ORIGINAL RESEARCH ARTICLE

ELSEVIER

Pharmacodynamic Effects of Depot-Medroxyprogesterone Acetate (DMPA) Administered to Lactating Women on Their Male Infants

P. Virutamasen,* S. Leepipatpaiboon,* R. Kriengsinyot,* P. Vichaidith,* P. Ndavi Muia,† C.B. Sekadde-Kigondu,† J.K.G. Mati,† M.G. Forest,‡ L.D. Dikkeschei,§ B.G. Wolthers,§ and C. d'Arcangues#

Normal postpartum women, who had a spontaneous vaginal delivery of one full-term male infant, free of congenital abnormalities and other diseases, were recruited for this study. Thirteen women received 150 mg depot-medroxyprogesterone acetate (DMPA), intramuscularly on days 42 + 1 and 126 + 1 postpartum. Infants of nine mothers, who did not receive DMPA, served as controls. Blood samples were collected from treated mothers on days 44, 47, 74, 124, 128, and 130 postpartum for medroxyprogesterone acetate (MPA) measurements. Four-hour urine collections were obtained from all 22 infants in the morning on days 38, 40, 42, 44, 46, 53, 60, 67, 74, 88, 102, 116, 122, 124, 126, 128, 130, and 137. Urinary follicle stimulating hormone (FSH), luteinizing hormone (LH), unconjugated testosterone, and unconjugated cortisol were measured by radioimmunoassay, and serum MPA and urinary MPA metabolites were measured by gas chromatography-mass spectrometry (GC-MS). No MPA metabolites could be detected in the urine of the infants from the DMPA-receiving mothers. Hormonal profiles in the urine samples were not suppressed in comparison with those of the control infants. The present study demonstrates that DMPA, administered to the mother, does not influence the hormonal regulation of the breast-fed normal male infant. CONTRACEPTION 1996;54: 153–157

KEY WORDS: pharmacodynamics, depot-medroxyprogesterone acetate, DMPA, lactating, male infants

Introduction

Injectable Depo-Provera (depot-medroxyprogesterone acetate, DMPA), a microcrystalline suspension, has proven to be an effective hormone in pregnancy prevention by depressing pituitary gonadotrophic hormone release.[1] It has been used for contraceptive purposes in more than 90 countries.[2] In many countries, injectable contraceptives are often used to supplement the contraceptive effect of lactation. DMPA does not have any deleterious effects on the quantity of breast milk.[3] Medroxyprogesterone acetate (MPA) and its metabolites can be detected in breast milk. The MPA concentration was 400 pg/mL after eight weeks and was still measurable 12 weeks after administration.[3–6] Therefore, it is conceivable that DMPA administered to the mother could influence the hormonal regulation of the breast-fed infant. For this reason, we considered it worthwhile to undertake a controlled study on the possible effects of DMPA on urinary follicle stimulating hormone (FSH), luteinizing hormone (LH), unconjugated testosterone, and unconjugated cortisol patterns of normal male infants.

Materials and Methods

Normal postpartum women who had spontaneously delivered a healthy male infant were recruited for this study in two different centers, namely, the Department of Obstetrics and Gynecology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand, and the Child Welfare Clinic in Machakos, Kenya. The mothers who entered the study had previously delivered at least one healthy infant and breast-fed

*Department of Obstetrics and Gynecology, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand; †Department of Obstetrics and Gynecology, University of Nairobi, College of Health Sciences, Nairobi, Kenya; ‡INSERM U.329, Hospital Debrousse, Lyon, France; §Central Laboratory for Clinical Chemistry, University Hospital, Groningen, The Netherlands; and #Special Programme of Research in Human Reproduction, World Health Organization, Geneva, Switzerland

Name and address for correspondence: Dr. Pramuan Virutamasen, WHO CCR in Human Reproduction, Department of OB & GYN, Chulalongkorn Hospital, Medical School, Rama IV Rd., Bangkok, 10330, Thailand. Fax: 66 2 254 9292

Submitted for publication July 7, 1995
Revised June 7, 1996
Accepted for publication June 7, 1996

655 Avenue of the Americas, New York, NY 10010

ISSN 0010-7824/96/$15.00
PII S0010-7824(96)00170-9

successfully for more than three months. All women in this study agreed to breast-feed their newborn infants for a six-month period. The clinical characteristics of the infants are shown in Table 1. Thirteen postpartum women (six from Bangkok and seven from Machakos) received 150 mg of DMPA intramuscularly on days 42 + 1 and 126 + 1. The effects of DMPA on the hormonal regulation in the children of these mothers were studied. Nine other infants (six from Bangkok and three from Machakos), whose mothers did not receive DMPA, served as controls.

