Original Article

COX4I2 is a novel biomarker of blood supply in adrenal tumors

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Background: Previous study has been reported that COX4I2 expression level demonstrated a positive correlation with microvessel density in pheochromocytomas (PCC) samples, suggesting that the expression of COX4I2 maybe related to blood supply level in other adrenal tumors as well. The aim of this study is to clarify the correlation of COX4I2 expression and blood supply in adrenal tumors.

Methods: A total of 84 patients were recruited, among which 46 was diagnosed as adrenocortical adenoma (ACA) and 38 was diagnosed as PCC. Contrast-enhanced CT values were used to evaluate the blood supply levels in those patients. The expression of mRNA was examined by quantitative real-time polymerase chain reaction (qPCR) and protein was detected by immunohistochemistry (IHC).

Results: The COX4I2 expression level in PCC group is significantly higher than that in ACA group ($P<0.01$). The expression of angiogenesis-related genes EPAS1, VEGFA and KDR mRNA in PCC group is higher than that of ACA group ($P<0.05$). Correlation analysis shows COX4I2 expression level is correlated with CT values ($P<0.001$), intraoperative blood loss ($P<0.05$) and operation time ($P<0.05$), and the expression of COX4I2 mRNA is correlated with EPAS1, VEGFA and KDR mRNA ($P<0.01$).

Conclusions: The results displayed a distinct expression level of COX4I2 between ACA and PCC, suggesting that COX4I2 is a novel biomarker of blood supply in adrenal tumors. This research also opens the possibility for further research on COX4I2 as a novel target for anti-tumor angiogenesis.

Keywords: COX4I2; adrenal tumors; blood supply; angiogenesis; biomarker

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Introduction

Adrenocortical adenoma (ACA) and pheochromocytoma (PCC) account for the majority of adrenal tumors (1,2). As the two most common tumor types in adrenal surgery, there are numerous studies on them, but few studies have explored tumor vascular abundance and related biomarkers. Our research team once selected a group of PCC samples and divided them into rich blood supply group and poor blood supply group according to different classification methods. Using high-throughput proteomics analysis, we found that the differentially expressed protein COX4I2 was significantly up-regulated in the rich blood supply group and down-regulated in the poor blood supply group.

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Subsequently, another batch of PCC samples were used for quantitative real-time polymerase chain reaction (qPCR) and immunohistochemistry (IHC) to confirm the proteomic results (3).

Cytochrome c oxidase subunit 4 isoform 2 (COX4I2) is the terminal enzyme in the mitochondrial electron transport chain (ETC) that drives oxidative phosphorylation and catalyzes the transfer of electrons from reduced cytochrome c to oxygen. It is the second isoform of subunit IV encoded by the nuclear gene of cytochrome C oxidase (4-7). Studies believe that the COX4 subunit can optimize the respiratory chain function by controlling the expression of its isomers COX4I1 and COX4I2 (8). The differential expression of COX4I2 in PCC of different blood supply stimulated our interest. We boldly hypothesized that COX4I2 may also show different expression levels between adrenal tumors with abundant blood supply and insufficient blood supply. In clinical practice, we found that ACAs often appear as smaller tumors with a lack of blood supply, while PCCs can usually be observed with more abundant blood vessels than other adrenal tumors.

Medical imaging technology is one of the commonly used methods to determine whether the blood supply of tumors is abundant in clinical practice. The use of computed tomography (CT) provides great convenience for the diagnosis of tumors, especially the CT scan with contrast enhancement can be used to reflect the richness of blood vessels. Previous studies have shown that there is a positive correlation between enhanced CT values and microvessel density (3,9,10). Before the examination, the examiner needs to inject an iodine contrast agent into the blood vessel of the examinee. The principle that enhanced CT scans can reflect the abundance of blood vessels is: after intravenous injection of iodine contrast agent, these contrast agent molecules are transported to the tissue through the bloodstream, diffuse and exchange between the intravascular and extravascular matrix, and CT images of different periods can be obtained over time (11). This quick and relatively economical examination provides clinicians with sufficient information in tumor evaluation, especially the blood supply status of the tumor, the adjacency and location of important organs and blood vessels, which facilitates preoperative preparation.

