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Lack of correlations between plasma concentration of medroxyprogesterone acetate, hypothalamic-pituitary function, and tumour response in patients with advanced breast cancer

David W. Hedley[1], Macdonald Christie[2]*, Robert P. Weatherby[2], and Ian D. Caterson[3]

[1] Ludwig Institute for Cancer Research (Sydney Branch), Blackburn Building, University of Sydney, Sydney, N.S.W. 2006, Australia

[2] Department of Pharmacology, University of Sydney, Sydney, N.S.W. 2006, Australia

[3] Department of Endocrinology, Royal Prince Alfred Hospital, Missenden Road, Camperdown, N.S.W. 2050, Australia

Summary. *Plasma medroxyprogesterone acetate (MPA) concentrations were measured in 61 patients with advanced breast cancer, after 3 weeks' treatment using 200 mg PO 8-hourly, to determine whether the previously reported wide interpatient variations correlated with tumour response or toxicity. Seventeen patients (28%) responded to the drug, and their mean plasma MPA concentration was 97 ng/ml ± 68 SD, compared with 115 ng/ml ± 87 SD for the patients whose disease progressed. Side-effects attributed to MPA were seen in 18 patients, who had a mean drug concentration of 113 ng/ml ± 104 SD. This was not significantly higher than that of the patients who did not experience drug toxicity.*

Because of a suggestion that some of the antitumour activity of the drug could be mediated via an effect on the hypothalamic-pituitary axis, we also measured plasma FSG, LH, and prolactin concentrations after the 3-week treatment with MPA, but found no correlations with either drug concentration or tumour response. These results indicate that with the present treatment schedule the monitoring of plasma MPA concentrations has no role in routine practice and suggest that the inherent sensitivity of the tumour to progesterone is probably the major determinant of response.

Introduction

Recent trials of high-dose (i.e., 500–1,500 mg daily) medroxyprogesterone acetate (MPA) given by IM depot injections have yielded response rates of up to 40% in patients with advanced breast cancer [4, 10]. These are higher than were previously reported with lower doses given PO [3, 18], but because it has now been shown that equivalent plasma concentrations can be achieved using high-dose oral MPA [16, 19], it seems likely that these improved tumour response rates were due more to the high doses used than to the route of administration. We therefore performed a phase-II study of oral high-dose MPA, treating patients with advanced breast cancer [6].

Because the published pharmacokinetic data had shown wide interpatient variation in plasma drug concentrations [2, 16], we measured these after 3 weeks' treatment, at a time when the earlier studies had shown plasma concentration to have reached a plateau. During the course of this study it was suggested that the antitumour activity of MPA may in part be mediated via suppression of the hypothalamic-pituitary axis [1], and we therefore measured plasma FSH, LH, and prolactin concentrations in addition to MPA during the later part of the trial.

* *Present address:* University Department of Medicine, Austin Hospital, Heidelberg, Victoria 3084, Australia

Offprint requests to: D. W. Hedley

Materials and methods

Patients. All patients had measurable advanced breast cancer, and were treated in a single institution as part of a phase-II trial that has been reported in full elsewhere [6]. Treatment comprised MPA (Provera, Upjohn) 200 mg PO 8-hourly. Heparinised blood was obtained from 61 patients 3 weeks from the start of treatment, and the plasma was stored at −70° C. Because of logistic problems and a previously reported long plasma half-life, we did not venesect the patients at a fixed time after the preceding dose of MPA.

The patients' renal and liver functions were assessed by measurement of plasma creatinine, bilirubin, albumin, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, and γ-glutamyl transpeptidase 3 weeks after the start of treatment.

MPA assay. MPA was measured by a radioimmunoassay modified from that of Shrimanker et al. [17]. Volumes of 200–500 µl plasma were equilibrated for 30 min with 1,000 dpm [^{3}H]MPA internal standard (60 Ci/mmol, New England Nuclear, Boston, Mass., dissolved in 1% ethanol), then extracted twice for 30 min with 5 ml redistilled petroleum ether (40–60°). Extracts were evaporated to dryness under a stream of nitrogen and dissolved in 1 ml phosphate-gelatin-methanol buffer (PGM: 0.05 *M* phosphate, 0.05% gelatin, 10% methanol, pH 7.0 at 20° C) by heating for 30 min at 35° C. Of each plasma extract, 400 µl was counted by liquid scintillation spectroscopy and extraction efficiency estimated from comparison with unextracted internal standard added to PGM buffer. It was found to be 69% ± 1% (SEM, n = 140).