Blood samples were collected from DMPA-receiving mothers on day 44, 46, 74, 124, 128, and 130 postpartum for MPA measurement. Serum was separated and kept frozen until analysis.

Morning urine was collected from all infants during a 4-h period at the age of 38, 40, 42, 44, 46, 53, 60, 67, 74, 88, 102, 116, 122, 124, 126, 128, 130, and 137 days. It is unknown whether the bladder was empty before starting the urine collection. However, it has been documented that a timed urine collection is representative of a 24-h urine.[7] Urine was thoroughly mixed and total volume was measured. Two 5-ml aliquots of urine were immediately submitted to acetone precipitation and the extract containing the gonadotrophins frozen until analysis. Another 10-ml aliquot of urine was immediately frozen for the determination of unconjugated testosterone and unconjugated cortisol. Likewise, a 5-ml portion of the urine was frozen for MPA metabolite measurements. Urinary LH, FSH, unconjugated testosterone, and unconjugated cortisol were determined at the laboratory Pathologic Hormonale Moleculaire of the Hospital Debrousse, Lyon, France. Serum MPA and urinary MPA metabolites were determined at the Central Laboratory for Clinical Chemistry, University Hospital, Groningen, The Netherlands.

LH and FSH measurements

LH and FSH were measured in the acetone precipitates of the urine samples by a previously described RIA method.[8] In brief, two duplicate 5-ml urine samples were acidified to pH 4–5 with glacial acetic acid and then extracted with 10 ml acetone. The mixtures were kept overnight at 4°C and centrifuged. The pellet containing FSH and LH was allowed to dry at room temperature and the residue dissolved in 1.5 ml phosphate buffer (0.05 M, pH 7.5). The solution containing FSH and LH was kept frozen at -20°C until analysis. Finally, LH and FSH were measured by commercially available methods (IRE, Saclay, France). The interassay coefficient of variation including the extraction step was <7.7% and <10% for LH and FSH, respectively.

Testosterone measurements

Urinary unconjugated testosterone was measured by a previously described RIA procedure.[9,10] Mean recoveries were 76% (range 66–95%). The detection limit was 5 pg/tube. Intra-assay and interassay coefficients of variation were <5% and <9%, respectively.

Cortisol measurements

Extracted urinary unconjugated cortisol was measured by RIA using ^{125}I-cortisol as tracer according to

Table 1. Demographic characteristics of infants

Bangkok					
		Case (n = 6)		**Control (n = 6)**	
		Mean	**SD**	**Mean**	**SD**
Infant's weight (g)	0 day*	3158	330	3055	141
	38 days	4063	571	4267	486
	60 days	4980	538	5040	531
	88 days	5828	612	5886	659
	122 days	6543	738	6390	781
	137 days	6795	768	6592	804
Machakos					
		Case (n = 7)		**Control (n = 3)**	
		Mean	**SD**	**Mean**	**SD**
Infant's weight (g)	0 day*	—	—	4900	0
	38 days	4465	513	5100	10
	60 days	4925	531	5600	0
	88 days	5225	311	6405	405
	122 days	5875	507	—	—
	137 days	6300	0	—	—

a previously described procedure.[9,10] The SB-CORT kit was obtained from International-CIS, France. The detection limit was 625 pg/tube. Intra-assay and interassay coefficients of variation were <5% and <10%, respectively.

