Method-specific serum cortisol responses to the adrenocorticotropin test: comparison of two generations of Roche automated immunoassays using *monoclonal* versus *polyclonal* antibodies

***Klose M*[1], *Hilsted L*[2] *and Feldt-Rasmussen U*[1]

Department of Medical Endocrinology[1] and Clinical Biochemistry[2], Copenhagen University Hospital, Rigshospitalet

OBJECTIVES

The plasma cortisol response to the adrenocorticotropin (ACTH) test is known to vary significantly by assays. An automated cortisol immunoassays with increased specificity due to the shift from polyclonal to monoclonal antibodies, and standardized against mass spectrometry was recently introduced, with an expected decrease in cortisol concentrations by 20%. Cut-offs used in clinical practice for patient evaluation will thus have to be adjusted.

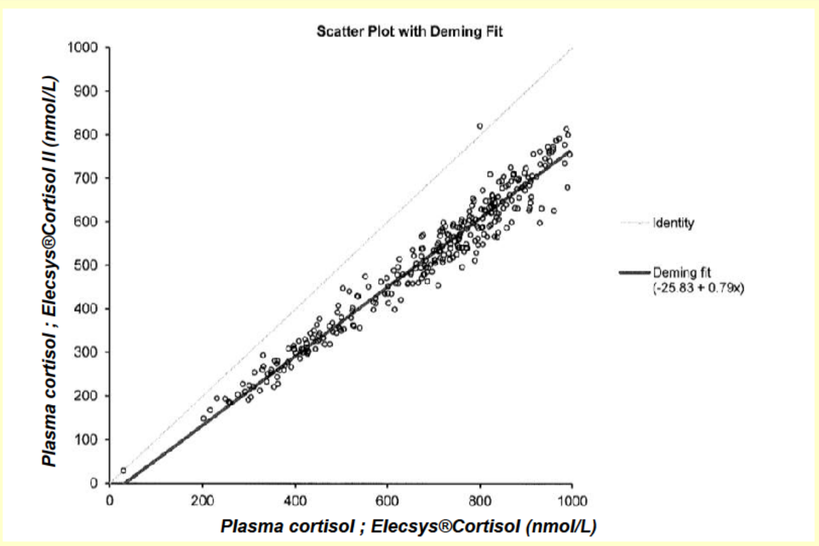
We aimed to assess the normal cortisol response to ACTH stimulation measured by Elecsys®Cortisol II.

METHODS

An ACTH test (250 µg iv $ACTH_{1–24}$) was undertaken in 100 healthy volunteers (age, 18–70 years; 50 women) and 13 women on oral contraceptives[1].

Plasma cortisol was measured by two commercially available immunoassays: Elecsys®Cortisol (polyclonal Ab) and Elecsys®Cortisol II (monoclonal Ab)(Roche).

The estimated lower reference limit for the 30 min plasma cortisol response to ACTH was derived from the adjusted 2.5th percentile, defined as: *2.5th percentile-1.96*SE.*



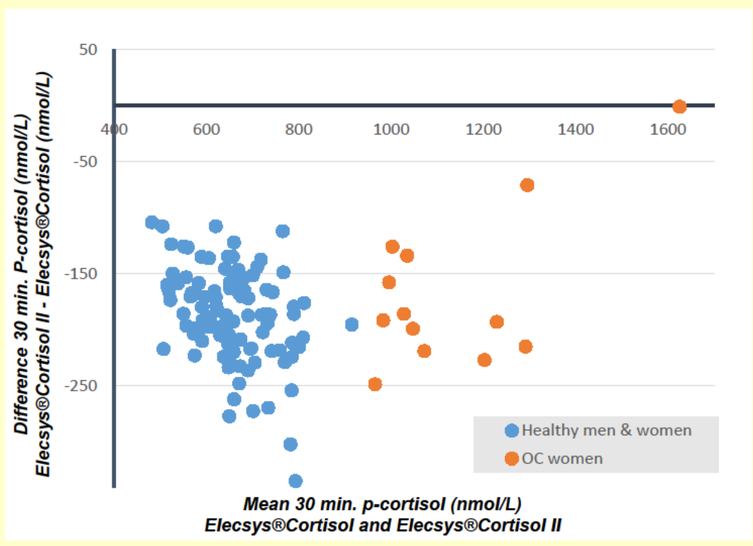


Table 1. 30 min p-cortisol in response to the 250 microg ACTH test

	Elecsys®Cortisol nmol/L	Elecsys®Cortisol II nmol/L
Min	534	399
STDV	94	79
SE	9.44	7.89
5th percentile	602	450
2.5th percentile	590	434
Adj. 2.5th percentile*	572	420

**2.5th percentile - 1.96*SE*

RESULTS

Cortisol concentrations measured by Elecsys®Cortisol II were approximately 20% lower relative to Elecsys®Cortisol. The 30 min cortisol response was normally distributed in males but not in females, with no significant gender difference at baseline nor post-ACTH (p = 0.1). The cortisol concentrations including the lower reference limit for the 30 min cortisol response was method-specific (Table 1); Elecsys®Cortisol (range: 534–1013; adj. 2.5th percentile: 572 nmol/L) and Elecsys®Cortisol II (range: 399-817; adj. 2.5th percentile: 420 nmol/L), and remained significantly elevated by both methods in women on oral contraceptives.

CONCLUSIONS

The cutoff defining a normal 30 min. cortisol response to the ACTH test is influenced significantly by assay employed as well as oestrogen treatment. New cutoffs should be introduced with the introduction of new generation immunoassays with higher specificity.

Elecsys®Cortisol II derived a cutoff of 420 nmol/L, which is significantly lower than the commonly recommended cutoff of 500 nmol/L, but similar to the cutoff previously suggested for mass spectrometry[2].

References

[1] *Klose M et al. J Clin Endocrinol Metab. 2007; 92(4):1326*
Factors influencing the adrenocortico-tropin test: role of contemporary cortisol assays, body composition, and oral contraceptive agents.

[2] *El-Farhan N et al, Clin Endocrinol (Oxf). 2013; 78(5):673*
Method-specific serum cortisol responses to the adrenocorticotrophin test: comparison of gas chromatography-mass spectrometry and five automated immunoassays.