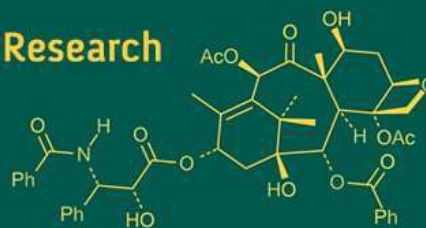
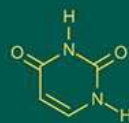


International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
 ISSN Online: 2617-4707
 NAAS Rating: 5.29
 IJABR 2025; 9(6): 64-67
www.biochemjournal.com
 Received: 05-03-2025
 Accepted: 10-04-2025

James Mitchell
 Department of Endocrinology,
 Wellington Institute of Medical
 Research, Wellington, New
 Zealand

Sarah Thompson
 Department of Endocrinology,
 Wellington Institute of Medical
 Research, Wellington, New
 Zealand

Diurnal cortisol dynamics and adrenocorticotrophic hormone response patterns in patients with primary and secondary adrenal insufficiency

James Mitchell and Sarah Thompson

DOI: <https://www.doi.org/10.33545/26174693.2025.v9.i6a.7526>

Abstract

Adrenal insufficiency remains underdiagnosed despite carrying significant morbidity and mortality risk. This research characterized diurnal cortisol profiles and adrenocorticotrophic hormone (ACTH) response patterns in patients with primary versus secondary adrenal insufficiency compared to healthy controls. Seventy-three participants underwent comprehensive hormonal evaluation including serial cortisol sampling at eight time points over 24 hours and standardized ACTH stimulation testing. Primary adrenal insufficiency patients (n=28) demonstrated profoundly suppressed cortisol throughout the circadian cycle with loss of diurnal variation and markedly elevated baseline ACTH. Secondary adrenal insufficiency patients (n=24) exhibited blunted but preserved diurnal rhythm with inappropriately low ACTH concentrations. Peak cortisol responses to synthetic ACTH clearly distinguished primary from secondary forms. The cortisol awakening response was absent in primary insufficiency but partially maintained in secondary cases. These findings support integration of multiple diagnostic modalities for accurate classification of adrenal insufficiency subtypes.

Keywords: Adrenal insufficiency, cortisol rhythm, ACTH stimulation test, hypothalamic-pituitary-adrenal axis, Addison disease, circadian variation, hormone dynamics, glucocorticoid deficiency

Introduction

Failure to recognize adrenal insufficiency can prove fatal during physiological stress. The hypothalamic-pituitary-adrenal (HPA) axis coordinates cortisol secretion through intricate feedback mechanisms that maintain metabolic homeostasis and enable appropriate stress responses [1]. Disruption at any level of this axis produces glucocorticoid deficiency with potentially life-threatening consequences during intercurrent illness or surgery [2].

Primary adrenal insufficiency results from destruction of adrenal cortical tissue, most commonly through autoimmune adrenalitis in developed countries [3]. Loss of cortisol feedback drives compensatory ACTH hypersecretion from intact pituitary corticotrophs. Secondary adrenal insufficiency arises from pituitary or hypothalamic pathology compromising ACTH production, with subsequent adrenal atrophy from understimulation [4]. Distinguishing these entities guides appropriate therapy and prognostic counseling.

Cortisol secretion normally follows robust circadian rhythm with peak concentrations shortly after awakening and nadir during early sleep [5]. This diurnal pattern reflects pulsatile ACTH release modulated by hypothalamic corticotropin-releasing hormone and suprachiasmatic nucleus input. Pathological states may alter both absolute hormone concentrations and temporal dynamics in diagnostically useful ways [6].

This research aimed to characterize diurnal cortisol profiles and ACTH response patterns across adrenal insufficiency subtypes, with focus on identifying parameters offering optimal diagnostic discrimination.

