Detection of Classic Serum Tumor Markers (CSTMs) Group Benefits to Differential Diagnosis and Disease Assessment of UTUC

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Research

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Abstract

Background: To explore the potential relationship of changes in classic serum tumor markers (CSTM) with differential diagnosis and disease assessment of upper tract urothelial carcinoma (UTUC).

Methods: 60 UTUC (56 operated), 44 RCCC and 36 NTHN, were included into this retrospective analysis. The initial classic serum tumor markers (CSTMs), including CA242, CA199, CA125, CEA, AFP, SCC and CA724, were compared among the three groups. The preoperative, 1 month postoperative and 1 year postoperative/PD parameters (value and abnormal rate) of CSTMs were compared in UTUC group. A recommend test strategy was given and rechecked. The pathological manifestations of tumoral tissues and paracancerous tissues were analyzed.

Results: The value of CA242, CA199 and CEA were higher in UTUC than in RCCC. The value of CEA was higher in UTUC than in NTHN. The value of CA199 and CEA were higher in UTUC than in RCCC+NTHN. The AR of CA199, CEA, SCC and CA724 were higher in UTUC than in RCCC. The AR of CEA and CA724 were higher in UTUC than in NTHN. The AR of CA199, CEA, SCC and CA724 were higher in UTUC than in RCCC+NTHN. The preoperative value of CA242 and CA199 were higher than 1 month postoperative ones in UTUC. The 1 year postop/PD value of CA242 and AFP were higher than 1 month postoperative ones in UTUC. For postop PD patients, the value during PD of CA242, CA199 and AFP were higher than 1 month postop ones. The preoperative AR of CA199 and CEA were higher than 1 month postoperative ones in UTUC. The recommended test strategy was given: CA242+CA199+CEA+AFP+SCC+CA724, and was rechecked. The P values were almost the lowest and the positive results covered all the comparisons. In 56 operated cases, the NAC in CSTMs by the recommended test strategy was statistically related with the tumor load. It appeared positive labelling by Ab of CA199, CA125, CEA, AFP and CA724 in UTUC tissues, while negative in paracancerous tissues.

Conclusions: CSTMs may help to make differential diagnosis and disease assessment of UTUC. Group test (CA242+CA199+CEA+AFP+SCC+CA724) was recommended and more valuable.

Trial registration: Not applicable

Background

Urinary epithelial transitional cell carcinoma (TCC) includes renal pelvic cancer, ureteral cancer, bladder cancer, and urinary tract cancer. Among them, most studies aimed at finding a diagnostic and prognostic index focused on bladder carcinoma. Some indicators like CxBladder monitor, UroVysion, NMP-22 and bladder tumor antigen were utilized in diagnosis and monitoring of TCC, however, few biomarkers achieve high sensitivity and specificity [1]. And other biomarkers, such as FGFR3, p53, pRb, p21, Ki67 and VEGF, were used as prognostic factors for bladder cancer in previous studies [2–6]. And even some genomic biomarkers were proposed in recent researches [7, 8]. However, only a few established prognostic factors were found to effectively assess the tumor progression of upper tract urothelial carcinoma (UTUC) [9]. On the other hand, some classic serum tumor markers (CSTM) such as carbohydrate associated cancer antigen 199 (CA199), cancer antigen 125 (CA125), carcinoembryonic antigen (CEA) and alpha-fetoprotein (AFP), which were widely used for the diagnosis of different types of gastrointestinal cancer[10], seem to have more evaluation functions. In this study, in term of differential diagnosis and disease assessment of UTUC, we analyzed 7 CSTMs commonly utilized in clinical detection, including CA242, CA199, CA125, CEA, AFP, SCC and CA724, which partially presented abnormal in majority of cases. We attempted to explore the potential relationship between CSTMs changes and the UTUC changes.

Methods

Ethics statement

All procedures performed in this study involving human participants were in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by institutional ethics committee of Shanghai Ruijin Hospital and informed consent was taken from all the patients.