Creatinine measurements
Urinary creatinine was determined by an automated chemistry method utilizing the Jaffe reaction.[11]

MPA metabolite measurements in urine
In order to detect MPA metabolites in urine, a GC-MS procedure was applied. A 1-ml sample of urine from a woman who used DMPA as a contraceptive was processed according to a method described for urinary steroid profiling.[12,13] In short, after hydrolysis, steroids were extracted and derivatized. Samples were analyzed by gas chromatography. In the gas chromatographic profiles, four peaks could be assigned to MPA metabolites by means of mass spectrometric identification. The mass spectrum of the major metabolite, 6α-methyl-3 ,17α-dehydroxy-5 -pregnan-20-one-17-acetate, showed significant mass fragments at m/z 269 and 359. Using this method, urine samples from infants of the DMPA-receiving mothers were screened by single ion monitoring of both mass fragments.

MPA measurements in serum
MPA in serum was measured after extraction and derivatization by means of GC-MS, as described previously.[14] Interassay and intra-assay coefficients of variation were 5.0 and 5.5%, respectively.

Data analysis
The data were analyzed using unpaired t-test.

Results

Mean urinary creatinine values of the infants of DMPA-receiving mothers and control infants were comparable (Figure 1). Creatinine values tend to rise with the age of the infants. This has been reported for older children previously.[15] As can be seen in Figure 2, unconjugated cortisol levels showed large intra-individual differences, but were not suppressed after maternal DMPA injection. In both groups, cortisol levels remained constant during the period of investigation, but showed marked day-to-day variations. Marked day-to-day variations and intra-individual differences were likewise observed for unconjugated testosterone, LH, and FSH (Figures 3, 4, and 5, respectively). Nevertheless, it is obvious that basal levels of these three hormones show a remarkable parallel decline within each group of infants. The decline appears somewhat steeper in the controls compared to the treatment group, but this was not significant (unpaired t-test, $p < 0.05$). In any case, the data indicate that the function of the hypothalamo-pituitary-gonadal axis was not impaired in the treatment group. Since this decline is observed in both groups, it must be independent of DMPA. No differences were found in LH, FSH, and unconjugated testosterone levels between infants from DMPA-treated mothers and control infants.

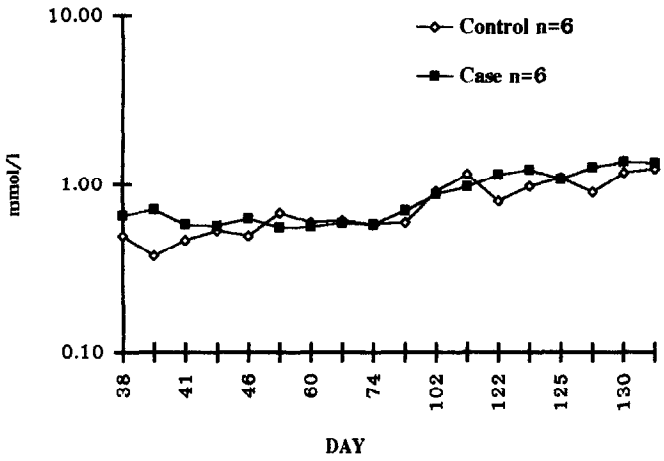

Figure 1. Mean urinary creatinine of male infants.

Maternal serum MPA concentrations rose sharply after administration of the drug, and gradually dropped to levels below 4 ng/ml (Figure 6). MPA metabolites were not detected in any of the urine samples of the infants from these mothers.