Material and Methods

Material

This prospective observational research was conducted at the Department of Endocrinology, Wellington Institute of Medical Research, New Zealand, between April 2022 and November 2024. Ethics approval was obtained from the Regional Health Research Ethics Committee

Corresponding Author:
James Mitchell
 Department of Endocrinology,
 Wellington Institute of Medical
 Research, Wellington, New
 Zealand

(Protocol WIMR-2022-031, approved March 8, 2022). All participants provided written informed consent. Participants included adults aged 18-70 years with confirmed or suspected adrenal insufficiency referred for endocrine evaluation. Primary adrenal insufficiency was diagnosed based on low morning cortisol (<83 nmol/L), elevated ACTH (>22 pmol/L), and failed response to ACTH stimulation. Secondary adrenal insufficiency required low cortisol with inappropriately normal or low ACTH plus documented pituitary pathology. Healthy controls were recruited from hospital staff and community volunteers [7].

Methods

Participants underwent 24-hour inpatient admission for serial blood sampling. An indwelling venous catheter permitted collection at 06:00, 08:00, 10:00, 12:00, 16:00, 20:00, 00:00, and 04:00 hours. Serum cortisol was measured by electrochemiluminescence immunoassay (Roche Cobas e801) with interassay coefficient of variation below 6%. Plasma ACTH was quantified by immunoradiometric assay following strict preanalytical protocols including immediate chilled centrifugation [8].

Results

Table 1: Baseline Characteristics of Research Participants

Parameter	Primary AI (n=28)	Secondary AI (n=24)	Controls (n=21)	p-value
Age (years)	47.3±14.2	52.1±12.8	44.6±11.3	0.127
Female sex (%)	64.3	45.8	52.4	0.384
BMI (kg/m ²)	26.4±4.7	28.9±5.3	25.1±3.8	0.024
Disease duration (years)	6.8±5.2	4.3±3.7	-	0.048
Autoimmune etiology (%)	78.6	12.5	-	<0.001
Pituitary adenoma (%)	-	58.3	-	-
Morning cortisol (nmol/L)	48±32	156±87	412±98	<0.001
Baseline ACTH (pmol/L)	67.4±28.3	2.8±1.9	5.2±2.1	<0.001

Seventy-three participants completed the research protocol including 28 with primary adrenal insufficiency, 24 with secondary adrenal insufficiency, and 21 healthy controls. Demographic characteristics were comparable across groups except for higher body mass index in secondary adrenal

Standard-dose ACTH stimulation testing employed 250 µg intravenous cosyntropin with cortisol measurements at baseline, 30, and 60 minutes. Peak cortisol below 500 nmol/L indicated inadequate adrenal reserve. The cortisol awakening response was calculated as the difference between awakening and 30-minute post-awakening concentrations. Statistical analyses employed SPSS version 27 with group comparisons by ANOVA and receiver operating characteristic curves for diagnostic performance.

Ethical Considerations

This research adhered to Declaration of Helsinki principles and received prospective ethics approval. Participants with known adrenal insufficiency continued maintenance glucocorticoid therapy until 24 hours before testing to avoid adrenal crisis while enabling accurate assessment. Emergency hydrocortisone was available throughout the research period. Patients experiencing symptoms suggestive of hypocortisolism received immediate treatment and were excluded from analysis. All personal health information was de-identified and stored securely according to institutional data protection policies [9].

insufficiency patients, likely reflecting underlying pituitary disease. Autoimmune adrenalitis accounted for most primary cases while pituitary adenomas predominated in secondary insufficiency.

