Clinical and pathological characteristics of hypertensive and normotensive adrenal pheochromocytomas

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CONTEXT AND OBJECTIVES

- Pheochromocytoma/Paraganglioma (PPGL) present with an extremely variable clinical picture which ranges from dramatic, to mild, to silent, depending on tumor attitude to release catecholamines. Hypertension is the hallmark of these tumors but is not always present [1].
- Distinct differences of clinical manifestations exist in hypertensive pheochromocytomas (HP) and normotensive pheochromocytomas (NP), however the comparative analysis is lacking.
- With the progress of imaging technology, an increasing number of asymptomatic pheochromocytomas were gradually acknowledged, and more than 25 percent of pheochromocytomas were accidentally discovered [2].
- The objective was to assess the clinical symptoms, hemodynamics, metabolism, radiological and histological features of patients with HP and NP.

METHODS

- This retrospective study included 104 patients who underwent a unilateral adrenalectomy with a diagnosis of pheochromocytoma by preoperative examinations of urinary catecholamines and postoperative investigations of pathological morphology and immunohistochemical staining at the Drum Tower Hospital Affiliated to Nanjing University Medical School from January 2004 to December 2014.
- 104 pheochromocytoma patients were categorized into HP (n=69) and NP (n=35) groups. Biochemical examinations of 95 subjects with primary hypertension(PH) were recorded for comparative study. All clinical records were reviewed. Tumor samples were examined to determine the Adrenal Gland Scale Score and were available for measurement of gene transcriptions.
- Patients with bilateral pheochromocytomas, familial history of pheochromocytomas or disease associated with chromaffin tumor were excluded. Known Pheochromocytoms –predisposing gene mutations and those who received chemotherapy during the observation period were excluded.
- All patients provided written informed consent. The study was approved by the Institutional Review Board of Drum Tower Hospital Affiliated to Nanjing University Medical School.

RESULTS 1.1

Table 1. The overall clinical characteristics of HP, NP and PH groups. [($\bar{x}$± s), n (%)]

	n	HP	n	NP	n	PH	P		
							HP vs. NP	HP vs. PH	NP vs. PH
Number	69	-	35	-	95	-	-	-	-
Male/Female	40/29	-	15/20	-	48/47	-	0.153	0.368	0.424
Age (years)	69	48.3±14.6	35	42.2±10.0	95	44.4±17.0	0.058	0.125	0.381
Height (cm)	65	168.0±7.2	32	159.5±0.7	95	160.1±1.8	0.141	0.134	1.200
Weight (kg)	65	61.4±13.7	32	56.9±7.8	95	65.6±5.4	0.052	0.032	0.060
BMI (kg/m^2)	65	22.5±3.8	32	19.9±0.5	95	24.8±2.5	0.061	0.035	0.065
SBP (mmHg)	69	146.4±25.7	35	122.2±14.8	93	160.4±28.9	0.000	0.002	0.000
DBP (mmHg)	69	92.3±17.2	35	76.4±9.7	93	99.8±19.1	0.000	0.013	0.000
IGR or T2DM (%)	69	23 (36%)	35	5 (15%)	93	9 (10%)	0.036	0.000	0.570
Hemoglobin (g/L)	69	136.6±16.1	35	127.4±22.1	93	120.2±15.0	0.021	0.015	0.890
FPG (mmol/L)	69	6.4±2.0	35	5.4±0.9	93	5.3±0.7	0.002	0.000	0.761
Uric acid (umol/L)	69	283.4±87.5	34	238.9±94.7	93	352.7±78.7	0.023	0.028	0.002
Creatinine (umol/L)	69	65.0±14.4	34	57.4±13.1	93	67.6±18.8	0.014	0.385	0.002
ALT (IU/L)	69	39.6±58.1	34	28.2±19.8	93	32.9±19.3	0.099	0.722	0.511
r-GT (IU/L)	69	49.2±71.6	34	26.1±21.1	93	24.4±10.6	0.020	0.515	0.385
Ur NE (<20ug/24h)	65	195.3±409.5	27	59.1±110.0	77	21.9±66.4	0.020	0.000	0.001
Ur E (<90ug/24h)	65	651.8±947.1	27	263.1±493.0	77	42.5±139.4	0.010	0.000	0.000
Ur DA (<600ug/24h)	65	479.0±825.4	27	312.9±300.9	77	169.9±86.5	0.340	0.000	0.068

RESULTS 1.2

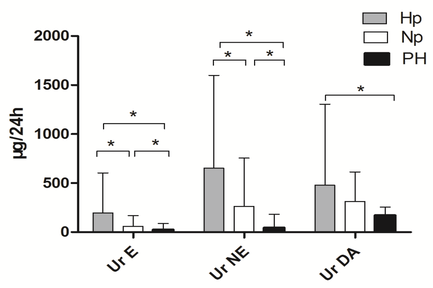

Fig 1. The levels of urinary catecholamines metabolites in HP, NP and PH groups.