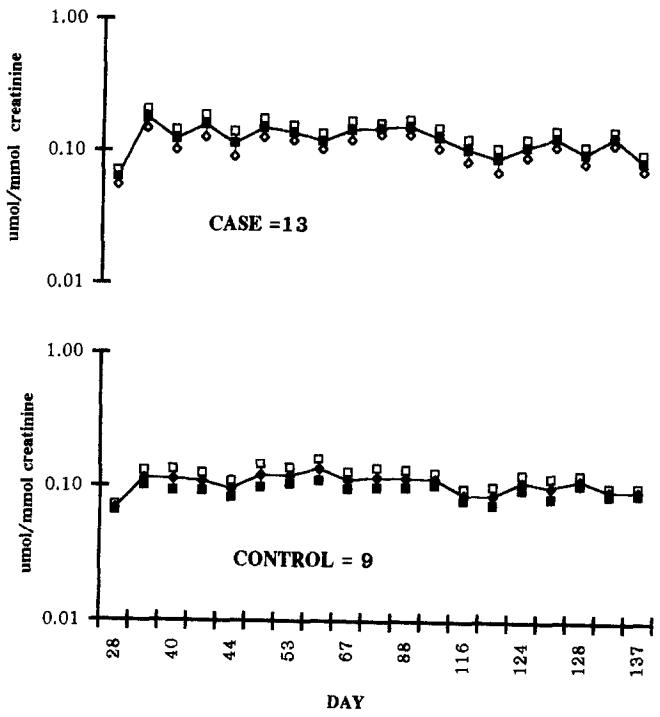

Figure 2. Mean ± SE urinary cortisol of male infants.

Contraception
1996;54:153–157

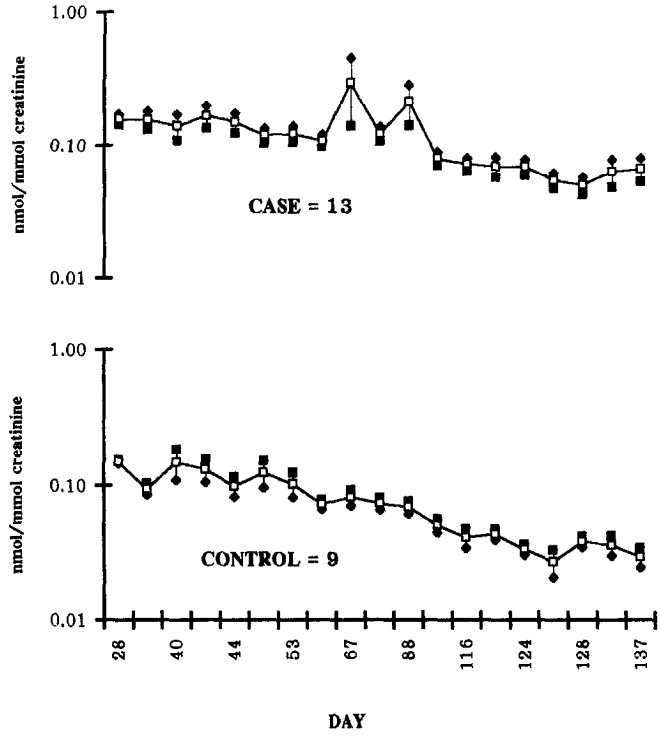

Figure 3. Mean ± SE urinary testosterone of male infants.

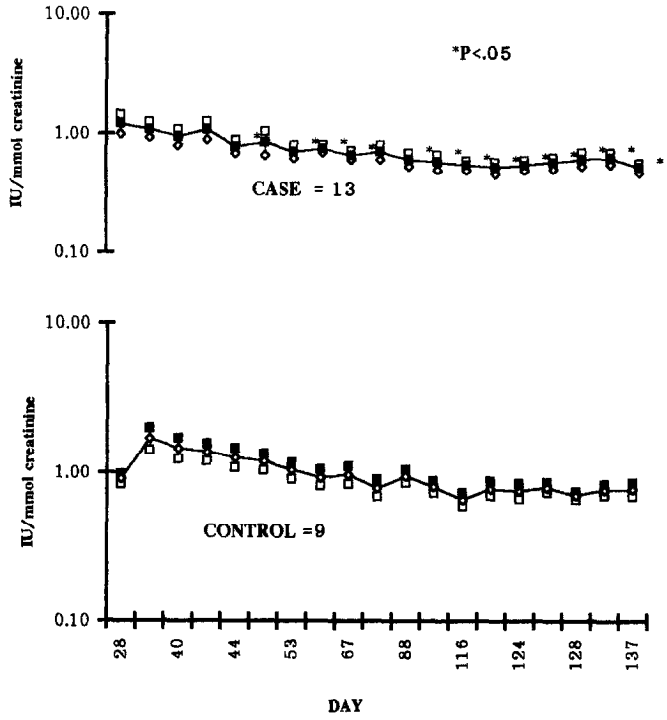

Figure 5. Mean ± SE urinary FSH of male infants.

