Cancer Chemother Pharmacol (1985) 14 : 229–231

Cancer Chemotherapy and Pharmacology

Medroxyprogesterone acetate (MAP) and tamoxifen (TMX) plasma levels after simultaneous treatment with 'low' TMX and 'high' MAP doses

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Summary. *Plasma levels of medroxyprogesterone acetate (MAP), tamoxifen (TMX) and its major metabolites, 4-hydroxy TMX and desmethyl TMX, were determined in five patients with advanced breast cancer following simultaneous MAP (2,000 mg/day) and TMX (20 mg/day) oral therapy.*

The interindividual variance in MAP plasma levels was wide; the mean plasma levels of both drugs were nearly identical, despite the large difference in the administered doses.

Introduction

Both medroxyprogesterone acetate (MAP) and tamoxifen (TMX) are currently used in advanced breast cancer management. Objective response rates and survival have been found to be similar for the two drugs in controlled clinical trials if MAP is used in high daily doses [6, 10].

No significant differences in response rates have been established after TMX treatment at different dosages (20–200 mg/day) [7]. MAP is scarcely effective if used in low doses [5, 8, 11]. We report here the plasma levels of the two drugs recorded in a clinical trial where 'high' oral doses (2,000 mg/day) of MAP and 'low' oral doses (20 mg/day) of TMX were simultaneously administered to patients with advanced breast cancer.

Materials and methods

The subjects of this study were five hospital inpatients with advanced mammary cancer resistant to chemotherapy.

All had normal liver and renal functions. MAP (2,000 mg/day) and TMX (20 mg/day) were administered simultaneously for 30 days. The treatment was then discontinued for 1 week to record the plasma decay of both drugs, and was eventually resumed until progession. Blood samples were drawn into heparinized tubes before the treatment and on days 4, 8, 12, 15, 21, 28, 30, 31, 33, 35, and 37 of the therapy.

Plasma samples obtained by centrifugation were submitted to drug and metabolites analysis in our laboratory.

MAP analysis [4, 9]. A known amount of internal standard (17-α-hydroxyprogesterone caproate) was added to plasma samples (1 ml) and extracted twice with cyclohexane (2 × 3 ml) in a vortex mixer for 5 min. After centrifugation, the organic phase was evaporated under vacuum (Buchler Vortex evaporator), and the residue, diluted with acetone : acetonitrile 1 : 1 (500 µl), was treated with heptafluorobutyric anhydride (Carlo Erba, Milan, 50 µl).

The solution was incubated with stirring for 1 h at 30° C, and then evaporated under vacuum. The residue was reconstitued with 1 ml cyclohexane and 1-µl fractions were analyzed by gas chromatography (Varian model 4600 automated instrument equipped with autosampler model 8000, Ni63 Electron Capture detector and Vista 401 data system; 3% SP2250 on 100–120 mesh Supelcoport glass column; injection port temperature: 270° C; detector temperature, 300° C; column oven program: 230–270° C, 2 degoc/min; carrier gas, nitrogen, 30 ml/min.

Retention times (min) were 13.6 (MAP) and 19.6 (internal standard). Calibration curves were obtained by spiking blood bank plasma samples with known amounts of MAP (1–1,000 ng/ml) and 17α-hydroxyprogesterone caproate (100 ng/ml).

Typical accuracy was 5%–8% for MAP plasma levels > 2 ng/ml. Analysis of blank samples gave MAP concentration values of 0–1.3 ng/ml (mean value on 20 samples = 0.3 ng/ml). Tamoxifen is not detected by this analytical method.

Tamoxifen and metabolites analysis. Plasma levels of TMX and its major active metabolites, *N*-desmethyl TMX and 4-hydroxy TMX, were determined by a specific method involving HPLC separation, online photocyclization of drug and metabolites to phenantrenes, and fluorimetric detection [2].

Plasma samples (1 ml) were deproteinized with methanol (1 ml) after the addition of a known amount of internal standard (*N*-acetyl-*N*-desmethyl TMX) and distilled water (1 ml).

After centrifugation, the supernatant was filtered through a Sep-Pak C18 cartridge (Waters Assoc., USA) previously washed with methanol (5 ml) and water (5 ml). The cartridge was then eluted with water (5 ml), water-acetonitrile 1 : 1 (1 ml), and acetonitrile (0.5 ml). These eluates were discarded, and TMX, internal standard, and metabolites were finally eluted from the Sep-Pak cartridge with 5 ml 0.3 *M* phosphoric acid in acetonitrile. The solution was concentrated under vacuum to 500 µl; after addition of 500 µl 10 m*M* KH2PO4, 100 µl of this sample was injected into the chromatographic system (Rheodyne 7105 injector, Perkin-Elmer 3B HPLC pumps, model 650/10LC spectrofluorimeter, and Sigma 10 data system).

