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DIFFERENCES IN THE URINARY EXCRETION OF STEROID METABOLITES AFTER DEHYDROEPIANDROSTERONE-SULFATE ADMINISTRATION, IN FEMINIZED AND NON FEMINIZED CIRRHOTIC PATIENTS

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SUMMARY

Twenty four hours urinary excretion of 11-dO-17KS and his 16 α-Hidroxylated metabolites (16 α-OH-DHA and A-triol) has been studied in cirrhotic patients with and without signs of feminization, before and after 100 mg DHA-S administration. Results have been compared with those found in normal control subjects. Under basal conditions, feminized cirrhotics excreted a smaller amount of 11-dO-17KS compared with controls, being this difference enhanced after DHA-S administration. Feminized cirrhotics presented a decreased excretion of Et, DHA and 11-dO-17KS in comparison with both controls and non feminized patients. On the other hand a higher excretion of 16 α-OH-DHA after DHA-S administration was found in cirrhotic patients when compared with controls, being the increase significant only in the non feminized group. This could suggest the presence of an increased 16 α-hydroxylating capacity in the cirrhotic patients. In the feminized group this 16 α-hydroxylated metabolites could be further converted into other steroids, possibly estrogens.

A reduced urinary excretion of Androsterone (A), Ethiocholanolone (Et) and Dehydroepiandrosterone (DHA) in male cirrhotic patients, together with enhanced 16 α-hidroxy-DHA (16 α-OH-DHA) after a 50 mg. DHA sulfate (DHA-S) administration, has been reported by our group.[1]

In order to test the possible different metabolic pathways in the feminized and non feminized cirrhotic patients, 100 mg of DHA-S have been administrated to a group of feminized and a group of non feminized cirrhotic male patients, estimating the urinary excretion of A, Et, DHA (11-deoxy-17-Ketosteroids) (11-dO-17KS), and 16 α-hydroxylated compounds: 16 α-OH-DHA and 16 α-Androstentriol (A-triol).

Both these latter metabolites are rather low in the normal adult but they are present in the newborn [2, 3] reflecting special characteristics of fetal metabolism showing high 16 α-hidroxylating capacity.[4]

PATIENTS AND METHODS

Ten male cirrhotic patients and 5 controls between 45 and 68 years of age have been used in this study. Consent to perform the experiments was obtained from each subject. From the 10 patients with liver cirrhosis, assesed by laparoscopy and hepatic biopsy, 5 with a mean age of 58.6 years showed clinical signs of feminization.

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(Gynecomastia of more than 10 cm of diameter and testicular atrophy with less than 3 cm of longitudinal diameter). The other 5 patients and the control group, had the same mean age: 54.2 years.

Al subjects discontinued whatever treatment at least 7 days before the experiment.

After a 24 hours urine collection, 100 mg DHA-S (ro-6-6827-6 Lab. Hoffman La Roche) dissolved in saline were infused intravenously. The infusion was started between 9 and 10 am. and continued over a 4 hour period. Urine was again collected over 24 hours. A, Et, DHA (11-dO-17KS), 16 α-OHDHA and A-triol were extracted from the barium acetate purified urine after enzymatic hydrolysis with β-Glucoronidase — aryl--sulfatase as described elsewere [5] and measured by Gas-liquid chromatography. This measurement was performed as Trimetyl-Silyl (TMSi) and Methoxime-trimetyl-Silyl (MoTMSi) derivatives on OV-1 and OV-225 columns as previously described [1] in order to avoid any possible interference.

RESULTS

A good separation between all steroid derivates is achieved by double derivatization (TMSi) and (MoTMSi) of urinary extracts. On basal conditions feminized cirrhotic patients have a reduced excretion of Et ($p<0.05$), DHA ($p<0.01$) and 11-dO-17KS ($p<0.02$) in comparison with control subjects (table 1).

