate twice weekly. With the latter formulation there were several patients with values at 3.7 to 3.9 ng/ml. With MVI-12 administration the values were at 5.0 ng/ml or higher for all patients with the exception of a single value (3.9) in a patient who previously had had values consistently in the range of 6.8 to 8.8 ng/ml.

DISCUSSION

The AMA-FDA formula is now available from a number of manufacturers. Its price has been reduced so that it is only modestly higher than the multiple preparations

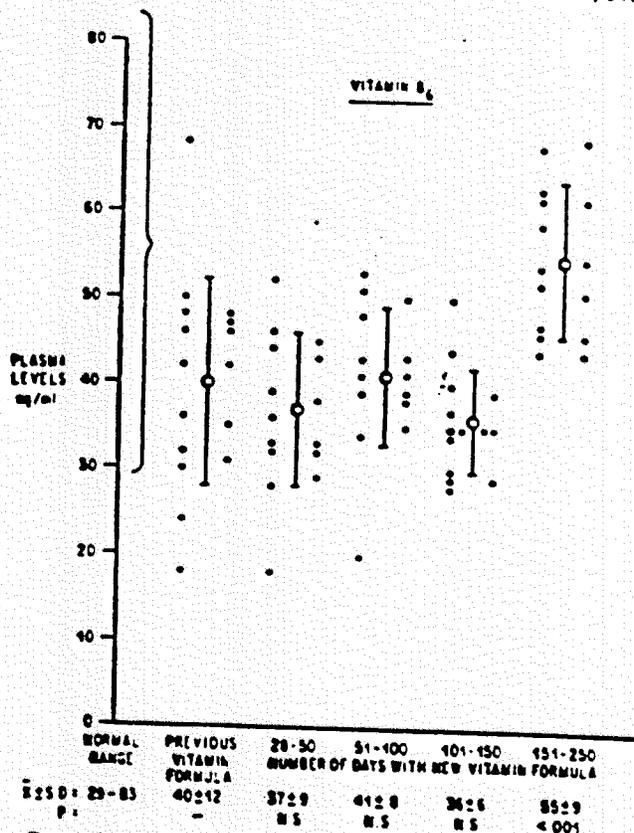


FIG. 5. Plasma level of vitamin B₆ in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See legend of Figure 1 for description of data presentation.

used in our original formula. It has the advantage of providing all known vitamins in one preparation with the exception of vitamin K and it provides appreciably more vitamin E than is available from MVI concentrate (5 ml) given twice weekly.

In the original AMA publication recommending this formulation, it was stated that this preparation was intended as a daily maintenance dose for stable patients receiving TPN and that there may be patients who need higher dosages.³ In this study the vitamins were given over an 8 to 16 hr period. For those patients who are initially depleted of one or more vitamins or may have increased requirements for some reason while in hospital, the nutrients in TPN are usually given over 24-hr periods which would permit more efficient reabsorption of water-soluble vitamins by the renal tubules if further retention is physiologically possible.¹⁷

Measurements of blood levels of individual vitamins give no information on their modification into their biologically active forms. Nevertheless, the finding of normal values in most of the determinations over a period of many months—with the exception of ascorbic acid—in all but an occasional patient is reassuring. In addition, all patients were clinically very well.

It has been reported that appreciable amounts of vitamin A added to TPN solutions may not be recoverable¹⁸⁻²⁰ and that vitamins D and E may similarly be lost.²⁰ It is apparent from our data that these three fat-soluble vitamins were sufficiently stable and non-