Abbreviations: HP, hypertensive pheochromocytomas; NP, normotensive pheochromocytomas; PH, primary hypertension. Ur E, urinary epinephrine; Ur NE, urinary norepinephrine; Ur DA, urinary dopamine. *, P<0.05.

RESULTS 2

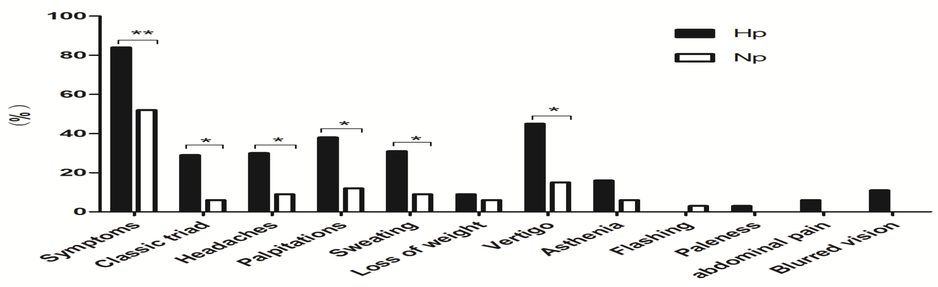

Fig 2. HP and NP of clinical symptoms comparison. **, P<0.01; *, P<0.05

RESULTS 3

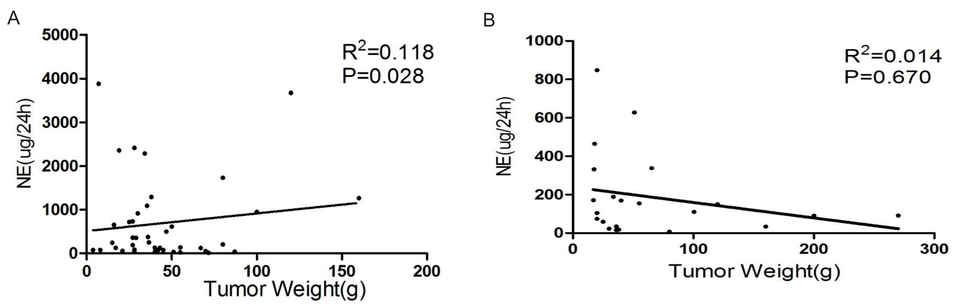

Fig 3. The linear regression of tumor weight and urinary norepinephrine in HP (A) and NP (B).
(A) Linear regression equation: Y=1.376+0.653×TW; (B) Linear regression equation: Y=1.551+0.073×TW
Abbreviations: TW: Tumor weight; NE: norepinephrine.

RESULTS 4.1

Table 2. CT characteristics of the HP and NP groups. [($\bar{x}$± s), n (%)]

	n	HP	n	NP	P
Location of tumor (L/R)	34	12/22	16	6/10	0.576
Complete capsule (%)	34	97%	16	93%	0.851
Necrosis (%)	34	14%	16	17%	0.490
Cystic change (%)	34	71%	16	78%	0.521
Tumor diameter (cm)	34	5.3±2.2	16	5.0±1.9	0.734
Unenhanced CT attenuation values (HU)	34	36.8±6.9	16	42.3±16.3	0.294
Aterial phase dynamic enhanced CT value (HU)	34	79.2±27.7	16	97.5±30.9	0.321
Venous phase dynamic enhanced CT value (HU)	34	58.6±22.2	16	59.9±13.8	0.892

