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Influence of secretory phenotype and preoperative preparation on surgical outcome in pheochromocytoma

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Abstract:

Objectives: Surgery of pheochromocytomas (PCs) still carries a high risk of hemodynamic complications during the perioperative period. We aimed to evaluate the influence of their secretory phenotype and preoperative alpha-blocker treatment on surgical outcome.

Design: a retrospective monocentric study at a tertiary medical centre.

Patients: 80 consecutive patients operated by the same team for a PC between 1988 and 2018.

Results: Diagnosis was based on typical symptoms and signs in 58 patients, genetic testing in 12 and work-up of an adrenal incidentaloma in 9. It was made during surgery in one patient. A genetic predisposition was found in one third of index cases (21/62). The majority of the patients (73/79) had a secreting PC; more than 2/3 had an adrenergic phenotype and less than 1/3 a noradrenergic phenotype. The rate of perioperative hemodynamic complications was not influenced by the secretory phenotype, but persistent hypertension after surgery, recurrence and malignancy were more frequently observed in patients with a noradrenergic tumour. Preoperative alpha-blocker treatment was given for ≥ 14 days in 29 patients and, although being more symptomatic at diagnosis, these patients had less hemodynamic complications (3/29 vs. 12/51 non-treated patients, p=0.05).

Conclusions: The occurrence of hemodynamic complications during surgery was not significantly affected by the secretory phenotype in our study, but noradrenergic tumours show a worse post-surgical outcome. Our data also provide additional support in favor of a sufficient preoperative alpha-blockade in patients with pheochromocytoma.

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Introduction

Clinical presentation of pheochromocytomas and paragangliomas (PPGL) is highly variable, making early diagnosis often challenging. The pattern and hemodynamic effects of tumoural catecholamine secretion may indeed vary from a biochemically and clinically silent chromaffin adrenal tumour to a highly expressive pheochromocytoma inducing severe hypertension with or without superimposed bursts (1, 2, 3). Severe cardiovascular complications may ensue, such as a Takotsubo-like acute myocardial syndrome or chronic hypertrophic or dilated cardiomyopathy (4).

For decades, the diagnosis of PPGL has been most commonly based on the presence of paroxysmal or sustained hypertension, associated with suggestive symptoms and signs such as palpitations, pallor, tremor, hyperhidrosis and nausea (5). Over the last years, however, pheochromocytomas have been increasingly diagnosed during the work-up of an incidentally discovered adrenal mass (6, 7, 8) or as a result of genetic case detection testing, as these tumours carry a very high rate of germline mutations, up to 35-40% in recent series (9, 10, 11).

Surgery represents the primary treatment of PPGL (2, 3). Thanks to improvements in medical treatment, anaesthesia and surgical techniques, the mortality has dropped markedly to an acceptable range of 0-3%, but the risk of cardio-vascular complications during the perioperative period remains high (12, 13). Preoperative medical treatment with alpha-blockers is currently recommended, at least in hormonally functional PPGL, as it might improve surgical outcome (2, 14). However, controversy persists regarding the true clinical benefits of such preparation, partly due to the lack of a well-designed placebo-controlled trial (14) and the optimal preparation has not been yet clearly defined (12, 15). For instance, some

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authors have proposed that only patients with severe and complicated hypertension require blood pressure normalization prior to surgery (16, 17).

Both perioperative manifestations and surgical outcome seem to be related to the severity and phenotype of catecholamine secretion. In particular, the amount of preoperative total urinary metanephrine excretion has been reported as an independent risk factor for morbi-mortality during surgical resection of the tumour (18). In addition, predominant norepinephrine secretion is characterized by a more sustained hypertension and an absence or reversal of nocturnal blood pressure dipping that may lead to a higher prevalence of target organ damage (19).

The aims of our study were to characterize a large series of patients treated in our tertiary center for an adrenal pheochromocytoma (PC) and to determine whether the secretory phenotype could influence surgical complications and long-term outcome. We also thought to determine whether an adequate preoperative medical preparation with alpha-blockers improved surgical outcome.

Materials and methods:

Patients

We retrospectively analyzed the data from 80 consecutive patients who were managed by the same clinical team and who had undergone adrenal surgery between 1988 and 2018 in our academic centre to remove an immunohistochemically confirmed pheochromocytoma, Twelve patients were operated before 2000 and 68 thereafter, while four other patients had to be excluded because of insufficient follow-up data. We collected patient's characteristics, symptoms and complications at diagnosis, hormonal results, non-invasive imaging techniques used to localize PC, tumour size, preoperative medical preparation, type of surgery, complications that occurred during and within 24h after surgical intervention, as well as

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postoperative outcome including evolution of hypertension, PC relapse, malignant transformation and death. The median duration of follow-up was 53 months [P5-P95 interval: 6-248 months].

The circumstances of diagnosis were available in 74 patients only, whereas preoperative urinary catecholamine excretion was available in 79 patients (24-hour urine fractionated metanephrines in 74 patients, 24-hour urinary epinephrine and norepinephrine in 5 patients diagnosed before 1996, and no data in one patient with a PC diagnosed at the time of surgery). The patients were divided into three subgroups according to the tumour secretory phenotype, as proposed by Eisenhofer and colleagues (20, 21): (i) adrenergic tumours (PCs secreting metanephrine and/or epinephrine, alone or in combination with normetanephrine/norepinephrine), (ii) noradrenergic tumours (PCs secreting only norepinephrine and normetanaphrine), and (iii) biochemically silent tumours (both urinary metanephrine and normetanephrine concentrations in the normal range). Blood genetic testing was routinely performed in every PPGL patient since 1998 and data were available in 70/80 patients. Only the first family member diagnosed with PC was taken into consideration to calculate the proportion of genetic disease.

