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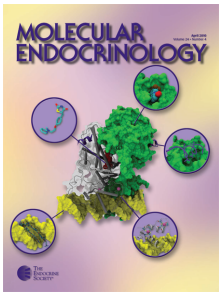

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Normal Cortisol Response on Low-Dose Synacthen (1 μg) Test in Children with Prader Willi Syndrome

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Introduction: It has been postulated that central adrenal insufficiency (CAI), resulting from hypothalamic dysfunction, may contribute to the increased unexplained death rates in Prader Willi syndrome (PWS). A study using the overnight metyrapone test reported a 60% prevalence of CAI in children with PWS. We used a low-dose Synacthen test to screen for CAI in children with PWS.

Methods: We studied 41 children with genetic diagnosis of PWS [20 males; mean age, 7.68 (±5.23) yr] in five pediatric endocrinology centers in Australasia. All participants were randomly selected, and none had a history of Addisonian crisis. Ten of the cohort were receiving sex hormone therapy, 19 were receiving GH, and four were receiving T_4. Their mean body mass index z-score was +1.48 (±1.68). Baseline morning ACTH and cortisol levels were measured, followed by iv administration of 1 μg Synacthen. Post-Synacthen cortisol levels were measured at 30 min, and a cortisol level above 500 nmol/liter was considered normal.

Results: The mean baseline ACTH and cortisol were 15 (±14) ng/liter and 223 (±116) nmol/liter, respectively. The mean 30-min plasma cortisol was 690 (±114) nmol/liter, and the average increase from baseline was 201%.

Conclusions: Our result suggests that CAI is rare in children with PWS. (***J Clin Endocrinol Metab*** **95: E464–E467, 2010)**

Prader Willi syndrome (PWS) is a chromosomal disorder characterized by hypotonia, obesity, short stature, hypogonadotropic hypogonadism, behavior problems, and sleep-related breathing disorders (1, 2). PWS is commonly due to paternal deletion or uniparental disomy of chromosome 15q11–13 (3–5), whereas a small percentage of PWS is due to imprinting center defect (6). Many of the typical features of PWS can be explained by hypothalamic dysfunction, and this hypothesis is supported by functional imaging studies (7–10).

There have been reports of unexplained deaths in patients with PWS (11–13). Most deaths were thought to be related to a combination of GH therapy, disordered breathing, and respiratory infections. Central adrenal insufficiency (CAI) due to hypothalamic dysfunction is a plausible alternative explanation that may have caused or

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Abbreviations: CAI, Central adrenal insufficiency; HPA, hypothalamo-pituitary-adrenal; IIH, insulin-induced hypoglycemia; LDSST, low-dose Synacthen test; PWS, Prader Willi syndrome; SST, standard Synacthen test.

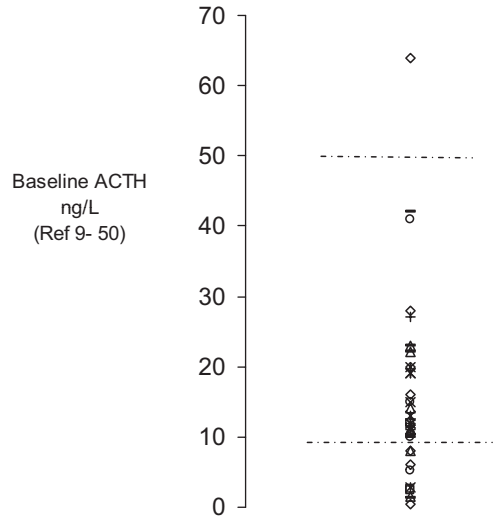

FIG. 1. Baseline plasma ACTH levels of the cohort (normal reference, 9 to 50 ng/liter). (To convert ACTH to picomoles per liter, multiply the value in nanograms per liter by 0.2222.)

contributed to the unexplained deaths. A study reported a 60% prevalence of CAI among children with PWS based on the result of an overnight metyrapone test (14). An earlier mortality report of eight children and two adults with PWS, who had unexplained death, demonstrated less than normal adrenal weight on postmortem examination, suggesting adrenal atrophy probably secondary to CAI (15).

