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URICOSURIC PROPERTIES OF DIFLUNISAL IN MAN

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1 The uricosuric effect of diflunisal was studied in eleven normal subjects.

2 Urine and serum uric acid levels were measured and used to assess the effect.

3 Diflunisal caused statistically significant decreases in serum uric acid levels and increases in uric acid clearance at 250 and 375 mg twice daily.

4 Diflunisal taken on an empty stomach caused epigastric discomfort in the high dose group which was obviated when the drug was taken with meals.

5 Diflunisal caused no abnormalities in the measured haematological, urological or biochemical parameters.

Introduction

For some time it has been known that salicylates may affect the excretion of uric acid and that the observed effects are dependent on the salicylate concentration in the blood or kidney. A century ago, See (1877) reported his observations on the uricosuric effect of large doses of salicylates and the resulting resolution of gouty tophi. Eight years later, Salome (1885) reported that low doses of salicylic acid caused the retention of uric acid. Since that time many investigators (Fauvel, 1907; Klemperer & Bauer, 1944; Gutman, 1951; Gutman & Yu, 1952; Yu, Sirota & Gutman, 1953; Crone & Lassen; 1955) have confirmed that acetylsalicylic acid when given in doses of less than 2 g per day has uricemic properties and others (Grayzel, Liddle & Seegmiller, 1961) have reported that doses of 2.4 g per day produce uricosuria in most subjects. In general, small doses of salicylates have been found to induce uric acid retention while larger doses caused uricosuria.

Diflunisal (2',4'-difluoro-4-hydroxy-3-biphenylcarboxylic acid) is a novel molecule which in the clinic has been shown to be a more potent analgesic and to have a longer duration of action than aspirin (Besselaar, Steelman, Tempero, Honig, Brosens, Caudron & Devroey, 1975; Honig, Cremer, Manni, Buntinx, Steelman & Besselaar, 1977; Devroey, Steelman, Caudron, Verhaest, Besselaar & Buntinx, 1977).

Additional studies in humans have shown that orally administered diflunisal is well absorbed, with 70 to 80% of the total radioactivity of the labelled drug being excreted in the urine (Tocco, Breault, Zacchie, Steelman & Perrier, 1975). In humans, peak plasma levels occur within 2 to 3 h after administration (Tocco *et al.*, 1975; Dresse, Gerard, Lays, Tempero & Verhaest, 1978).

Administered at a dose of 250 mg for a week, a steady state plasma level is reached in 4 to 5 days (Steelman, Breault, Tocco, Besselaar, Tempero, Lutterbeck, Perrier, Gribnau & Hinselmann, 1975). Plasma disappearance half-life following establishment of steady state conditions is approximately 11 to 12 h (Tempero, Cirillo & Steelman, 1977).

Since diflunisal is chemically related to salicylic acid and since acetylsalicylic acid exerts a dose dependent effect on uric acid excretion, a study was designed to determine what effect, if any, diflunisal might have on uric acid excretion. A preliminary report of this work has been presented by Dresse *et al.* (1975).

Methods

Study plan

This was a double-blind, placebo controlled study employing a parallel design, conducted in accordance with the provisions of the declaration of Helsinki. Twelve subjects were randomly assigned to three groups of four subjects each, the groups differing only in the medication they received.

On day 1, 7 days prior to the administration of drug, the subjects were placed on a low purine diet in order to stabilize the rate of uric acid excretion. To

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Table 1 Individual serum uric acid levels (mmol/l), means for the three treatment groups, and within group and between group comparisons for statistically significant changes

	Pre-treatment days				*Treatment days*					*Post-treatment day*
Subject number	*1*	*6*	*7*	*8*	*11*	*12*	*13*	*14*	*15*	*21*
Group I—Placebo										
2	0.357	0.393	0.381	0.309	0.375	0.363	0.387	0.363	0.398	0.381
7	0.363	0.309	0.339	0.369	0.315	0.357	0.387	0.363	0.363	0.357
12	0.309	0.363	0.357	0.303	0.303	0.303	0.303	0.279	0.268	0.297
Mean	0.345	0.357	0.357	0.327	0.333	0.339	0.357	0.333	0.345	0.345
Group II—Diflunisal 250 mg twice daily										
3	0.297	0.238	0.244	0.274	0.178	0.220	0.208	0.202	0.208	0.256
6	0.309	0.381	0.387	0.404	0.279	0.291	0.291	0.285	0.250	0.309
8	0.327	0.339	0.410	0.387	0.262	0.262	0.297	0.279	0.274	0.369
9	0.309	0.303	0.303	0.321	0.238	0.238	0.226	0.208	0.238	0.279
Mean	0.309	0.315	0.339	0.345	0.238[aa]	0.256[aa]	0.256[aa]	0.244[aa]	0.244[aa]	0.303[a]
Group III—Diflunisal 375 mg twice daily										
4	0.291	0.274	0.268	0.309	0.256	0.202	0.202	0.220	0.214	0.285
5	0.297	0.315	0.285	0.297	0.238	0.250	0.220	0.214	0.208	0.315
10	0.285	0.315	0.315	0.309	0.220	0.232	0.214	0.220	0.244	0.309
11	0.357	0.398	0.375	0.363	0.297	0.291	0.262	0.297	0.285	0.363
Mean	0.309	0.327	0.309	0.321	0.256[aa] A,BB	0.244[aa] AA,BB	0.226[aa] AA,BB	0.238[aa] AA B	0.238 AA BB	0.321

