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The relationship between mild autonomous cortisol secretion and metabolic diseases in cases with adrenal incidentaloma

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Abstract

Introduction: This study investigates the link between mild autonomous cortisol secretion (MACS) in adrenal incidentaloma (AI) patients and the occurrence and severity of cardiovascular and metabolic comorbidities. It aims to provide a detailed overview of this relationship, highlight gaps in current research, and propose directions for future studies.

Material and methods: We conducted a retrospective analysis at Ankara City Hospital's Endocrine Department outpatient clinic, reviewing 627 AI patients from February 2019 to May 2021. The study involved a detailed analysis of clinical records, hormonal evaluations, and imaging, focusing on differentiating MACS from non-functioning adrenal incidentalomas (NFAI) and examining the impact of MACS on associated health conditions.

Results: The study found that MACS patients had a statistically higher incidence of diabetes mellitus (35% *vs*. 20%), hypertension (60% *vs*. 45%), hyperlipidaemia (40% *vs*. 25%), and coronary artery disease (30% *vs*. 15%) compared to the NFAI group. Independent predictors of MACS included the presence of bilateral adrenal masses, larger adrenal mass diameter (with a cutoff value of ≥ 18.5 mm, showing 83% sensitivity and 56% specificity for predicting MACS, and lower dehydroepiandrosterone sulphate (DHEAS) levels (≤ 49.31 μg/dL predicting MACS, with 61% sensitivity and 73% specificity).

Conclusion: This research underscores the critical clinical implications of detecting MACS in AI patients, particularly its association with increased cardiovascular and metabolic risks. It calls for vigilant screening and a comprehensive management approach for affected patients. Additionally, the findings highlight the need for further studies to improve patient care and outcomes in this population.

Key words: *adrenal incidentalomas; mild autonomous cortisol secretion; cardiovascular risk; metabolic comorbidities; endocrinology*

Introduction

The detection of adrenal incidentalomas (AIs), adrenal masses identified unintentionally during radiological examinations for unrelated medical reasons, has become an increasingly common clinical scenario, paralleling the advancements and expanded use of cross-sectional imaging technologies. These incidental findings pose a significant challenge for endocrinologists, necessitating careful evaluation to discern their clinical significance and manage potential hormonal excesses. Among these, mild autonomous cortisol secretion (MACS), recently defined by the European Society of Endocrinology/European Network for the Study of Adrenal Tumours (ESE/ENSAT) guidelines, is notably concerning due to its insidious impact on patient morbidity and mortality [1, 2]. It has been observed that the incidental discovery of adrenal masses occurs in approximately 1% to 5% of all abdominal computed tomography (CT) scans. This incidence rate can vary depending on the sensitivity of the imaging modality used and the reasons for which scans were performed. The prevalence of AIs has been reported to increase with age. The prevalence is estimated to be around 2% to 3% in the general population, but this rate can rise to 6% or higher in older populations [3–5].

MACS, in the context of AIs, represents a spectrum of cortisol hypersecretion, distinct from overt Cushing's syndrome by its lack of classic clinical features yet associated with a range of subtle metabolic and cardiovascular derangements. The pathophysiological mechanisms underlying MACS involve dysregulated cortisol production, independent of the hypothalamic-pituitary-adrenal axis, leading to an array of clinical consequences, including hypertension, type 2 diabetes mellitus, dys-

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lipidaemia, and obesity, which collectively contribute to increased cardiovascular risk [6, 7].

Cardiovascular disease (CVD) remains the leading cause of death globally, with metabolic syndrome — a constellation of interrelated metabolic risk factors—significantly elevating the risk of developing CVD. The intricate relationship between cortisol and metabolic syndrome components underscores the hormone's pivotal role in metabolic regulation and highlights the potential for cortisol dysregulation, as seen in MACS, to exacerbate or precipitate metabolic syndrome, thereby amplifying cardiovascular risk [8–10].

Despite the acknowledged association between hypercortisolism and metabolic dysfunction, the specific impact of subclinical cortisol elevations, characteristic of MACS in AIs, on the development and progression of cardiovascular and metabolic comorbidities is not fully understood. Research has primarily concentrated on the metabolic consequences of overt hypercortisolism, with less attention devoted to the nuanced effects of MACS, particularly within the context of AIs [11]. This gap in knowledge underscores the need for focused investigation into how MACS contributes to cardiovascular and metabolic pathologies, the potential for reversibility of such conditions with normalisation of cortisol levels, and the implications for clinical management of AIs [12].

Therefore, this article seeks to elucidate the relationship between MACS in patients with AIs and the prevalence and severity of cardiovascular and metabolic comorbidities. By integrating insights from recent epidemiological studies, clinical trials, and biochemical investigations, we aim to provide a comprehensive overview of the current understanding of this relationship, identify unresolved questions, and suggest future research directions. This endeavour is critical for refining risk stratification, guiding therapeutic interventions, and ultimately improving outcomes for patients with AIs and MACS.

Material and methods

Study design and participants

This observational study employed a retrospective, monocentric design, conducted at the outpatient clinic of the Endocrine Department, Ankara City Hospital, between February 2019 and May 2021. We reviewed 627 patients diagnosed with AI identified during routine scheduled monitoring visits. Eligibility criteria included males and females aged 18 years or older, with unilateral or bilateral AI detected by CT or magnetic resonance imaging (MRI). We meticulously collected and analysed data from clinical records, adhering to ethical standards for retrospective research.