We considered that patients had received adequate preoperative medical preparation if they had been treated with an alpha-adrenergic receptor blocker for at least 14 days before surgery. This two week-period was selected based on current guidelines (2) and procedures adopted by most other centers (12). Two patients receiving preoperative alpha-blockers for only 4 and 6 days were therefore not included in this subgroup. Major hypertensive peaks (systolic blood pressure $\geq$ 300 mmHg), cardiac arrhythmia, acute pulmonary edema, cardiac arrest and vasoplegic shock during or early after surgery were all considered as severe hemodynamic perioperative complications. We also included as outcome measures a prolonged total surgical stay (> 10 days) and a prolonged stay (> 24h) in the intensive care

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unit (ICU). Indeed, several patients with no perioperative complications were systematically monitored in the ICU during the first postoperative day. Postoperative normalisation of blood pressure was defined as repeated normal ambulatory blood pressures without any anti-hypertensive drug both at home and during outpatient visits for at least 6 months after surgery.

Hormone assays

Epinephrine, norepinephrine and fractionated metanephrines were measured on 24-hour urine collection. Urine samples were collected in polyethylene containers with 15 mL of HCl 6N used as a preservative and stored at 4°C. Assays were always performed within 2 weeks. Until the end of 2018, urinary catecholamines and metanephrines concentrations were determined by high-performance liquid chromatography method with electrochemical detection. An internal standard (dehydroxylbenzylamine/DHBA) was included with each extraction to monitor recovery and facilitate quantification. Since 2019, in order to increase throughput, reduce solvent use and enhance resolution, urinary catecholamines and metabolites were measured using ultra-performant liquid chromatography on Acquity® systems with electrochemical detector ECD 2465 (Waters corporation, Milford, MA, USA).

Statistics

Statistical analyses were performed using the SPSS Statistics version 25.0 software (SPSS Inc., Chicago, IL). The continuous variables were reported as mean ± standard deviation, or as median and (P5-P95) interval. The normality of the data was assumed for a sample size greater than or equal to 30 according to the strong law of large numbers. Continuous variables were compared using Student's unpaired t-tests whereas analysis of variance was used for multiple comparisons. Discrete variables were compared by the Chi-square test. For multiple comparisons, the significance threshold was adjusted by the Bonferroni method. Correlations were calculated using Pearson’s regression analysis. A

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multivariate logistic regression analysis was also performed to determine factors independently predicting peroperative hemodynamic complications. A p value of less than 0.05 was considered statistically significant. The study was approved by the Ethics Committee of the Cliniques Universitaires Saint Luc of Brussels.

Results

General characteristics at diagnosis

The study population included 80 patients (39 men and 41 women, mean age at diagnosis: 47±18 years) (**Table 1**). A significant proportion of patients (58/79) had hypertension and more than half had abnormal glucose metabolism, either diabetes (n=21) or impaired fasting glycaemia (n=22). Clinical manifestations at diagnosis were present in 53/74 patients. Thirty-seven subjects had one or more typical manifestations of PC (paroxystic crises of headaches, palpitations, diaphoresis, and/or pallor associated with hypertension), while the other patients presented with Takotsubo-like cardiomyopathy (n=5), abdominal pain (n=5), malignant hypertension (n=3) or other symptoms related to tumor or metastases (n=3). In the 21 asymptomatic patients, diagnosis was made during familial genetic screening in 12 or following incidental discovery of an adrenal mass in nine.

The most effective imaging techniques to localize PC were abdominal CT-scan and MRI, both techniques identifying the adrenal tumour in all cases they were performed (61 and 43 patients, respectively). ^{123}I meta-iodo-benzyl-guanidine (MIBG) scintigraphy was performed in 54 patients and correctly identified the tumor(s) in 49, whereas ^{18}F-deoxyglucose positron emission tomography (PET) fused with CT was also regularly performed during the last ten years and showed hypermetabolism of the tumour in 15/17 cases with a maximum SUV value of 6.1 ± 3.9. Six patients had a bilateral PC at diagnosis, whereas

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two other patients developed a metachronous controlateral PC during follow-up. Two patients had liver metastases at diagnosis.

Genetic testing

Genetic testing was performed in 70 patients and a germline mutation in one of the known susceptibility genes was found in 29 of them. When only the first family member was considered, one third of the tested patients (21/62) had a hereditary disease. The mutated genes were RET (n=9), VHL (n=5), SDHB (n=3), SDHD (n=1), NF1 (n=2) and MAX (n=1). Compared to patients with a sporadic pheochromocytoma, index patients harbouring a germline mutation were diagnosed at a younger age (40 ± 18 vs 51 ± 17 years; p=0.02), were more often men (3:1 vs 1:1.3 ratio, p=0.015), and tended to be less often symptomatic at diagnosis (15/29 vs 38/51; p=0.08). Notably, all bilateral PCs, belonged to the hereditary group.

