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Comparison of the Gastrointestinal Absorption of Aluminum Acetylsalicylate and Acetylsalicylic Acid in Man

By GERHARD LEVY and BERNICE A. SAHLI

The gastrointestinal absorption rate and biological availability of aluminum acetylsalicylate have been studied in human subjects by the urinary excretion method. Acetylsalicylic acid absorption from orally administered aluminum acetylsalicylate was found to be less rapid than from aspirin, probably due to the very slow dissolution of the aluminum salt in gastrointestinal fluids. In the form used in this study, aluminum acetylsalicylate was incompletely absorbed after oral administration. Dosage forms containing this drug must be carefully evaluated with respect to absorption rate and biological availability.

THE ALUMINUM salt of acetylsalicylic acid has been admitted recently to the National Formulary (1) and is used currently in a number of proprietary pharmaceuticals. According to the background information which accompanied the proposed monograph on aluminum acetylsalicylate (Al.ASA) at the time it was considered for admission to the National Formulary (2), the drug has several significant advantages over acetylsalicylic acid (ASA). These are greater palatability, stability, and absence of astringent taste or acetic odor which is often found with ASA. As a result of its relative inertness, Al. ASA is said to be compatible with many more drugs than is ASA.

Neither the official monograph, nor the introductory statement which accompanied it at the time of submission, made reference to any possible difference in the rate or extent of gastrointestinal absorption of Al.ASA as compared with ASA. Al.ASA is a constituent of certain proprietary pharmaceuticals which are intended, among others, for the relief of acute pain and fever. Since Al.ASA is practically insoluble in water (1, 3), the possibility suggests itself that the absorption rate of the drug from the gastrointestinal tract of man may be considerably less than the absorption rate of ASA. If this is the case, there is the further possibility that Al.ASA is absorbed only partially, and that a portion of the undissolved drug is excreted in the feces. Accordingly, the gastrointestinal absorption rate and biological availability of Al.ASA have been investigated and compared with those of ASA. The present communication reports the results of this investigation and also presents certain physical-chemical data pertinent to the interpretation of the biological observations.

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EXPERIMENTAL

Materials.—Acetylsalicylic acid U.S.P., aluminum acetylsalicylate N.F. With each drug, only the fraction passing through a 100-mesh sieve was used in the investigation. Both compounds were assayed colorimetrically (4) after alkaline hydrolysis and were found to contain 100 ± 0.5% of the theoretical amount of salicylate.

Absorption Tests.—Healthy male adults served as test subjects. Their weights and ages are listed in Tables I and II. In the initial absorption test utilizing nine subjects, the drugs were administered

TABLE I.—URINARY EXCRETION OF APPARENT SALICYLATE[a] AFTER ORAL ADMINISTRATION OF EQUIVALENT DOSES[b] OF ASPIRIN AND ALUMINUM ACETYLSALICYLATE

Subject	Age, yr.	Weight, lb.	One Hour		Two Hours	
			ASA	Al.-ASA	ASA	Al.-ASA
A	31	155	7.29	4.69	29.4	17.0
B	26	218	10.2	5.63	55.1	35.3
C	22	152	21.1	4.98	55.9	16.3
D	21	172	9.03	8.43	36.7	14.5
E	27	170	20.2	3.62	70.9	13.7
F	27	166	15.7	4.02	45.9	14.7
G	22	155	11.2	3.93	67.6	22.8
H	22	187	8.99	6.34	30.8	19.9
I	22	150	28.2	5.42	66.1	23.0
Average			14.7	5.23	50.9	19.7

[a] Expressed as mg. of salicylic acid. [b] 1.00 Gm. of aspirin, or 1.12 Gm. of aluminum acetylsalicylate.

in crossover fashion 30 to 60 minutes after the noon meal with exactly 100 ml. of water. A small amount of urine was collected just prior to drug administration and served as a blank. Urine samples were then collected exactly 1 and 2 hours after drug administration. The crossover test was carried out after one week. The test subjects were instructed to eat essentially the same type and quantity of food, and at the same time, as in the previous week.

The second absorption test involved five subjects, and was initiated in the morning on empty stomachs. The test subjects were instructed to empty their bladder of overnight urine and to drink one glass of water. One hour later, the urine blank was collected and the drug was administered with 100 ml. of water. Exactly 1 hour later, the first urine sample was collected. From then on, urine samples were obtained at intervals of approximately 3 hours (except for the greater night-time interval) for about 36 hours. The time of urine collection and the volume were carefully noted.