Study subjects

A total of 140 unilateral affected patients, in which 60 UTUC, 44 RCCC and 36 NTHN (with ipsilateral renal dysfunction), were included into this retrospective, descriptive analysis. In UTUC patients, 56 of them accepted the surgical therapy (radical nephroureterectomy for 53 patients and radical nephrectomy for the rest 3 patients due to the misdiagnose as kidney carcinoma) during the year 2014–2020 in Department of Urology in Ruijin Hospital. The RCCC and NTHN cases were collected as control (in 2018–2019, Ruijin Hospital). All UTUC diagnoses and recurrence, implantation or metastasis diagnoses were confirmed by pathological analysis (biopsy or surgical specimens). There were 1 multiple distant metastasis case and 2 cases of lymphatic metastasis and multiple distant metastasis in preop examinations, to whom the surgical therapy was not suitable. There was also 1 patient without detectable metastasis refused to accept the operation. None of the patients was combined with bladder or urethral transitional cell carcinoma (TCC) in UTUC group. None of the UTUC patients had received radiotherapy, chemotherapy, or immuno-therapy before the initial test of CSTMs. Preoperative metastatic RCCC occurred in 1 patient (iliac joint metastasis) and one case of tumor thrombus in main renal vein.

Methods

The value and the AR of CSTMs were both counted. The comparisons of CSTMs were done among the UTUC, RCCC, NTHN and RCCC + NTHN group, also between preop and 1 month postop, 1 month postop and 1 year postop or progression of disease (PD), 1 month postop and postop PD in UTUC cases. Once a CSTMs had revealed a statistical value in any of the comparisons in this study, it would be gathered into a recommended test strategy. The strategy obtained based on these comparisons would be reanalyzed in all comparisons to compare with the single CSTMs for testing its application value.

The pathological analysis was also collected in 56 UTUC cases which was non-metastatic and have accepted the surgical treatment. The tumor load, grade and infiltration were statistically observed with the value and the NAC of preop CSTMs. The analysis of pathological sections were also specifically stained for detection of the expression of CA724, CA125, CEA, AFP and CA199, the antibodies (Ab) for immunohistochemical stain of CA242 and SCC were not available. The tumoral tissues and the paracancerous tissues were pair-compared.

Biochemical detection of classic serum tumor markers

The CSTMs test is as a routine preoperative examen for all patients in our department and also as a routine follow-up examen of the UTUC patients recent years. The serum CA242, CA199, CA125, CEA, AFP, SCC and CA724 levels were detected by electrochemiluminescence immunoassay (Cobas; Roche Diagnostics, Germany) at the Department of clinical laboratory of Ruijin Hospital affiliated to Shanghai Jiao Tong University School of Medicine, Shanghai, China. The normal reference values were as follows: CA199 < 35 U/ml, CA242 < 20 IU/ml, CA724 < 8.2 U/ml, CA125 < 35 U/ml, CEA < 5 ng/ml, AFP < 9 ng/ml, SCC < 1.5ng/ml.

In this study, 39 UTUC patients had detected the CSTMs 1 month postoperative and 23 of them had the CSTMs results 1 year postoperative (without recurrence and metastasis) or during PD (with recurrence and metastasis).

Statistical analysis

The status of the level of each CSTM were expressed as mean ± standard error of the mean (SEM). Measurement data between groups were compared with the group t-test, paired t-test, chi-square test (χ²) of a four-fold table, Fischer exact test, Wilcoxon two-sample test, and linear correlation analysis. Statistical analysis was performed using SPSS version 23. All tests were two-tailed and P < 0.05 was considered statistically significant.

Results

General parameters

The comparisons of the gender, age, side, BMI, affected renal function and metastasis rate in 3 groups were presented in Table 1.

Table 1
General data statistics in UTUC, RCCC and NTHN patients

Parameters		UTUC (n = 60)	RCCC (n = 44)	NTHN (n = 36)	RCC + NTHN (n = 80)	P value (UTUC v.s. RCCC)	P value (UTUC v.s. NTHN)	P value (UTUC v.s. RCCC + NTHN)
Gender	Male (n, %)	42 (70.00)	34 (77.27)	17 (47.22)	51 (63.75)	0.128	0.015	0.107
	Female (n, %)	18 (30.00)	10 (22.73)	19 (52.78)	29 (36.25)			
Age (y)		67.23 ± 9.74	56.14 ± 11.89	53.97 ± 15.62	55.16 ± 13.65	<0.001	<0.001	<0.001
Side	Left (n, %)	36 (60.00)	24 (54.55)	22 (61.11)	46 (57.50)	0.136	0.170	0.132
	Right (n, %)	24 (40.00)	20 (45.45)	14 (38.89)	34 (42.50)			
BMI (kg/m ²)		23.92 ± 3.08	25.60 ± 3.34	23.02 ± 3.15	24.44 ± 3.51	0.010	0.175	0.360
Affected renal function (mL/min)		26.99 ± 13.00	40.30 ± 12.66	/	/	<0.0001	/	/
Metastasis (n, %)		3 (5.00)	1 (2.27)	/	/	0.328	/	/

*UTUC = Upper tract urothelial carcinoma; RCC = Renal cell carcinoma; NTHN = non-tumoral hydronephrosis.