VITAMIN LEVELS IN HTPN PATIENTS

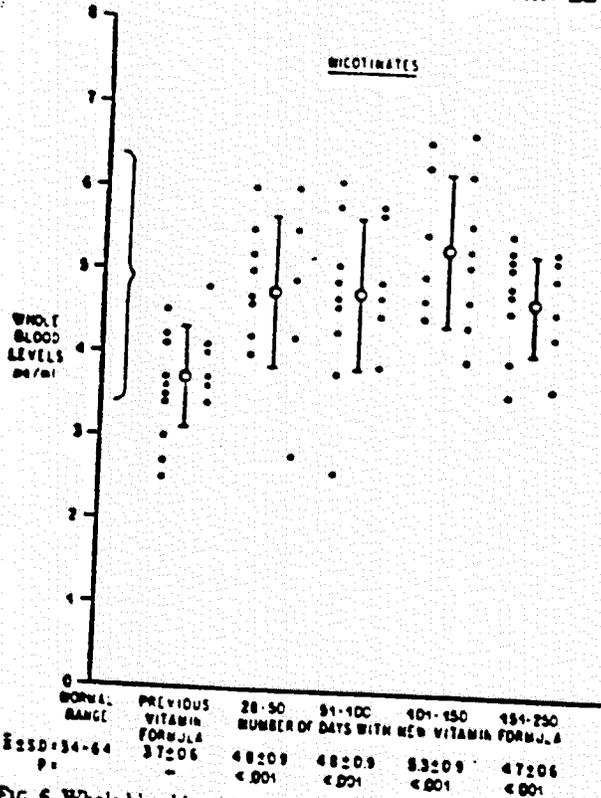


FIG. 6. Whole blood levels of niacin in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

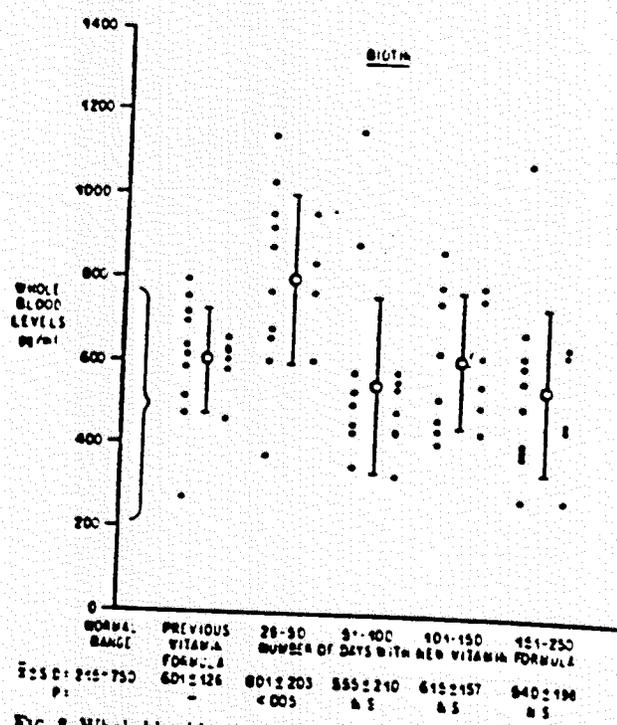


FIG. 8. Whole blood levels of biotin in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