Characteristics according to the secretory phenotypes

The vast majority of PCs (73/79) were secreting at diagnosis, although a significant proportion of them (n=18) had no suggestive symptoms. Fifty-two had an adrenergic phenotype, with tumours secreting excess of metanephrine/epinephrine, alone (n=6) or most often in combination with normetanephrine/norepinephrine (n=46), while 21 patients had a noradrenergic phenotype, secreting only normetanephrine/norepinephrine (**Table 2**). Biochemically silent PCs were found in 6 patients, all being asymptomatic and normotensive. When comparing adrenergic and noradrenergic tumours, most features at diagnosis were not significantly different between the two subgroups (**Table 2**). The prevalence of hereditary forms was not different between the adrenergic and noradrenergic groups, but was higher in the biochemically silent patients, reflecting early diagnosis by genetic screening in this group.

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Interestingly, tumour size differed according to the secretory PC phenotype. Biochemically silent tumours were the smallest ones, while noradrenergic PC were the largest ($p<0.01$). Furthermore, considering the whole cohort of patients, tumour size significantly correlated with 24 hour-urinary metanephrine or normetanephrine excretion (**Figure 1a and 1b**).

Preoperative medical treatment

Prolonged (≥ 14 days) preoperative alpha-adrenergic receptor blocker treatment had been given in 29 of the 80 patients, never in patients with a biochemically silent PC and less often in patients with a noradrenergic phenotype ($p=0.03$; **Table 2**). Interestingly enough, alpha-blockade was much more frequently administered to the patients operated during the more recent period 2013-2018 (20/32) than in those with surgery performed during the period 1988-2012 (9/58; $p<0.01$). Most patients (n=27) were treated with prazosin at a median steady-state dose of 1.5 mg/day (range, 1 to 7 mg/day). The median duration of alpha-adrenergic receptor blocker treatment in these patients was 30 days (range, 14 to 90). Additional beta-adrenergic blockade was provided to 23 patients after a median interval of 5 days [2-12 days] while 15 other patients were already treated with beta-blockers since several months or years.

When comparing patients with and without preoperative alpha- blocker treatment (**Table 3**), there was no difference in age, prevalence of hypertension or tumour size between the two subgroups, but a higher proportion of patients with alpha-blockade were symptomatic at diagnosis or had paroxysmal attacks ($p=0.01$). In addition, these patients had 2-fold higher urinary metanephrine levels, although the difference did not reach statistical significance. Regarding surgical outcome, peri-operative hemodynamic complications occurred more frequently in patients without medical preparation (12/51 vs 3/29, $p=0.05$) while these patients had also undergone more frequently laparotomy than patients with preoperative

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blockade. The number of patients hospitalized in ICU for more than one day tended to be higher in patients without preoperative alpha-blockers (**Table 3**).

Surgical outcome

All our patients underwent surgical adrenalectomy, 49 patients by a laparoscopic approach and 31 patients by open laparotomy. The mean time delay between diagnosis and surgery was 56 ± 46 days. There was no perioperative death but 15 patients (one fifth) had severe hemodynamic complications during or within 24h after surgery. These subjects had a more severe and symptomatic hypertension at diagnosis, a 2.7 fold-higher excretion of normetanephrines (but not of metanephrines) and a larger tumour (**Table 4**). They had also undergone more frequently a laparotomy than a laparoscopic adrenalectomy and had more frequently a final diagnosis of malignant PC. In contrast, age, BMI, glucose metabolism and the secretory phenotype (**Table 2**) as well as the presence and type of a genetic mutation were not associated with the incidence of hemodynamic complications. Interestingly, while the proportion of patients with alpha-blocker preparation was not significantly different between the two subgroups, patients with severe complications had been treated for a much shorter period before surgery (median of 18 vs 30 days, p=0.017). As expected, the mean duration of ICU stay and total duration of surgical stay were longer in the complicated group (**Table 4**). We also performed a multivariate analysis, which showed that tumor size was the main preoperative factor independently predicting the risk of hemodynamic complications, while a shorter duration of preoperative alpha-blocker treatment was also a significant risk factor when tumor size was not considered (**Table 5**).

Long-term follow-up

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The patients were followed for a median period of 53 months [6-248 months]. All patients except the two with metastases were in remission after surgery (78/80). Patients with a noradrenergic PC showed less often normalization of their blood pressure after surgery as opposed to patients with an adrenergic phenotype ($p< 0.05$; **Table 2**). A relapse of the tumour was observed in 5 cases, after a median delay of more than 10 years. During follow-up, liver, bone and/or peritoneal metastases developed in 5 patients so that a malignant PC was finally identified in a total of 7 patients of the cohort. The relapsing and malignant PCs had usually a noradrenergic phenotype and were never diagnosed as a biochemically silent tumor (**Table 2**). The proportions of patients in remission, with relapse, or with a malignant PC were not different between hereditary and sporadic cases (data not shown). At the end of the follow up, 5 patients had died, but only 2 deaths could be attributed to complications of a metastatic pheochromocytoma.