Radioimmunoassay tubes contained a 1 : 50,000 dilution of MPA antiserum (prepared in methanol-free PGM buffer, batch 9980-JCC-112, Upjohn Company, Kalamazoo, Mich), 22,000 dpm [^{3}H]MPA, and 10–10,000 pg (10 concentrations) of unlabelled MPA (Upjohn, Rydalmere, N.S.W.) or 20–100 µl plasma extracts in a final volume of 0.9 ml. Following 16–20 h incubation at 4° C, 100 µl ice-cold charcoal suspension (2.5% activated charcoal, 0.25% dextran T-70) was added to each tube. After 10 min the tubes were centrifuged (2,500 *g* 10 min, 4° C), and radioactivity remaining in the supernatant determined. Plots of log C_{MPA} versus logit $B/_{BO}$

Table 1. Pretreatment characteristics of the 61 patients used in the MPA plasma concentration – therapeutic response study

	Response (17 patients)	No response (44 patients)
Median age (years)	63	56
Range (years)	44–80	35–85
Premenopausal	3 (18%)	11 (25%)
Stage III or IV at initial diagnosis	4 (24%)	9 (21%)
Median disease-free survival of remainder (months)	60	14
Range (months)	10–132	4–96
ER +ve	1	9
ER −ve	2	12
ER unknown	14	23
Prior endocrine therapy	10 (59%)	29 (66%)
Endocrine response	5	7
Endocrine non-response	4	20
Adjuvant endocrine therapy	1	2
Dominant sites of disease		
Visceral	6	18
Bone	5	14
Soft tissue	6	12

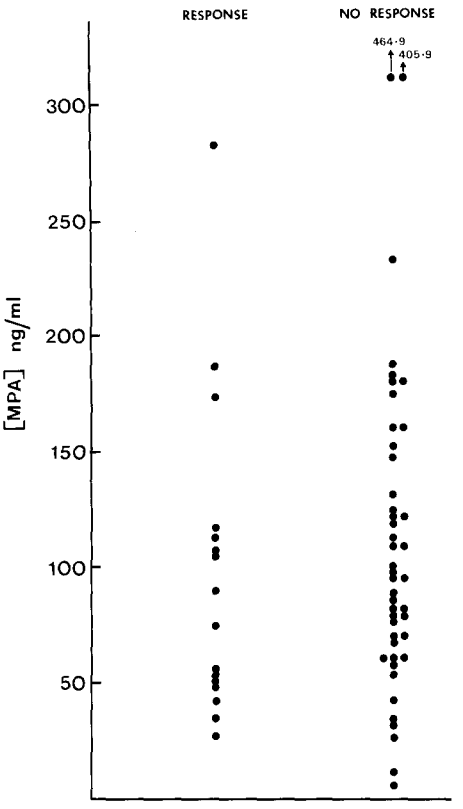

Fig. 1. Plasma MPA concentrations after 3 weeks' treatment with 200 mg t.d.s. PO; responders and nonresponders

were linear in the range 50–5,000 pg per tube and when MPA-free plasma was extracted, 63% ± 4% of [^{3}H]MPA was bound to the antiserum. The plasma extracts were assayed in duplicate and reassayed if the coefficient of variation exceeded 5% and all assay procedures were carried out using silanized glassware.

Hormone assays. All hormones were assayed by double-antibody radioimmunoassay. Antibodies, standards, and reagents for LH and prolactin assays were obtained from Diagnostic Products Corporation, Los Angeles, Calif 90045, USA and those for FSH from Bio-RIA, Montreal, Quebec, Canada. Interassay coefficients of variation were 7.1% for LH, 6.2% for FSH, and 10.1% for prolactin. Relevant reference standards were MRC 68/40 for LH, MRC 69/104 for FSH, and MRC 75/504 for prolactin.

Results

Of the 61 patients included in this study, 17 (28%) had evidence of a partial response which met the criteria proposed by Haywood et al. [5]. Pretreatment characteristics of responders and nonresponders are shown in Table 1. Three-week plasma MPA concentrations showed a wide range (Fig. 1) with a mean of 97 ng/ml ± 68 SD for the patients who showed an objective response and 115 ng/ml ± 87 SD for the patients whose disease progressed. This difference is not statistically significant according to Student's *t*-test. Side-effects of varying severity were attributed to MPA in 18 patients, and are detailed in Table 2. Mean plasma MPA concentration for these patients was 113 ng/ml ± 104 SD and for those not experiencing toxicity, 108 ng/ml ± 74 SD. Again, this difference is not statistically significant.