Exclusion criteria

Patients were excluded if they had conditions or were on medication known to affect steroid hormone secretion or metabolism (n = 11), if their radiological evaluations were initiated due to suspected cancer (adrenocortical carcinoma [ACC] n = 3, adrenal metastasis n = 2), or if they presented with adrenal masses smaller than 1 cm (n = 11). Additional exclusions encompassed individuals with overt hormonal excess syndromes, such as Cushing's syndrome (n = 3), primary hyperaldosteronism (n = 31), pheochromocytoma (n = 7), non-classic congenital adrenal hyperplasia (n = 5), myelolipoma (n = 8), or incomplete data in the initial dexamethasone suppression test (DST) (n = 85) (Fig. 1).

Data collection

Retrospective data collection from electronic medical records included demographic information (age, gender, disease duration) and comorbidities (hypertension, type 2 diabetes, dyslipidaemia, cardiovascular disease, and osteoporosis). Blood pressure measurements and definitions for MACS-related comorbidities followed established guidelines, with hypertension categorised by systolic blood pressure ≥ 140 mmHg and/or diastolic ≥ 90 mmHg or the use of antihypertensive medications. Diabetes mellitus was defined as per the American Diabetes Association criteria, dyslipidaemia was identified based on specific lipid thresholds or medication use, cardiovascular disease included ischaemic heart disease or heart failure, and osteoporosis was determined by dual-energy X-ray absorptiometry (DEXA) scan results, using World Health Organisation criteria for classification.

Hormonal evaluation and diagnostic criteria

A comprehensive hormonal evaluation was conducted, including measurements of morning serum cortisol, plasma adrenocorticotropic hormone (ACTH), and dehydroepiandrosterone sulphate (DHEA-S) at 8 a.m., 24-hour urinary free cortisol, plasma metanephrines, aldosterone, and plasma renin activity, alongside a 1 mg dexamethasone suppression test (DST). Laboratory tests for fasting

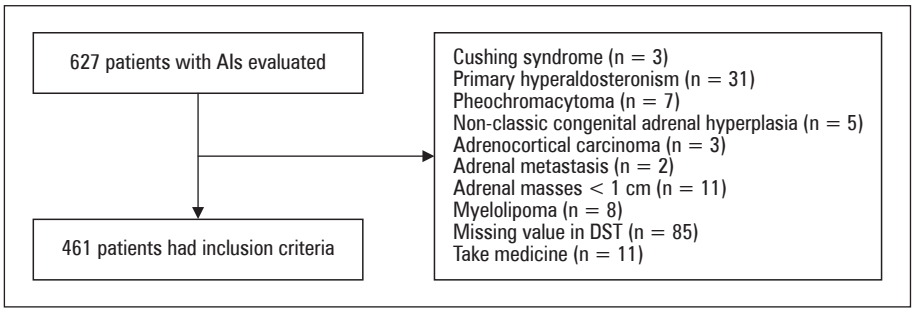

Figure 1. *Study population and exclusion study population. AIs — adrenal incidentalomas; DST — dexamethasone suppression test*

plasma glucose, glycated haemoglobin, serum electrolytes, liver function, and lipids were performed after an 8-hour fast.
AIs were classified as nonfunctioning without biochemical evidence of adrenal hormone hyperactivity. For suspected endocrine abnormalities, confirmatory tests were executed, including a 48-hour, 2 mg/day, low-dose DST (LDDST) for MACS, saline loading test for primary hyperaldosteronism, and iodine-123-metaiodobenzylguanidine scintigraphy or gallium-68 (Ga-68) positron emission tomography computed tomography (PET-CT) for pheochromocytoma. MACS diagnosis was based on serum cortisol levels >1.8 μg/dL following 1 mg DST, without overt clinical signs of Cushing's syndrome.

Imaging and laboratory assays

All participants underwent abdominal or adrenal magnetic resonance imaging (MRI) to assess tumour size, location, and laterality. Hormonal determinations were consistently performed in the same laboratory throughout the study, utilising assays from reputable manufacturers. The analytical precision of each assay was documented, including inter-assay and intra-assay coefficients of variation for cortisol, ACTH, aldosterone, plasma renin activity, urinary metanephrines, and other relevant analytes.

Statistical analysis

Data analysis was conducted via IBM SPSS Statistics for Windows, version 25.0 (IBM Corp., Armonk, N.Y., USA). The normality of the distribution of continuous variable was assessed using the Kolmogorov Smirnov test. Continuous variables with a normal distribution were expressed as mean ± standard deviation, while those with a non-normal distribution were expressed as median (interquartile range). Categorical variables were presented as frequency (percentage). Student's t-test was used to compare normally distributed continuous variables, and the Mann-Whitney U test was used for non-normally distributed ones. Categorical variables were compared using the chi-square test or Fisher's exact test. A binary univariate logistic regression analysis was performed to determine independent predictors of MACS, followed by binary multivariable logistic regression analysis that included the variables with a p-value ≤ 0.1 in univariate logistic regression analysis. The results were given as the odds ratio (OR), 95% confidence interval (CI), and p-value. Receiver operating characteristic (ROC) curve analyses were performed for continuous variables that were found to be independent predictors of MACS. The results are presented as the area under curve (AUC), 95% CI, p-value, cut-off value, sensitivity, and specificity. Also, ROC curve analyses were performed for cortisol levels after an overnight DST in predicting diabetes mellitus (DM), hypertension (HT), hyperlipidaemia (HL), and coronary artery disease (CAD) in patients with MACS. Statistical significance was defined as a p-value < 0.05.