Discussion

In this study, we report the characteristics of a monocentric cohort of 80 patients with an adrenal pheochromocytoma. These characteristics are consistent with the features usually observed in large series of PCs in the literature (5, 8, 18, 20, 22, 23, 24). In particular, we found no sex predominance, a mean age at diagnosis of 47 years and a mean tumour size of more than 50 mm. However, in contrast to recently reported series (7, 8), the diagnosis in our cohort was still mostly driven by the presence of symptoms or complications typical of a pheochromocytoma. In addition, in agreement with previous publications (18, 22, 23, 24), three quarters of our subjects had a history of hypertension and more than half had diabetes or impaired fasting glucose level at diagnosis.

In the remaining 21 patients, the tumour was found either as the result of mutation-based case detection testing or following identification of an adrenal incidentaloma on

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imaging. This mode of presentation has become increasingly prevalent over the last years, due to frequent routine use of abdominal imaging leading to the diagnosis of less active and less symptomatic tumours in older individuals (7, 8). It is worth mentioning that PC is a frequent finding in the work-up of an incidentally discovered mass, accounting for 17% of them in a recent study (6). The differences in the mode of presentation of PC between our study and more recent series may be explained by the extended period of inclusion and by recruitment bias, as severe and symptomatic tumours were more often referred to our center.

Case detection testing for patients with genetic predisposition has also become a common way of diagnosing PC, as it was the case for more than half of the asymptomatic patients in our cohort. In total, we found a pathogenic germline mutation in one third of all index patients, a proportion similar to those reported by previous genetic studies on PPGL, ranging from 25 % to 40% of patients (2, 9, 10, 11). As expected, familial cases were diagnosed at a younger age, had frequently bilateral tumours (25), and, as reported in other studies, were not different compared to sporadic PC in terms of surgical complications (18, 26).

Amongst the 73 patients with secreting PC at diagnosis, more than 2/3 were oversecreting epinephrine, most often together with norepinephrine, and less than 1/3 had a noradrenergic tumour. This is consistent with previous findings on PC secretory phenotypes based on plasma metanephrines (19, 20, 21, 27). We also observed a significant correlation between tumour size and the amount of urinary normetanephrine and metanephrine excretion. This suggests that as much as plasma metanephrines, 24-hour urinary metabolites reflect well tumour activity and secretory phenotype. Moreover, noradrenergic PCs were the largest ones, as shown in other studies (19, 28).

We also observed that the noradrenergic tumours showed less often normalization of blood pressure after surgery compared to the adrenergic phenotype. This is well in line with

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the recent observation that patients with a noradrenergic PC have more severely impaired diurnal blood pressure variability, permanent damage to the arterial vessels and persistent hypertension (19). In contrast, patients with significantly elevated levels of epinephrine often have paroxysmal hypertension and orthostatic hypotension (29).

It is interesting to note that in 6 cases we found a biochemically silent PC. All these patients were clinically asymptomatic and showed neither paroxysmal attacks nor hypertension at diagnosis. However, even though their clinical presentation was mild, one patient showed perioperative hemodynamic complications, because of catecholamine surges induced by mobilization and surgical excision of the tumour. Such incidentally discovered normotensive pheochromocytomas with normal metanephrines values have indeed been reported previously and may show hemodynamic instability at surgical excision (8, 30).

Surgery is the primary treatment for patients with PCs but still carries a high risk of hemodynamic complications during the perioperative period (12, 13). Current guidelines support the use of alpha-adrenergic receptor blockers for at least 10 to 14 days prior to surgery to minimize perioperative complications (2, 14, 17, 31). This recommendation is mainly based on retrospective studies and there is no randomized controlled study that has definitively confirmed the benefits of preoperative medical preparation (12, 14, 15). Data are even weaker regarding the duration of such medical treatment.

In our study, only 36% of the patients were treated for at least 14 days with alpha-adrenergic receptor blockers. There was however a significant trend to use more frequently preoperative alpha-blockade over the last years. The treated patients were also more symptomatic at diagnosis and had more frequently paroxysmal attacks. Their pre-treatment systolic and diastolic blood pressure also tended to be higher than in those without medical preparation. Despite this selection bias and a more severe clinical presentation at diagnosis, hemodynamic complications during surgery were less frequent in patients who had received

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preoperative alpha-adrenergic blocker treatment. Notably also, in the few patients who developed hemodynamic complications despite having received preoperative alpha-blockers, the median duration of treatment was shorter than in those without complications. These observations support the use of preoperative alpha-blocker treatment for a sufficient period of time before PPGL surgery and this recommendation might even be extended to the normotensive patients.

Perioperative complications resulting from severe hemodynamic instability occurred more frequently in patients with a symptomatic PC, higher levels of normetanephrine, a larger or malignant tumour, and a need for open laparotomy. Similar results were found in the study reported by Bruynzeel et al. (26). However, in the latter, there was no difference between those who underwent open surgery and laparoscopic surgery. Our results may be biased by the fact that a laparotomy was usually preferred in the more complex patients. Nevertheless, other studies have also reported that laparotomy was associated with more surgical complications (32, 33).

During follow-up, we observed a recurrence in 5 patients, a recurrence rate close to the rate of 0.98 events/100 person-years reported by Amar et al. (34). The proportion of malignant PCs was close to the 10% figure found in the literature (35). As reported previously (20, 34), recurrence and malignancy were associated with initial tumour size, but also with a noradrenergic phenotype which seems therefore to confer a worse prognosis to the tumour.

