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SERUM PHOSPHOLIPIDS IN TUMORS OF THE URINARY TRACT

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Serum phospholipids were evaluated in renal cell carcinoma, renal pelvic tumor, solitary renal cyst and bladder tumor. There was a trend that renal cell carcinoma showed lowered serum total phospholipids with decreased percentage of phosphatidyl choline in the composition, while phosphatidyl ethanolamine was increased in percentage. This trend was not noted in renal pelvic tumor and solitary renal cyst. The fact might be valuable in differentiating space occupying lesions of kidney.

Key words: serum phospholipids, renal cell carcinoma, tumors of the urinary tract

INTRODUCTION

The difference between tumor and normal tissue has been mainly pointed out in glycolysis, synthesis of nucleic acids and immunological aspects in experimental and clinical subjects[5]. Recently it has again been found that there is a difference in lipid metabolism between them[6]. However, clinical study on lipid has been rarely reported in malignant tumors[3,4].

This paper reports the estimation of serum total phospholipids along with their composition in 45 healthy adults as control, 7 cases of renal cell carcinoma, 7 cases of renal pelvic tumor, 2 cases of solitary renal cyst and 19 cases of bladder tumor.

MATERIAL AND METHODS

The method[2] used to estimate serum total phospholipids (TPL) is shown in the table 1. Lipids were extracted from serum 1.0 ml with 19 volumes of chloroform: methanol (2:1). Two phases were separated by adding H_2O (20% in volume). The upper phase was removed and the lower phase was washed twice with the addition of the theoretical upper phase (chloroform: methanol: H_2O=4:48:47) to the original volume. After the phase separation, the upper phase was again discarded. The lower phase was dried with the rotary evaporator. One aliquot of the extracted lipids was digested with perchloric acid to separate phosphorus from lipids, which was then reacted with 2.5% ammonium molybdate and 10% ascorbic acid for colorimetry. The amount of phosphorus was measured with the spectrophotometer under the wave length 850 mμ following Allen's method[1]. The value of phospholipid was calculated as 25 times the amount of phosphorus.

Table 1. The process of total phospholipids estimation

0.5ML	SERUM
9.5ML	FOLCH (CH_3CL : CH_3OH = 2 : 1)
2.5ML	H_2O
↓	
SHAKE AND DISCARD THE UPPER PHASE	
↓	
WASH WITH THE THEORETICAL UPPER PHASE	
↓	
EVAPORATE THE LOWER PHASE TO DRYNESS	
↓	
PHOSPHORUS ASSAY (ALLEN'S METHOD)	

The other aliquot of extracted lipids was used for thin layer chromatography (TLC) with 250 μ thick silica gel plate to study the composition of phospholipids (Table 2). The TLC plate was developed in the solvent system consisting of chloroform: methanol: ammonium hydroxide=65:35:5. The com-

Table 2. The process of estimation of phospholipids composition

250 THICK SILICA GEL-G PLATES
THE SOLVENT SYSTEM - CH_3CL : CH_3OH : NH_4OH = 65:35:5
1) SCRAPE AND ELUTE EACH COMPONENT OF PHOSPHOLIPIDS
2) DRY UNDER N_2 STREAM
3) ADD 4ML FOLCH AND 1ML H_2O
4) SEPARATE INTO PHASES AND DISCARD THE UPPER PHASE
5) WASH WITH 1.5ML THEORETICAL UPPER PHASE
6) DRY LOWER PHASE UNDER N_2 STREAM
7) PHOSPHORUS ASSAY

ponents of phospholipids were identified with standards of dipalmitoyl phosphatidyl choline (PC), lysophosphatidyl choline (LL) sphyngomyelin (SM) and phosphatidyl ethanolamine (PE).

Serum total phospholipids were estimated in 45 healthy adults as control, 7 cases of renal cell carcinoma, 7 cases of renal pelvic tumor and 2 cases of solitary renal cysts. In addition to them, the estimation was also performed in 19 cases of bladder tumor.

The composition of serum phospholipids was studied in 5 healthy adults as control, 5 cases of renal cell carcinoma, 2 cases of renal pelvic tumor and 2 cases of bladder tumor.

RESULT

1. *Serum total phospholipids*

The values in 45 healthy adults, 7 cases of renal cell carcinoma, 7 cases of renal pelvic tumor, 2 cases of solitary renal cyst and 19 cases of bladder tumor are shown in the figure 1.