Discussion

Breast-fed infants may be exposed to the suppressive effect of DMPA given to the lactating mother as a contraceptive. MPA is known to suppress the hypothalamic-pituitary-gonadal axis and the hypothalamic-pituitary-adrenal axis.[16–18] Since the sensitivity of the infant's pituitary gland for MPA is unknown, the present study was initiated to measure MPA transfer from the mother to the infant and possible effects of this transfer on the infant's pituitary-gonadal and pituitary-adrenal axis. It has been estimated that following a single dose of MPA, the hormone transferred over a period of three months is about 0.5% of the maternal dose. Since the highest concentrations of MPA are reached immediately after maternal injection of this hormone, most is transferred within the first 28 days.[5–7] The total dose received by the infants appears to be directly proportional to the dose administered to the mother.[5]

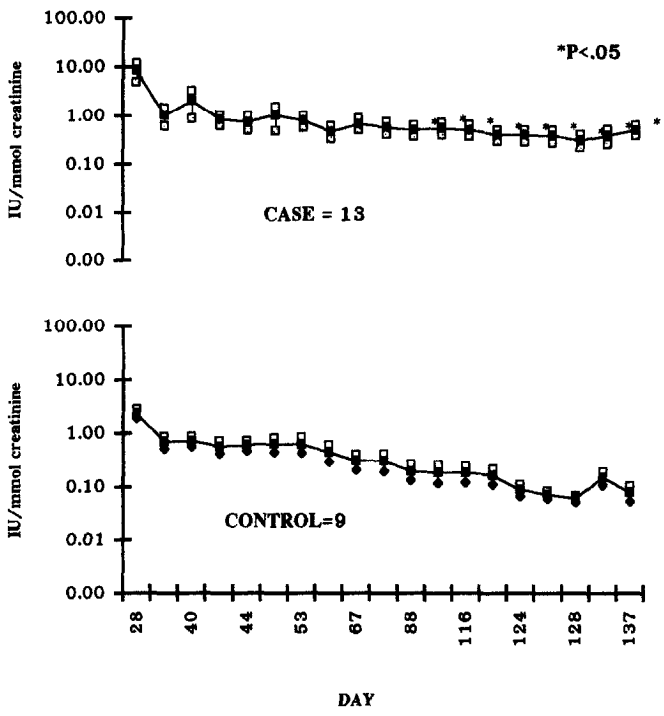

Figure 4. Mean ± SE urinary LH of male infants.

The present study showed a sharp rise of maternal serum MPA concentrations immediately after its administration, which gradually declined to levels below 4 ng/ml. No MPA metabolites could be measured in the urine of the infants of the mothers receiving DMPA.

Therefore, it can be concluded that only small amounts of MPA are transferred to the infants and that these amounts are not expected to have any influence, since effective dosages are known to be much higher.[16,17] This is confirmed by the observation that LH, FSH, unconjugated testosterone, and cortisol are not measurably suppressed by DMPA administered to the mother. It should be remembered, however, that the drug is administered every three months so that the second or third injection will be given at a time

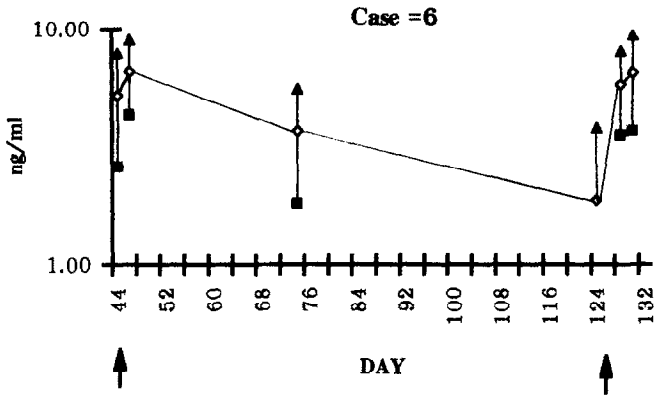

Figure 6. Mean ± SD of serum MPA of DMPA-treated mothers. ↑ = DMPA injection

when the activity of the hypothalamic-pituitary-testicular axis is naturally declining.

It is, therefore, difficult to detect small differences possibly induced by MPA. In conclusion, the present study shows that DMPA can be used as a long-term contraceptive by lactating women without suppressing the adrenal and gonadal function of the breast-fed infant.