Because of the recent evidence and its potential fatal complications, we screened for CAI in children with PWS in Australasia. Rather than using an overnight metyrapone test, we have used a low-dose Synacthen test (LDSST) (1 μg Synacthen test) to screen for CAI. LDSST examines the stress response of the adrenal cortex to a low dose of synthetic ACTH. The adrenal cortex requires sufficient endogenous ACTH to maintain its ability to mount a stress response during Synacthen tests. LDSST is considered to be a robust, sensitive, and practical screening test for assessment of ACTH deficiency (16–18). In two studies, LDSST was found to be equally sensitive, if not more sensitive than the standard Synacthen test (SST; 250 μg Synacthen) and insulin-induced hypoglycemia (IIH) (16, 17). Abdu *et al.* (16) studied 64 children with proven or suspected pituitary disease and compared the cortisol responses on LDSST and SST to IIH, and LDSST to SST. Using IIH as a standard and the cortisol cutoff level of 500 nmol/liter, the sensitivity of LDSST was 100%, and the specificity was 93.3%. The sensitivity and specificity of SST against IIH were 100 and 90%, respectively. The authors concluded that LDSST can replace SST or IIH for assessment of the hypothalamo-pituitary-adrenal (HPA) axis in patients with ACTH deficiency (16). Weintrob *et al.* (17) compared the cortisol responses on IIH, SST, and LDSST in children with pituitary disease with or without impaired HPA axis, as well as cortisol responses on SST and LDSST in the control group. In the study, both SST and LDSST were found to be equivalent to IIH in screening for integrity of the HPA axis. Furthermore, in a meta-analysis, LDSST was found to be superior to SST in diagnosing chronic CAI (18). In the study, cortisol area under the curve in LDSST was larger than that of SST.

Subjects and Methods

Our study was undertaken because of the publication that suggested high prevalence of CAI in PWS and its potential fatal complication (14). Forty-one children from five pediatric endocrinology units in Australia and New Zealand were included in this study. All children were genetically confirmed to have PWS and were randomly selected. The LDSST was performed in the morning on children who were inpatients for the day of testing. All children had rest for not less than 30 min while waiting for cutaneous anesthetic topical cream to be effective before venipuncture. An indwelling iv catheter was inserted, and the baseline blood sample for ACTH and cortisol was taken. Then 0.1 ml of Synacthen (250 μg in 1 ml) was drawn and added to 24.9 ml of normal saline. Special care and attention were paid to ensure that only 0.1 ml of Synacthen was drawn from the vial. One milliliter of the diluted solution (1 μg of Synacthen) was administered iv, followed by a 5-ml flush of normal saline. The blood samples for cortisol were collected at 30 min after Synacthen, and a serum cortisol level above 500 nmol/liter was considered a normal response (16). The serum cortisol cutoff level of 500 nmol/liter was shown to have 100% sensitivity and 93.3% specificity against IIH (16). The cutoff level, although arbitrary, is based on previous studies and is widely accepted.

ACTH and cortisol were measured by the respective laboratories at the different centers. ACTH was measured by either Immulite 1000 or Immulite 2000 (Siemens, Los Angeles, CA). Cortisol was measured by the Access (Beckman Coulter, Fullerton CA) and Architect (Abbott Diagnostics Division, Lisnamuck, Longford, Ireland) in Queensland; Immulite 1000 and Immulite 2000