Like adrenal tumors, most solid tumors can see increased angiogenesis (12,13). On the one hand, the new vessels of tumor bring great challenge to surgery. The new blood vessels increase the possibility of bleeding during operation, lead to excessive blood loss of patients, and at the same time affect the exposure of the surgical field, prolong the operation time, and bring inconvenience to the operation. On the other hand, non-benign tumors have dense angiogenesis and grow rapidly. The neovascular structure of tumor tissue is abnormal and its function is not perfect, which provides great convenience for tumor cells to invade the inside of blood vessels and cause distant metastasis with blood flow, which can easily lead to poor prognosis. Therefore, it will be beneficial to patients if we can find biomarkers of adrenal tumor-specific angiogenesis process, whether it is used for anti-adrenal tumor angiogenesis treatment before or after surgery. In this study, we chose ACA as the representative of adrenal tumors in the poor blood supply group and PCC as the representative of adrenal tumors in the rich blood supply group. The CT value of the arterial phase of the tumor was used as a reference to judge the blood supply of the tumor. The main purpose of this study is to explore whether COX4I2 can be used as a biomarker of the blood supply level of adrenal tumors by comparing the expression level of COX4I2 in ACA and PCC and its correlation with the CT value of arterial phase.

We present the following article in accordance with the MDAR reporting checklist (available at https://dx.doi.org/10.21037/tau-21-229).

Methods

Sample collection

The study conformed to the provisions of the Declaration of Helsinki (as revised in 2013). The institutional Committee abandoned the ethical review because it did not affect the treatment strategy. Informed consent was obtained from each patient prior to surgery. The inclusion criteria of this study were based on the pathological diagnosis, that is, patients whose pathological result was certainly diagnosed to be ACA or PCC. Samples with ambiguous or with other pathological diagnostic results were excluded. From year 2016 to 2019, 46 ACA specimens and 38 PCC specimens met the inclusion criteria were collected from the Urology Department of Ruijin Hospital. The specimens were divided into two parts and stored in liquid nitrogen (–196 ℃) and 4% paraformaldehyde. Detailed patients' information, including name, inpatient number, gender, age, clinical diagnosis, intraoperative blood loss, operation time and other specific conditions, was recorded by the Electronic medical record system. The pathological diagnosis

Table 1 Primers used for qPCR

Gene		Primer sequences (5'-3')
GAPDH	Forward	GCATTGCCCTCAACGACCAC
	Reverse	CCACCACCCTGTTGCTGTAG
COX4I2	Forward	ACTACCCCATGCCAGAAGAG
	Reverse	TCATTGGAGCGACGGTTCATC
EPAS1	Forward	TTGCTCTGAAAACGAGTCCGA
	Reverse	GGTCACCACGGCAATGAAAC
VEGFA	Forward	AGGGCAGAATCATCACGAAGT
	Reverse	AGGGTCTCGATTGGATGGCA
KDR	Forward	GTGATCGGAAATGACACTGGAG
	Reverse	CATGTTGGTCACTAACAGAAGCA

qPCR, quantitative real-time polymerase chain reaction.

information of the patients was inquired by the pathological report system about two weeks after surgery.

Computed tomography (CT)

After intravenous injection of 80–100 mL non-ionic contrast agent at a rate of 2.0–3.0 mL/s, the patient received a contrast-enhanced CT scanning in the supine position of the tumor area. The maximum diameter of the adrenal tumor was recorded and the average CT values of unenhanced and arterial phase in region of interest (ROI) could be measured from the CT image. The ROI is defined by selecting a measurement plane located in the center of the tumor. When measuring, try to include most of the lesions but avoid the edges of the lesions to evade partial volume effects, with obvious calcification, necrosis and artifacts areas are also avoided (14).

Quantitative real-time polymerase chain reaction (qPCR)

Total RNA was extracted from ACA and PCC tissues by Eastep® Super Total RNA Extraction Kit (Promega, China). Reverse-transcription total RNA was converted into first-strand complementary DNA with a reverse-transcription kit (Promega, China). Quantitative real-time PCR was then performed with a QuantStudio™ Dx Real-Time PCR Instrument (Thermo Fisher Scientific, USA). Primer name and sequences were listed in *Table 1*. Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used as a reference control. The experiment was repeated three times and the average Ct by the values obtained for each reaction was calculated. Fold-change of target genes against the reference gene was calculated from $2^{-\triangle\triangle Ct}$ values. And was expressed as mean ± standard deviation.

IHC

The specimens fixed with 4% paraformaldehyde were embedded in paraffin and sectioned with 5 μm thickness. The paraffin-embedded specimens were deparaffinized and hydrated with xylene and gradient ethanol. IHC was performed with UltraSensitive™ SP (Mouse/Rabbit) IHC Kit (Maixin Biotechnology Development Co., Ltd., China) according to user's manual. The sections were incubated with COX4I2 Rabbit Polyclonal antibody (Diluted 1:100, Cat. No. 11463-1-AP, Proteintech Group Inc., China) at 4 ℃ overnight, then incubated with biotin-labeled secondary antibody at room temperature for 10 minutes. Finally, the sections were stained by diaminobenzidine, counterstained with hematoxylin, fixed with neutral gum, and observed under an optical microscope (Olympus, Japan).