CSTMs value in UTUC, RCCC and NTHN patients

The value of CA242 (8.42 ± 8.82 U/mL v.s. 5.07 ± 4.68 U/mL, P = 0.014), CA199 (24.73 ± 36.60 U/mL v.s. 11.16 ± 10.96 U/mL, P = 0.008) and CEA (4.38 ± 6.41 ng/mL v.s. 2.25 ± 1.05 ng/mL, P = 0.022) were significantly higher in UTUC group than in RCCC group. The value of CEA (4.38 ± 6.41 ng/mL v.s. 2.39 ± 1.32 ng/mL, P = 0.014) was significantly higher in UTUC group than in NTHN group. The value of CA199 (24.73 ± 36.60 U/mL v.s. 13.49 ± 16.37 U/mL, P = 0.028) and CEA (4.38 ± 6.41 ng/mL v.s. 2.33 ± 1.20 ng/mL, P = 0.017) were much higher in UTUC group than in RCCC + NTHN group. (Table 2)

Table 2
Comparison of CSTMs in UTUC, RCCC and NTHN patients

CSTMs and recommended test strategy		tUTUC (n = 60)	nmUTUC (n = 56)	RCCC (n = 44)	NTHN (n = 36)	RCCC + NTHN (n = 80)	P value (tUTUC v.s. RCCC)	P value (tUTUC v.s. NTHN)	P value (tUTUC v.s. RCCC + NTHN)
CA242	---X±SD (U/mL)	8.42 ± 8.82	7.96 ± 8.44	5.07 ± 4.68	8.80 ± 15.63	6.75 ± 11.12	0.014	0.878	0.339
	Abnormal rate (n, %)	5 (8.33)	3 (5.36)	0 (0.00)	2 (5.56)	2 (2.50)	0.059	0.289	0.096
CA199	---X±SD (U/mL)	24.73 ± 36.60	22.38 ± 30.50	11.16 ± 10.96	16.33 ± 21.04	13.49 ± 16.37	0.008	0.206	0.028
	Abnormal rate (n, %)	11 (18.33)	10 (17.56)	2 (4.55)	2 (5.56)	4 (5.00)	0.027	0.054	0.010
CA125	---X±SD (U/mL)	11.47 ± 8.16	10.98 ± 8.03	11.33 ± 4.58	13.15 ± 8.12	12.15 ± 6.44	0.919	0.330	0.582
	Abnormal rate (n, %)	2 (3.28)	2 (3.57)	0 (0.00)	0 (0.00)	0 (0.00)	0.331	0.388	0.182
CEA	---X±SD (ng/mL)	4.38 ± 6.41	3.31 ± 3.93	2.25 ± 1.05	2.39 ± 1.32	2.33 ± 1.20	0.022	0.014	0.017
	Abnormal rate (n, %)	10 (16.67)	7 (12.50)	2 (4.55)	0 (0.00)	2 (2.50)	0.041	0.007	0.003
AFP	---X±SD (ng/mL)	2.83 ± 1.32	2.84 ± 1.35	2.98 ± 1.61	2.45 ± 1.31	2.74 ± 1.50	0.591	0.176	0.728
	Abnormal rate (n, %)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1.000	1.000	1.000
SCC	---X±SD (ng/mL)	4.29 ± 13.33	2.05 ± 5.31	1.21 ± 0.89	0.98 ± 0.49	1.08 ± 0.70	0.059	0.080	0.067
	Abnormal rate (n, %)	13 (21.67)	10 (17.56)	3 (6.82)	5 (11.36)	8 (10.00)	0.026	0.142	0.032
CA724	---X±SD (U/mL)	3.35 ± 3.83	3.04 ± 3.77	3.02 ± 3.22	2.57 ± 2.45	2.82 ± 2.89	0.652	0.278	0.355
	Abnormal rate (n, %)	8 (13.33)	6 (10.71)	0 (0.00)	0 (0.00)	0 (0.00)	0.010	0.019	0.001
Strategy	According to value			CA242 + CA199 + CEA	CEA	CA199 + CEA	-	-	-
	According to abnormal rate			CA199 + CEA + SCC + CA724	CEA + CA724	CA199 + CEA + SCC + CA724	-	-	-
	NAC by the recommended strategy (CA242 + CA199 + CEA + AFP + SCC + CA724) (Nonparametric, rank mean)			59.07 v.s. 43.55	53.33 v.s. 40.44	81.90 v.s. 61.95	0.003	0.013	0.001
*CSTM = Classic serum tumor marker; UTUC = Upper tract urothelial carcinoma; tUTUC = Total upper tract urothelial carcinoma (non-metastatic and metastatic UTUC); nmUTUC = Non-metastatic upper tract urothelial carcinoma; RCCC = Renal clear cell carcinoma; NTHN = Non-tumoral hydronephrosis; NAC = Number of changes (in CSTMs).									