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The end of the chromatographic column (Bondapak CN, 10 μm, 30 cm × 3.9 mm ID, Waters Assoc.) was connected to a PTFE capillary tube 0.3 mm ID × 5 m long, wound around a Philips HPK-125W high-pressure mercury lamp at a distance of 10 cm. The outlet was directly connected to the fluorescence detector set at ex = 260 nm (slit 10 nm), em = 375 nm (slit 10 nm).

Chromatographic separation and photocyclization of TMX, metabolites, and internal standard to the corresponding fluorescent phenantrene derivatives were achieved with a mobile phase of acetonitrile-0.3 *M* phosphoric acid-10 m*M* KH2PO4 (190 : 50 : 280) at a flow rate of 1.5 ml/min.

Retention times (min) in our experimental conditions were 7.4 for 4-hydroxy TMX, 11.0 for desmethyl TMX, 11.9 for TMX, and 14.9 for internal standard. Calibration factors were obtained by adding known amounts of TMX to blood bank plasma samples, metabolites, and internal standard. The mean coefficient of variation on multiple determinations was 11.5%; the minimum detectable concentration was 1 ng/ml. MAP is not detectable in these analytical conditions.

Results

The mean plasma levels of TMX, metabolites, and MAP determined in our patient sample are reported in Fig. 1.

After the last administration, the apparent half-lives were 2.7 days for MAP, 6.8 days for TMX, and 17.1 days for desmethyl TMX. The hydroxylated metabolite – 4 hydroxy TMX – was present in low concentration (less than 10 ng/ml). Three other TMX metabolites, of unknown structure but retaining the triphenylethylene moiety, were present in all the patients in low concentration (5–20 ng/ml).

Figure 2 shows the plasma levels of MAP and TMX observed in the individual patients. The intersubject spread of MAP bioavailability is high.

Discussion

Notwithstanding the large difference in the administered doses (100 : 1), the mean plasma levels of MAP and TMX reached during the simultaneous treatment are nearly the same.

Differences in the proportion of drug absorbed, in the half-life of each drug (easily recognizable in the terminal part of the concentration-time course (Fig. 1), and possibly in the volumes of distribution, are responsible for this behavior [13]. In a clinical trial we demonstrated that MAP and TMX give similar response rates in advanced breast cancer therapy if given in a dose ratio of 100 : 1 [10]. The data reported in this work may be an indication of the equipotency of the two drugs – if plasma levels rather than administered doses are compared.

Historical comparison with data obtained in our laboratory shows that the apparent half-life of MAP [1, 9] is not influenced by simultaneous TMX administration. The half-lives of TMX and desmethyl TMX also seem not to be heavily influenced by simultaneous MAP treatment. A slower

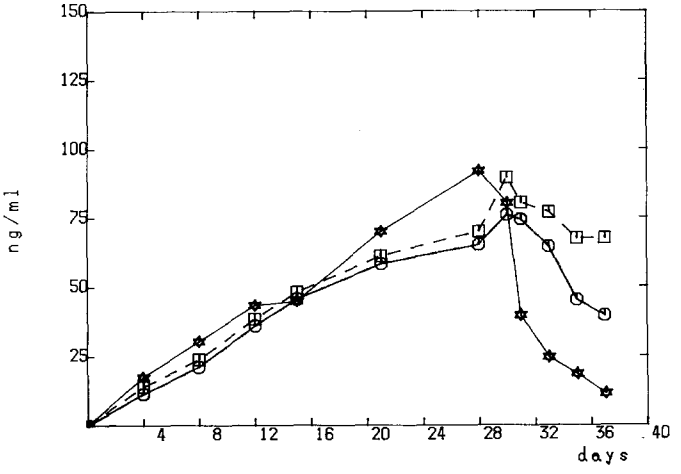

Fig. 1. Mean plasma levels of TMX, *N*-desmethyl TMX and MAP after simultaneous daily oral administration of 20 mg TMX and 2,000 mg MAP, for 30 days. ✿———✿ = MAP; ⊙———⊙ TMX; ⊡———⊡ DSM-TMX

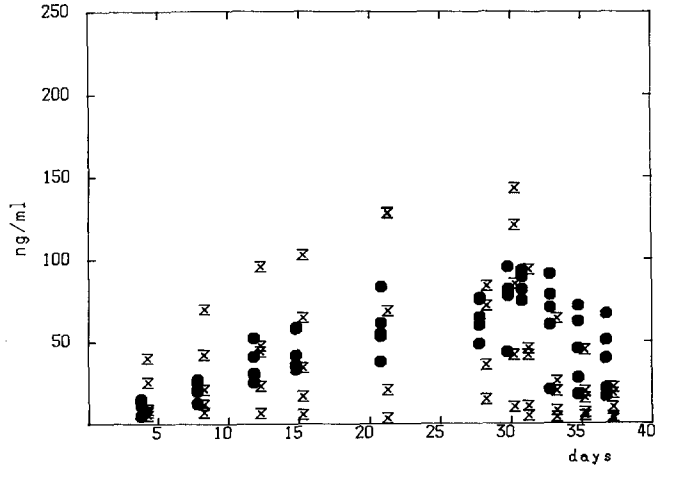

Fig. 2. Plasma levels of TMX and MAP observed in the individual patients. x x x MAP; ● ● ● TMX

increase in MAP concentrations was observed in this study than was recorded during our previous experience with MAP alone [1]. However, the wide intersubject spread in MAP plasma levels observed in both studies prevents the assigning of statistical significance to this difference.