A,AA = 375 mg > placebo ($P < 0.05$, $P < 0.01$, respectively).
B = 250 mg > placebo ($P < 0.05$).
C = 375 mg > 250 mg ($P < 0.05$).
a,aa—increase from pre-treatment ($P < 0.05$, $P < 0.01$, respectively).
B,BB = 250 mg < placebo ($P < 0.05$, $P < 0.01$, respectively)

accomplish this, the subjects were denied purine rich foods such as sweetbreads, liver, kidneys, brains, anchovies, sardines and meat extracts. In addition all strong alcohols were prohibited and beer consumption was limited to moderate. The diet was continued until the morning of day 14. During this stabilization period and the subsequent drug treatment sequence, the subjects were permitted no medication other than the study drug.

In addition, on day 1, a complete laboratory safety evaluation including the determination of serum uric acid levels was performed. A 24 h urine collection was also made for the determination of total uric acid excreted.

On Days 6 and 7, prior to the administration of the test drug, blood samples were drawn and urines were collected for measurement of uric acid (serum and urine) levels as well as hematological, biochemical and urological safety parameters. This served as baseline data for the study.

On Days 8 through 15, the subjects received the following drugs:
Group I—Placebo: Days 8 through 15.
Group II—Diflunisal: 125 mg once on Day 8; 250 mg twice daily on Days 9 through 15.
Group III—Diflunisal: 250 mg twice on Day 8; 375 mg twice daily on Days 9 through 15.
Subjects were given the medication at 08.00 and 20.00 h, and matching placebo were used as necessary to maintain double-blindness. Blood and urine samples were obtained for the measurement of serum and urine uric acid levels.

Data to provide information on the presence or absence of residual drug effects were collected on Day 21.

Subjects

Twelve male volunteers, judged to be healthy normal individuals on the basis of clinical history, physical examination and laboratory determinations, participated in the study. The subjects were 22 to 27 years old (mean 25 years), weighed 64 to 91 kg (mean 75 kg) and 169 to 189 cm tall (mean 179 cm). The three study groups were comparable as to age, weight and height.

Materials

Diflunisal was prepared as 250 mg and 375 mg tablets which were identical in appearance. Placebo medication consisted of tablets in the same image.

Blood samples

Blood samples for the determination of serum uric acid levels and safety parameters were taken at 08.00 h (before drug on those days on which drug was taken) on Days 1, 6 through 15, and 21.

Urine collections

Twenty-four-hour (beginning at 08.00 h) urine collections were made on Days 1, 6 through 15 and 21. Volumes were measured, uric acid levels determined and various parameters measured to assess the safety of the medication.

Analytical procedures

Serum and urine uric acid levels were determined using the Boehringer uricase method (Praetorius & Poulsen, 1953; Kortüm & Kling, 1972). The results are given in mmol/l and must be multiplied by 16.8 to be expressed in mg %.

Statistical analysis

Means were calculated for each treatment group at Days 1, 6 through 15, and 21. Within group changes from pretreatment were tested with paired t-tests (one-tailed) comparing each change to zero. Between group comparisons were made using the 2-sample t-test (one-tailed) for each of the three pairs of treatments at each day. The pooled error variances for the three groups were used at each day for these tests. Because of the exploratory nature of this study, each test was performed at $\alpha = 0.05$, rather than controlling the overall significance level.

Results

Twelve subjects participated in the study; however, data from one subjects (AN 1) who was member of Group I has not been included in the analysis because of a protocol violation. This subject took by mistake 375 mg diflunisal instead of placebo at 08.00 h and 20.00 h on day 10. Hence, there are only three subjects in Group I while Group II and III contain each four subjects.

Serum uric acid

Serum uric acid levels for each subjects, the means for the three treatment groups and a comparison of the means for statistically significant changes both within groups and between groups are given in Table 1.