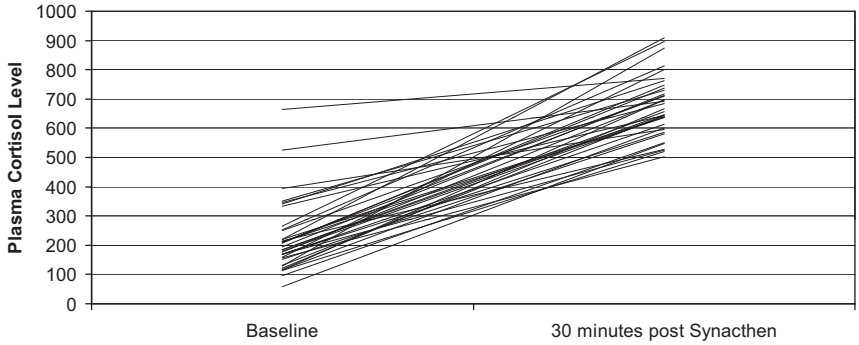

FIG. 2. The baseline plasma cortisol level and 30 min post-Synacthen plasma cortisol levels in children with PWS. (To convert cortisol to micrograms per deciliter, divide the value in nanomoles per liter by 27.5862.)

(Siemens) in New South Wales; and E170 (Roche, Basel, Switzerland) in Auckland. Results from the RCPA Quality Assurance Program (RCPA Quality Assurance Program Pty. Limited, Chemical Pathology QAP Group, Endocrine Program Cycle 32, 6 July to 23 November 2009) do not show significant biases between these cortisol methods.

Results

The mean age was 7.68 (±5.23) yr for the 41 patients, and 20 of them were male. The mean body mass index z-score was +1.48 (±1.68), and 10 of the patients were in early puberty induced by sex hormone replacement therapy. Nineteen of them were on GH, and four were on T_4 at the time of testing. None had a past history of adrenal crisis, and none were treated with glucocorticoid replacement. The mean baseline serum ACTH and cortisol for the whole cohort were 15 (±14) ng/liter (normal range, 9–50) (Fig. 1) and 223 (±116) nmol/liter (normal range, 150–700), respectively. At 30 min after Synacthen, the mean serum cortisol level was 690 (±114) nmol/liter, and the average increase of serum cortisol from the baseline was 201% (Fig. 2). All children had serum cortisol higher than 500 nmol/liter at 30 min after Synacthen.

Discussion

Our result, using LDSST, showed that the cortisol response in children with PWS was normal in the entire cohort, indicating that there must have been sufficient ACTH secretion to maintain health of the adrenal cortex. Our finding is a contrast to the recent finding of 60% prevalence of abnormal metyrapone test response in children with PWS (14). In the study, the authors examined the diurnal salivary cortisol levels in children with PWS. The level of salivary cortisol 30 min after wake-up in the CAI group was similar to the non-CAI group, which was consistent with a normal adrenocortical response to the physiological stress of waking up. The possibility is that the definition of an abnormal test in the overnight metyrapone test in the study was not based on changes in blood 11-deoxycortisol but on ACTH level, which is an unstable compound (19). In the study, seven patients had evidence of CAI because 11-deoxycortisol levels were less than 200 nmol/liter. The other possibility is that the cutoff level of ACTH for the diagnosis of CAI is high, resulting in overdiagnosis of CAI. However, unlike LDSST, the overnight metyrapone test assesses the whole HPA axis. It is also possible that some of our patients may have recently had ACTH deficiency but produced enough cortisol to have falsely normal results on LDSST.

The ACTH levels at baseline are variable in our cohort, depending on the time in the morning when the testing was done (Fig. 1). It is also possible that the abnormality lies in the other aspects of control of the HPA axis, such as circadian rhythm disturbance. It is supported by an animal study. Mage-like 2 gene, which falls in the PWS region, and null mice have defective circadian rhythm as well as symptoms similar to PWS (20).

Conclusion

Our study showed normal cortisol response to stress by using LDSST in all of the randomly selected children with PWS. Our result contradicts the 60% prevalence of CAI in children with PWS, and further testing is needed to evaluate the difference in prevalence—for example, an overnight metyrapone test or insulin-induced hypoglycemia on those who had normal response to LDSST to confirm the prevalence of CAI.

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Disclosure Summary: The authors have nothing to disclose.

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