For each slice, we randomly choose five fields of vision to take photos, and the final result is the average of the five images. Two researchers analyzed the results independently. The sections were scored according to the staining intensity and the number of positive cells (15). The staining intensity was graded as 0 (no color), 1 (light yellow), 2 (light brown), or 3 (brown), and the number of positive cells was graded as 0 (<5%), 1 (5–25%), 2 (25–50%), 3 (51–75%), or 4 (>75%). Add the two grades together to get the final intensity grade below: 0–1 (negative, intensity grade 0), 2 (low-positive, intensity grade 1), 3–4 (positive, intensity grade 2), more than 5 (high-positive, intensity grade 3).

Statistical analysis

Statistical analysis was conducted using SPSS software version 22 (IBM Corporation, USA) and GraphPad Prism software version 8 (GraphPad Software Inc., USA). The Student's *t*-test was used to compare the differences between the means of two groups of data. Correlation analysis was performed to demonstrate the relationship between the COX4I2 expression level and CT value of arterial phase or among COX4I2, EPAS1, VEGFA and KDR mRNA expression. The correlation analysis of COX4I2 expression level with intraoperative blood loss and operation time was also implemented. All statistical analyses were two-sided,

Table 2 Clinical parameters of patients

Variables	ACA (n=46)	PCC (n=38)	P value
Gender (n, %)			
Male	18 (21.43)	11 (13.10)	–
Female	28 (33.33)	27 (32.14)	–
Age (year)	49.80±13.46	39.66±14.75	<0.01
Tumor diameter (cm)	2.77±1.49	5.23±2.00	<0.0001
≤3 cm	2.12±0.59	2.62±0.41	0.08
>3 cm	4.81±1.60	5.65±1.82	0.2
CT value of arterial phase (HU)	50.10±30.38	89.10±26.55	<0.0001
≤3 cm	44.96±23.07	120.30±33.81	<0.0001
>3 cm	63.19±42.54	83.72±21.53	<0.05

The measurement data is displayed as mean ± standard deviation. ACA, adrenocortical adenoma; PCC, pheochromocytoma; CT, computed tomography.

and a value of P<0.05 (*), P<0.01 (**), P<0.001 (***) and P<0.0001 (****) were considered statistically significant.

Results

Patients' clinical characteristics

A total of 84 specimens were collected, including 46 ACA tissues and 38 PCC tissues. The general clinical parameters of these 84 patients are shown in *Table 2*. The table shows the patient's gender, age, tumor diameter and the average CT value of tumor ROI in arterial phase. The diameters of all tumors ranged from 0.5–10 cm. In the ACA group, the average tumor diameter was 2.77±1.49 cm, and the average CT value was 50.10±30.38 HU. The average diameter of the tumor in the PCC group was 5.23±2.00 cm, and the average CT value was 89.10±26.55 HU. The average diameter and CT value of the two groups were significantly different (P<0.0001, P<0.0001). In order to eliminate the bias that tumor diameter may be associated with CT value, we divided all tumors into two subgroups with 3 cm as the threshold. After grouping, the tumor diameter of ACA and PCC had no statistical difference in the two subgroups, but the CT value of tumor still showed significant difference, which showed that the CT value of PCC group was higher than that of ACA group.

Observation of gross specimen

After the tumor is removed, take pictures to record the tumor morphology. By observation, the ACA tumor has poor blood supply, while the blood supply of PCC tumor is rich. ACA usually has a yellow cut surface, the tumor is small and lacks blood supply; while PCC usually has a brown-black cut surface with abundant blood supply. Large PCC tumors often have hemorrhage, necrosis, and cystic degeneration. *Figure 1* shows the general view of typical ACA and PCC tumors.

CT value in ROI

The average CT values in ROIs were determined from the CT scan images. In contrasted-enhanced CT images, most of ACA tumors showed no enhancement or mild enhancement, while PCC tumors showed obvious enhancement. *Figure 2A,B* was a representative CT image from an ACA patient. The patient is a 64-year-old male. The size of his tumor was 5.3 cm by 4.0 cm. The average CT value of the unenhanced and contrasted-enhanced arterial phase in ROI was –6.7 and 28.3 HU. *Figure 2C,D* was a representative CT image from a PCC patient. The patient was a 55-year-old female. The size of tumor was 4.4 cm by 4.0 cm. The average CT value of the unenhanced and contrasted-enhanced arterial phase in ROI was 31.5 and 122.7 HU. PCC showed significantly higher CT values than ACA in contrasted-enhanced arterial phase (P<0.0001) (*Figure 2E*).