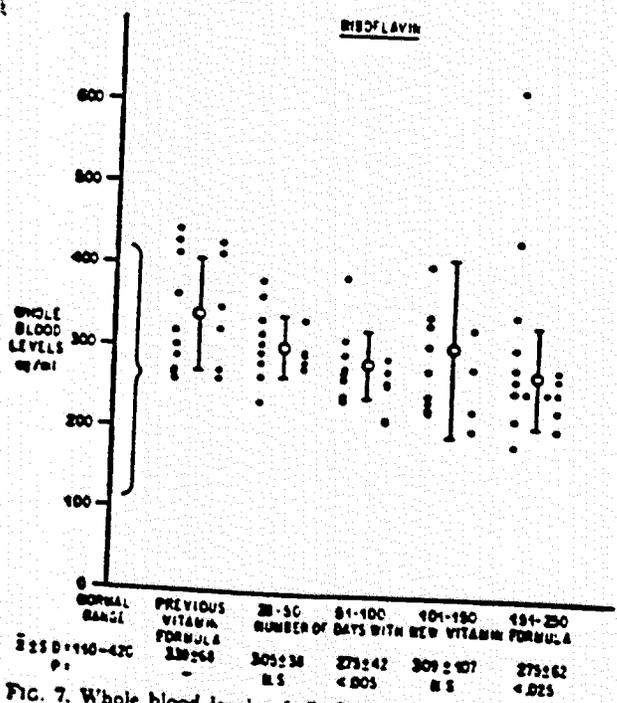


FIG. 7. Whole blood levels of riboflavin in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

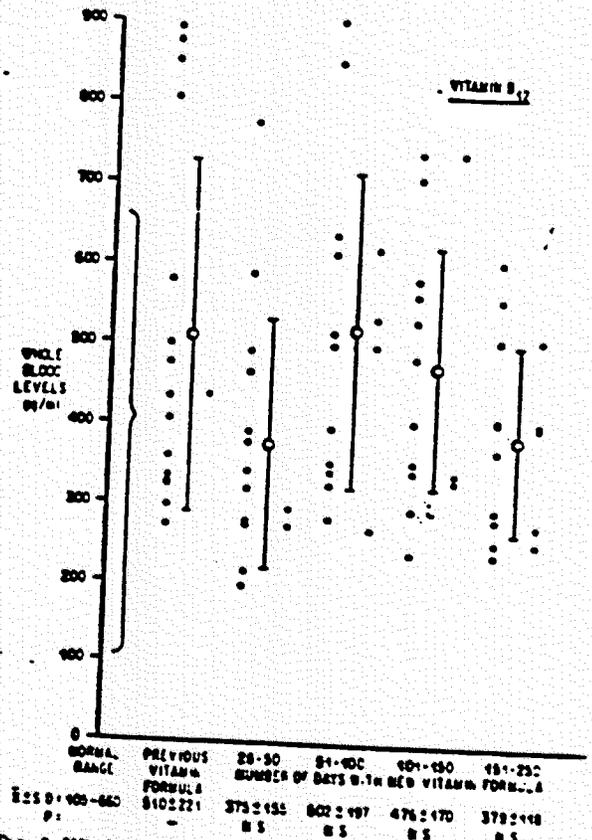


FIG. 9. Whole blood levels of vitamin B₁₂ in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

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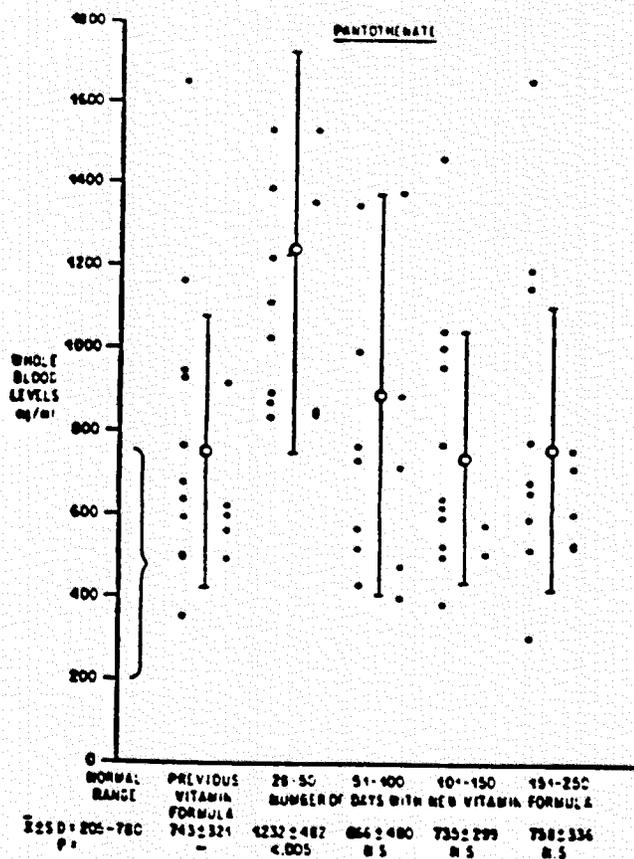


FIG. 10. Whole blood levels of pantothenate in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See legend of Figure 1 for description of data presentation.