Our study has several limitations. Due to its retrospective design, some data at diagnosis were missing for a few patients. In addition, this study was carried out in a monocentric cohort and the number of patients may have been insufficient to reach statistical significance for some comparisons. The inclusion period spanned three decades and recommended practice evolved over that time period. Plasma metanephrines were measured only in a minority of patients and, in contrast to the pivotal study by Eisenhofer and

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colleagues (20), we rather used 24 h-urinary factionated metanephrines to determine the secretory phenotype. We believe however that this approach has also been strongly validated to evaluate tumoural catecholamine secretion (2). The strengths of this study are that all patients were diagnosed and managed by the same multidisciplinary team and followed for a long period. It therefore provides valuable results on the real-life management of adrenal PCs in a tertiary center.

In conclusion, we show in this monocentric series of patients with an adrenal pheochromocytoma that the secretory phenotype does not significantly influence the rate of hemodynamic complications during surgery but may affect long-term post-surgical outcome. While biochemically silent PCs are usually of small size and good prognosis, noradrenergic tumours show a worse post-surgical outcome than adrenergic ones, with persistent hypertension despite remission and a higher rate of recurrence or malignant behavior. Our data also add support to the current recommendation of a prolonged alpha-adrenergic blocker preoperative preparation in patients with PPGL, even in those with normal blood pressure.

Declarations of interest : We declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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References:

1. Lenders JW, Eisenhofer G, Mannelli M, Pacak K. Phaeochromocytoma. *Lancet* 2005 **366** 665-675.

2. Lenders JW, Duh QY, Eisenhofer G, Gimenez-Roqueplo AP, Grebe SK, Murad MH, Naruse M, Pacak K, Young WF Jr. Endocrine Society. Pheochromocytoma and paraganglioma: an endocrine society clinical practice guideline. *Journal of Clinical Endocrinology and Metabolism* 2014 **99** 1915-1942.

3. Nölting S, Ullrich M, Pietzsch J, Ziegler CG, Eisenhofer G, Grossman A, Pacak K. Current Management of Pheochromocytoma/Paraganglioma: A Guide for the Practicing Clinician in the Era of Precision Medicine. *Cancers (Basel)* 2019 **11** 1505-1531.

4. Santos Jenn RU, Brofferio A, Viana B, Pacak K. Catecholamine-Induced Cardiomyopathy in Pheochromocytoma: How to Manage a Rare Complication in a Rare Disease? *Hormone and Metabolic Research* 2019 **51(07)** 458-469.

5. Geroula A, Deutschbein T, Langton K, Masjkur J, Pamporaki C, Peitzsch M, Fliedner S, Timmers HJLM, Bornstein SR, Beuschlein F, et al. Pheochromocytoma and paraganglioma: clinical feature-based disease probability in relation to catecholamine biochemistry and reason for disease suspicion. *European Journal of Endocrinology* 2019 **181** 409-420.

6. Ye YL, Yuan XX, Chen MK, Dai YP, Qin ZK, Zheng FF. Management of adrenal incidentaloma: the role of adrenalectomy may be underestimated. *BMC Surgery* 2016 **16** 41.

7. Falhammar H, Kjellman M, Calissendorff J. Initial clinical presentation and spectrum of pheochromocytoma: a study of 94 cases from a single center. *Endocrine Connections* 2018 **7** 186-192.

Copyright © 2019 the authors

8. Gruber LM, Hartman RP, Thompson GB, McKenzie TJ, Lyden ML, Dy BM, Young WF, Bancos I. Pheochromocytoma Characteristics and Behavior Differ Depending on Method of Discovery. *Journal of Clinical Endocrinology and Metabolism* 2019 **104** 1386-1393.

9. Gimenez-Roqueplo AP, Dahia PL, Robledo M. An update on the genetics of paraganglioma, pheochromocytoma, and associated hereditary syndromes. *Hormone and Metabolic Research* 2012 **44** 328–333.

10. Fishbein L, Merrill S, Fraker DL, Cohen DL, Nathanson KL. Inherited mutations in pheochromocytoma and paraganglioma: why all patients should be offered genetic testing. *Annals of Surgical Oncology* 2013 **20** 1444–1450.

11. Buffet A, Burnichon N, Favier J, Gimenez-Roqueplo AP. An overview of 20 years of genetic studies in pheochromocytoma and paraganglioma. *Best Practice & Research Clinical Endocrinology & Metabolism* 2020 **34** 1014-1016.

12. Challis BG, Casey RT, Simpson HL, Gurnell M. Is there an optimal preoperative management strategy for phaeochromocytoma/paraganglioma? *Clinical Endocrinology (Oxford)* 2017 **86** 163-167.

13. Groeben H, Walz MK, Nottebaum BJ, Alesina PF, Greenwald A, Schumann R, Hollmann MW, Schwarte L, Behrends M, Rossel T, et al. International multicentre review of perioperative management and outcome for catecholamine-producing tumours. *British Journal of Surgery* 2020 **107** e170-e178.

14. Berends AMA, Kerstens MN, Lenders JWM, Timmers HJLM. Approach to the Patient: Perioperative Management of the Patient with Pheochromocytoma or Sympathetic Paraganglioma. *Journal of Clinical Endocrinology and Metabolism* 2020 1**05** 3088-3102.