COX4I2 expression level

The pictures showed in *Figure 3* were taken by a light

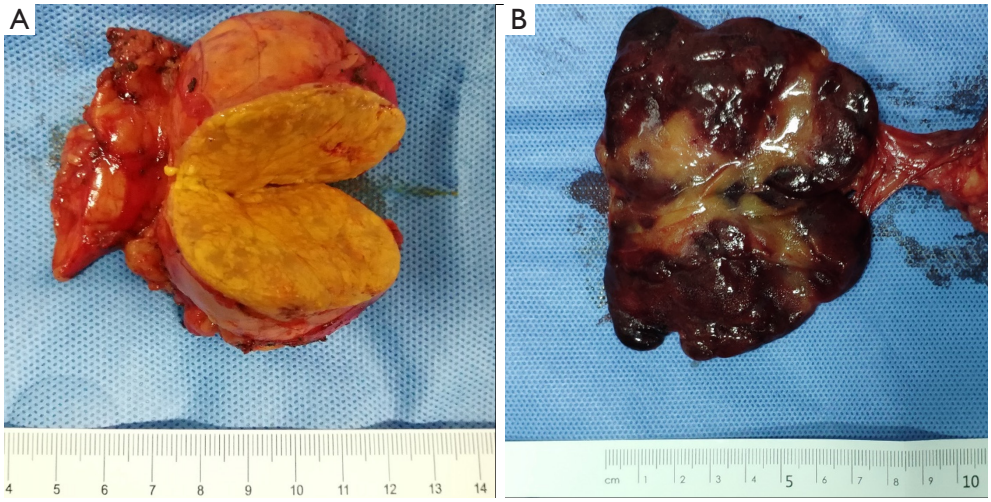

Figure 1 General observation of ACA and PCC tumors. (A) A typical ACA tumor. The tumor is a golden yellow mass with a small amount of adipose tissue on the surface, about 4 cm in diameter. The transverse section of the tumor was golden yellow with fine texture and no obvious blood vessels. (B) A typical PCC tumor. The tumor is a dark brown soft mass with adipose tissue on the surface, about 7.5 cm in diameter, surrounded by a complete capsule. The tumor was dark brown in cross section, with local fat infiltration and abundant blood supply. ACA, Adrenocortical adenoma; PCC, Pheochromocytoma.