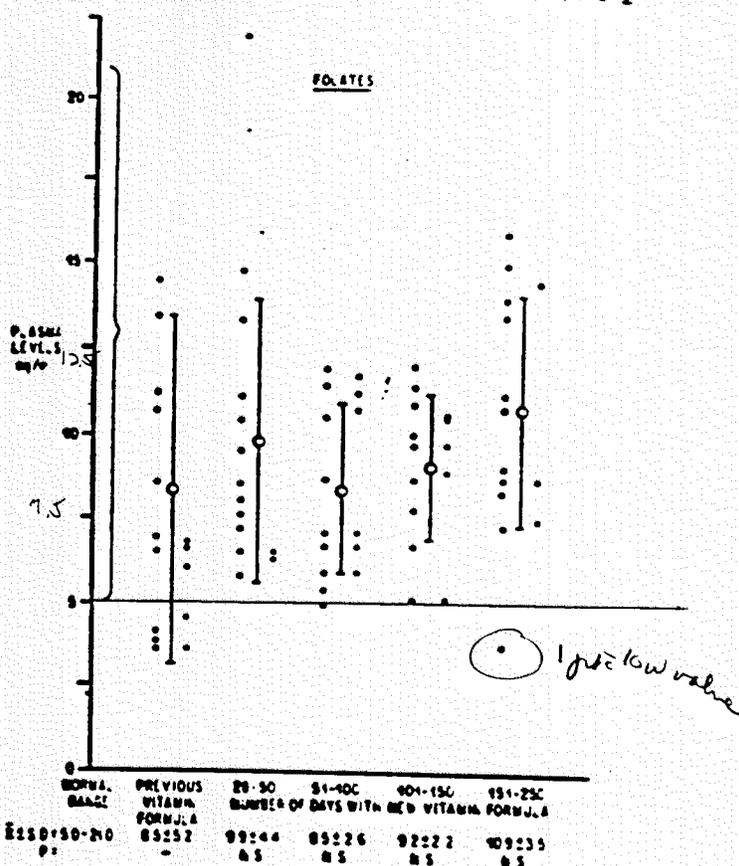


FIG. 11. Plasma levels of folate in subjects on long-term HTPN in the course of daily administration of the adult AMA-FDA multivitamin parenteral formulation (MVI-12). See the legend of Figure 1 for description of data presentation.

adherent to the TPN container and tubing to provide normal value and, in the instance of vitamin A, high blood levels despite an average contact time of the vitamins with container and TPN solution of 30 hr (averaging the 2-day batches). Exposure to sunlight and artificial light was quite limited since the infusions were done at night and the prepared solutions kept in the refrigerator until use.

It is well known that thiamin is very sensitive to sodium bisulphite (present in the Aminosyn) which splits the molecule in two.²¹ The bisulphite, until very recently, has been a component of all amino acid solutions and an occasional electrolyte solution used in the United States. Care was taken to avoid mixing the vitamin solution with the undiluted amino acid solutions. Our data indicate that normal levels were attained.

Our data suggest that ascorbic acid stability may be more of a problem than is the case for thiamin. Our unpublished data indicate that, when cupric ion is present in the TPN solution, and when the solutions are allowed to stand at room temperature for 24 hr after being refrigerated for 48 hr, there is approximately 60% loss of ascorbic acid. Hence, the time of contact of ascorbic acid with other constituents of the TPN solutions should be kept to a minimum. Accordingly, since the conclusion of this study, it has been the practice to

have patients add the vitamins to TPN just before starting each TPN infusion.