Results

Comparative analysis of clinical, radiological, and laboratory characteristics

Table 1 delineates the clinical, radiological, and laboratory data across the entire cohort, further stratified into MACS and non-functioning adrenal incidentaloma (NFAI) subgroups, alongside the outcomes of intergroup comparisons. The cohort's mean age was 54.8 ± 10.19 years, with a predominance of female

Table 1. ***Clinical, radiological, and laboratory data of the study group, mild autonomous cortisol secretion (MACS), and nonfunctioning adrenal incidentaloma (NFAI) subgroups, and intergroup comparisons****

	Study group (n = 461)	MACS group (n = 77)	NFAI group (n = 384)	p
Age [year]s	54.8 ± 10.19	56.87 ± 10.67	54.39 ± 10.05	0.051
Sex, female, n (%)	309 (67)	56 (72.7)	253 (65.9)	0.244
Diabetes mellitus, n (%)	144 (31.2)	33 (42.9)	111 (28.9)	**0.016**
Hypertension, n (%)	247 (53.6)	53 (68.8)	194 (50.5)	**0.003**
Hyperlipidaemia, n (%)	141 (30.6)	32 (41.6)	109 (28.4)	**0.022**
Coronary artery disease, n (%)	53 (11.5)	16 (20.8)	37 (9.6)	**0.005**
Heart failure, n (%)	6 (1.3)	2 (2.6)	4 (1)	0.264
Mass laterality, n (%)				
Unilateral	356 (77.2)	48 (62.3)	308 (80.2)	
Right	117 (25.4)	21 (27.3)	96 (25)	**0.001**
Left	239 (51.8)	27 (35.1)	212 (55.2)	
Bilateral	105 (22.8)	29 (37.7)	76 (19.8)	
Mass diameter [mm]	19 (13–26)	26 (20–34)	17 (12–24)	**< 0.001**
Suppression after overnight DST, n (%)	384 (83.3)	–	384 (100)	**< 0.001**
Cortisol 8 a.m. after overnight DST [mcg/dL]	1.2 (0.87–1.57)	2.53 (2.09–3.6)	1.09 (0.8–1.3)	**< 0.001**
ACTH [pg/mL]	14.8 (9.9–20.3)	11.2 (7.2–14.7)	15.7 (10.8–21.5)	**< 0.001**
Cortisol [mcg/dL]	13.9 (10.8–17.7)	14.8 (11.3–17.4)	13.65 (10.6–17.83)	0.415
DHEAS [μg/dL]	70.78 (40.92–118.77)	41.51 (20.69–74.14)	76.67 (47.36–122.4)	**< 0.001**
Fasting blood glucose [mg/dL]	92 (86–104)	95 (86.5–113.5)	92 (85–103)	0.206
Creatinine [mg/dL]	0.76 (0.65–0.88)	0.77 (0.63–0.93)	0.76 (0.65–0.88)	0.871

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Table 1. *Clinical, radiological, and laboratory data of the study group, mild autonomous cortisol secretion (MACS), and non-functioning adrenal incidentaloma (NFAI) subgroups, and intergroup comparisons*[x]

	Study group (n = 461)	MACS group (n = 77)	NFAI group (n = 384)	p
GFR [mL/min/1.73 m^2]	94 (83–103)	93 (74.5–104)	95 (84–103)	0.155
AST [U/L]	19 (15–23)	18 (14–23)	19 (15–24)	0.190
ALT [U/L]	22 (17–29)	20 (15–28.5)	22 (17–30)	0.071
ALP [U/L]	80 (65–99)	80 (64–103)	80 (65–99)	0.761
GGT [U/L]	21 (16–29)	21 (15–27)	22 (16–29)	0.683
Albumin [g/L]	46 (44–47)	45 (42–47)	46 (44–47)	**0.008**
Calcium [mg/dL]	9.5 (9.2–9.9)	9.5 (9.2–9.76)	9.5 (9.2–9.9)	0.463
Potassium [mmol/L]	4.4 (4.2–4.6)	4.3 (4.13–4.5)	4.4 (4.2–4.6)	0.312
Total cholesterol [mg/dL]	194 (170.5–218)	189 (161–216)	195 (172.25–219)	0.237
LDL [mg/dL]	117 (96–136)	110 (88.5–132.5)	117 (96–137)	0.270
HDL [mg/dL]	46 (40–55)	47 (41–53.5)	46 (40–55.75)	0.818
Triglyceride [mg/dL]	135 (100.25–179.75)	132 (101.5–187)	136 (99–179)	0.850
HbA_{1c} (%)	6 (5.68–6.6)	6.3 (5.8–7)	5.9 (5.6–6.6)	**0.010**
25-hydroxy vitamin D [ng/mL]	18 (11.78–25)	19.45 (10.23–28.58)	17.6 (12–25)	0.765
Haemoglobin [g/dL]	13.84 ± 1.58	13.64 ± 1.69	13.88 ± 1.55	0.218
Platelets [10^9/L]	270.05 ± 66.91	273.5 ± 58.31	269.36 ± 68.56	0.623
Follow-up [years]	3 (3–5)	4 (3–7)	3 (2–5)	**0.004**
Follow-up continuity, n (%)	173 (37.5)	32 (41.6)	141 (36.7)	0.423