RESULTS 4.2

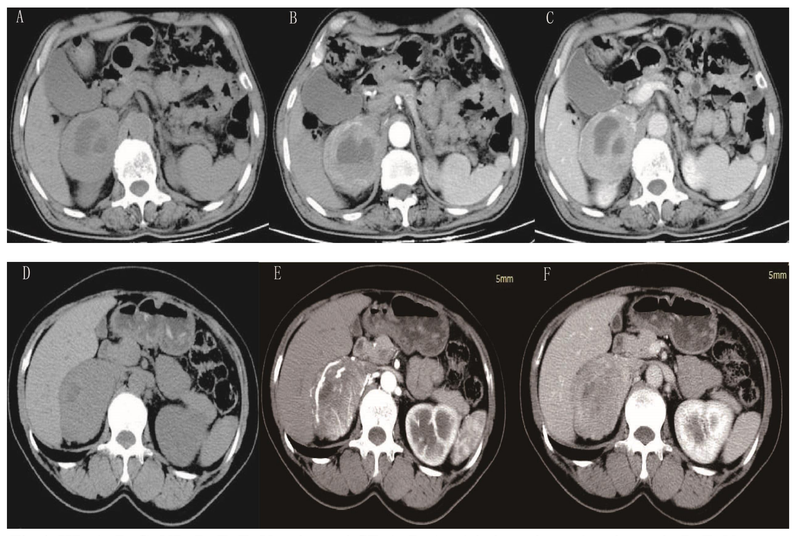

Fig 4. HP: A, B, C; NP: D, E, F; Unenhanced CT: A, D; Aterial phase dynamic enhanced: B, E; Venous phase dynamic enhanced: C, F.

RESULTS 5.1

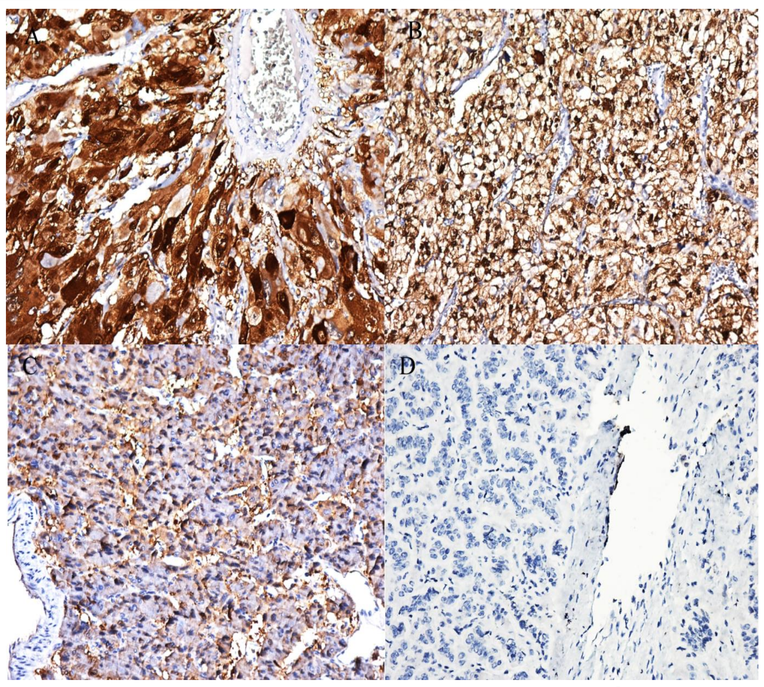

Fig 5. Immunohistochemical staining of PNMT in adrenal phechromocytoma [3] .
A (×200): PNMT (+++). B (×200): PNMT (++). C (×200): PNMT (+). D(×200) : PNMT (-).

RESULTS 5.2

Table 3. Two diameter comparisons according to PNMT immunohistochemical staining. ($\bar{x}$± s)

	PNMT immunohistochemical staining				P
	n	Negative (-)	n	Positive (+)	
Tumor diameter in HP	21	5.7±2.4	42	5.9±2.2	0.781
Tumor diameter in NP	9	7.1±2.7	22	4.8±1.8	0.011

NOTE: NP, Y=0.940-0.356X, R2=0.494, F=11.736, P=0.005.

RESULTS 6.1

Table 4. The pathological characteristics of the HP and NP groups. [($\bar{x}$± s), n (%)]

	n	HP	n	NP	P
Envelope (Complete)	46	38 (83%)	21	17 (81%)	1.000
Nature (Soft/Moderate/Tough)	51	35/14/2 (69%/28%/3%)	19	10/9/0 (52%/47%/0%)	0.159
Texture (Solid/Cystic/Both)	54	44/3/7 (82%/6%/13%)	27	19/6/2 (70%/22%/4%)	0.432
Tumor Weight (g)	44	45.9±30.4	21	44.3±34.2	0.860
Tumor diameter (cm)	60	5.9±2.3	29	5.6±2.3	0.509