After the 100 mg of DHA-S administration this excretion pattern is maintained in cirrhotic patients with a significant decrease of DHA $p<0.005$) and 11-dO-17KS ($p<0.005$) specially in the feminized group, which also present a decreased excretion of Et ($p<0.05$). On the other hand higher excretions of 16 α-hydroxylated compounds are observed in cirrhotic patients after administering the substrate, being the increases only statistically significant in the non feminized group for 16 α-OH-DHA ($p<0.05$) and A-triol ($p<0.05$).

The excretion of 16 α-OH-DHA and A-triol altogether is three times higher in the non feminized group ($p<0.005$) and twice as much in the feminized group ($p<0.05$) than in the control subjects.

DISCUSSION

The G. L. C. behaviour of the TMSi and MoTMSi urinary steroid derivatives on OV-1 and V-225 columns allow a quantitative measurement for A, DHA, Et, 16 α-OH-DHA and A-triol.[1] It has been generally accepted that male patients with liver cirrhosis have a reduced 17-Ketosteroid (17-KS) excretion compared with normal males. Sonka [6] reported a positive significant correlation and the reduction in A, Et and DHA excretion and Oseko [7], showed modifications in the metabolism of exogenous DHA-S by the cirrhotic liver. However, as far as we know the possible differences in the metabolic pathways of the feminized and non feminized cirrhotics both under basal conditions and after a DHA-S administration have not been further investigated. Results obtained in this paper show again a reduced 11-dO-17KS excretion under basal conditions in both groups with liver cirrhosis, with the lowest excretion levels found in the feminized group. After administering 100 mg of DHA-S the excretion patterns remain similar, with higher excretions of 16 α-OH-DHA and A-triol found in the non feminized group. This may lead to the suggestion that the feminized patients are able to further convert 16 α-OH-metabolites to estrogens as has been

Table 1

Basal urinary excretion

(μg/d)

	A	Et	DHA	11 do 17 KS	16-OH DHA	A-triol	ξ16αOH
CONTROLS							
F. A.	901	1.169	68	2.138	190	160	350
L. G.	948	1.807	175	2.930	< 20	1.370	1.390
T. F.	502	513	< 20	1.025	120	70	190
V. C.	142	250	< 20	402	< 20	40	60
L. B.	1.046	1.094	75	2.215	270	60	330
$\bar{X}$	707	966	71	1.746	124	338	464
sem	169	272	28	450	48	256	240
FEMINIZED CIRRHOTICS							
C. P.	19	378	< 20	408	40	50	90
B. G.	1.335	183	37	1.555	160	490	650
C. C.	308	192	21	512	40	400	440
M. P.	677	271	33	981	670	320	990
E. M.	143	202	< 20	355	140	160	300
$\bar{X}$	496	245	26	786	246	284	494
sem	237	37	4	225	91	79	155
NON FEMINIZED CIRRHOTICS							
R. C.	632	263	253	1.148	40	210	250
F. H.	175	119	36	330	70	50	120
G. S.	781	1.268	< 20	2.059	540	1.040	1 580
A. C.	252	477	< 20	739	751	740	1 490
$\bar{X}$	471	527	77	1.075	210	450	732
sem	114	198	44	286	117	188	332

Table 2

Post DHA-S urinary excretion

(μg/d)