AR of CSTMs in UTUC, RCCC and NTHN patients

The AR of CA199 (18.33 % v.s. 4.55 %, P = 0.027), CEA (16.67 % v.s. 4.55 %, P = 0.041), SCC (21.67 % v.s. 6.82 %, P = 0.026) and CA724 (13.33 % v.s. 0.00 %, P = 0.010) were significantly higher in UTUC patients than in RCCC patients. The AR of CEA (16.67 % v.s. 4.55 %, P = 0.007) and CA724 (13.33 % v.s. 0.00 %, P = 0.019) were significantly higher in UTUC patients than in NTHN patients. The AR of CA199 (18.33 v.s. 5.00 %, P = 0.010), CEA (16.67 % v.s. 2.50 %, P = 0.003), SCC (21.67 % v.s. 10.00 %, P = 0.032) and CA724 (13.33 % v.s. 0.00 %, P = 0.001) were significantly higher in UTUC patients than in RCCC + NTHN patients. (Table 2)

Preoperative and postoperative CSTMs value

The preoperative value of CA242 (8.44 ± 8.66 U/mL v.s. 4.41 ± 3.05 U/mL, P = 0.004) and CA199 (21.23 ± 26.08 U/mL v.s. 10.16 ± 8.08 U/mL, P = 0.009) were significantly higher than 1 month postoperative ones in UTUC patients. The 1 year postop/PD value of CA242 (6.87 ± 4.77 U/mL v.s. 4.66 ± 3.16 U/mL, P = 0.007) and AFP (3.71 ± 1.74 ng/mL v.s. 3.08 ± 1.42 ng/mL, P = 0.032) were significantly higher than 1 month postoperative ones in UTUC patients. For the postop PD patients, the value during PD of CA242 (7.11 ± 5.93 U/mL v.s. 5.13 ± 4.11 U/mL, P = 0.034), CA199 (27.40 ± 25.12 U/mL v.s. 11.59 ± 6.58 U/mL, P = 0.034) and AFP (4.23 ± 2.12 ng/mL v.s. 2.87 ± 1.37 ng/mL, P = 0.024) were significantly higher than 1 month postop ones. (Table 3)