In the case of "sustained" ASA administration, the drug was given in divided doses at 0, 3, 6, and 9 hours. The participants of the second study were cautioned not to eat until the first-hour urine sample had been collected. The interval between successive tests was 3 days or more. The drugs were administered in random order.

Analytical Method.—Salicylate in the urine was determined colorimetrically with ferric nitrate reagent (4, 5), using a Bausch and Lomb Spectronic 20 colorimeter. All readings were corrected for blank values. For first and second-hour urine samples, the zero hour blank values were used. For all other urine samples, average blank values were obtained from 24-hour urine collections of each subject.

For the determination of dissolution rates, the drug contained in aliquots of the dissolution medium was subjected to alkaline hydrolysis. The solution was then acidified, appropriately diluted with 0.1 *N* hydrochloric acid, and assayed spectrophotometrically with a Beckman model DU spectrophotometer in terms of salicylic acid.

Excretion Rates.—Individual cumulative plots of the amount of apparent salicylate excreted *vs.* time were constructed. Excretion rates were determined graphically at 1, 3, 6, 9, 12, and 15 hours over the 2-hour period having the cited time as the midpoint. The individual excretion rates were then averaged and used to construct the excretion rate *vs.* time plot.

In the case of Al.ASA, the 4-hour average excretion rate was also calculated in order to characterize the excretion rate *vs.* time curve more adequately.

Determination of Dissolution Rates.[1]—Flat-faced, one-half inch diameter tablets of the pure drugs were prepared by means of a specially modified Carver model B hydraulic press. The compression pressure was 10,000 lb. (approximately 50,000 lb. per square inch). These tablets were mounted on Plexiglas holders with the aid of paraffin wax in such a manner that only one surface of the tablet was exposed (Fig. 1). The holder with the tablet was attached to a metal shaft which, in turn, was connected to an electronically controlled precision stirring motor.[2]

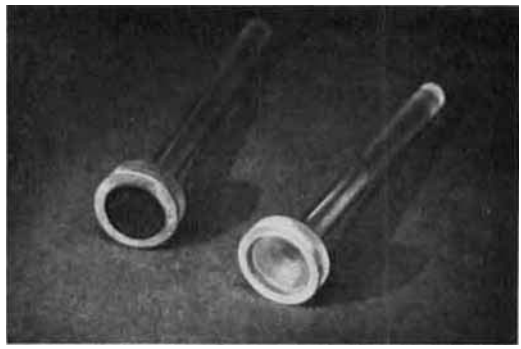

Fig. 1.—Plexiglas tablet holder for dissolution rate determinations. Empty holder (right) and holder on which a tablet has been mounted (left). The tablet shown in the picture was colored with charcoal to improve visualization.

A 200-ml. quantity of the dissolution medium was placed in a 500-ml. three-neck round-bottom flask which was immersed in a constant temperature bath adjusted to 37 ± 0.1°. After temperature equilibration was attained, the tablet attached to the Plexiglas holder was immersed in the dissolution medium to a depth of 1 inch. Under these conditions, the metal shaft did not come into contact with the solution. Just prior to immersion of the tablet, the stirrer was turned on at 555 r.p.m. The speed of rotation could be monitored by means of a continuous reading tachometer attached to a second shaft which reflected stirring speed at a fixed ratio.

Aliquots of 5 or 10 ml. were removed from the flask at appropriate intervals of time for analysis. A similar volume of dissolution medium was added each time to maintain constant volume. The tablet in its holder was weighed before and after the dissolution run as a check on the assay. The conditions of the dissolution experiment were such that the concentration of drug in the dissolution medium was a small fraction of its total solubility in order to maintain the quantity $(C_s - C)$ in the Noyes-Whitney dissolution rate expression almost constant (6, 7).

[1] The procedure is based in principle on a method developed and suggested to us by Dr. Eino Nelson, University of California, School of Pharmacy, San Francisco.

[2] Gerald K. Heller Co., Las Vegas, Nev.