*Results are expressed as mean ± standard deviation, median (interquartile range) or frequency (%). Significant P values are in bold. DST — dexamethasone suppression test; ACTH — adrenocorticotropic hormone; DHEAS — dehydroepiandrosterone sulphate; GFR — glomerular filtration rate; AST — aspartate aminotransferase; ALT — alanine aminotransferase; ALP — alkaline phosphatase; GGT — gamma-glutamyl transferase; LDL — low-density lipoprotein; HDL — high-density lipoprotein; HbA1c — glycated haemoglobin

participants, constituting 67% (n = 309) of the sample. MACS was diagnosed in 77 patients (16.7%), whereas NFAI was present in 384 patients (83.3%). HT emerged as the predominant comorbidity, affecting 53.6% (n = 247) of the cohort, followed by DM, which was reported in 31.2% (n=144) of the cases. Intergroup analyses revealed no significant differences in age, gender, and heart failure incidence (p = 0.051, p = 0.244, and p = 0.264, respectively). Conversely, DM, HT, HL, and CAD were significantly more prevalent in the MACS subgroup compared to the NFAI subgroup (p = 0.016, p = 0.003, p = 0.022, and p = 0.005, respectively). The majority of adrenal masses were unilateral, accounting for 77.2% (n = 356) of cases. The incidence of bilateral adrenal masses was significantly higher in the MACS subgroup at 37.7% (n = 29) compared to 19.8% (n = 76) in the NFAI subgroup (p = 0.001). Furthermore, the median adrenal mass diameter was significantly larger in the MACS subgroup (26 mm, range 20-34 mm) than in the NFAI subgroup (17 mm, range 12–24 mm) (p < 0.001). Post-DST applied at night, complete suppression was observed in the NFAI subgroup, in contrast to the absence of suppression in the MACS subgroup (p < 0.001). Morning cortisol levels post-DST were significantly elevated in the MACS subgroup compared to the NFAI subgroup (2.53 mcg/dL, range 2.09-3.6 mcg/dL *vs*. 1.09 mcg/dL, range 0.8–1.3 mcg/dL, p < 0.001). Laboratory analysis indicated significantly higher levels of ACTH, dehydroepiandrosterone sulphate (DHEAS), and albumin in the NFAI subgroup compared to the MACS subgroup (p < 0.001, p < 0.001, and p = 0.008, respectively), whereas haemoglobin A1c (HbA1c) levels were significantly elevated in the MACS subgroup (p = 0.010). No significant differences were observed in other laboratory parameters (p > 0.05 for all parameters). The median follow-up duration for the entire cohort was 3 years (range 3-5 years), with the MACS subgroup experiencing a significantly longer follow-up period compared to the NFAI subgroup (4 years, range 3–7 years *vs*. 3 years, range 2-5 years, p = 0.004). Follow-up compliance did not significantly differ between the subgroups (p = 0.423).

Predictive variables for autonomous cortisol secretion

The outcomes of univariate and multivariable logistic regression analyses, aimed at identifying independent predictive variables for MACS, are summarised in Table 2. The univariate logistic regression analy-

Table 2. *Univariate and multivariable logistic regression analyses of predictors for mild autonomous cortisol secretion (MACS)*