Copyright © 2019 the authors

 Accepted Manuscript published as EC-20-0537. Accepted for publication: 14-Dec-2020

15. Schimmack S, Kaiser J, Probst P, Kalkum E, Diener MK, Strobel O. Meta-analysis of α-blockade versus no blockade before adrenalectomy for phaeochromocytoma. *British Journal of Surgery* 2020 **107** e102-e108.

16. Lentschener C, Gaujoux S, Thillois J.M. Increased arterial pressure is not predictive of haemodynamic instability in patients undergoing adrenalectomy for phaeochromocytoma. *Acta Anaesthesiologica Scandinavica* 2009 **53** 522–527.

17. Isaacs M, Lee P. Preoperative alpha-blockade in phaeochromocytoma and paraganglioma: is it always necessary? *Clinical Endocrinology (Oxford)* 2017 **86** 309-314.

18. Plouin PF, Duclos JM, Soppelsa F. Factors Associated with Perioperative Morbidity and Mortality in Patients with Pheochromocytoma: Analysis of 165 Operations at a Single Center. *Journal of Clinical Endocrinology and Metabolism* 2001 **86** 1480– 1486.

19. Petrák O, Rosa J, Holaj R, Štrauch B, Krátká Z, Kvasnička J, Klimová J, Waldauf P, Hamplová B, Markvartová A, et al. Blood Pressure Profile, Catecholamine Phenotype, and Target Organ Damage in Pheochromocytoma/Paraganglioma. *Journal of Clinical Endocrinology and Metabolism* 2019 **104** 5170-5180.

20. Eisenhofer G, Lenders JW, Goldstein DS, Mannelli M, Csako G, Walther MC, Brouwers FM, Pacak K. Pheochromocytoma catecholamine phenotypes and prediction of tumour size and location by use of plasma free metanephrines. *Clinical Chemis*try 2005 **51** 735–744.21. Eisenhofer G, Timmers HJ, Lenders JW, Bornstein SR, Tiebel O, Mannelli M, King KS, Vocke CD, Linehan WM, Bratslavsky G, et al. Age at diagnosis of pheochromocytoma differs according to catecholamine phenotype and tumour location. *Journal of Clinical Endocrinology and Metabolism* 2011 **96** 375-384.

22. Kebebew E, Duh QY. Benign and malignant pheochromocytoma: diagnosis, treatment, and follow-up. *Surgical Oncology Clinics of North America* 1998 **7** 765–789.

Copyright © 2019 the authors

23. Amar L, Servais A, Gimenez-Roqueplo AP, Zinzindohoue F, Chatellier G, Plouin PF. Year of diagnosis, features at presentation, and risk of recurrence in patients with pheochromocytoma or secreting paraganglioma. *Journal of Clinical Endocrinology and Metabolism* 2005 **90** 2110-2116.

24. Guerrero MA, Schreinemakers JM, Vriens MR, Suh I, Hwang J, Shen WT, Gosnell J , Clark OH, Duh QY. Clinical spectrum of pheochromocytoma. *Journal of the American College of Surgeons* 2009 **209** 727-732.

25. Kittah NE, Gruber LM, Bancos I, Hamidi O, Tamhane S, Iñiguez-Ariza N, Babovic-Vuksanovic D, Thompson GB, Lteif A, Young WF, et al. Bilateral pheochromocytoma: Clinical characteristics, treatment and longitudinal follow-up. *Clinical Endocrinology (Oxford)* 2020 May 14. doi: 10.1111/cen.14222.

26. Bruynzeel H, Feelders RA, Groenland TH, van den Meiracker AH, van Eijck CH, Lange JF, de Herder WW, Kazemier G. Risk Factors for Hemodynamic Instability during Surgery for Pheochromocytoma. . *Journal of Clinical Endocrinology and Metabolism* 2010 **95** 678-685.

27. Petrák O, Klímová J, Mráz M, Haluzíková D, Doležalová RP, Kratochvílová H, Lacinová L, Novák K, Michalsky D, Waldauf P, et al. Pheochromocytoma With Adrenergic Biochemical Phenotype Shows Decreased GLP-1 Secretion and Impaired Glucose Tolerance. *Journal of Clinical Endocrinology and Metabolism* 2020 **105** 1878–1887.

28. Funahashi H, Imai T, Tanaka Y, Tobinaga J, Wada M, Matsuyama T, Tsukamura K, Yamada F, Takagi H, Narita T, et al. Discrepancy between PNMT presence and relative lack of adrenaline production in extra-adrenal pheochromocytoma. *Journal of Surgical Oncology* 1994 **57** 196-200.

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29. Zuber SM, Kantorovich V, Pacak K. Hypertension in pheochromocytoma: characteristics and treatment. *Endocrinology and Metabolism Clinics of North America* 2011 **40** 295-311.

30. Lafont M, Fagour C, Haissaguerre M, Darancette G, Wagner T, Corcuff JB, Tabarin A. Per-operative hemodynamic instability in normotensive patients with incidentally discovered pheochromocytomas. *Journal of Clinical Endocrinology and Metabolism* 2015 **100** 417-421

31. Pacak K. Preoperative management of the pheochromocytoma patient *Journal of Clinical Endocrinology and Metabolism* 2007 **92** 4069-4079.

32. Janetschek G, Finkenstedt G, Gasser R, Waibel UG, Peschel R, Bartsch G, Neumann HP. Laparoscopic surgery for pheochromocytoma: adrenalectomy, partial resection, excision of paragangliomas. *The Journal of Urology* 1998 **160** 330–334.