Tests for liver function were also carried out 3 weeks after commencement of MPA therapy. Twenty-nine patients showed impairment of liver function in one or more of the tests (bilirubin, albumin, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, and γ-GT). However, no significant difference in the mean MPA plasma concentrations was found (no impairment, mean = 105 ng/ml ± 86 SD; impaired liver function, mean = 116 ng/ml ± 84 SD). Individual patients with severe impairment of their liver function exhibited MPA plasma concentrations close to the mean. Renal function, measured by serum creatinine, was found to be normal in all but three patients. These three patients had plasma MPA concentrations within the mean range.

Plasma FSH, LH, and prolactin concentrations were measured in 40 patients, after the 3-week treatment with MPA. Mean plasma FSH concentration was 14.5 IU/l ± 9.2 SD for the patients who responded and 13.7 IU/l ± 10.0 SD for nonresponders. For LH these values were 8.5 IU/l ± 6.4 SD

Table 2. Incidence of side-effects to MPA (found in 18 patients)

Weight gain	6
Muscle cramps	5
Tremor	4
Leg oedema	2
Dyspnoea	2
Muscle aches	1
Insomnia	1
Euphoria	1
PV bleeding	1
Facial swelling	1
Hair loss	1

NB: Some patients had multiple side-effects

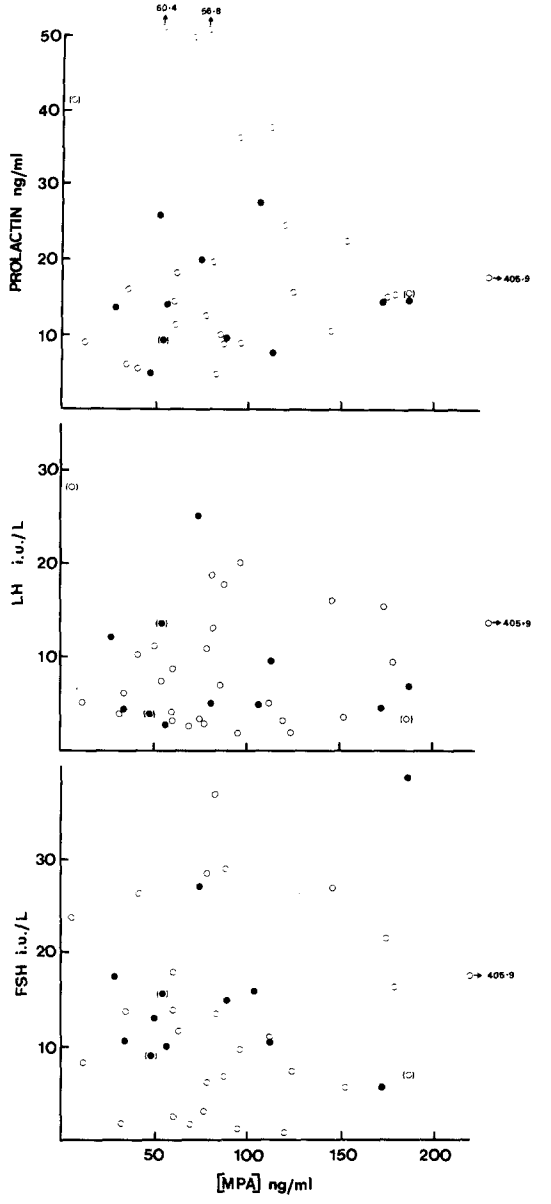

Fig. 2. Comparison of plasma MPA concentration and plasma prolactin, LH, and FSH concentrations after 3 weeks' treatment. *Solid symbols,* patients showing tumour regression; open symbols, patients showing progressive disease. *Symbols* in *parentheses* indicate premenopausal patients

and 8.6 IU/l ± 6.7 SD, respectively, and for prolactin 14.0 ng/ml ± 7.5 SD and 22.1 ng/ml ± 16.7 SD. None of these differences approaches statistical significance according to Student's *t*-test. Figure 2 shows the relationships between MPA and pituitary hormone concentrations for responders and nonresponders. It can be seen that the hormone and MPA concentrations bear no relationship to each other.