The inclusion of biotin at 60 µg/day in the AMA-FDA formulation was based on data indicating that this quantity would maintain normal serum biotin levels for long periods in TPN patients (Shils ME, unpublished data). This amount given daily appears to have rapidly cleared signs and symptoms of apparent biotin deficiency in a patient who had been on TPN without added biotin.²² Further research is needed to determine whether this level can be significantly reduced for maintenance purposes.

Despite a constant intake of the AMA-FDA formula the observed range of blood values was very great. For pantothenic acid and for vitamins A and C, the range was more than four times from the lowest to the highest value; more than three times for B₁₂, folate, biotin, and riboflavin and two times or less for thiamin, ascorbic acid, B₆, and E. The rates of infusion, uptake, metabolic utilization, and renal excretion could be involved.

A number of reports have been concerned with the adequacy of various vitamin formulations added to the TPN solutions. The patients observed have usually been in a postoperative state or otherwise ill in relative short-term studies. Widely varying amounts of vitamin were administered from study to study with differ-

intervals between the time of vitamin infusion and the time of blood sampling.²²⁻²⁴ There is little relevance of such reports to our study in terms of patient types, duration of study, or vitamin formulations used. However, it is of interest that in some of the reports the amounts administered of certain of the vitamins were either similar to those of the AMA-FDA formulation. These findings suggest that, although patients may be hypercatabolic, adequate blood levels or related enzyme activities may be attained at these infusion dosages and hypercatabolic states are not necessarily associated with elevated requirements for specific vitamins. More definitive studies on this point are needed in view of the increasing hospital use of the AMA-FDA formula.

Data on vitamin levels in long-term stable TPN patients are very limited. Jeejeebhoy et al²⁷ reported on blood levels in six patients on TPN for at least 1 yr who were given a Canadian version of MVI (MVI 1000) and Soluzyme on alternate days. No information was given on the time of blood sampling in relation to the time of the discontinuation of the TPN (but, based on other sampling times listed, was probably in the range of 2-4 hr later) or the presence or absence of vitamins in the TPN on the sampling day. Vitamin E was given almost entirely as a natural constituent of Intralipid. The average daily input was 2500 IU vitamin A, 250 IU D₂; 52.5 mg of E, 5.5 mg of pyridoxine; 500 mg of C, 16.3 mg of thiamin, 7.5 mg of riboflavin; 150 mg of niacinamide, 2.5 mg of folate, 29 mg of pantothenate, and 12.5 µg of B₁₂. Niacin and thiamin were measured in whole blood; the others in plasma or serum. B₆ was slightly low in two subjects; the levels of thiamin, niacin, and pantothenate were markedly elevated in all. Vitamin E was in the range of 0.48 to 0.92 mg/dl with the lower normal limit being 0.8. Vitamins A, C, B₁₂, and folate were at normal levels as was 25-OH vitamin D. No data on riboflavin were given. Biotin was not administered and its serum levels were far below the normal range.

Howard et al²⁸ studied six stable patients who had been on HTPN for 0.5 to 7 yr. Three ingested significant amounts of food orally. All infused vitamins twice weekly. The vitamin preparations consisting of MVI concentrate, folate, B₁₂, and biotin with an average daily intake (according to label) of ascorbic acid of 140, niacin 28.6 mg, pyridoxine HCl 4.3 mg, and folic acid 1.4 mg. The average daily ascorbic acid intake (by label) was 40% higher, niacin was approximately 25% less, and pyridoxine was at about the same label amount as the AMA-FDA formula. Blood sampling occurred approximately 14 hr after the vitamin infusion was completed. The levels of these vitamins were well maintained in these patients.