Table 3
Preoperative and postoperative CSTMs in non-metastatic UTUC patients

CSTMs and recommended test strategy		Preop (n = 39)	1 month postop (n = 39)	P value	1 month postop (n = 23)	1 year postop/PD (n = 23)	P value	1 month postop (n = 10)	Postop PD (n = 10)	P value
CA242	---X±SD (U/mL)	8.44 ± 8.66	4.41 ± 3.05	0.004	4.66 ± 3.16	6.87 ± 4.77	0.007	5.13 ± 4.11	7.11 ± 5.93	0.034
	Abnormal rate (n, %)	2 (5.13)	0 (0.00)	0.247	0 (0.00)	0 (0.00)	1.000	0 (0.00)	0 (0.00)	1.000
CA199	---X±SD (U/mL)	21.23 ± 26.08	10.16 ± 8.08	0.009	10.60 ± 6.46	17.32 ± 18.87	0.055	11.59 ± 6.58	27.40 ± 25.12	0.034
	Abnormal rate (n, %)	8 (20.51)	1 (2.56)	0.013	0 (0.00)	3 (13.04)	0.117	0 (0.00)	3 (30.00)	0.105
CA125	---X±SD (U/mL)	11.82 ± 9.03	16.77 ± 17.64	0.072	16.50 ± 18.42	17.50 ± 30.51	0.762	20.20 ± 26.79	29.39 ± 44.50	0.172
	Abnormal rate (n, %)	2 (5.13)	3 (7.69)	0.321	1 (4.35)	1 (4.35)	0.511	1 (10.00)	1 (10.00)	0.526
CEA	---X±SD (ng/mL)	3.55 ± 4.65	2.21 ± 0.95	0.080	2.32 ± 1.11	2.45 ± 1.21	0.327	2.31 ± 1.05	2.45 ± 1.23	0.370
	Abnormal rate (n, %)	6 (15.38)	0 (0.00)	0.013	1 (4.35)	2 (8.70)	0.383	0 (0.00)	1 (10.00)	0.500
AFP	---X±SD (ng/mL)	2.83 ± 1.45	2.81 ± 1.38	0.877	3.08 ± 1.42	3.71 ± 1.74	0.032	2.87 ± 1.37	4.23 ± 2.12	0.024
	Abnormal rate (n, %)	0 (0.00)	0 (0.00)	1.000	0 (0.00)	1 (4.35)	0.500	0 (0.00)	1 (10.00)	0.500
SCC	---X±SD (ng/mL)	2.18 ± 6.06	1.14 ± 0.42	0.294	1.13 ± 0.49	1.25 ± 0.52	0.336	1.13 ± 0.49	1.33 ± 0.70	0.414
	Abnormal rate (n, %)	7 (17.95)	4 (10.26)	0.163	3 (13.04)	5 (21.74)	0.216	2 (20.00)	3 (30.00)	0.348
CA724	---X±SD (U/mL)	2.72 ± 3.06	2.52 ± 1.70	0.610	2.41 ± 1.76	4.43 ± 7.53	0.181	2.08 ± 1.44	6.31 ± 10.28	0.202
	Abnormal rate (n, %)	3 (7.69)	0 (0.00)	0.120	0 (0.00)	3 (13.04)	0.117	0 (0.00)	2 (2.00)	0.237
Strategy	According to value	CA242 + CA199			CA242 + AFP			CA242 + CA199 + AFP		
	According to abnormal rate	CA199 + CEA			/			/		
	NAC by the recommended strategy (CA242 + CA199 + CEA + AFP + SCC + CA724) (Rank mean)	9.65	7.00	0.0003	0.00	6.00	0.001	0.00	4.50	0.005

*CSTM = Classic serum tumor marker; UTUC = Upper tract urothelial carcinoma; NAC = Number of changes (in CSTMs).

Preoperative and postoperative AR of CSTMs

The preoperative AR of CA199 (20.51 v.s. 2.56 %, P = 0.013) and CEA (15.38 % v.s. 0.00 %, P = 0.013) were significantly higher than 1 month postoperative ones in UTUC patients. (Table 3)

Strategy of CSTMs test in UTUC

According to the results as above, 6 CSTMs: CA242, CA199, CEA, AFP, SCC and CA724 were recommended to be tested for the UTUC patients. We rechecked the test strategy CA242 + CA199 + CEA + AFP + SCC + CA724

in the comparisons and found that by this strategy, the NAC of CSTMs in UTUC group was significantly higher than that in RCCC (P = 0.003), NTHN (P = 0.013) and RCCC + NTHN group (P = 0.001). The preop NAC of CSTMs in UTUC group was significantly higher than 1 month postop one (P = 0.0003). The 1 year postop/PD NAC of CSTMs in UTUC group was much higher than 1 month postop one (P = 0.001). The postop PD NAC of CSTMs in UTUC group was significantly augmented than 1 month postop in the same patients (P = 0.005). (Table 2, Table 3)