	A	Et	DHA	11 do 17 KS	16-OH DHA	A-triol	§16αOH
CONTROLS							
F. A.	1.939	2.055	5.285	11.219	1.145	910	2 055
L. G.	1.298	3.133	981	5.412	2.650	3.795	6 445
T. F.	1.386	1.040	4.595	7.021	1.210	955	1 755
V. C.	397	530	6.414	7.452	1.210	545	1 650
L. B.	1.016	1.482	9.400	12.425	1.430	180	2 165
$\bar{X}$	1.027	1.648	5.435	8.705	1.717	1.237	2 814
sem	252	448	1.439	1.330	293	645	919
FEMINIZED CIRRHOTICS							
C. P.	206	1.651	2.529	4.386	1.215	530	1 745
B. G.	937	269	175	1.371	4.940	2.875	7 815
C. C.	911	704	290	1.906	1.200	1.285	2 485
M. P.	1.091	429	1.082	2.602	9.275	2.185	11 460
E. M.	167	138	182	487	2.795	1.180	3 975
$\bar{X}$	662	638	852	2.150	3.885	1.611	5 495
sem	197	270	452	657	1.511	411	1 835
NON FEMINIZED CIRRHOTICS							
R. C.	1.064	177	886	2.141	10.030	4.590	14 620
F. H.	208	107	192	507	3.300	860	4 160
G. S.	1.026	1.886	697	3.614	3.565	2.795	6 360
A. C.	351	714	2.164	3.230	13.799	3.015	16 814
$\bar{X}$	961	959	839	2.763	7.673	2.223	10 488
sem	346	397	356	666	2.567	760	2 778

shown before in breast cancer [8] and in hepatoma.[9] It could be further suggested that non feminized cirrhotic patients mantain a higher sulfatase activity together with the possibility of using the free DHA substrate for Androstenedione formation.

16 α-hydroxylation could be an alternative route for the metabolism of the DHA-S substrate as shown by [10] in slices of cirrhotic liver. In our series these metabolites are excreted in higher amounts in the nom feminized patients suggesting an enhanced 16 α-hydroxylating system in this special group or a better utilization of these metabolites for estrogen formation by the feminized group.

The DHA-S substrate is better metabolized in the cirrhotic liver because of its sulfate form [10, 12] and in this situation is better accessible to the 16 α-hydroxylase, remembering an activity which is highly present in the newborn liver.[13, 14, 15]

THE FOLLOWING ABREVIATIONS AND TRIVIAL NAMES HAVE BEEN USED

Androsterone (A) = 3 α-hydroxy-5 α-androstan-17-one; Aetiocholanolone (Et) = 3 α-hidroxy-5 β-androstan-17-one; Dehydroepiandrosterone (DHA) = 3 β-hidroxy-5- androstan-17-one; Dehydroepiandrosterone-sulfate (DHA-S) = 5-androsten-17-one-3 β-yl--sulfate; 16-hydroxy-dehydroepiandrosterone (16 α-OH-DHA) = 3 β, 16 α-dihydroxy-5-androsten-17-one; Androstentriol (A-triol) = 5-Androsten-3 β, 16 α, 17 β-triol; Androstenedione = 4, androsten-3, 17 dione.

RESUMO

Procedeu-se à avaliação da excreção urinária de 24 horas de 11-dO-17KS e dos seus metabolitos 16 α-Hidroxilados (16 α-OH-DHA e A-triol), antes e após a administração de 100 mg de DHA-S, em doentes cirróticos com e sem sinais de feminização. Os resultados são comparados com os obtidos num grupo controlo. Em condições basais, os cirróticos com sinais de feminização excretam uma menor quantidade de 11-dO-17KS em comparação com os indivíduos normais. Este diferente grau de excreção acentua-se após a administração de DHA-S. Os doentes cirróticos com sinais de feminização apresentavam ainda, quando comparados com indivíduos normais e com doentes cirróticos sem sinais de feminização, uma menor excreção de Et, DHA e 11-dO-17KS. Por outro lado, a excreção urinária de 16 α-OH-DHA após administração de DHA-S era mais elevada nos doentes cirróticos que nos indivíduos normais, sendo no entanto a diferença apenas significativa em relação aos doentes sem sinais de feminização. Este facto sugere-nos hipoteticamente uma maior capacidade 16 α-hidroxilante nos doentes cirróticos. No grupo de doentes com evidência de feminização estes metabolitos 16 α-hidroxilados poderiam ser ainda convertidos em outros esteroides, provavelmente estrogénios.

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