	Univariate analysis			p	Multivariable analysis			p
		95% CI				95% CI		
	OR	Lower	Upper		OR	Lower	Upper	
Age, years	1.025	1	1.051	0.051	0.993	0.956	1.030	0.697
Gender, female	1.381	0.801	2.379	0.245	–	–	–	–
Diabetes mellitus	1.845	1.116	3.048	**0.017**	1.312	0.638	2.700	0.460
Hypertension	2.163	1.283	3.645	**0.004**	1.505	0.754	3.005	0.246
Hyperlipidaemia	1.794	1.083	2.972	**0.023**	1	0.473	2.114	1
Coronary artery disease	2.460	1.289	4.696	**0.006**	1.400	0.594	3.296	0.442
Heart failure	2.533	0.456	14.082	0.288	–	–	–	–
Mass laterality, bilateral	2.448	1.449	4.139	**0.001**	2.111	1.124	3.966	**0.020**
Mass diameter [mm]	1.102	1.071	1.133	**< 0.001**	1.094	1.059	1.131	**< 0.001**
ACTH [pg/mL]	0.986	0.961	1.012	0.287	–	–	–	–
Cortisol [mcg/dL]	1.011	0.967	1.057	0.624	–	–	–	–
DHEAS [μg/dL]	0.987	0.980	0.993	**< 0.001**	0.992	0.986	0.998	**0.013**
Fasting blood glucose [mg/dL]	1.002	0.996	1.009	0.521	–	–	–	–
Creatinine [mg/dL]	1.522	0.910	2.544	0.109	–	–	–	–
GFR [mL/min/1.73m^2]	0.982	0.969	0.995	**0.006**	0.987	0.969	1.006	0.177
AST [U/L]	0.983	0.950	1.016	0.314	–	–	–	–
ALT [U/L]	0.987	0.966	1.008	0.220	–	–	–	–
ALP [U/L]	1.002	0.993	1.011	0.678	–	–	–	–
GGT [U/L]	1.001	0.992	1.010	0.865	–	–	–	–
Albumin [g/L]	0.879	0.810	0.954	**0.002**	0.937	0.844	1.040	0.221
Calcium [mg/dL]	0.932	0.599	1.451	0.757	–	–	–	–
Potassium [mmol/L]	0.871	0.471	1.610	0.659	–	–	–	–
Total cholesterol [mg/dL]	0.997	0.990	1.003	0.280	–	–	–	–
LDL [mg/dL]	0.996	0.988	1.003	0.285	–	–	–	–
HDL [mg/dL]	0.991	0.972	1.010	0.335	–	–	–	–
Triglyceride [mg/dL]	1	0.997	1.003	0.816	–	–	–	–
HbA1c [%]	1.159	0.936	1.434	0.175	–	–	–	–
25–Hydroxy Vitamin D [ng/mL]	1.005	0.978	1.033	0.696	–	–	–	–
Haemoglobin [g/dL]	0.908	0.779	1.059	0.218	–	–	–	–
Platelets [10^9/L]	1.001	0.997	1.005	0.622	–	–	–	–

Significant P values are in bold. OR — odds ratio; CI — confidence interval; DST — dexamethasone suppression test; ACTH — adrenocorticotropic hormone; DHEAS — dehydroepiandrosterone sulphate; GFR — glomerular filtration rate; AST — aspartate aminotransferase; ALT — alanine aminotransferase; ALP — alkaline phosphatase; GGT — gamma-glutamyl transferase; LDL — low-density lipoprotein; HDL — high-density lipoprotein; HbA_{1c} — glycated haemoglobin

sis highlighted significant associations of DM, HT, HL, CAD, bilateral adrenal mass, adrenal mass diameter, DHEAS, glomerular filtration rate (GFR), and albumin with MACS (p values ranging from < 0.001 to 0.006). Multivariable logistic regression analysis pinpointed bilateral adrenal mass, adrenal mass diameter, and DHEAS levels as independent predictors for MACS (p = 0.020, p < 0.001, and p = 0.013, respectively).

Receiver operating characteristic (ROC) curve analysis for predictive variables

Table 3 displays ROC curve analysis results for continuous variables identified as independent predictors in the multivariable logistic regression analysis. An adrenal mass diameter of ≥ 18.5 mm predicted MACS with 83% sensitivity and 56% specificity (AUC = 0.746, 95% CI: 0.687–0.806, p < 0.001) (Fig. 2). Similarly, DHEAS levels ≤ 49.31 μg/dL predicted MACS with 61% sensitiv-

Table 3. *Receiver operating characteristic (ROC) curve analyses of continuous variables with statistically significant p-values in multivariable logistic regression analyses in predicting mild autonomous cortisol secretion (MACS)*

	AUC	95% CI		p	Cut-off value	Sensitivity	Specificity
		Lower	Upper				
Mass diameter [mm]	0.746	0.687	0.806	**< 0.001**	18.5	0.831	0.560
DHEAS [μg/dL]	0.704	0.636	0.771	**< 0.001**	49.31	0.614	0.725

Significant p-values are in bold. AUC — area under curve; CI — confidence interval; DHEAS — dehydroepiandrosterone sulphate

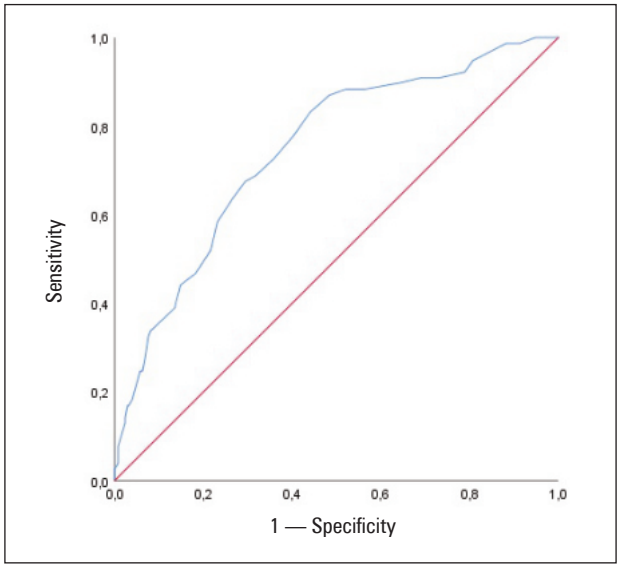

Figure 2. *Receiver operating characteristic (ROC) curves of mass diameter for predicting mild autonomous cortisol secretion (MACS)*