In two studies on TMX pharmacokinetics after daily administration of 40 mg, plasma levels of *N*-desmethyl TMX were found to be consistently higher than those of the parent drug (481 ng/ml vs 310 ng/ml [12] and 462 ng/ml vs 300 ng/ml [3]. In this study, in contrast, TMX and *N*-desmethyl TMX plasma concentrations were nearly identical. MAP is known to induce liver enzymes, and a stimulation of hepatic clearance of desmethyl TMX may therefore not be surprising.

Current trends in the hormonal treatment of mammary cancer are slanted toward sequential rather than simultaneous use of TMX and MAP. Our data clearly indicate that the long half-life of TMX and of its active metabolite desmethyl TMX (6.8 and 17.1 days, respectively) requires a rather long washout period before administration of the second drug is started if a true sequential treatment is wanted. Progesterone receptor priming can probably be achieved better with drugs characterized by shorter biological half-lives.

References

1. Camaggi CM, Strocchi E, Giovannini M, Angelelli B, Costanti B, Zebini E, Ferrarri P, Pannuti F (1983a) Medroxyprogesterone acetate (MAP) plasma levels after multiple high dose administration in advanced cancer patients. Cancer Chemother Pharmacol 11: 19–22
2. Camaggi CM, Strocchi E, Canova N, Pannuti F (1983b) High-performance liquid chromatographic analysis of tamoxifen and major metabolites in human plasma. J Chromatogr 275: 436–442
3. Daniel P, Gaskell SJ, Bishop H, Campbell C, Nicholson RI (1981) Determination of tamoxifen and biologically active metabolites in human breast tumors and plasma. Eur J Cancer Clin Oncol 17: 1183–1189
4. Kaiser DG, Carlson RG, Kirton KT (1974) GLC determination of medroxyprogesterone acetate in plasma. J Pharm Sci 63: 420–424
5. Loeber J, Mouridsen HT, Rose C (1983) Oral or intramuscular treatment of advanced breast cancer with medroxyprogesterone acetate: a review. In: Campio L et al. (eds) Role of medroxyprogesterone in endocrine-related tumors, Vol 2. Raven Press, New York, pp 105–114
6. Mattson W, von Eyben F, Hallsten L, Tennvall L (1983) A trial of tamoxifen versus high-dose medroxyprogesterone acetate in advanced postmenopausal breast cancer. A final report. In: Cavalli F et al. (eds) Proceedings of the International Symposium on Medroxyprogesterone Acetate, Excerpta Medica, Amsterdam, pp 276–284
7. Mouridsen H, Palshof T, Patterson J, Battersby L (1978) Tamoxifen – a review of its efficacy in advanced breast cancer. Cancer Treat Review 5: 131
8. Pannuti F, Martoni A, Camaggi CM, Strocchi E, Di Marco AR, Rossi AP, Tomasi L, Giovannini M, Cricca A, Fruet F, Lelli G, Giambiasi ME, Canova N (1982a) High-dose medroxyprogesterone acetate in oncology. History, clinical use and pharmacokinetics. In: Cavalli F et al. (eds) Proceedings of the International Symposium on Medroxyprogesterone Acetate. Excerpta Medica, Amsterdam, pp 5–43
9. Pannuti F, Camaggi CM, Strocchi E, Giovannini M, Di Marco AR, Costanti B (1982b) Medroxyprogesterone acetate (MAP) relative bioavailability after single high-dose administration in cancer patients. Cancer Treat Rep 66: 2043–2049
10. Pannuti F, Martoni A, Fruet F, Burroni P, Canova N, Hall S (1982c) Oral high dose medroxyprogesterone acetate versus tamoxifen in postmenopausal patients with advanced breast cancer. In: Jacobelli S et al. (eds) The role of tamoxifen in breast cancer. Raven Press, New York, pp 85–92
11. Robustelli Della Cuna, G, Bernardo-Strada MR, Ganzina F (1983) High-dose medroxyprogesterone acetate in metastatic breast cancer. A critical review. In: Cavalli F et al. (eds) Proceeding sof the International Symposium on Medroxyprogesterone acetate. Excerpta Medica, Amsterdam, pp 290–305
12. Strocchi E (1983) Tamoxifen pharmacokinetics after single- and multiple administration in advanced breast cancer patients. 13th International Congress of Chemotherapy, Vienna, Plenary Session 12.8.2, Proceedings part 289
13. Wagner JG (1975) Fundamentals of clinical pharmacokinetics. Drug Intelligence, Hamilton

Received March, 1, 1984/Accepted September 11, 1984