Pathological manifestations and CSTMs in UTUC

In 56 patients with surgical specimens, the NAC in CSTMs by the recommended test strategy was statistically related with the tumor load, which was $31935.96 \pm 72663.83 \text{ mm}^3$ (P < 0.001). The single CSTM was not linear correlated with the tumor load (P > 0.05 for all of them). All other pathological

parameters, like grade, submucosa invasion, muscle invasion, adventitia invasion, renal parenchyma invasion and perirenal fat invasion had no significant influence on the values and the AR of CSTMs, also the NAC in CSTMs by the recommended test strategy, except three statistical differences: (1) the value of CA125 in muscle invaded cases (13.41 ± 10.44 U/mL *v.s.* 8.56 ± 3.19 U/mL) was much higher than that in muscle non-invaded ones ($P = 0.025$); (2) the value of AFP in submucosa non-invaded cases (3.77 ± 1.75 ng/mL *v.s.* 2.66 ± 1.20 ng/mL) was significantly higher than that in submucosa invaded ones ($P = 0.022$); (3) the AR of SCC in adventitia non-invaded cases (25.00 % *v.s.* 0.00 %) was significantly higher than in adventitia invaded ones ($P = 0.024$). (Table 4)

Table 4
Pathology and CSTMs in nmUTUC (n = 56)

Pathological parameters		CA242		CA199		CA125		CEA		AFP		SCC	
		Value (U/mL)	AR (%)	Value (U/mL)	AR (%)	Value (U/mL)	AR (%)	Value (ng/mL)	AR (%)	Value (ng/mL)	AR (%)	Value (ng/mL)	AR (%)
Tumor load ($31935.96 \pm 72663.83\text{mm}^3$) (LCA, P value)		0.366	/	0.063	/	0.966	/	0.505	/	0.334	/	0.709	/
Grade	Low grade (n = 6)	12.79 ± 17.52	1 (16.67)	31.69 ± 54.78	1 (16.67)	9.49 ± 3.27	0 (0.00)	6.93 ± 11.50	1 (16.67)	2.81 ± 1.45	0 (0.00)	7.65 ± 15.22	3 (50.00)
	High grade (n = 50)	7.38 ± 6.42	2 (4.00)	21.26 ± 26.95	9 (18.00)	11.16 ± 8.42	2 (4.00)	2.88 ± 1.44	6 (12.00)	2.85 ± 1.35	0 (0.00)	1.38 ± 1.94	7 (14.00)
	P value	0.486	0.265	0.664	0.422	0.634	0.796	0.427	0.411	0.948	1.000	0.359	0.056
Submucosa	Non-invaded (n = 9)	12.54 ± 15.14	2 (22.22)	31.14 ± 47.75	2 (22.22)	8.35 ± 1.94	0 (0.00)	2.45 ± 1.56	1 (11.11)	3.77 ± 1.75	0 (0.00)	1.13 ± 0.53	2 (22.22)
	Invaded (n = 47)	7.08 ± 6.36	1 (2.13)	20.70 ± 26.42	8 (17.02)	11.49 ± 8.65	2 (4.26)	3.48 ± 4.23	6 (12.77)	2.66 ± 1.20	0 (0.00)	2.23 ± 5.79	8 (17.02)
	P value	0.316	0.061	0.540	0.318	0.286	0.702	0.481	0.417	0.022	1.000	0.576	0.318
Muscle	Non-invaded (n = 28)	8.07 ± 9.51	2 (7.14)	18.02 ± 29.45	3 (10.71)	8.56 ± 3.19	0 (0.00)	3.84 ± 5.37	4 (14.29)	3.09 ± 1.38	0 (0.00)	2.58 ± 7.14	7 (25.00)
	Invaded (n = 28))	7.85 ± 7.39	1 (3.57)	26.74 ± 31.44	7 (25.00)	13.41 ± 10.44	2 (7.14)	2.78 ± 1.46	3 (10.71)	2.60 ± 1.29	0 (0.00)	1.53 ± 2.45	3 (10.71)
	P value	0.923	0.382	0.289	0.109	0.025	0.246	0.317	0.289	0.177	1.000	0.466	0.109
Adventitia	Non-invaded (n = 40)	7.82 ± 9.40	3 (7.50)	21.49 ± 32.53	6 (15.00)	9.83 ± 5.68	1 (2.50)	3.52 ± 4.55	5 (12.50)	2.99 ± 1.37	0 (0.00)	2.49 ± 6.26	10 (25.00)
	Invaded (n = 16)	8.31 ± 5.60	0 (0.00)	24.61 ± 25.56	4 (25.00)	13.86 ± 11.84	1 (6.25)	2.78 ± 1.54	2 (12.50)	2.47 ± 1.25	0 (0.00)	0.97 ± 0.31	0 (0.00)
	P value	0.844	0.356	0.733	0.196	0.090	0.416	0.529	0.341	0.186	1.000	0.339	0.024
Renal parenchyma	Non-invaded (n = 44)	8.09 ± 9.14	3 (6.82)	21.10 ± 31.21	7 (15.91)	10.01 ± 5.60	1 (2.27)	3.53 ± 4.36	6 (13.64)	2.83 ± 1.41	0 (0.00)	2.37 ± 5.97	10 (22.73)
	Invaded (n = 12)	7.47 ± 5.40	0 (0.00)	27.07 ± 28.55	3 (25.00)	14.54 ± 13.49	1 (8.33)	2.52 ± 1.44	1 (8.33)	2.88 ± 1.45	0 (0.00)	0.88 ± 0.25	0 (0.00)
	P value	0.824	0.478	0.552	0.237	0.278	0.343	0.435	0.365	0.906	1.000	0.392	0.070
Perirenal fat	Non-invaded (n = 52)	8.16 ± 8.70	3 (5.77)	23.30 ± 31.46	10 (19.23)	10.20 ± 5.40	1 (1.92)	3.37 ± 4.08	7 (13.46)	2.90 ± 1.36	0 (0.00)	2.14 ± 5.51	10 (19.23)
	Invaded (n = 4)	5.38 ± 2.97	0 (0.00)	10.18 ± 5.43	0 (0.00)	21.15 ± 23.19	1 (25.00)	2.51 ± 0.37	0 (0.00)	2.11 ± 1.02	0 (0.00)	0.93 ± 0.30	0 (0.00)
	P value	0.531	0.797	0.412	0.444	0.415	0.135	0.679	0.577	0.264	1.000	0.664	0.444

