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Glycose aminoglycane excretion and concentration in the urine of patients with frequently recurrent calcium-oxalate lithiasis prior to and following Diclofenac-Na therapy

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Summary. Excretion and concentration of glycose aminoglycans were measured in 19 patients with frequent relapses of calcium-oxalate lithiasis, prior to and two weeks after onset of Diclofenac-Na therapy. In most patients, markedly anhanced glycose aminoglycan concentration and excretion could be demonstrated. Elevation of the high-molecular inhibitor potential in the urine results in reduced risk of calcium-oxalate lithogenesis, which may explain the therapeutic successe of nonsteroidal anti-inflammatory drugs in the treatment of therapy resistent calcium-oxalate lithiasis.

Key words: Glycose aminoglycanes – Diclofenac-Na therapy – calcium-oxalate lithiasis

In 1986 and 1987, we reported on the first successful short- and long-term administration of nonsteroidal anti-inflammatory drugs (NSAID) in therapy resistent calcium-oxalate stone formers [4, 5].

This type of therapy was initiated as a result of the effect on calcium metabolism by prostaglandin synthetase inhibitors. Another stimulus was the casual remark of a patient with recurrent stone disease that calculus episodes had been suppressed during and following long-term Diclofenac-Na therapy of rheumatism [7, 11, 14, 23].

In structure, the phenyl acetic acid derivative Diclofenac-Na (Rewodina®, Voltaren®) is an ampiphilic acid combining hydrophilic and lipophilic properties [20]. Diclofenac-Na is a cyclo-oxygenase inhibitor with optimal biological effectiveness [24, 27].

In comparison to other NSAID, Diclofenac-Na has the highest therapeutic index. It does not accumulate and is not contraindicated in the elderly, nor in patients with reduced renal function.

NSAID belong to different classes of chemical substances. But their anti-inflammatory-analgesic effect is always due to the inhibition of certain steps in the so-called arachidonic acid cascade [4, 16, 17, 25]. The inhibition of the cylclo-oxygenase pathway reduces the formation of prostaglandins, thromboxane and prostacycline. Moreover, inhibition of cell proliferation as well as of lysosomal enzymes (mediators of inflammation) by stabilization of the lysosomal membrane have been described for several antirheumatics [19, 21]. Structural and functional alteration in cartilage metabolism due to impaired protein, collagen and proteoglycane synthesis must be taken into consideration when administering several NSAID [15].

Proteoglycanes are formed from glycose aminoglycanes bound to proteins. Glycose aminoglycanes (GAG) are polysaccharide chains of recurrent identical disaccharide units [28]. These units form the main constituent in the interstitial substance of the connective tissue. In the renal tissue, GAG is mainly found in the tips of the papillae, in the medulla and the cortex. GAG are excreted by glomerular filtration in the kidney [10]. Metabolic changes in connective tissue, biosynthesis and degradation rate are reflected by GAG excretion in the urine.

A quantitative relation between GAG in the urine and in the tissue depends on its postmetabolic tissue concentration, which may be influenced by various pathological conditions (e.g. inflammation, urothelial and hepatic disease etc.) [15]. Even in the midsixties, GAG were thought to be crystallization promoting factors in lithogenesis [17]. Various in-vivo and in-vitro investigations, however, successfully demonstrated that the GAG inhibit calcium-oxalate crystallization [26]. Chondroitine-6-sulfate and chondroitine-4-sulfate are the main GAG constituents, which can be detected in the analysis of the uronic acid component. They contribute most effectively to inhibitory activity by preventing aggregation through adsorption to calcium-oxalate crystals [13].

GAG concentration and excretion were measured in the urine of calcium-oxalate stone patients prior to and during Diclofenac-Na therapy. Both the present knowledge of possible influences of NSAID on connective tissue metabolism parameters and still unknown reasons of an unfavourable influence on therapyresistant calcium-oxalate stone diathesis were taken into consideration.

Table 1. Age, sex, glycose aminoglycane excretion and concentration in calcium-oxalate lithiasis patients prior to and during Diclofenac-Na therapy

No.	Patient	Sex	Age	GAG (mg/100 ml)		GAG (mg/d)	
				prior to therapy	after 2 weeks therapy	prior to therapy	after 2 weeks therapy
1.	L. D.	m	49	2.84	4.55	42.6	50.1
2.	H. M.	m	57	3.12	2.7	39.0	41.0
3.	S. C.	f	63	5.64	6.0	109.0	108.0
4.	W. W.	m	63	2.2	3.2	38.5	51.2
5.	Z. C.	f	54	2.1	2.68	52.5	46.9
6.	K. K.	m	54	3.1	1.85	85.3	57.4
7.	R. H.	m	59	2.9	5.1	50.7	66.3
8.	W. H.	m	59	2.9	4.9	31.0	49.0
9.	P. B.	m	46	2.8	2.9	37.8	42.6
10.	K. R.	m	47	3.1	5.2	52.7	78.0
11.	A. K.	m	63	3.2	3.6	57.6	46.8
12.	K. O.	m	54	3.2	4.7	64.6	78.0
13.	B. W.	m	59	2.1	2.4	42.0	54.0
14.	W. W.	m	53	3.05	2.44	57.9	51.2
15.	L. K.-H.	m	57	6.4	7.2	83.2	68.4
16.	S. G.	f	63	3.2	4.1	56.6	73.0
17.	K. C.	f	45	3.5	4.5	73.5	112.5
18.	D. H.	m	51	4.9	7.1	110.3	152.6
19.	S. K.	f	48	3.2	5.1	72.0	91.8
				$\bar{x}$ 3.34 $\pm$ s 1.12 $\alpha < 0.1$	$\bar{x}$ 4.22 $\pm$ s 1.56	$\bar{x}$ 60.9 $\pm$ s 23.0 ns	$\bar{x}$ 69.4 $\pm$ s 29.2