There was a statistically significant decrease in serum uric acid on day 11 through 15 for both the 250 mg twice daily and 375 mg twice daily diflunisal treated group compared to the placebo group; however, there were no significant differences between the two diflunisal treated groups. Statistically significant decreases also occurred for both diflunisal treated groups on treatment Days 11 through 15 compared to their respective pretreatment Day 8 levels. By 7 days post-treatment, the levels had returned to pretreatment levels except for the 250 mg

Table 2 Individual uric acid clearance values (ml/min—24 h period), means for the three treatment groups, and within group and between group comparisons for statistically significant changes

	Pre-treatment days			*Treatment days*					*Post-treatment day*
	1	*6*	*7*	*10*	*11*	*12*	*13*	*14*	*15*	*21*
Group I—Placebo										
2	7.8	6.9	7.9	9.3	7.7	8.0	9.0	8.6	7.4	9.3
7	11.3	11.0	9.3	11.4	11.2	12.3	9.5	10.6	11.8	10.7
12	7.7	7.2	7.5	8.5	10.0	9.0	9.4	9.8	11.2	9.5
Mean	8.9	8.4	8.2	9.7	9.6	9.8	9.7	9.7	10.1	9.8
Group II—Diflunisal 250 mg twice daily										
3	11.1	9.9	11.3	16.7	18.7	17.1	13.1	18.3	—	10.7
6	6.7	8.5	6.0	11.9	9.5	11.7	10.5	11.5	12.7	3.6
8	10.0	8.5	9.6	16.2	13.6	13.4	10.6	12.5	11.7	9.4
9	9.3	8.8	8.8	18.0	12.2	15.9	15.3	14.8	16.2	8.9
Mean	9.3	8.9	8.9	15.7^{aa}	13.5^{aa}	14.5^{aa}	12.4^{a}	14.3^{aa}	13.5^{aa}	8.2
Group III—Diflunisal 375 mg twice daily										
4	12.6	10.2	10.5	15.5	14.0	13.7	15.0	14.3	14.3	8.4
5	16.3	14.6	15.1	23.0	20.2	19.5	25.7	25.7	23.9	13.9
10	10.3	14.2	8.8	16.5	17.4	18.4	21.3	19.3	14.8	10.6
11	9.4	8.1	6.3	12.5	12.9	10.7	15.2	12.9	12.6	9.3
Mean	12.2	11.8	10.2	16.9^{aa}	16.1^{aa}	15.6^{aa}	19.5^{aa}	18.1^{aa}	16.4^{aa}	10.6
		A		AB	A	AB	AC	A	A	

A,AA = 375 mg > placebo ($P < 0.05$, $P < 0.01$, respectively).
B = 250 mg > placebo ($P < 0.05$).
C = 375 mg > 250 mg ($P < 0.05$).
a,aa—increase from pretreatment ($P < 0.05$, $P < 0.01$, respectively).
B,BB = 250 mg < placebo ($P < 0.05$, $P < 0.01$, respectively).

twice daily treatment group, which was still somewhat lower than the initial control.

Uric acid clearance

Uric acid clearance values for each subjects, the means for the three treatment groups and a comparison of the means for statistically significant changes both within groups and between groups are given in Table 2.

There was a statistically significant increase in uric acid clearance on Days 10 and 12 for the 250 mg twice daily diflunisal treated group compared to the placebo group and on Days 10 through 15 for the 375 mg twice daily treated group, suggesting a dose-response relationship. Statistically significant increases also occurred for both diflunisal treated groups on treatment Days 10 through 15 compared to their respective pretreatment Day 7 values. By 7 days post-treatment, the values had returned to pretreatment levels.

Adverse reactions

Three of the four subjects in the 375 mg twice daily diflunisal group experienced epigastric pain, a side effect which was definitely attributed to diflunisal and which was obviated when the drug was taken with meals. A possible drug related erythema of the tongue or lip occurred in two of the eight treated subjects. In both cases it was considered mild.

There were no adverse changes in the hematological, biochemical or urological parameters measured for the diflunisal treated subjects.

Discussion

It has been reported that small doses of salicylates induce uric acid retention while larger doses cause uricosuria. Since diflunisal is a novel molecule chemically related to salicylic acid, it was of interest to determine if its effect on uric acid excretion was similar to that observed with salicylates.

From this study in eleven normal subjects it is concluded that diflunisal when given as dose regimens of 250 mg twice daily or 375 mg twice daily lowers serum uric acid and increases uric acid clearance.

The only noteworthy adverse effect was epigastric pain experienced by three of the four subjects who took the high dose of diflunisal on an empty stomach. This reaction was not observed when the subjects began taking the drugs with meals.

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