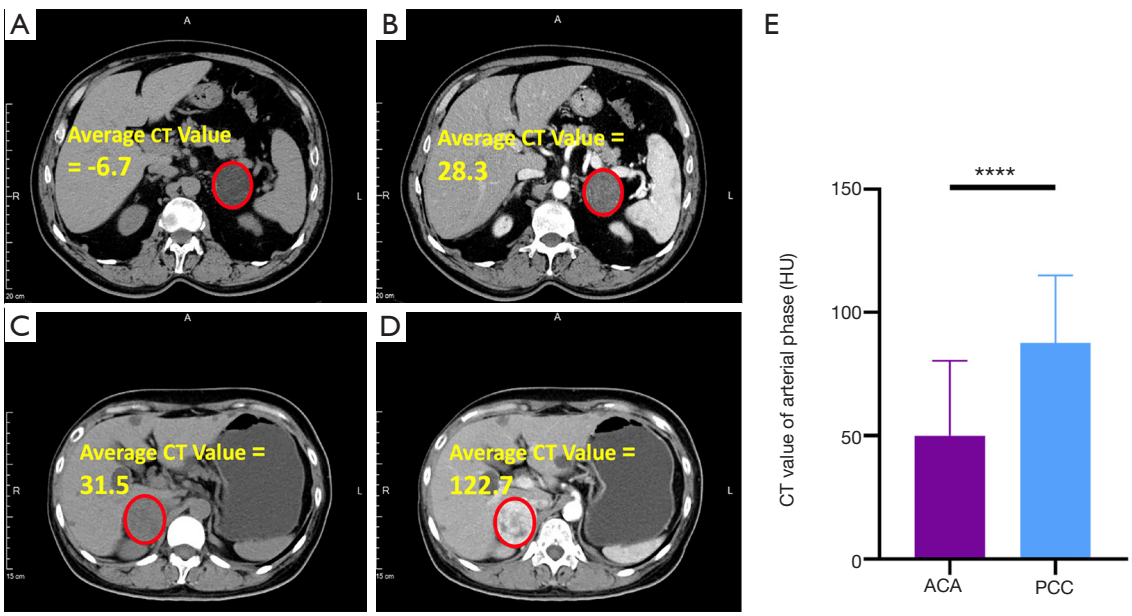

Figure 2 The CT images of ACA and PCC. (A,B) A representative CT image from an ACA patient. ROI was marked with a red circle. The average CT value of the unenhanced (A) and arterial phase (B) in ROI was −6.7 and 28.3 HU. (C,D). A representative CT image from a PCC patient. ROI was marked with a red circle. The average CT value of the unenhanced (C) and arterial phase (D) in ROI was 31.5 and 122.7 HU. (E) The average CT values in ACA and PCC samples. (****, P<0.0001; ACA, adrenocortical adenoma; PCC, pheochromocytoma; CT, computed tomography; ROI, region of interest.

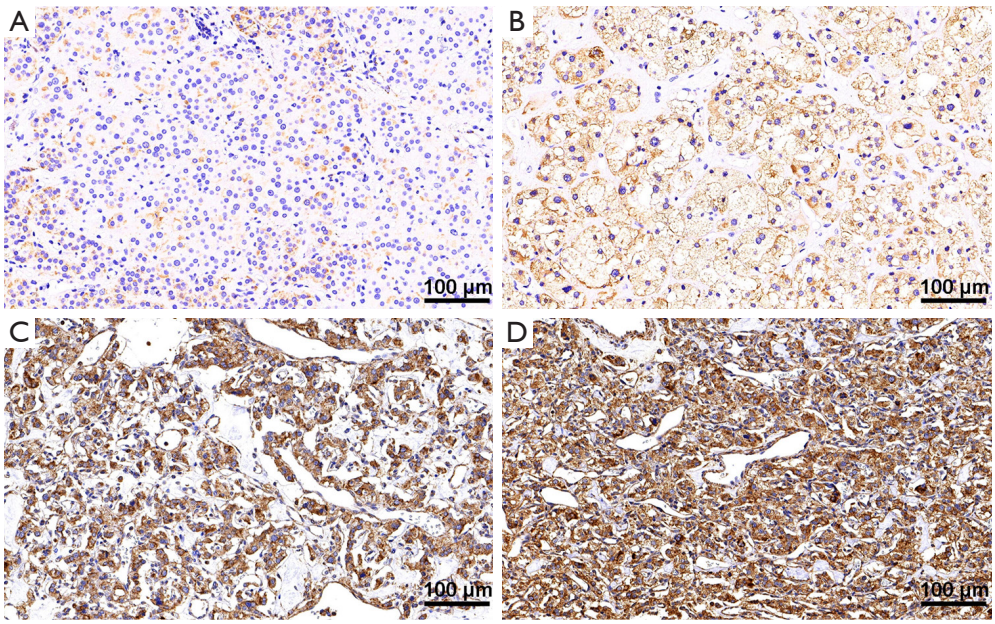

Figure 3 IHC staining of COX4I2 in ACA and PCC (scale bars, 100 μm). The sections were incubated with COX4I2 Rabbit Polyclonal antibody (Diluted 1:100) at 4 ℃ overnight. IHC intensity grade is used to measure the expression level of COX4I2 in tumor tissues of ACA or PCC. (A) The expression of COX4I2 protein is negative in ACA, IHC intensity grade: 0. (B) The COX4I2 protein is weakly expressed in ACA, IHC intensity grade: 1. (C) The positive expression of COX4I2 protein in PCC, IHC intensity grade: 2. (D) The expression of COX4I2 protein is strongly positive in PCC, IHC intensity grade: 3. ACA, adrenocortical adenoma; PCC, pheochromocytoma; IHC, immunohistochemistry.

microscopy with the magnification of 200-fold. The relative expression of COX4I2 mRNA in PCC group was 39.14 times that of ACA group (P<0.01) (*Figure 4A*). The average intensity grade of COX4I2 protein was 1.13±0.61 in ACA group and 2.13±0.55 in PCC group (P<0.0001) (*Figure 4B*).

Correlation analysis

To explore whether COX4I2 expression level can reflect the blood supply of adrenal tumors, we performed a correlation analysis between COX4I2 expression level and average CT value of arterial phase in ACA and PCC samples. The relative expression of COX4I2 mRNA is correlated with CT value of arterial phase (P<0.0001), and Pearson correlation coefficients is 0.611 (*Figure 4C*). The expression of COX4I2 protein is correlated with CT value of arterial phase as well (P<0.001), and Spearman's rank correlation coefficient is 0.554 (*Figure 4D*).