We are aware that it is common manufacturing practice to add more vitamins at the time of preparation of the solutions than is on the label. At least one pharmaceutical company routinely adds overages of 25 to 50% depending on the vitamin. The stated reason is an expected decline in storage prior to the expiration date. Hence, the amounts of vitamins given at any time to a patient will depend on the storage conditions and the interval since production. Many batches of MVI were

used during this study. The results of analysis of one sample in Table II indicate that the variation between analytic and label contents varied greatly among the vitamins with the majority being at or within 5% of the label averages. Analyses of all batches used should be performed at the time of patient use to determine actual intakes. While this was not done in this study, hindsight suggests that this practice is desirable even though it is difficult to monitor in long-term studies on many outpatients.

The following statements appear warranted from this study with stable long-term HTPN patients infused with the AMA-FDA adult vitamin formulation.

(1) The formulation was capable of maintaining plasma and whole blood levels of its 11 constituent vitamins and of the vitamin D metabolites over many months consistently above the lower limits of normal in all but a few of the patients who had low vitamins C and E.

(2) Because the mean values for vitamin A at each sampling were near or above the upper limit of normal (with 23 of 57 determinations were more than 100 µg/dl), questions must be raised as to whether the concentration of this vitamin in the formulation is too high or whether the elevated levels are, in fact, without clinical significance. Clinically, there were no symptoms suggestive of vitamin A toxicity. Preliminary studies on the relation of vitamin A levels to retinol-binding protein (RBP) and prealbumin levels were performed by two of us (HB and OF) on the plasma of 15 of the original 16 patients (obtained between December 1983 and February 1984) who remained on HTPN and who had received the AMA-FDA formulation daily for additional periods extending from 28 months to 4½ months. Twelve patients had vitamin A levels with the normal range (25-84 µg/dl) and three had levels of 123, 132, and 132, respectively. These three had significantly elevated RBP (9.8, 15.8, and 10.3 mg/dl; normal range 3-6). Another patient with an RBP of 9.4 had a vitamin A level of 65 µg/dl but this patient had been taking the MVI-12 only three times per week for the previous 14 months. These four patients had renal dysfunction (serum creatinine in the range 1.7-8.1 mg/dl); a fifth patient with mild renal failure (creatinine 1.5) had an RBP of 10.3 with a vitamin A level of 61. Thus the high vitamin levels were associated with elevated RBP and renal dysfunction confirmatory to a previous report.²⁹ The two lowest vitamin A levels (25 and 31) were associated with RBP levels of 2.8 and 1.9, respectively. The prealbumin levels were normal (10-40 mg/dl) in 13 of the 15 patients with two being slightly low (8 and 9). These preliminary data suggest that under the circumstances of this study, the intake of the vitamin plays an essential but secondary role in plasma levels.

(3) Elevated pantothenate and biotin levels were also associated with the presence of significant renal dysfunction. There was a tendency for B₁₂ levels to be in a high normal range in the presence of kidney disease. Elevation of blood levels of certain B complex vitamins in patients with renal failure has been reported.³⁰

(4) Vitamin E levels tended to be clustered between 0.5 and 0.95 (normal range 0.5-1.6 mg/dl). A direct

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relationship between vitamin E levels and those of serum lipids has been noted.¹⁴ Our patients tended to have low serum cholesterol levels (x̄ 117.2 mg/dl, range 69-168 with only two being above 145 mg/dl) as well as low glycerides (x̄ 65.5 mg/dl, range 39-108 with only 3 being above 88 mg/dl). The low circulating lipids may be expected to decrease the circulating E levels.

(5) Thiamin and ascorbic acid levels also tended to be clustered toward the lower half of the normal range. As indicated earlier, ascorbic acid was found to be unstable on standing in TPN solutions in the presence of cupric ion and this may have been a contributory factor. Addition of the vitamin solution to the TPN should be made just before starting the infusion.

(6) There is now a definite move to the elimination of bisulfite from amino acid mixtures for TPN and this change may result in better availability of thiamin.