33. Fernandez-Cruz L, Taura P, Saenz A, Saenz A, Benarroch G, Sabater L. Laparoscopic approach to pheochromocytoma: hemodynamic changes and catecholamine secretion. *World Journal of Surgery* 1996 **20** 762–768.

34. Amar L, Lussey-Lepoutre C, Lenders JW, Djadi-Prat J, Plouin PF, Steichen O. Recurrence or new tumours after complete resection of pheochromocytomas and paragangliomas: a systematic review and meta-analysis. *European Journal of Endocrinology* 2016 **175** R135-R145.

35**.** Pacak K, Eisenhofer G, Ahlman H, Bornstein SR, Gimenez-Roqueplo AP, Grossman AB. Pheochromocytoma: recommendations for clinical practice from the First International Symposium. *Nature Clinical Practice Endocrinology* 2007 **3(2)** 92-102.

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Figure 1. Correlations observed between tumour size and 24-hour urinary metanephrine (A) and normetanephrine excretion (B)

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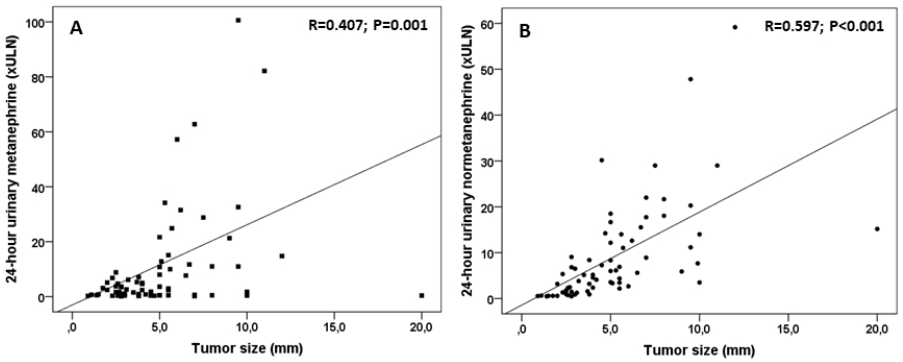


Figure 1. Correlations observed between tumour size and 24-hour urinary metanephrine (A) and normetanephrine excretion (B)%

254x190mm (96 x 96 DPI)

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Table 1. General characteristics of the 80 patients operated for an adrenal pheochromocytoma

	All patients n=80
Mean age at diagnosis (years)	47.0 ± 18.4
Patients aged < 50 years	40/80
Sex ratio (men/women)	39/41
Mean BMI (kg/m²)	24.8 ± 5.0
Mean tumour diameter (cm)§	5.4 ± 3.4
Family history of PC	17/74
Patients with hypertension	58/79
Systolic blood pressure at diagnosis (mmHg)	160 ± 40
Diastolic blood pressure at diagnosis (mmHg)	92 ± 20
Patients with abnormal blood glucose level	43/79

Data are shown as mean ± SD, medians and [P5-P95] intervals or proportions. BMI: body mass index; PC: pheochromocytoma; §: mean diameter based on pathology report.

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Table 2. Characteristics of patients at diagnosis (data available for 74 patients), urinary fractionated metanephrines, tumour size and post-surgical outcome in relation to the biochemical profile of their pheochromocytoma (n=79)

	Adrenergic phenotype (n=52)	**Noradrenergic phenotype (n=21)**	**Biochemically silent (n=6)**	**P-value**
Mean age at diagnosis (years)	49.1 ± 17.3	40.4 ± 19.7	49.0 ± 20.9	0.18
Symptomatic PC at diagnosis	38/50	15/19	0/5*	0.01
Paroxysmal crises at diagnosis	32/50	9/19	0/5*	0.03
Nb of patients with hypertension	40/50	16/19	0/5*	<0.01
Systolic blood pressure (mm Hg)	163 ± 42	164 ± 35	126 ± 12	0.09
Diastolic blood pressure (mm Hg)	94 ± 21	91 ± 18	78 ± 5	0.19
Nb of patients with abnormal blood glucose	32/50	8/19	1/5	0.17
Urinay metanephrine (xULN)	7.4 [1.7-94.1]	0.4 [0.2-0.9]*	0.7 [0.3-0.9]*	<0.01
Urinary normetanephrine (xULN)	5.2 [0.6-41.2]	8.4 [2.1-22.0]	0.6 [0.5-0.8]*	0.04
Nb of patients with mutation	19/45	6/19	4/5	0.21
Type of mutation (RET/VHL/SDHX/NF1/MAX)	12/4/1/2/0	0/3/2/0/1	1/3/0/0/0	0.02
Tumour size (cm)	5.3 ± 2.6	6.6 ± 4.5	1.7 ± 0.8*	<0.01
Preoperative alpha-blocker treatment[$]	24/52	5/21	0/6	0.03
Hemodynamic complications	8/52	6/21	1/6	0.42
ICU stay > 1 day	9/52	7/21	1/6	0.30
Remission	52/52	19/21	6/6	0.06
Normal blood pressure after Sx	35/40	8/16*	6/6	<0.01
Recurrence	1/52	4/21*	0/6	0.01
Malignancy	2/52	5/21*	0/6	0.02

Data are shown as mean ± SD, medians and [P5-P95] intervals or proportions. PC: pheochromocytoma; ULN: upper limit of normal range; Sx: surgery; Nb: number. The P value is derived from one-way ANOVA comparing the three subgroups.
*:indicates subgroups with values significantly different from other subgroups without an asterisk. [$]:refers to patients with alpha-blocker treatment for at least 14 days before adrenalectomy.