In order to explore the possibility why COX4I2 expression level is correlated with blood supply in adrenal tumors, qPCR analysis was performed on EPAS1, VEGFA and KDR mRNA level in ACA and PCC samples. As shown in *Figure 4E*, the relative expression of EPAS1, VEGFA and KDR mRNA were all elevated in PCC samples. The fold change was 1.70 (P<0.01), 1.85 (P<0.01), 1.50 (P<0.05), respectively.

We performed a correlation analysis among COX4I2, EPAS1, VEGFA and KDR mRNA expression level in ACA and PCC samples (*Figure 4F,G,H*). The relative expression of COX4I2 mRNA is correlated with EPAS1, VEGFA, and KDR mRNA (P<0.0001, P<0.0001, P<0.01, respectively), and Pearson correlation coefficients are 0.657, 0.549, 0.438, respectively.

The expression level of COX4I2 has a significant positive correlation with intraoperative blood loss and operation time regardless of mRNA level or protein level. As shown in *Figure 4I,J*, COX4I2 mRNA is positively correlated with intraoperative blood loss (P<0.05) and operation time (P<0.05), with Pearson correlation coefficients of 0.275 and 0.304, respectively. *Figure 4K,L* shows that IHC intensity grade is positively correlated with intraoperative blood loss (P<0.0001) and operation time (P<0.001), and the Spearman's rank correlation coefficient are 0.589 and 0.483, respectively.