RESULTS 6.2

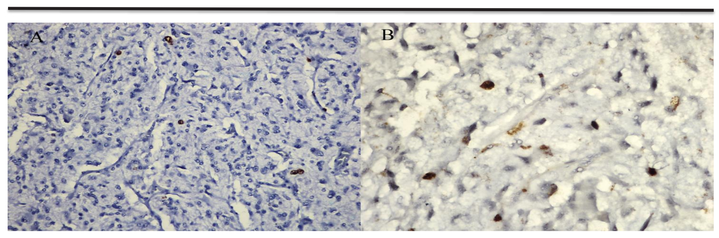

Fig 6. Immunohistochemical staining of Ki 67 in adrenal phechromocytoma.
A (×200): Ki 67<1% (+). B (×200): Ki 67>3% (+).

RESULTS 6.3

Table 5. Grinding scores of the HP and NP groups. [($\bar{x}$± s), n (%)]

	n	HP	n	NP	P
Zellballen (Y) (%)	59	44 (75%)	30	20 (67%)	0.635
Large and irregular sized nest (Y) (%)	59	22 (37%)	30	11 (37%)	1.000
Pseudorosette forming (Y) (%)	59	8 (14%)	30	2 (7%)	0.485
Cellularity (High)	61	10 (16%)	32	7 (22%)	0.577
Cellularity (Moderate)	61	31 (51%)	32	22 (69%)	0.124
Cellularity (Low)	61	20 (33%)	32	3 (9%)	0.013
Coagulation necrosis (Y) (%)	61	11 (18%)	31	4 (13%)	0.566
Vascular invasion (Y) (%)	61	2 (3%)	31	0	0.544
capsule invasion (Y) (%)	61	5 (8%)	31	4 (13%)	0.489
Ki 67 (<1%/1-3%/ >3%)	64	17/41/6 (27%/64%/9%)	32	9/18/5 (28%/56%/16%)	0.737
Types of catecholamine (E-type/NE-type or No function)	65	34/31(52%/48%)	27	6/18(22%/78%)	0.002
Scores	64	3.2±1.6	31	3.0±1.8	0.669

Abbreviations: E-type, Epinephrine-type; NE-type, Norepinephrine-type.
NOTE: Cellularity (low): <150 cells/mm^2, (Moderate): 150-250 cells/mm^2, (High): more than 250 cells/mm^2. Ki 67 immunoreactivity (avg. count per 200 ×field, based on 20 fields with highest labeling). Metastases were reported in 13% of tumors with scores 1-2, 63% with 3-6, and 100% with 7-10. Excerpted from Kimura et al[4].

RESULTS 7

Table 6. Quantitative Real-time PCR assays[5]

Gene symbol	n	HP C_T	n	NP C_T	P
PNMT	5	28.9± 1.6	3	25.0± 2.2	0.038
SGII	5	25.9± 1.1	3	21.5± 1.7	0.040
VAMT1	5	33.5± 1.4	3	30.8± 3.7	0.053
NPY	5	24.6± 2.8	3	23.1± 2.7	0.032

NOTE: PNMT: Phenylethanolamine-N-methyltransferase; SGII: Secretogranin II; VMAT1: Vesicular monoamine transporter 1; NPY: Neuropeptide Y.

RESULTS

1. Patients with NP showed lower proportion of clinical triad, inapparent metabolic disorders and lower urinary catecholamine levels than HP, but higher than PH.
2. Tumor weight positively correlated with 24h urinary norepinephrine level in patients with HP (P=0.028), and tumor diameter negatively correlated with Phenylethanolamine-N-methyltransferase (PNMT) immunohistochemistry (P=0.011) in NP but not in HP.
3. The Adrenal Gland Scale Score of NP group was similar to that of HP group.
4. The positive percentage of epinephrine type (E-type) of catecholamine in HP group was higher than that in the NP.
5. The transcript gene levels of PNMT, Secretogranin II (SGII) and Neuropeptide Y (NPY) from tissue samples were significantly lower in NP than in HP, while Vesicular monoamine transporter 1 (VMAT1) had no difference between HP and NP.

CONCLUSIONS

This study analyzed differences in HP and NP from clinical manifestations to molecular level, and suggested that HP and NP have distinct differences in clinical, biochemical, pathological and molecular phenotypes, which are closely related with the catecholamine pathway productions involved in tumor occurrence and development. Additionally, the change of quantity and phenotype of catecholamines has been clarified as the cause of biochemical and pathological phenotype change.

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