Discussion

Early trials of MPA in the treatment of advanced breast cancer used low doses (i.e., 100–400 mg daily) given PO, and yielded response rates of about 10% [3, 18], while the more recent studies have usually been conducted with IM depot injections in the dose range 500–1,500 mg daily and have yielded response rates of 30–40% [4, 9, 10, 13]. Taken together, these studies suggest that tumour response increases with the dose of MPA up to several hundred milligrams per day, beyond which level little further improvement is seen, while side-effects become increasingly common.

Since the development of a sensitive RIA for MPA it has become possible to study plasma drug concentrations using different treatment schedules, and as expected there is an overall correlation between dose given and drug level [2, 16, 19]. With continuous oral administration, plasma concentrations reach a plateau between 4 and 16 days and after cessation of treatment decline in two apparent phases with half-lives of approximately 20 hours and 4 days [1]. Although the build-up and decline of plasma MPA concentrations are much slower with the IM depot preparations [19], equivalent levels can be maintained with either route of administration, suggesting that the improved response rates seen in the more recent trials resulted from the overall higher doses given. Because wide interpatient variations are seen it has been suggested that plasma MPA levels should be monitored during the course of treatment [13], and this was examined in the present phase II trial, in which a fixed dose of MPA, 200 mg PO 8-hourly, was used. Plasma was obtained after 3 weeks' treatment, and although we were unable to venesect the patients at a constant interval between doses, this probably had little effect on the plasma concentrations determined. After 3 weeks, the plasma concentrations have reached a steady state [19], and peak plasma concentrations resulting from oral doses of 200–1,000 mg have been shown to be 1–25 ng/ml [7, 11]. Therefore our doses of 200 mg have a relatively small effect on steady-state levels in the order of 100 ng/ml.

We confirmed a wide interpatient variation in drug levels. The mean plasma concentration of 110 ng/ml is in the same range as that reported using similar dosage levels by IM depot injections [2, 19]. The partial response rate of our patients (28%) is less than that reported in some series [4, 9, 10], but as shown in Table 1 our patients were in general a poor-risk group, and the criteria for objective response were rigidly adhered to. We were unable to demonstrate any difference in the plasma MPA levels between responders and nonresponders. The side-effects and their incidence were similar to those previously reported, but again were not related to the 3-week plasma drug concentration.

Impairment of liver function did not appear to affect the 3-week MPA plasma concentrations. There was no significant difference in the means between patients with impaired liver function and those without impairment. MPA is known to induce liver drug metabolism and enhance protein synthesis [14] and has been used to improve the clinical condition of patients with liver diseases [12]. This suggests that significant induction of drug metabolising enzymes may have occurred in the 3 weeks of MPA administration and that liver damage is not responsible for the variation in the plasma concentrations. Impaired kidney function also does not give rise to the elevated plasma concentrations found in some patients. We cannot define an optimum therapeutic range for MPA, but the present data suggest that it is very wide, and routine monitoring of drug levels in patients treated with MPA does not seem indicated.

MPA suppresses the pituitary secretion of FSH and LH [8, 15], and it has been suggested that this may be responsible for some of its effects on breast cancer growth [1]. The effects of MPA on pituitary function are well established by 3 weeks [15], and we therefore examined FSH, LH, and prolactin concentrations in the 3-week plasma samples of 40 patients. Although

we did not have pretreatment samples, and therefore were unable to document any changes in response to MPA in the present series, there were no correlations between any of the hormone levels and either MPA concentration or tumour response.

As shown in Table 1, the characteristics of the patients whose tumours responded to MPA are similar to those previously reported for other hormonal manipulations, i.e., long disease-free interval and documented response to prior endocrine therapy. It seems probable that the inherent tumour sensitivity to progesterone is the major determinant of response to MPA, that adequate plasma concentrations of the drug were achieved with the present schedule, and that any indirect antitumour effect that may be mediated via the hypothalamic-pituitary axis is minor.