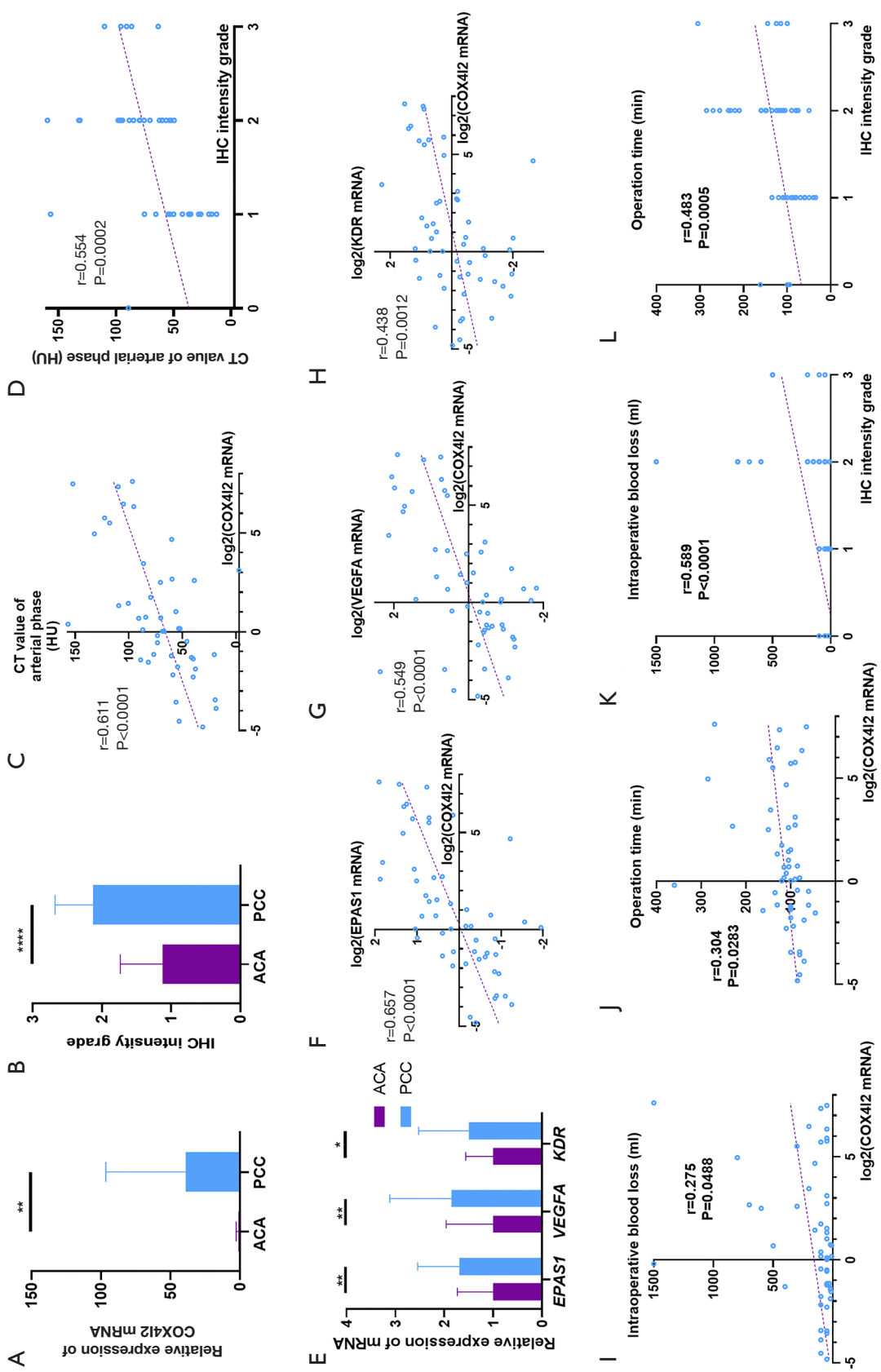

Figure 4 Comparison and correlation analysis of tumor related parameters between ACA and PCC. (A) The relative expression of COX4I2 mRNA in ACA and PCC samples. (B) The IHC intensity grade of COX4I2 protein expression in ACA and PCC samples. (C,D) Correlation analysis between COX4I2 mRNA (C) OR protein (D) expression level and average CT value. (E) The relative expression of EPAS1, VEGFA and KDR mRNA in ACA and PCC samples. (F,G,H) Correlation analysis between COX4I2 mRNA expression level and EPAS1 (F), VEGFA (G) and KDR (H) mRNA expression level. (I,J) Correlation analysis between COX4I2 mRNA expression level and intraoperative blood loss (I) and operation time (J). (K,L). Correlation analysis between COX4I2 IHC intensity grade and intraoperative blood loss (K) and operation time (L). * , P<0.05; ** , P<0.01; *** , P<0.001; **** , P<0.0001; r, correlation coefficient; CT, computed tomography; ACA, adrenocortical adenoma; PCC, pheochromocytoma; IHC, immunohistochemistry.

Discussion

Generally speaking, the results of enhanced CT scan can be used to reflect the vascular abundance, which is based on the number of blood vessels in the tumor (3,9,10). When the contrast agent is injected intravenously, it begins to pass through the blood vessels and then begins to develop (16). Traditional imaging studies often describe that tumors have a rich and complex vascular network, especially in large tumors that have a more abundant supply of blood vessels (9,10). It has been observed in imaging and surgical procedures that most PCCs are tumors with high vascular abundance. Due to the rich lipids in adenoma cells, ACA often appears as low-density nodules on CT. However, in PCC, the solid part of the tumor is enhanced on CT due to the abundant capillary network (17). The higher the average CT value, the more abundant the blood supply. In this study, we used CT scan results as a control to reflect the difference in vascular abundance between adrenocortical adenoma and pheochromocytoma. Our findings confirm that PCC has a richer blood supply than ACA.

The expression of COX4I2 demonstrated significant difference between rich blood supply group and non-rich blood supply PCC tumors according to the proteomics screening result (3). However, being the most common cause of adrenal cortical tumors, the expression of COX4I2 in ACA has not yet been investigated. As a kind of tumor with low blood supply, whether there is a difference in the expression level of COX4I2 between ACA and blood supply rich PCC is one of our questions after the previous study. Therefore, by examining the expression level of COX4I2 in ACA and comparing it with PCC, we can more clearly understand the relationship between COX4I2 and the blood supply of adrenal tumors. The analysis shows that the expression level of COX4I2 in all tumors is positively correlated with the CT enhancement value of arterial phase, which provides strong evidence that COX4I2 can be used as a biomarker of the blood supply of adrenal tumors.