TABLE II.—FIRST HOUR AND TOTAL URINARY EXCRETION OF APPARENT SALICYLATE[a] AFTER ORAL ADMINISTRATION OF ASPIRIN AND ALUMINUM ACETYLSALICYLATE

Subject	Age, yr.	Weight, lb.	One Hour After Drug Administration— ASA, 1.00 Gm.	ASA, 0.50 Gm.	Al.ASA, 1.12 Gm.[b]	Total Excretion[c] "Sustained" ASA	Al.ASA
B	26	218	26.6	20.6	13.2	652	536	458
D	21	172	18.9	10.4	7.96	545	600	484
J	33	168	27.9	8.24	13.6	538	564	532
K	22	200	19.0	9.29	1.34	399	563	358
L	21	150	39.5	17.0	4.59	547	520	374
Average			26.4	13.1	8.14	536	557	441

[a] Expressed as mg. of salicylic acid. [b] Equivalent to 1.00 Gm. of aspirin. [c] ASA = 1 Gm. given in one dose. "Sustained" ASA = 0.5 Gm. initial dose, then 0.167 Gm. every 3 hours for three doses. Al.ASA = 1.12 Gm. given in one dose.

RESULTS AND DISCUSSION

The results of the initial comparative study of Al.ASA and ASA absorption are shown in Table I. In this study, the drugs were administered after the noon meal rather than on empty stomachs so that the results may reflect absorption under conditions of normal use. The doses given were 1.00 Gm. of ASA powder and the equivalent (1.12 Gm.) of Al.ASA, respectively. The apparent salicylate excretion 1 hour after ASA administration was about three times greater than that from the aluminum salt. After 2 hours, the difference in mean amounts of salicylate excreted cumulatively was still two and one-half fold. In both instances, the differences were statistically significant ($P < 0.005$). These observations indicate that Al.ASA is absorbed much more slowly than the acid form of the drug, and that it cannot be expected to be as efficacious as ASA in the treatment of acute conditions where a reasonably rapid onset of drug action is desirable.

There is always the possibility of incomplete biological availability when a drug is absorbed from the gastrointestinal tract at an unusually slow rate since part of the drug may not be absorbed during the time in which it is in contact with the absorption sites and hence be excreted in the feces. On the other hand, poorly soluble compounds frequently afford a type of sustained action which may be desirable under certain circumstances. Accordingly, the second experiment was designed to determine salicylate excretion rate as a function of time, as well as the total amount of salicylate excreted. The rationale for using urinary excretion rates to reflect absorption rates and drug plasma levels is indicated by the following relationship (8)

$$dA/dt = K \cdot f \cdot V \cdot C \qquad \text{(Eq. 1)}$$

where A is the amount of drug excreted, K is a constant, f is the fraction of absorbed drug which is excreted eventually in the urine, V is the apparent volume of distribution, and C the drug concentration in the plasma at time t. In the case of the drugs used in this study, the values for K, f, and V are identical. Therefore, differences in excretion rate are a direct indication of differences in C, the concentration of drug in the plasma.[3] The described relationship is limited to cases where the urinary excretion rate is proportional to plasma concentration.

[3] Good comparative correlation between plasma salicylate levels and urinary excretion rates of salicylate has recently been demonstrated by Levy and co-workers (9).

In theory at least, there is the possibility that a comparative study of the urinary excretion of salicylate in the manner described may give results that are misleading. Equation 1 assumes that the ratio of the various metabolites of salicylic acid which are excreted in the urine is the same with both drugs. While this assumption appears reasonable as long as the drugs do not differ in their effect on urinary pH, it has been shown that when large doses of a drug are given, the endogenous mobilization rate of a conjugating compound may become the limiting factor in determining the rate of formation of a given metabolite (8). Thus there is a theoretical possibility that a greater fraction of the more rapidly absorbed ASA is excreted as the glucuronide of salicyclic acid than would be the case when the slowly absorbed Al.ASA is used. Since the analytical method serves to determine free salicylic acid and salicyluric acid, but not the glucuronide (10), and since the overall salicylate excretion rate is the sum of the excretion rates of each of the various metabolites and of the free drug, it is possible to obtain comparative data which do not reflect adequately the true relationship between the absorption rates or total excretion rates after administration of the two drugs.

The problem described above was overcome by the administration of ASA on a "sustained" dosage schedule. One-half gram of the drug was given at zero hour, and 0.167 Gm. was then administered every 3 hours for three doses. The dosage schedule was based on a 6-hour half-life of salicylate.[4] In this manner, any significant difference in the ratio of the excreted metabolites due to differences in absorption rate would become apparent by a comparison of the amount of salicylate (as free salicylate and salicyluric acid) excreted after single dose and "sustained" ASA administration, respectively.