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References

1. Blossey HC, Bartsch HH, Kanne D, Koebberling J, Nagel GA (1982) The pharmacokinetics of high-dose medroxyprogesterone acetate (MPA) in the therapy of advanced breast cancer. Cancer Chemother Pharmacol 8: 77–81
2. Camaggi CM, Strocchi E, Giovannini M, Angelelli B, Costanti B, Zebini E, Ferrari P, Pannuti F (1983) Medroxyprogesterone acetate (MPA) plasma levels after multiple high-dose administration in advanced cancer patients. Cancer Chemother Pharmacol 11: 19–22
3. Cooperative Breast Cancer Study group (1969) Clinical trial of testololactone, medroxyprogesterone acetate and oxylone acetate in advanced female breast cancer. Cancer 23: 109–112
4. DeLena M, Brambilla C, Valagussa P, Bonadonna G (1979) High-dose medroxyprogesterone acetate in breast cancer resistant to endocrine and cytotoxic therapy. Cancer Chemother Pharmacol 2: 175–180
5. Haywood JL, Rubens RD, Carbone PP, Heuson J-C, Kaumaoka S, Segaloff A (1978) Assessment of response to therapy in advanced breast cancer. Eur J Cancer 14: 1291–1292
6. Hedley D, Dalgleish A, Raghavan D, Tattersall M, Coates A, Fox R (1984) Advanced breast cancer: response to high-dose oral medroxyprogesterone acetate. Aust NZ J Med 14: 251–254
7. Hesselius I, Johansson EDB (1981) Medroxyprogesterone acetate (MPA) plasma levels after oral and intramuscular administration in a long-term study. Acta Obstet Gynecol Scand [Suppl] 101: 65–70
8. Izuo M, Iino Y, Tominaga Y, Nomura O, Abe K, Enomoto K, Takatani O, Kubo K (1982) Oral high-dose medroxyprogesterone acetate therapy in advanced breast cancer: clinical and endocrine studies. In: Proceedings of the International Symposium on Medroxyprogesterone Acetate. Excerpta Medica, Amsterdam, pp 250–263
9. Mattson W (1980) A phase III trial of treatment with tamoxifen versus treatment with high dose medroxyprogesterone acetate in advanced postmenopausal breast cancer. In: Iacobelli S, DiMarco A (eds) Role of medroxyprogesterone in endocrine-related tumors. Raven, New York, pp 65–71
10. Pannuti F, Martoni A, Lenaz GR, Piana E, Nanni P (1978) A possible new approach to the treatment of advanced breast cancer: massive doses of medroxyprogesterone acetate. Cancer Treat Rep 62: 499–504
11. Pannuti F, Camaggi CM, Strocchi E, Giovannini M, Di Marco AR, Constanti B (1982) Medroxyprogesterone acetate (MAP) relative bioavailability after single high-dose administration in cancer patients. Cancer Treat Rep 66: 2043–2049
12. Rautio A, Sotaniemi EA, Pelkonen RO, Luoma PV (1980) Treatment of alcoholic cirrhosis with enzyme inducers. Clin Pharmacol Ther 28: 629–647
13. Robustelli Della Cuna G, Bernado-Strada MR, Garzina F (1982) High-dose medroxyprogesterone acetate in metastatic breast cancer. A critical review. In: Proceedings of the International Symposium on Medroxyprogesterone Acetate. Excerpta Medica, Amsterdam, pp 290–305
14. Saarni HV, Stengard J, Karki NT, Sotaniemi EA (1983) Medroxyprogesterone acetate improvement of the hepatic drug-metabolizing enzyme system in rats after chemical liver injury. Biochem Pharmacol 32: 1075–1081
15. Sala G, Iannotta F, Facchinetti A (1982) Endocrinological properties of medroxyprogesterone acetate. In: Proceedings of the International Symposium on Medroxyprogesterone Acetate. Excerpta Medica, Amsterdam, pp 125–138
16. Salimtschik M, Mouridsen H, Loeber J, Johansson E (1980) Comparative pharmacokinetics of medroxyprogesterone acetate administered by oral and intramuscular routes. Cancer Chemother Pharmacol 4: 267–269
17. Shrimanker K, Saxena BN, Fotherby K (1978) A radioimmunoassay for serum medroxyprogesterone acetate. J Steroid Biochem 9: 359–363
18. Stoll BA (1967) Progestin therapy of breast cancer: comparison of agents. Br Med J III: 338–341
19. Tamassia V, Battaglia A, Ganzina F, Isetta AM, Sacchetti G, Cavalli F, Goldhirsch A, Brunner K, Bernardo G, Robustelli Della Cuna G (1982) Pharmacokinetic approach to the selection of dose schedules for medroxyprogesterone acetate in clinical oncology. Cancer Chemother Pharmacol 8: 151–156

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