*CSTM = Classic serum tumor marker; nmUTUC = Non-metastatic upper tract urothelial carcinoma; RCC = Renal cell carcinoma; NTHN = Non-tumoral hydrone Rank mean; NAC = Number of changes (in CSTMs); LCA = Linear correlation analysis.

Immunohistochemical stain by Ab of 5 CSTMs in tissues

It appeared in UTUC tissues an intense labelling by Ab of CA199 and CEA, a mild labelling by Ab of AFP and CA724, and feeble by Ab of CA125, while the labelling in paracancerous tissues was all invisible (Fig. 1–10).

Discussion

Usually, the UTUC diagnosis is easy, but it occurred sometimes that the UTUC could be misdiagnosed as renal carcinoma, or NTHN. In this study, there were 3 cases (5%) which were diagnosed as renal carcinoma. They accepted the radical nephrectomy instead of classic surgical therapy for UTUC, radical nephrectourectomy. For them, the prognosis was obviously different. And for the UTUC patients, the progression like recurrence and metastasis are not easily detected or definite diagnosed, even through an imaging examination. So it is needed some simple indicators to differentiate UTUC with other upper urinary tract diseases and to reveal the development of UTUC and curative effect.

Although some indicators like CxBladder monitor, UroVysion, NMP-22 and bladder tumor antigen were used in diagnosis and monitoring of TCC, few biomarkers achieve high sensitivity and specificity[1]. And only a few established prognostic factors were found to effectively assess the tumor progression of UTUC[9]. The expensive and time-consuming nature also restricted new methods' developments. Upon these, there is no clinical recognized bio-indicator for diagnosis, differential diagnosis and status assessment of UTUC.

The classic serum tumor makers (CSTMs, including CA242, CA199, CA125, CEA, AFP, SCC and CA724) are the most commonly used indicators for predicting and monitoring the residual and recurrent of tumors. SCC were used for squamous cell carcinoma predicting[12, 13], all other CSTMs were related with digestive system cancer or reproductive cancer[10, 14–18]. However, some unconventional roles were played in CSTMs: CEA and SCC were used for lung cancer diagnosis and prognosis[19, 20]; CA125 and CA199 was reported as significant markers of endometrium pathology[21]; SCC had a fair diagnostic value for hepatocellular carcinoma[22]. These meant that CSTMs might have more unique values for diagnosis and prognosis of cancerous diseases. However, among these 7 CSTMs, only CEA had been reported as early sign of malignancy in urotheliomas of the upper urinary tract and closely related to the recurrence and survival[23, 24].