One cluster of PCC related mutant genes is highly related to hypoxia pathway (18). These genes activate hypoxia inducible factor (HIF) and then transcribe several genes related to angiogenesis and cell growth. VEGF is one of the main cytokines that stimulate tumor angiogenesis. It is the most important target gene of HIF. It is generated by HIF binding with hypoxia response element and downstream transcription (19). In the existing studies, the mechanism of COX4I2 is less directly related to angiogenesis. It has been found that increasing the expression of COX4I2 can increase COX activity, promoting electron transport chain (ETC) activity, and affecting reactive oxygen species (ROS) production (20). ROS has been reported to be related to angiogenesis. The downstream molecule PAK regulated by ROS is an effector of Rac-mediated cytoskeletal remodeling, responsible for cell migration and angiogenesis (21). Increased ROS can stabilize HIF-1α, which helps to increase VEGF (22,23). Increased levels of ROS in the retina can trigger an inflammatory cascade. Impairment of ROS homeostasis can lead to the occurrence of hereditary retinal dystrophy, including retinitis pigmentosa, accompanied by choroidal neovascularization (CNV) (24). The apoptosis of COX inhibits the oxygen consumption of human retinal pigment epithelial (RPE) cells, resulting in a decrease in the vitality of RPE cells (25). Studies have shown that the condition of hypoxia can up-regulate the expressions of COX4I2 in liver, cervical, colorectal, and lung cancer cell lines (26). MNRR1 is a transcription activator of COX4I2, which is stimulated by hypoxia. It specifically binds to a 13 bp highly conserved element in the COX4I2 promoter, which we call oxygen-responsive element (ORE) (27,28). MNRR1 has been shown to promote the angiogenesis of liver cancer (29). When knocked out, it can inhibit the migration and angiogenesis of human renal cell carcinoma (30). In a study on endometriosis (8), it was found that COX4I2 is targeted by MLL1 and is essential for the process of endometrial decidualization. Enzymes containing COX4I2 reduce the oxygen affinity, leading to hypoxia in the endometrial microenvironment, which is essential for successful endometrium implantation and angiogenesis. During the *in vitro* decidualization process, the expression of HIF-2α increases, but if COX4I2 is knocked down, the expression of HIF-2α is blocked. This study suggests that the expression of HIF-2α in the matrix can be regulated by COX4. In our study, the correlation analysis showed that the expression of COX4I2 mRNA was significantly correlated with the mRNA levels of angiogenesis-related genes, such as EPAS1, VEGFA and KDR. These results suggest that COX4I2 may be involved in the angiogenesis of adrenal tumors. Based on the existing research results, we hypothesize that in adrenal tumors, COX4I2 may act as an upstream or downstream factor of HIF to promote the cascade reaction and contribute to the angiogenesis process in some way. Subsequent research deserves to be advanced to further explore the interesting mechanisms that may exist.

The abundant blood supply affects the surgical

treatment and prognosis of adrenal tumors. Due to its extremely important role in the growth and progression of tumors, anti-angiogenesis has gradually become one of the important treatment options to control tumor growth. The existing therapeutic drugs such as sunitinib, axitinib, pazopanib, sorafenib and bevacizumab are metabolized in the liver by cytochrome P450 3A4 enzyme (12). Different patients have different clinical responses after using drugs. Some patients' tumor growth can be controlled by taking drugs, the tumor size is reduced, and the survival time is prolonged. However, due to the inevitable cardiovascular toxicity and tumor drug resistance, the curative effect of other patients is limited.

Intraoperative hemorrhage has always been a serious challenge in adrenal tumor surgery. Especially when laparoscopic technique is used, bleeding will greatly prolong the operation time and increase the operation difficulty. Therefore, anti-angiogenic therapy as an adjuvant treatment of adrenal surgery has a very far-reaching significance. For adrenal tumors, it is of great significance to find and study the biomarkers of its specificity. The result of differential expression of COX4I2 in different blood supply adrenal tumors provides us a new angle for tumor blood supply abundance and a new idea for further study of tumor formation mechanism. The limitation of this study is that it has not been widely verified. In the future, larger sample sizes and prospective studies need to be invested to further test the specificity and sensitivity of COX4I2 as a blood supply biomarker for adrenal tumors. It is believed that with the deepening of the research, we can determine that COX4I2 alone or in combination with other molecules can be used as a biomarker for the diagnosis of adrenal tumor blood supply for treatment selection and prognosis determination. COX4I2 inhibitors may be considered for preoperative preparation to inhibit angiogenesis and growth of adrenal tumors, and ultimately benefit patients.

Conclusions

In conclusion, the results displayed a distinct expression level of COX4I2 between ACA and PCC, suggesting that COX4I2 is a novel biomarker of blood supply in adrenal tumors. This research also opens the possibility for further research on COX4I2 as a novel target for anti-tumor angiogenesis.

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Footnote

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Data Sharing Statement: Available at https://dx.doi.org/10.21037/tau-21-229

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://dx.doi.org/10.21037/tau-21-229). The authors have no conflicts of interest to declare.

Ethical Statement: 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. The study conformed to the provisions of the Declaration of Helsinki (as revised in 2013). The institutional Committee abandoned the ethical review because it did not affect the treatment strategy. Informed consent was obtained from each patient prior to surgery.

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