The amounts excreted 1 hour after administration of ASA in single dose, of the same drug in divided doses, and of Al.ASA, as well as total amounts excreted, are listed in Table II. In this experiment, the drug was given on an empty stomach. As in the previous experiment, the first hour salicylate excretion as a result of ASA administration was about three times as high as that from Al.ASA. The difference was statistically significant ($p < 0.005$). The greater first hour absorption of both

[4] The maintenance doses, when determined by specific elimination rate constant × initial dose × hours between dosings, should actually be 0.173 Gm. A slightly lower dose (0.167 Gm.) was used as a matter of convenience in order to use an initial dose of 0.5 Gm. and provide a total of 1.00 Gm. in four doses.

drugs in the second experiment as compared with the first one is probably due to the fact that the drugs were given on empty stomachs and after the noon meal, respectively. With respect to total salicylate excretion, it is apparent that the effect of absorption rate on the ratio of the principal urinary metabolites of the drug (salicyluric acid and salicylic acid glucuronide) is negligible for the doses used in this study, since the average total apparent salicylate excretion after "single dose" and "sustained" ASA administration is essentially the same.

The total apparent salicylate excretion from Al.ASA is markedly less than that from ASA. Compared with the amount of drug excreted when ASA was given on the "sustained" dosage schedule, the biological availability of Al.ASA is only about 80% and this difference is statistically significant ($p < 0.02$). It must be concluded that Al.ASA in the form used in this study is incompletely absorbed. The degree of biological availability will of course vary depending on factors such as the particle size, the dosage form, and the experimental methods, inasmuch as the latter can affect gastrointestinal motility, pH, and gastric content. In the present study, Al.ASA was of a particle size smaller than 100 mesh, which is much more favorable than the larger particles or granules usually found in compressed tablets, as far as gastrointestinal absorption is concerned.

Figure 2 is a plot of urinary excretion rate of salicylate as a function of time. The biological half-life of salicylic acid as observed in the present study was about 6 hours. This is in excellent agreement with the 6.1-hour half-life reported by Brodie and Burns on the basis of plasma level studies (11), and is very similar to the 5 to 6-hour half-life determined by Chapman and co-workers (12), who used the urinary excretion method. The excretion rate curve obtained from "sustained" drug administration resulted in a period during which excretion rates were essentially constant. While the plateau is distinct, it is appropriate to point out that certain limitations inherent in the urinary excretion method can tend to idealize the data (13). The graphic determinations of excretion rates have been carried out with due consideration of these pitfalls and the linearity of the individual cumulative salicylate excretion curves is based, in each instance, on four experimental points. The excretion rate *vs.* time curve for Al.ASA is somewhat difficult to characterize after the 6th hour. As expected the area under the curve is smaller than that under the other two curves, reflecting the incomplete availability of the drug.

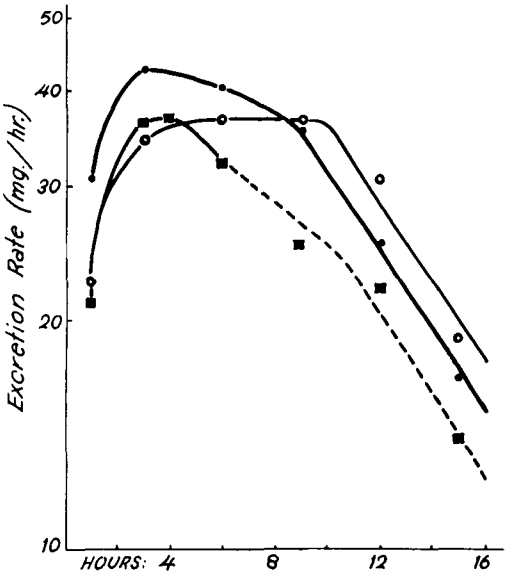

Fig. 2.—Average excretion rate of apparent salicylate as a function of time after administration of 1.00 Gm. aspirin or equivalent. ●, single dose of aspirin; ○, divided doses of aspirin; ■, single dose of aluminum acetylsalicylate.

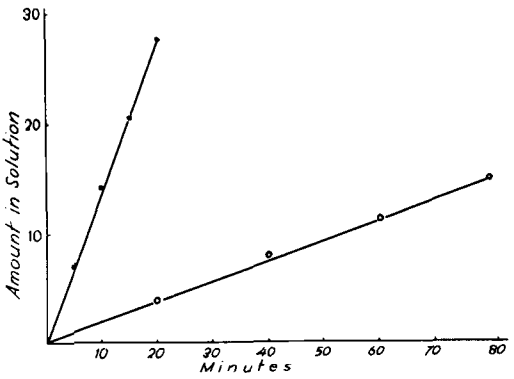

Fig. 3.—Dissolution of aspirin (●) and aluminum acetylsalicylate (○) in 0.1 *N* hydrochloric acid solution. The amount in solution is expressed as mg. salicylic acid.