Acknowledgment

This investigation received financial support from the Special Programme of Research, Development and Research Training in Human Reproduction of the World Health Organization.

References

1. Ortiz A, Hiroi M, Stanczyk FZ, et al. Serum medroxyprogesterone acetate (MPA) concentrations and ovarian function following intramuscular injection of depo-MPA. J Clin Endocrinol Metab 1977;44:32–8.
2. Population Crisis Committee on Injectable Contraceptives. Safe, effective but neglected population policy. Information Kit No. 7, November 1992.
3. Hefnawi F, Hassanyn S, Abdel-Kader M, Risk A, Hegazi E. Effect of DMPA on lactation. In: Zatuchni G,Goldsmith A, Shelton J, Sciarra J, eds. Long-Acting Contraceptive Delivery Systems. Philadelphia:Harper & Row, 1984:388–96.
4. Saxena B, Shrimanker K, Grudzinskas J. Levels of contraceptive steroids in breast milk and plasma of lactating women. Contraception 1977;16:605–13.
5. Koetsawang S, Nukulkarn P, Fotherby K, Shrimanker K, Mangalam M, Towobola K. Transfer of contraceptive steroids in milk of women using long-acting gestagens. Contraception 1982;25:321–31.
6. Fotherby K, Koetsawang S. Metabolism of injectable formulations of contraceptive steroids in obese and thin women. Contraception 1982;26:51–8.
7. Kulin H, Santner S. Timed urinary gonadotrophin measurements in normal infants, children, adults and on patients with disorders of sexual maturation. J Pediatr 1977;90:760–5.
8. Morel Y, La Selve H, Chatelain P, et al. Interet du dosage des gonadotrophines urinaires en endocrinologie pediatrique. Arch Fr Pediatr 1985;42:579–85.
9. Forest M, de Peretti E, Lecoq A, Cadillon E, Zabot M, Thoulon J. Concentration of 14 steroid hormones in human amniotic fluid of mid pregnancy. J Clin Endocrinol Metab 1980;51:816–22.
10. Forest M, Cathiard A, Bertrand J. Total and unbound testosterone levels in the newborn and in normal and hypogonadal children; use of a sensitive radioimmunoassay for testosterone. J Clin Endocrinol Metab 1973; 36:1132–42.
11. Chasson A, Grady H, Stanby M. Determination of creatinine by means of automated chemical analysis. Am J Clin Path 1961;35:83–8.
12. Kraan G, Wolthers B, van der Molen J, et al. Newly identified 15-beta-hydroxylated 21-deoxy-pregnanes in congenital adrenal hyperplasia due to 21-hydroxylase deficiency. J Steroid Biochem Mol Biol 1993;45:421–34.
13. Wolthers B, deVries I, Volmer M, Nagel G. Detection of 3-beta-hydroxy steroid dehydrogenase deficiency in a newborn by means of urinary steroid analyses. Clin Chim Acta 1987;169,109–16.
14. Dikkeschei L, van Veelen H, Nagel G, Willemse P, Wolthers B. Specific and sensitive determination of medroxyprogesterone acetate in human serum by gas chromatography-mass spectrometry. J Chromatog Biomed Appl 1985;345:1–10.
15. Jury D. Serum creatinine concentration in children; normal values for sex and age. N Z Med J 1979;90:453–6.
16. Reiter E, Kaplan S, Conte F, Grumbach M. Responsivity of pituitary gonadotropes to luteinizing hormone-releasing factor in idiopathic precocious puberty, precocious thelarche, precocious adrenarche and in patients treated with medroxyprogesterone acetate. Pediatr Res 1975;9:111–6.
17. Rifkind A, Kulin H, Cargille C, Rayford P, Ross G. Suppression of urinary excretion of luteinizing hormone (LH) and follicle stimulating hormone (FSH) by medroxyprogesterone acetate. J Clin Endocrinol Metab 1969;29:506–13.
18. Mathews J, Abrams C, Morishima A. Pituitary-adrenal function in ten patients receiving medroxyprogesterone acetate for true precocious puberty. J Clin Endocrinol Metab 1970;30:653–8.