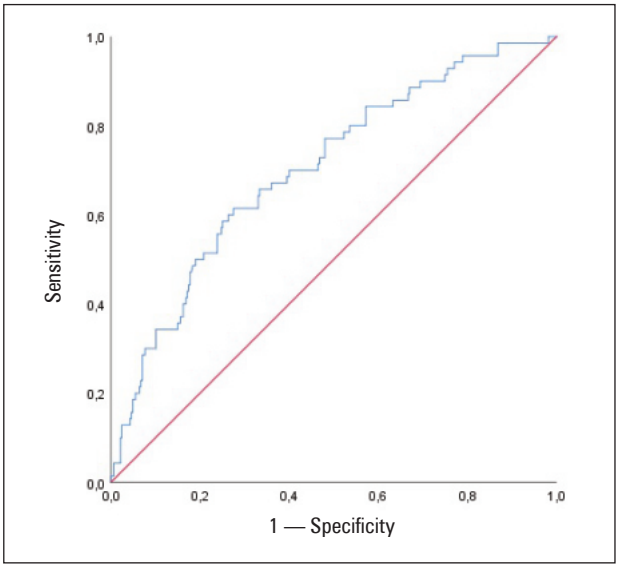

Figure 3. *Receiver operating characteristic (ROC) curves of dehydroepiandrosterone sulphate (DHEAS) for predicting mild autonomous cortisol secretion (MACS)*

ity and 73% specificity (AUC: 0.704, 95% CI: 0.636-0.771, $p < 0.001$) (Fig. 3).

Prediction of comorbidities using cortisol levels post-DST

ROC curve analysis, assessing the predictive capacity of morning cortisol levels post-night DST for DM, HT, HL, and CAD within the MACS subgroup, is presented in Table 4 and Figure 4. The analyses indicated that morning cortisol levels post-DST did not predict these comorbidities ($p > 0.05$ for all diseases).

Discussion

The detection and management of AIs, particularly those exhibiting MACS, have increasingly become a focal point of endocrinological practice. Our study contributes to the understanding of the clinical, radiological, and hormonal profiles of patients with AIs, emphasising the distinct characteristics and comorbidities associated with MACS. The outcomes of our investigation indicate that the incidence of DM, HT, HL, and CAD were found to be statistically significantly elevated in patients diagnosed with MACS in comparison to those in the NFAI cohort, aligning with findings reported in existing literature. Furthermore, logistic regression analyses revealed a statistically significant disparity in the prevalence of DM, HT, HL, and CAD among the cohorts studied. The research offers a comprehensive comparative analysis of clinical, radiological, and laboratory characteristics between patients with MACS and NFAI, elucidating significant distinctions and correlations with various comorbidities and adrenal mass characteristics. The study highlights the prevalence of HT and DM as the most common comorbid conditions within the cohort, with a marked predilection for these conditions in the MACS subgroup. This finding underscores the metabolic impact of cortisol dysregulation, aligning with previous literature that associates cortisol excess with metabolic syndromes. Reflecting on our findings, several key aspects warrant a nuanced discussion within the context of current knowledge and practice in endocrinology.

Table 4. *Receiver operating characteristic (ROC) curve analyses of cortisol 8 a.m. after overnight dexamethasone suppression test (DST) in predicting diabetes mellitus (DM), hypertension (HT), hyperlipidaemia (HL), and coronary artery disease (CAD) in patients with mild autonomous cortisol secretion (MACS)*

	AUC	95% CI		p	Cut-off value	Sensitivity	Specificity
		Lower	Upper				
DM							
Cortisol 8 a.m. after							
Overnight DST [mcg/dL]	0.498	0.368	0.627	0.971	–	–	–
HT							
Cortisol 8 a.m. after							
Overnight DST [mcg/dL]	0.512	0.356	0.668	0.869	–	–	–
HL							
Cortisol 8 a.m. after							
Overnight DST [mcg/dL]	0.601	0.473	0.729	0.133	–	–	–
CAD							
Cortisol 8 a.m. after							
Overnight DST [mcg/dL]	0.599	0.450	0.749	0.223	–	–	–

Significant p-values are in bold. AUC — area under curve; CI — confidence interval