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Table 3. Characteristics of patients with PC according to the administration or not of an adequate (≥ 14 days) preoperative alpha-blocker treatment

	No preoperative alpha-blocker treatment (n=51)	Preoperative alpha-blocker treatment (n=29)	P value
Mean age at diagnosis (years)	44.7 ± 19.1	51.1 ± 16.5	0.14
Symptomatic PC at diagnosis	27/50	25/29	<0.01
Paroxysmal crises	20/50	21/29	0.01
Nb of patients with hypertension	35/50	23/29	0.37
Systolic blood pressure (mm Hg)	155 ± 34	170 ± 48	0.11
Diastolic blood pressure (mm Hg)	89 ± 19	96 ± 23	0.14
Urinary metanephrine (xULN)	2.4 [0.2-34.0]	5.7 [0.3-110.0]	0.06
Urinary normetanephrine (xULN)	5.3 [0.5-29.0]	5.4 [0.7-39.9]	0.98
Tumour size (cm)	5.3 ± 3.9	5.4 ± 2.3	0.94
Nb of patients with laparotomy	24/51	7/29	0.04
Nb of patients with hemodynamic complications	12/51	3/29	0.05
Nb of patients with ICU stay > 24h	14/51	4/29	0.13

Data are shown as mean ± SD, medians and [P5-P95] intervals or proportions. PC: pheochromocytoma; ULN: upper limit of normal range; Nb: number; ICU: intensive care unit.

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Table 4. Characteristics of patients with PC according to the occurrence or not of severe hemodynamic complications during or within 24 hours after surgery

	No hemodynamic complications (n=65)	Hemodynamic complications (n=15)	P value
Mean age at diagnosis (years)	47.4 ± 18.7	45.3 ± 17.4	0.69
Mean BMI (kg/m²)	24.5 ± 4.8	26.1 ± 5.9	0.30
Symptomatic PC at diagnosis	40/64	13/15	0.06
Mean systolic blood pressure (mmHg)	156 ± 37	180 ± 48	0.04
Mean diastolic blood pressure (mmHg)	89 ± 17	102 ± 29	0.02
Abnormal blood glucose level at diagnosis	33/64	10/15	0.29
Urinary metanephrine (xULN)	0.9 [0.1-16.4]	0.4 [0.1-18.4]	0.54
Urinary normetanephrine (xULN)	2.7 [0.3-17.5]	7.3 [0.3-26.2]	0.05
Mean tumour size (cm)	4.9 ± 2.8	7.5 ± 4.8	0.04
Malignant PC (%)	3/65	4/15	0.01
Nb of patients with preoperative alpha-blocker treatment[$]	26/65	3/15	0.15
Duration of alpha-blocker treatment (days)#	30 [13-90]	18 [4-26]	0.02
Nb of patients with laparotomy (%)	17 (26%)	14 (93%)	<0.01
Mean ICU stay (days)	1 [0-5]	3 [1-7]	0.01
Mean surgical stay (days)	6 [4-12]	12 [8-16]	<0.01

Data are shown as mean ± SD, medians and [P5-P95] intervals or proportions. PC: pheochromocytoma; BMI: body mass index; ULN: upper limit of normal range; Nb: number; ICU: intensive care unit; [$]:refers to the 29 patients with alpha-blocker treatment for at least 14 days before adrenalectomy; #: refers to all 31 patients receiving preoperative alpha-blocker treatment.

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Table 5. Predictive factors for hemodynamic complications during or early after adrenal surgery for a pheochromocytoma according to univariate and multivariate analysis

Variables	Univariate analysis			Multivariate analysis*		
	odds ratio	IC 95%	*P value*	odds ratio	IC 95%	*P value*
Age at surgery (years)	0.994	[0.964-1.024]	0.690	-	-	-
Symptoms at diagnosis	4.065	[0.845-9.607]	0.180	-	-	-
Urinary normetanephrines (xULN)	1.048	[0.996-1 ?102]	0.069	0.999	[0.869-1.149]	0.991
Tumor size (cm)	1.230	[1.040-1.056]	0.016	2.074	[1.027-4.180]	0.042
Adequate preoperative alpha-blocker	0.357	[0.096-1.490]	0.157	-	-	-
Duration of alpha-blocker treatment (days)	0.861	[0.753-0.985]	0.030	0.858	[0.697-1.052]	0.141
Laparotomy	39.53	[4.83-323.72]	<0.001	-	-	-

* Only variables which were significant at p<0.010 in univariate analysis were introduced in the multivariate analysis. Laparotomy was intentionally not included as this factor outweighed by far all other variables and was highly correlated with both tumor size (r=0.488; p<0.001) and urinary normetanephrine excretion (r=0.430; p<0.001). When tumor size was removed from the analysis, only duration of preoperative alpha-blocker treatment remained significant with an odds ratio of 0.858 per day of treatment [0.747-0.987] (p=0.032).

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