The mean was 192±37 mg/100 ml in 45 healthy adults. When twice the standard error is counted as normal range, the control normal value ranges from 125 to 260 mg/100 ml.

In the cases of renal cell carcinoma, preoperative evaluation revealed abnormal low values in 4 out of 5. The case showing normal value has a well encapsulated small tumor measuring 4 cm in diameter and has remained tumor free postoperatively. Postoperative values were estimated in 6 cases. Normal values were obtained in all except

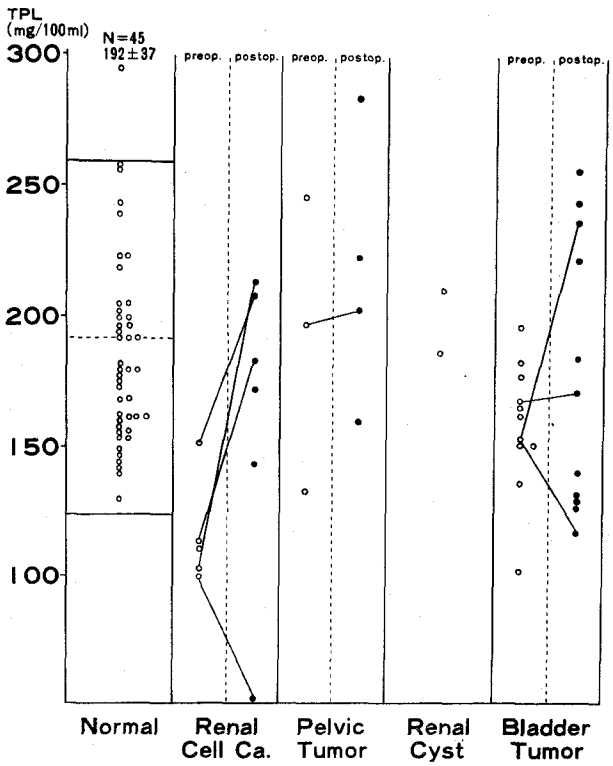

Fig. 1. Serum total phospholipids values in healthy adults and urinary tract tumors

one in whom the tumor was extremely advanced with metastases.

Estimations of total phospholipids in cases of renal pelvic tumor were within normal range except for one postoperative higher value 275 mg/100 ml.

Two cases of solitary renal cyst revealed normal values.

There were two pre- and post-operative abnormally low values among 19 cases of bladder tumor.

2. *The composition of serum phospholipids*

The control group of 5 healthy adults revealed lysolecithin approximately 10%, sphingomyelin 10 to 20%, phosphatidyl choline approximately 70%, and phosphatidyl ethanolamine lower than 10% (Table3).

This composition was changed in renal cell carcinoma (Table 4). Preoperative values revealed lowered percentage of phosphatidyl choline. Case No.3 showed decreased percentage of phosphatidyl choline 56% despite normal value of total phospholipids. In case No.5 with far advanced renal cell carcinoma, the percentage of sphingomyelin was elevated to 46. However, in all the other cases, elevated percentage was noted in phosphatidyl ethanolamine.

In cases of renal pelvic tumor, solitary renal cyst and bladder tumor, there were no marked deviations from the normal composition in the preoperative values (Table 5).

DISCUSSION

There are a few reports[3,4] concerning serum phospholipids in malignancy. Lee reported elevated serum total phospholipids in renal cell carcinoma. However, the presence or absence of hepatic metastasis and the stage of the tumor were not mentioned in the report.

It is a great problem to determine whether or not the decreased serum total phospholipids and the lowered percentage of phosphatidyl choline with increased phosphatidyl ethanolamine in the composition are specific to renal cell carcinoma. The decrease in serum total phospholipids

Table 3. Serum phospholipids composition in 5 healthy adults

CASES No.	Initials	Age	Sex	TPL (mg/100ml)	LL(%)	SM(%)	PC(%)	PE(%)	Others (%)
1.	C.K.	65	M	213	2	16	70	9	3
2.	H.U.	65	M	228	9	11	69	8	3
3.	S.K.	53	F	221	5.5	12	70	7	5.5
4.	Y.Y.	63	M	185	4	15	74	5	1
5.	Y.A.	54	F	196	8	17	70	4	1