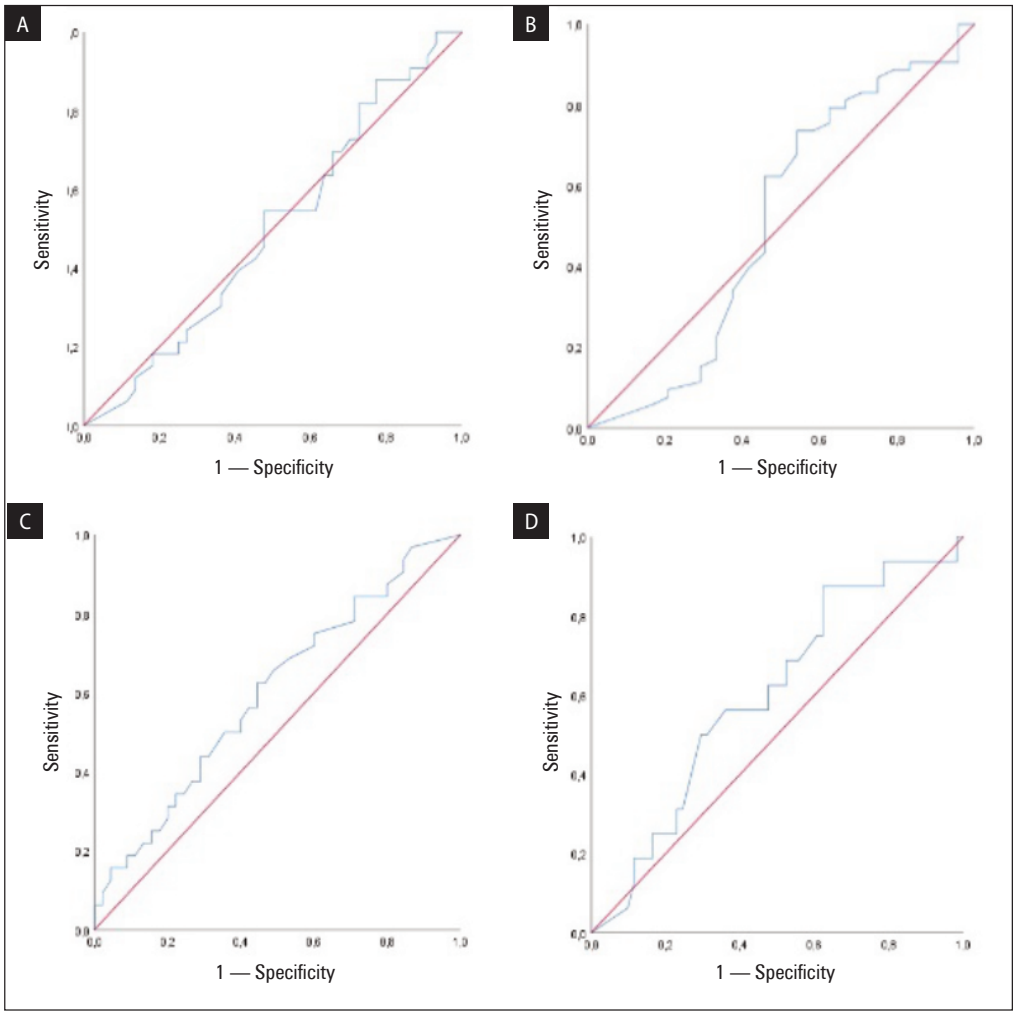

Figure 4. *Receiver operating characteristic (ROC) curves of cortisol 8 a.m. after overnight dexamethasone suppression test (DST) for predicting diabetes mellitus (**A**), hypertension (**B**), hyperlipidaemia (**C**), and coronary artery disease (**D**) in patients with mild autonomous cortisol secretion (MACS)*

Consistent with previous literature, our analysis underscores the prevalence of MACS within the AI population, highlighting its potential for significant clinical implications. Notably, the discernible metabolic and cardiovascular comorbidities associated with MACS align with the existing body of evidence that implicates cortisol in the pathogenesis of metabolic syndrome and its components. In the research conducted by Falcetta et al. [13], a retrospective follow-up of 310 patients diagnosed with adrenal incidentaloma was undertaken, monitoring endocrine function, lesion size progression, routine medical examinations, and comorbidity over an average duration of 31.4 months. The patient cohort was dichotomised into groups: those with non-functioning adrenal incidentaloma and those manifesting autonomous cortisol secretion. Existing literature suggests a correlation between autonomous cortisol secretion and various metabolic disorders, including dyslipidaemia, obesity, hypertension, and hyperglycaemia. However, the findings from this study revealed no significant differences in these metabolic parameters between the 2 groups. The research further highlighted that an increase in lesion size was associated with a rise in autonomous cortisol secretion. Additionally, a study by Emilia Sbardella et al. [14], examining cardiovascular involvement in 71 patients with adrenal incidentaloma, found that patients with potential autonomous cortisol secretion exhibited earlier cardiac and vascular dysfunction than those with non-functional adenomas. Another retrospective analysis by Patrova et al. [15] spanning 13 years and 365 weeks identified a higher prevalence of comorbidities, such as hypertension and osteoporosis, as well as an increased mortality rate among patients with autonomous cortisol secretion compared to their counterparts. The study also delineated age, tumour size, and autonomous cortisol secretion as significant determinants of mortality, with malignancies emerging as the leading cause of death. A recent international retrospective cohort study by Deutschbein et al. [16] investigated the age- and sex-dependent disparities in mortality among patients with adrenal incidentalomas and autonomous cortisol secretion. The study found that autonomous cortisol secretion was associated with increased morbidity and mortality, particularly in women under 65 years of age. Our study findings align with this observation, as we also noted a significantly higher incidence of diabetes mellitus, hypertension, hyperlipidaemia, and coronary artery disease in patients with MACS compared to those with NFAI. This further supports the notion that MACS contributes to the exacerbation of metabolic and cardiovascular comorbidities, necessitating vigilant screening and comprehensive management strategies for affected patients.

In our study, conditions such as DM, HT, HL, and CAD were significantly more common in patients with MACS, suggesting a direct or indirect role of cortisol in the exacerbation of these comorbidities. The study also highlights the anatomical and functional disparities, as shown by the higher incidence of bilateral adrenal masses and larger median adrenal mass diameter in the MACS group. These observations are critical for clinical practice, indicating that the size and bilaterality of adrenal incidentalomas can suggest hormonal activity, particularly mild autonomous cortisol secretion.