(7) In any event, the question must be raised as to whether patients with their vitamin levels in the lower range of normal are at a metabolic disadvantage as compared to those with higher values. Approximately one-half of normal individuals below the mean are still considered normal. Data on the thiamin effect on transketolase activity in individuals in the lower normal range indicate no difference from the response of those in the higher normal range.²¹

(8) Taking into account all of the factors reviewed above, it is concluded that the present AMA-FDA formulation appears reasonably satisfactory for the great majority of stable patients of the types studied who are on long-term HTPN. However, if further studies indicate that patients with renal kidney function consistently have low normal vitamin E, thiamin, and ascorbic acid levels, these vitamins may need to be increased. The use of elevated vitamin levels requires further study.

(9) The adequacy of the formulation for seriously ill patients with trauma and sepsis requires documentation.

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Drug Binding to Human Alpha-1-acid Glycoprotein in Health and Disease

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I. Introduction

THE PLASMA binding of drugs can have important pharmacokinetic implications, especially when the drugs are highly bound having a binding constant larger than approximately 10^7 M^{-1} and when their apparent volume of distribution is small (61, 136, 186, 266, 270, 307, 318, 347, 383, 391, 406, 446, 493, 497-499, 527, 537, 538, 573, 587).

Human serum albumin (HSA)[†] and alpha-1-acid glycoprotein (AGP) are the important drug binding proteins in plasma. HSA is the most abundant protein (4 g/100 ml of plasma), whereas the normal AGP level varies between about 50 and 100 mg/100 ml of plasma. The AGP level can vary considerably as a result of certain diseases, the use of drugs, and pregnancy. Values of up to 300 mg/100 ml of plasma have been found (396, 406,

426, 513, 538; see also table 6). HSA is largely responsible for the plasma binding of acidic drugs, whereas AGP binds mainly basic and neutral drugs. Although HSA has a greater binding capacity than AGP, especially for basic and neutral drugs, AGP can be the most important determinant in plasma binding, due to its greater drug affinity (44, 55, 75, 82, 83, 136, 194, 261, 358, 376, 385, 396, 450, 540, 544; see also table 8).

Drug monitoring is of increasing importance in clinical practice, especially in the case of drugs with a small therapeutic index. If such drugs are highly bound in the plasma and have a small volume of distribution, the free concentration of the drug in plasma will be a more reliable parameter for representing the intensity of the pharmacological effect than the total plasma concentration (3, 49, 71, 72, 79, 88, 189, 192, 202, 270, 277, 278, 308, 310, 321, 347, 395, 408, 408, 445, 518, 528, 538, 546, 556). If variations in the plasma levels of AGP occur, then the free plasma level of the drugs in question can vary considerably, whereas the total drug concentration of the drug in plasma will be only slightly affected (49, 108, 192, 210, 277, 278, 347).

A thorough review of the studies on the binding of drugs with AGP, both in vitro and in vivo, has been made in order to obtain a better understanding of the different factors which can affect the free plasma level of such drugs. These factors include the effects of exog-

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† Abbreviations used are: AGP, alpha-1-acid glycoprotein; B, bound fraction of a drug; B/F, binding ratio of a drug; c_{bound} , bound concentration of a drug; c_{free} , free concentration of a drug; F, free fraction of a drug; F_{AGP} , free fraction of a drug in an AGP solution; F_{HSA} , free fraction of a drug in a HSA solution; $F_{\text{AGP+HSA}}$, free fraction of a drug in a solution of a mixture of AGP and HSA; HSA, human serum albumin; K_{AGP} , drug-AGP binding constant; K_{HSA} , drug-HSA binding constant; LIPO, lipoprotein(s); n_{AGP} , number of binding sites on AGP; n_{HSA} , number of binding sites on HSA; P, plasma protein concentration; P_{AGP} , plasma concentration of AGP; P_{HSA} , plasma concentration of HSA; P_{LIPO} , plasma concentration of LIPO.