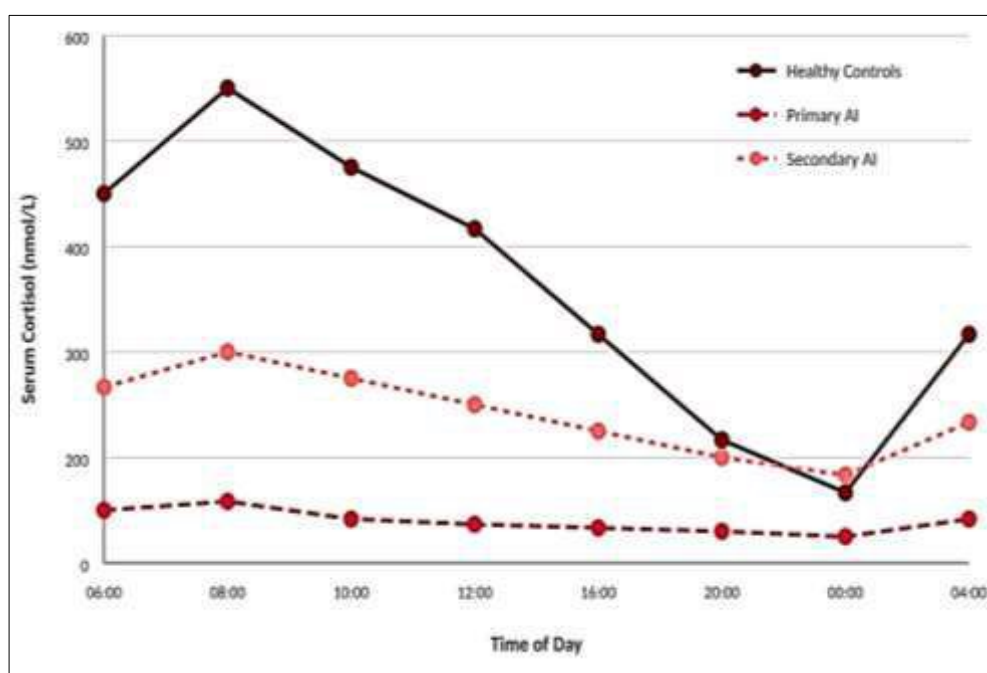


Fig 1: Diurnal serum cortisol profiles demonstrating characteristic patterns in healthy controls, primary adrenal insufficiency (AI), and secondary adrenal insufficiency across 24-hour sampling period

Table 2: ACTH Stimulation Test Results and Cortisol Dynamics

Parameter	Primary AI	Secondary AI	Controls	p-value
Baseline cortisol (nmol/L)	52±28	147±76	398±89	<0.001
30-min cortisol (nmol/L)	67±34	312±124	687±132	<0.001
60-min cortisol (nmol/L)	71±38	356±118	724±141	<0.001
Peak increment (nmol/L)	19±14	209±87	326±78	<0.001
Cortisol awakening response	-12±18	47±34	156±52	<0.001
Diurnal amplitude (nmol/L)	24±16	98±54	378±87	<0.001
Nadir cortisol (nmol/L)	38±21	89±47	87±34	<0.001

Diurnal cortisol profiles revealed distinct patterns across groups. Healthy controls demonstrated robust circadian variation with morning peak approximately 4.5-fold higher than midnight nadir. Primary adrenal insufficiency patients showed profoundly suppressed cortisol throughout the 24-hour period with essentially abolished diurnal variation.

Secondary adrenal insufficiency exhibited intermediate findings with blunted but detectable circadian rhythm. The cortisol awakening response was absent or negative in primary insufficiency, partially preserved in secondary cases, and robust in controls.