m = male; f = female; ns = statistically not significant

Material and methods

In 19 patients with frequent relapses of calcium-oxalate lithiasis (6 females, 55–63 years, 13 males, 46–63 years, number of average calculus recurrencies per year prior to NSAID therapy: 7), who had been treated with Diclofenac-Na for more than 6 months because of unsatisfactory therapeutic results, medication was interrupted, and GAG concentration and excretion in the urine were measurred after an interval of two weeks. Treatment was subsequently continued with 3×75 mg Rewodina® to register GAG concentration and excretion again after two weeks. Those with urinary tract infection were excluded. Neither fluid nor food supply was restricted. For the determination of GAG content in the urine, the Dische carbazole reaction modified by Teller [31] was employed, Student's two-tailed *t*-test [8] was used to find statistical significance.

Results

Tables 1 and 2 show age and sex of the patients glycose aminoglycane excretion and concentration as well as urinary excretion prior to and during Diclofenac-Na therapy. During the period of investigation, 16 out of 19 patients responded by an increase in GAG concentration, in 13 subjects GAG excretion increased. For both GAG concentration ($\alpha < 0.1$) and GAG excretion (statistically not significant), an increase in mean values were demonstrated, of about 40%. Urine excretion were slightly decreased and may have influenced GAG excretion values.

Discussion

Despite well known side effects, good therapeutic results in therapy-resistant calcium-oxalate stone patients justify Diclofenac-Na therapy. Nevertheless, no exact statement can be made concerning the mode of action of the drug [4, 5]. Influence on the prostaglandin-controlled membrane transport of calcium and on vitamin D metabolism are supposed to reduce the total calcium potential in the urine and thus the tendency to crystallization [7]. Observations on changes in calcium excretion, however, are controversial [7, 9].

Retrospective evaluations on calcium-oxalate lithiasis patients after long-term treatment with NSAID show a decrease of oxalic acid excretion in the urine, the reason for which is not clear [29]. Eventually, these results reflect the wide pharmacological spectrum of NSAID, which is known from rheumatism research. This wide spectrum has many undefined possible influence in various metabolic systems. The interrelations between connective tissue metabolism, particularly cartilage metabolism, and glycose aminoglycanes may be significant because GAG represent an essential part of the high-molecular potential in the urine [30].

According to Palmoski and Brandt [22], the balance between chondrocyte induced synthesis and degradation of matrix components in the cartilage is disturbed in arthrosis as well as following NSAID therapy. The

Table 2. Urine excretion of calcium-oxalate lithiasis patients prior to and during Diclofenac-Na therapy

No.	urine excretion per day	
	prior to therapy	after 2 weeks therapy
1.	1.5	1.1
2.	1.25	1.52
3.	1.95	1.80
4.	1.75	1.6
5.	2.5	1.75
6.	2.75	3.1
7.	1.75	1.3
8.	1.1	1.0
9.	1.35	1.47
10.	1.7	1.5
11.	1.8	1.3
12.	2.02	1.66
13.	2.0	2.25
14.	1.9	2.1
15.	1.3	0.95
16.	1.77	1.78
17.	2.1	2.5
18.	2.25	2.15
19.	2.25	1.80
	$\bar{x}$ 1.84 ± s 0.430 ns	$\bar{x}$ 1.72 ± s 0.535

ns = statistically not significant

suppression of GAG synthesis depends on the degree of demage to the cartilage and the dose of different NSAID. Sites of action for active substances are intracellular compartments and sections of GAG biosynthesis (transcription of the nuclear protein gene, ribosomal nuclear protein synthesis, synthesis of GAG chains, secretion, adsorptive pinocytosis and GAG degradation by proteinases, glycoside sulfatases and radicals) [18]. They also affect protein synthesis required for energy metabolism.

In contrast to other NSAID, Diclofenac-Na is said not to inhibit GAG synthesis and cell proliferation and thus to be chondroprotective [1].

First, the results of the present study show that both prior to and during Diclofenac-Na treatment the GAG concentration mean values are markedly above those found so far in normal persons and calcium-oxalate lithiasis patients [2, 3]. Like the excretion values, they rise during therapy. This means that the inhibitory potential in the urine is decisively enhanced, particularly when the risk of calcium-oxalate lithogenesis is reduced by concentrations of inorganic low-molecular urine components.

At present, it is not possible to distinguish whether the enhanced GAG values result from enhanced GAG synthesis, from reduced GAG degradation or from both processes.

Possibly the interval chosen between interruption of therapy and "normal value determination" was too brief to reproduce the initial situation prior to NSAID therapy. The high protein binding capacity allows the interpretation of a delayed induction of structural alterations in cartilage metabolism [14]. On the other hand, the age of the patients must be taken into consideration. With increasing age, rheumatoid problems are more likely [12].

The GAG profile may be affected by variation within the individual, and by diurnal and annual rhythms [21].

The frequently observed decrease of urinaryexcretions, which is also suggested by mean value calculations, is of particular interest. Excluding alimentary factors, the well known reduction of renal plasma flow and glomerular filtration rate with inhibition of prostaglandin synthesis by NSAID becomes effective here. This is said, however, to be of no clinical relevance with normal kidney function [16].

These results will help to fill another gap in the interpretation of present results in the NSAID treatment of therapy resistant calcium-oxalate lithiasis.

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