In our study, we found that the value or AR in the most of the single CSTMs and the NAC in CSTMs group were significantly augmented in UTUC than in RCCC, NTHN and RCCC + NTHN, which may help to differentiate the UTUC with other upper urinary tract common diseases, malignant or benign. This also suggested the potential compliant relationship between CSTMs and UTUC. The postop CSTMs parameters were significantly lower than the preop ones and have re-augmented in postop PD patients. This revealed that the changes of group of CSTMs complied with the changes of UTUC disease. For the differential diagnosis, CEA and CA724 presented the best ability, but for the disease assessment, CA242 had a best behavior. So it revealed that the single CSTMs could not sacrifice the both functions.

According to the results in all these comparisons, here we have got 6 CSTMs: CA242, CA199, CEA, AFP, SCC and CA724, which were related with UTUC. A test strategy based on these 6 CSTMs was recommended in the clinical detection. The relationship between this strategy and differential diagnosis and assessment of UTUC was re-check in the comparisons above, the P values were almost the lowest and the positive results covered all the comparisons. So the recommended strategy was considered practical, and the AR seemed to be more important than the value.

In pathological level, tumor load, grade, invasive depth, seemed like no significant relationship with most of the single CSTMs, the same to grade and invasive depth with CSTMs group. The NAC on CSTMs by the recommended strategy was linear correlated with tumor load. All these meant that the CSTMs were related with tumor cell quantity, the CSTMs were produced by tumor cell since perhaps the beginning, and the NAC on CSTMs might be a good indicator to assess the changes of disease.

The immunohistochemical labelling results confirmed the fact that the UTUC tissues, but not the paracancerous tissues, significantly contained the CSTMs, at least CA199, CA125, CEA, AFP and CA724. In which, CA199 and CEA were strongly expressed, while CA125 was expressed with very small amount (but also much more strongly than paracancerous tissues). These results were highly in accord with the previous biochemical results and the recommended strategy based on the biochemical results. According to these all above, the CSTMs were specially secreted by the UTUC cell, and the recommended test strategy was practical. This explained the compliance of the CSTMs changes with UTUC disease changes.

Conclusions

The CSTMs may help to make the differential diagnosis and the disease assessment of UTUC. Group test was shown to be more valuable. The recommended test strategy was CA242 + CA199 + CEA + AFP + SCC + CA724.

Abbreviations

CSTM
Classic serum tumor marker;
UTUC
Upper tract urothelial carcinoma;
RCCC
Renal clear cell carcinoma;
NTHN
non-tumoral hydronephrosis;
tUTUC

Total upper tract urothelial carcinoma (non-metastatic and metastatic UTUC);
nmUTUC
Non-metastatic upper tract urothelial carcinoma;
AR
Abnormal rate;
NAC
Number of changes (in CSTMs);
PD
progression of disease;
Ab
Antibody;
RM
Rank mean;
LCA
Linear correlation analysis.

Declarations

Ethics approval and consent to participate: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. All procedures performed in this study involving human participants were in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by Ruijin Hospital Ethics Committee and the informed consent was allowed to be exempted.

Consent for publication: Not applicable

Availability of data and material: The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests: The authors declare that they have no competing interests.

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Authors' contributions:

JWP and YS have drafted the protocol; JWP, FXL, XWJ, WCT, XJW, XZ, GLL, BXH and YZ have made substantive contributions to the project development; JWP and XWJ have put into effect the data collection or management, data analysis and manuscript writing/editing. All authors have read and approved the manuscript.

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Figures

Figure 1

Intense labelling of CA199-Ab in UTUC tissue (×200)

Figure 2
Invisible labelling of CA199-Ab in UTUC tissue (×100)

Figure 3

Feeble labelling of CA125-Ab in UTUC tissue (very small amount) (100)

Figure 4

Invisible labelling of CA125-Ab in UTUC tissue (100)

Figure 5
Intense labelling of CEA-Ab in UTUC tissue (200)

Figure 6
Invisible labelling of CEA-Ab in UTUC tissue (100)

Figure 7
Mild labelling of AFP-Ab in UTUC tissue (100)

Figure 8
Invisible labelling of AFP-Ab in UTUC tissue (100)

Figure 9

Intense labelling of CA724-Ab in UTUC tissue (100)

Figure 10
Invisible labelling of CA724-Ab in UTUC tissue (200)