Table 4. Serum phospholipids composition in renal cell carcinoma

CASES No.	Initials	Age	Sex	Stage of the Tumor	TPL (mg/100ml)	LL(%)	SM(%)	PC(%)	PE(%)	Others (%)
1.	K.M.	64	M (preop.)	C	105	6	12	51	21	10
2.	T.I.	63	M (Preop.)	B	112	12	15	56	12	5
3.	T.Y.	33	M (Preop.)	A	153	9	5	56	19	11
			(Postop.)		212	12	15	68	3	2
4.	A.Y.	45	F (postop.)	B	113	6	3	50	25	16
5.	Y.A.	62	M (postop.)	D	104	5	46	43	3	3
			(postop.)		48	11	10	73	4	2

Table 5. Serum phospholipids composition in various urinary tract lesions

	TPL(mg/100ml)	LL(%)	SM(%)	PC(%)	PE(%)	Others(%)
Renal Pelvic Tumor						
1. M.F. 73 M(preop.)	191	5	16	70	8	1
2. T.K. 61 M(preop.)	135	8	26	62	4	0
Renal Solitary Cyst						
1. M.S. 44 F(preop.)	165	9	12	68	9	2
2. S.I. 34 F(preop.)	188	9	11	72	4	4
Bladder Tumor						
1. E.K. 75 M(preop.)	151	6	2	87	2	3
2. Y.O. 62 M(preop.)	133	10	15	68	6	1
3. T.S. 78 F(preop.)	133	12	5	61	10	12

was more conspicuous in cases of renal cell carcinoma as compared to renal pelvic tumor and bladder carcinoma. The composition of serum phospholipids did not change markedly in cases of renal pelvic tumor, solitary renal cyst and cancer of bladder. It has been reported that serum phospholipids level becomes elevated in obstructive jaundice and decreases in marked damage of hepatic parenchyma. However, there was no significant hepatic dysfunction in the preoperative period in the reported 5 cases of renal cell carcinoma. Furthermore, their nutritional states were not so disturbed as to show abnormally low serum total protein or anemia. In case No.5 of renal cell carcinoma, generalized metastasis was observed postoperatively when serum phospholipid was measured. Therefore, marked decrease of postoperative serum phospholipids in this case might be due to disturbance of liver function and poor nutritional state. The authors assume that lowered values of serum phosphlipids in the other 4 cases are possibly due to the influence of the renal cell carcinoma, since their nutritional states were excellent and there was no evidence of liver dysfunction or liver lesions both clinically and in laboratory examination including liver scintiscan. Of interest is that preoperative lowered serum phospholipid levels returned to normal range postoperatively.

As for the composition of serum phospholipids, the percentage of phosphatidyl choline decreased and that of phosphatidyl ethanolamine increased in renal cell carcinoma, although in the other malignancies no marked difference was noted in the composition when compared to that of control. The change of serum total phospholipids and their composition suggests alteration of phospholipid metabolism specific to renal cell carcinoma. Geers et al.[7] reported that higher concentration of phosphatidyl choline of the tumor tissue of renal cell carcinoma compared to that of normal renal cortex, while the concentration of phosphatidyl ethanolamine of the tumor was lower than that of the remaining healthy cortex. The change of phospholipids composition of renal cell carcinoma might be related to the quite reverse alteration of serum phospholipids composition.

Taking advantage of the above fact, renal parenchymal tumor would be differentiated from renal pelvic tumor or solitary renal cyst. For the further study of phospholipid metabolism in renal parenchymal tumor, it would be necessary to investigate the *in vitro* incorporation of ^{32}P phosphatidic acid into phosphatidyl choline and phosphatidyl ethanolamine in tumor tissue along with tissue phospholipid composition.

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和文抄録

尿路系腫瘍における血清燐脂質

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腎癌7例，腎盂腫瘍7例，孤立性腎嚢胞2例，膀胱腫瘍19例，対照として健康成人45例において血清総燐脂質値を測定すると共に，腎癌5例，腎盂腫瘍2例，孤立性腎嚢胞2例，膀胱腫瘍3例において血清燐脂質の分画値について検討した．

腎癌症例では栄養状態良好で，肝障害など他に血清脂質値に影響を与える条件がない時期に血清総燐脂質の低下傾向がみられた．また分画では phosphatidyl choline が低下し，phosphatidyl ethanolamine が増加していた．これは Geers らの腎癌組織では正常腎皮質と比較して phosphatidyl cholineのしめる割合が高く phosphatidyl ethanolamineが低いという報告と考え合わせて興味深いことである．

また腎盂腫瘍，孤立性腎嚢胞では血清総燐脂質値，燐脂質分画においても異常はみられなかった．この事実は腎の space occupying lesions の鑑別にも役立つかもしれない．