Figure 3 shows the results of dissolution rate determinations using 0.1 *N* hydrochloric acid as the dissolution medium. Expressed in terms of salicylic acid, the dissolution rate of ASA is 65.1 mg./hr.-cm.^{2}, while that of Al.ASA is 8.88 mg./hr.-cm.^{2} under the conditions of the experiment. It is apparent that the dissolution rate of ASA is significantly greater than that of the aluminum salt. Similar differences were found when alkaline media were used. Since the absorption of salicylates is rate-limited by their dissolution rate in gastrointestinal fluids (5, 14), and since the dissolution rate of Al.ASA is considerably smaller than that of ASA, it is reasonable that the absorption and excretion rates of the former are also smaller than those of the latter. Dissolution of the Al.ASA used in this study in gastrointestinal fluids is evidently sufficiently slow to be incomplete, so that only part of the administered drug is eventually absorbed.

The ratio of the amounts of salicylate excreted 1 hour after administration of ASA and Al.ASA is not as unfavorable to Al.ASA as the *in vitro* dissolution data imply. This may be due to differences in the "functional" surface area of the two drugs. "Functional" surface area refers to the area actually in contact with gastrointestinal fluids and is related to particle size and shape, tendency of the particles to aggregate, and wettability of the drug solids, among others. Microscopic examination reveals that the average particle size of Al.ASA

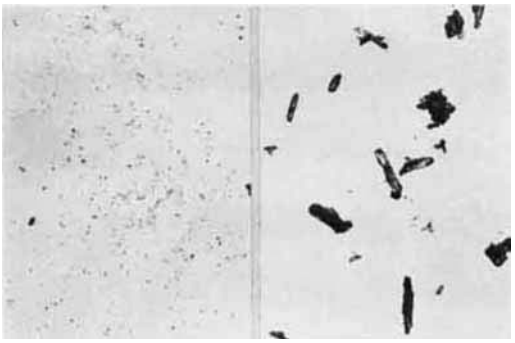

Fig. 4.—Photomicrograph of aluminum acetylsalicylate (left) and aspirin (right) used in this study. (22 × magnification).

used in this investigation was considerably smaller than that of ASA (Fig. 4). Better quantitative correlation between dissolution and absorption rates could possibly be obtained by administering thin, highly compressed, nondisintegrating tablets of the pure drugs so that surface area would be the same in each case. The contact-irritation liability of salicylates (14,15) makes such an experiment unfeasible.

The results of this investigation indicate that Al.ASA is inherently absorbed less rapidly than ASA, and that this difference is probably due to the slow dissolution of Al.ASA as compared with ASA. Administration of Al.ASA in the form used in this study would not bring about a pharmacologic effect as rapidly as administration of ASA. The use of Al.ASA may be limited markedly by its absorption pattern and by the possibility of incomplete biological availability. While it must not be excluded that the use of certain additives could enhance sufficiently the dissolution rate of this drug to overcome the shortcomings outlined above, it is necessary to subject Al.ASA preparations to careful clinical evaluation to assure complete availability of the drug and to ascertain that it is absorbed at a rate consistent with the medicinal use of the particular product.

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Paper Chromatographic Separation and Colorimetric Estimation of the Glycosides of *Digitalis ferruginea* Seeds

By AYHAN ULUBELEN

Digitoxin, gitoxin, digoxin, digilanide A, digilanide B, digilanide C, obtained from *Digitalis ferruginea*, were successfully separated by paper chromatographic techniques and estimated colorimetrically with alkaline 3,5-dinitrobenzoic acid.

A SURVEY of the literature to date would indicate that, the seeds of *Digitalis ferruginea* have not been investigated either qualitatively or quantitatively for their cardioactive glycoside content. Considerably less attention has been given to the phytochemical investigation of the seeds of the digitalis species than of their leaves. The phytochemical investigation of the seeds of digitalis species, therefore, has been initiated as part of a long range program. In these initial studies the air-dried seeds of *Digitalis ferruginea* that were collected from Uludag-Bursa, Turkey, on October 7, 1959, were used.

A number of authors have used different reagents for the colorimetric estimation of various cardiac glycosides. The most important methods are those based on the Baljet reaction (1–6), colors given by the xanthydrol reagent (7–10),

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