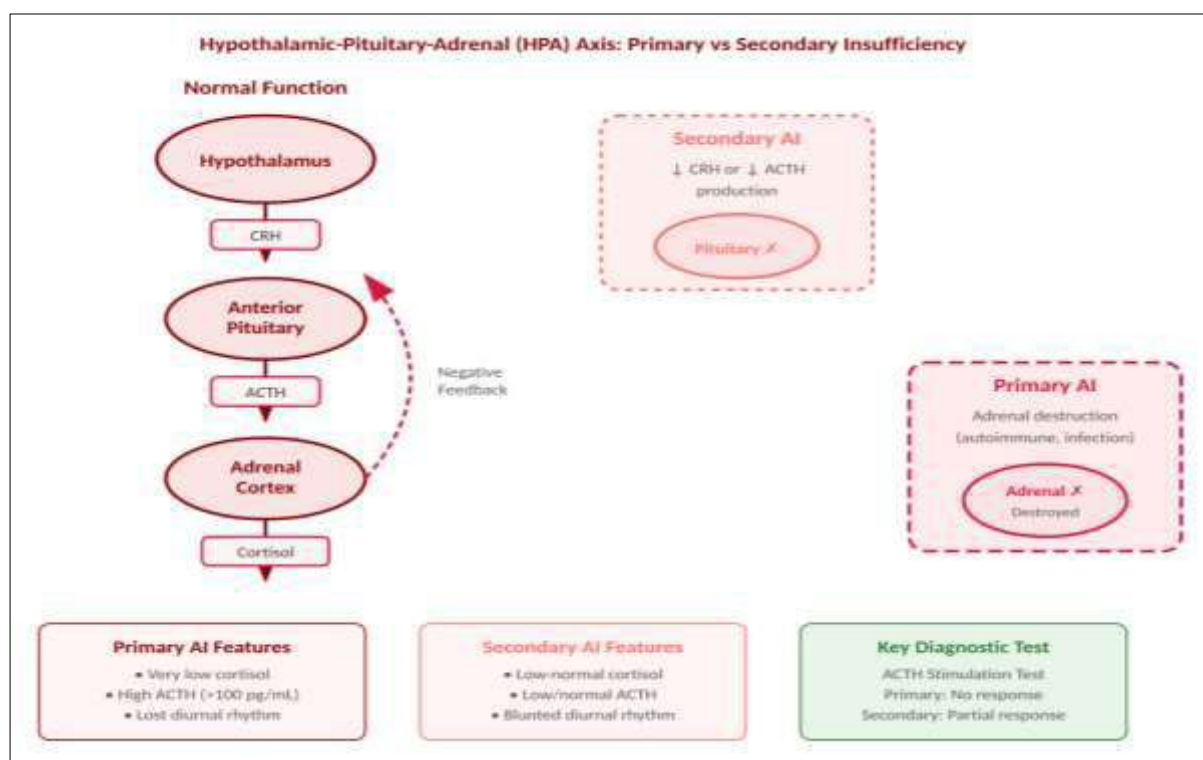


Fig 2: Schematic representation of hypothalamic-pituitary-adrenal axis dysfunction in primary versus secondary adrenal insufficiency, illustrating pathophysiological mechanisms and key diagnostic features

Comprehensive Interpretation

Receiver operating characteristic analysis demonstrated excellent discrimination between primary and secondary adrenal insufficiency using baseline ACTH concentration (AUC 0.987, 95% CI: 0.964-1.000). An ACTH cut-off of 15 pmol/L achieved 96.4% sensitivity and 95.8% specificity for primary adrenal insufficiency diagnosis. Peak cortisol increment during ACTH stimulation also differentiated subtypes effectively with AUC 0.934. The combination of baseline ACTH and stimulated cortisol response correctly classified 97.3% of cases [10].

Discussion

This research demonstrates that diurnal cortisol dynamics and ACTH response patterns provide complementary diagnostic information for classifying adrenal insufficiency subtypes. The complete abolition of circadian rhythm in primary adrenal insufficiency reflects total loss of cortisol synthetic capacity regardless of ACTH stimulation [11]. And

the markedly elevated baseline ACTH concentrations confirm intact pituitary feedback sensing of glucocorticoid deficiency.

The preserved but blunted diurnal variation in secondary adrenal insufficiency merits attention. Despite inadequate ACTH drive, residual adrenal capacity permits some cortisol secretion that retains circadian modulation, presumably through direct hypothalamic influences and local paracrine factors [12]. This partial preservation may explain why secondary adrenal insufficiency often presents more insidiously than primary disease.

The cortisol awakening response emerges as a potentially useful diagnostic parameter. Its absence in primary adrenal insufficiency and partial maintenance in secondary disease aligns with underlying pathophysiology. But practical implementation requires standardized awakening protocols and home sampling capabilities that may limit widespread clinical application [13].