The intergroup comparisons between MACS and NFAI subgroups in our study provide critical insights into the differential impact of cortisol hypersecretion. The increased prevalence of HT, DM, HL, and CAD in the MACS subgroup significantly underscores the pathophysiological role of cortisol dysregulation. These findings are pivotal because they highlight the need for comprehensive cardiovascular risk assessment and management in patients with MACS, beyond merely evaluating adrenal mass characteristics [10]. Upon reviewing the existing literature, it becomes evident that patients with MACS exhibit a heightened risk of developing DM, cerebrovascular disease, and CAD compared to those with NFAI [17, 18]. Furthermore, as the incidentaloma size and bilaterality increase while the DHEAS level decreases, the likelihood of an MACS diagnosis rises [19–22]. This correlation is corroborated by the findings of our study, which align with the data presented in the literature.

Our study further delineates the radiological and hormonal profiles characteristic of MACS in AIs, with larger adrenal mass diameter and bilateralism emerging as significant predictors of MACS. The lack of long-term follow-ups in patients and the absence of data on how comorbidities change in patients undergoing adrenalectomy for MACS are limitations in our study. These radiological features and the absence of cortisol suppression post-DST and altered hormonal levels provide a robust framework for identifying MACS. These markers should prompt a thorough endocrinological evaluation to mitigate the risk of associated comorbidities. Identifying bilateral adrenal mass, diameter, and DHEAS levels as independent predictors for MACS in our multivariable analysis offers valuable diagnostic insight. These variables, particularly when considered in conjunction with each other, enhance the predictive accuracy of MACS, thereby facilitating early diagnosis and intervention. The utility of these predictive variables underscores the importance of an integrated diagnostic approach that combines radiological, clinical, and laboratory data.

Our findings emphasise the critical need for a structured diagnostic and management pathway for patients with AIs, particularly those at risk for or presenting with MACS. The association of MACS with significant metabolic and cardiovascular comorbidities necessitates a multidisciplinary approach, involving endocrinologists, radiologists, and cardiologists, to optimise patient outcomes. This collaborative strategy should aim not only at hormonal normalisation but also at the comprehensive management of the associated comorbid conditions. While our study advances the understanding of MACS in AIs, it also highlights the necessity for further research. Prospective studies are needed to elucidate the long-term outcomes of patients with MACS following various therapeutic interventions.

One notable limitation of our study lies in its retrospective nature, which inherently restricts the ability to establish causal relationships between MACS and the observed comorbid conditions. Additionally, the study's dependence on previously collected clinical, radiological, and hormonal data may introduce biases related to the accuracy and completeness of these records. The cohort size, while substantial, may still not fully capture the heterogeneity of the patient population with AIs, potentially limiting the generalisability of our findings. Furthermore, the assessment of comorbid conditions relied on diagnostic codes and medical records, which might not reflect undiagnosed or subclinically managed conditions, thus underestimating the prevalence of certain comorbidities. Despite these limitations, our study presents several strong aspects. One advantage of our study is its extensive patient cohort, providing a comprehensive analysis of the clinical, radiological, and hormonal profiles of patients with AIs, with a particular focus on those exhibiting MACS, contributing valuable insights into the distinct characteristics and comorbidities associated with this condition. The use of logistic regression analyses to explore the prevalence of significant health conditions like DM, HT, HL, and CAD among different cohorts adds a robust statistical foundation to our findings. Moreover, our study reinforces the critical clinical implications of MACS, highlighting its association with a spectrum of metabolic and cardiovascular comorbidities. The identification of bilateral adrenal masses and larger median adrenal mass diameter as indicators of hormonal activity further enriches the diagnostic toolkit available to clinicians. By aligning with and expanding upon existing literature, our research underscores the necessity for vigilant monitoring for MACS in patients with AIs, advocating for a multidisciplinary approach to management that addresses both hormonal imbalances and associated comorbid conditions.

Conclusion

In conclusion, our study reaffirms the complex interplay between MACS and metabolic as well as cardiovascular comorbidities in patients with AIs. The insights gained from this research not only augment the existing knowledge base but also underscore the imperative for vigilant screening, accurate diagnosis, and comprehensive management of MACS in clinical practice.

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Informed consent
Written informed consent was obtained from all patients.

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Conflict of interest
The authors have no conflicts of interests to declare.

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Ethical approval and consent to participate
The study protocol was approved by the Ankara City Hospital Ethics Committee (E1-21-1924). Informed consent for the procedure was obtained from all patients. We confirm that all methods were carried out in accordance with relevant guidelines and regulations. Our study was performed in accordance with the Declaration of Helsinki.

Consent for publication
Informed consent was obtained from all subjects and/or their legal guardian(s) for publication of identifying information/images in an online open-access publication.

Availability of data and materials
The datasets generated and/or analysed during the current study are not publicly available due to privacy and ethical concerns but are available from the corresponding author on reasonable request.

Authors' contributions
B.T.E., B.E.O., C.A., O.T., R.E., and B.C. planned the conception and design of the study. B.T.E., B.E.O., and MS performed the data collection. B.T.E., B.E.O., and M.S. performed the data analyses. All authors interpreted the data. B.T.E. drafted the manuscript. All authors revised and approved the final version of the manuscript to be submitted.

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