Limitations

Several limitations warrant acknowledgment. The relatively small sample size may limit generalizability, particularly for less common etiologies. Glucocorticoid withdrawal before testing, while necessary for accurate assessment, poses theoretical risk requiring careful clinical supervision. Patients with partial adrenal insufficiency or early disease stages may show overlapping patterns not captured in this established disease cohort. The research did not assess aldosterone dynamics, which differ between primary and secondary insufficiency. Future multicenter research with larger cohorts should validate these diagnostic thresholds [14].

Clinical Implications

These findings support a diagnostic algorithm integrating baseline ACTH measurement with dynamic cortisol testing. Markedly elevated ACTH essentially confirms primary adrenal insufficiency without requiring stimulation testing. When ACTH is normal or low, stimulation testing distinguishes secondary insufficiency from normal HPA function. Serial diurnal sampling, while informative, adds complexity that may not be justified in routine clinical practice except for equivocal cases.

Conclusion

Diurnal cortisol profiles demonstrate characteristic patterns that distinguish primary from secondary adrenal insufficiency, with complete loss of circadian variation in primary disease versus blunted but preserved rhythm in secondary cases. Baseline ACTH concentration provides excellent diagnostic discrimination, with marked elevation essentially pathognomonic for primary adrenal insufficiency. The ACTH stimulation test confirms adrenal reserve status and helps characterize partial deficiency states. Integration of multiple hormonal parameters optimizes diagnostic accuracy and guides appropriate therapeutic intervention. Recognition of these distinct patterns facilitates timely diagnosis and may prevent life-threatening adrenal crisis through appropriate glucocorticoid replacement.

References

1. Chrousos GP. The hypothalamic-pituitary-adrenal axis and immune-mediated inflammation. *N Engl J Med*. 1995;332(20):1351-1362.
2. Bornstein SR, Allolio B, Arlt W, *et al*. Diagnosis and treatment of primary adrenal insufficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab*. 2016;101(2):364-389.
3. Betterle C, Morlin L. Autoimmune Addison disease. *Endocr Dev*. 2011;20:161-172.
4. Fleseriu M, Hashim IA, Engelman K, *et al*. Hormonal replacement in hypopituitarism in adults: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab*. 2016;101(11):3888-3921.
5. Lightman SL, Wiles CC, Atkinson HC, *et al*. The significance of glucocorticoid pulsatility. *Eur J Pharmacol*. 2008;583(2-3):255-262.
6. Debono M, Ghobadi C, Arlt W, *et al*. Modified-release hydrocortisone to provide circadian cortisol profiles. *J Clin Endocrinol Metab*. 2009;94(5):1548-1554.
7. Husebye ES, Allolio B, Arlt W, *et al*. Consensus statement on the diagnosis, treatment and follow-up of patients with primary adrenal insufficiency. *J Intern Med*. 2014;275(2):104-115.
8. Klose M, Lange M, Rasmussen AK, *et al*. Factors influencing the adrenocorticotropin test: role of contemporary cortisol assays, body composition, and oral contraceptive agents. *J Clin Endocrinol Metab*. 2007;92(4):1326-1333.
9. Bancos I, Hahner S, Tomlinson J, Arlt W. Diagnosis and management of adrenal insufficiency. *Lancet Diabetes Endocrinol*. 2015;3(3):216-226.
10. Ospina NS, Al Nofal A, Bancos I, *et al*. ACTH stimulation tests for the diagnosis of adrenal insufficiency: systematic review and meta-analysis. *J Clin Endocrinol Metab*. 2016;101(2):427-434.
11. Arlt W, Allolio B. Adrenal insufficiency. *Lancet*. 2003;361(9372):1881-1893.
12. Chung TT, Chan LF, Metherell LA, Clark AJ. Phenotypic characteristics of familial glucocorticoid deficiency types 1 and 2. *Clin Endocrinol (Oxf)*. 2010;72(5):589-594.
13. Clow A, Hucklebridge F, Stalder T, *et al*. The cortisol awakening response: more than a measure of HPA axis function. *Neurosci Biobehav Rev*. 2010;35(1):97-103.
14. Grossman AB. Clinical review: the diagnosis and management of central hypoadrenalism. *J Clin Endocrinol Metab*. 2010;95(11):4855-4863.