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Effects of Cytochrome P450 Inducers on Tamoxifen Genotoxicity in Female Mice *In Vivo*

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ABSTRACT. We recently reported that administration of the antiestrogen tamoxifen (TAM) gives rise to two groups of DNA adducts in female mouse liver *in vivo*, as measured by ^{32}P-postlabeling, and provided evidence that 4-hydroxytamoxifen and α-hydroxytamoxifen are proximate carcinogenic metabolites leading to group I and group II adducts, respectively (Randerath *et al.*, *Carcinogenesis* **15:** 2087–2094, 1994). Because cytochrome P450 (CYP) enzymes play an important role in TAM metabolism, in this investigation we tested the hypothesis that induction of liver CYP enzymes may affect TAM metabolism profoundly, resulting in increased or decreased TAM–DNA adduct formation *in vivo*. To this end, we treated female ICR mice with TAM either alone or in combination with one of several classic CYP inducers, i.e. phenobarbital (PB), β-naphthoflavone (BNF), and pregnenolone-16α-carbonitrile (PCN), and determined the levels of ^{32}P-postlabeled TAM–DNA adducts and the activities of several CYP-dependent enzymes. Each of the inducers greatly diminished levels of group II, but did not affect group I adducts. TAM elicited induction of benzphetamine *N*-demethylase activity in liver, while activities of other enzymes were not affected. TAM, when given in combination with BNF, elicited a synergistic induction of ethoxyresorufin O-deethylase (EROD) (CYP1A1) and methoxyresorufin O-demethylase (MROD) (CYP1A2) activities. Likewise, PCN given along with TAM caused synergistic induction of EROD and ethylmorphine *N*-demethylase activities. There was no synergism between PB and TAM, however. Overall, the results further support the existence of two pathways of TAM metabolism to DNA-reactive electrophiles and strongly suggest that the classic CYP inducers tested enhance detoxication of TAM to non-genotoxic metabolites. BIOCHEM PHARMACOL 53;5:663–669, 1997. © 1997 Elsevier Science Inc.

KEY WORDS. tamoxifen genotoxicity; mouse liver; DNA adducts; ^{32}P-postlabeling; cytochrome P450 inducers; *in vivo*

The antiestrogenic compound TAM† is widely used to treat breast cancer [1–3]. In addition to its established chemotherapeutic use, it is now being evaluated as a prophylactic agent in women at high risk for breast cancer [4, 5]. Recent studies have shown that women exposed to TAM have a 7.5-fold increased risk of endometrial cancer [6, 7]. TAM is a strong liver carcinogen in rats [8–11], and a high frequency of *p53* mutations is detected in hepatocarcinomas induced by TAM treatment [12]. Although the mechanisms that cause cancer have not been determined yet, several laboratories including our own have reported that administration of TAM results in the formation of DNA adducts in the liver and, to a lesser extent, in other organs of rats, mice, and hamsters [10, 13–19].

Metabolic activation of TAM is a prerequisite for DNA [17, 20–22] or protein [22–25] adduct formation. Several *in vitro* studies using rodent [26–30] or human [26, 30, 31] liver microsomes have shown that TAM undergoes extensive metabolism, major metabolites including 4-OH-TAM, *N*-desmethyl-TAM, TAM-*N*-oxide, and α-OH-TAM. Regarding the conversion of TAM to phase I metabolites, CYP enzymes have been shown to play a major role, being involved in the formation of *N*-desmethyl-TAM and 4-OH-TAM [30, 31]. On the other hand, flavin monooxygenase mediates the formation of TAM-*N*-oxide [32]. While TAM N-demethylation is catalyzed by CYP1A, CYP2C, and CYP3A isozymes in the rat [30] and primarily by CYP3A4 in humans [30, 31], 4-hydroxylation of TAM appears to be catalyzed by constitutive forms of CYP [25, 30].

Much has been learned recently about the chemical species of TAM that are responsible for DNA damage [16–18, 21, 29, 33–35]. Earlier, we showed that i.p. administration

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† *Abbreviations:* BNF, β-naphthoflavone; BZD, benzphetamine *N*-demethylase; CYP, cytochrome P450; EMD, ethylmorphine *N*-demethylase; EROD, ethoxyresorufin O-deethylase; 4-OH-TAM, 4-hydroxytamoxifen; α-OH-TAM, α-hydroxytamoxifen; MROD, methoxyresorufin O-demethylase; PB, phenobarbital; PCN, pregnenolone-16α-carbonitrile; RAL, relative adduct labeling; TAM, tamoxifen; and TO, trioctanoin.

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of TAM to female mice leads to the formation of two groups (I and II) of hepatic DNA adducts and that 4-OH-TAM, the hormonally active form of TAM [36], is a proximate metabolite leading to group I adduct formation [16]. Further, we provided evidence that 4-OH-TAM-quinone methide could be the ultimate carcinogenic metabolite responsible for group I adduct formation [21]. The other proximate carcinogen has been identified as α-OH-TAM, and Osborne *et al.* [35] recently characterized the structure of the major TAM adduct from this compound to be (*E*)-α-(N^2-deoxyguanosyl)tamoxifen. Although α-OH-TAM is capable of directly yielding this adduct [35], the fact that the rate of formation of the adduct is low at neutral pH led to the hypothesis that *in vivo*, α-OH-TAM undergoes further metabolic activation via sulfation or glucuronidation to form highly DNA-reactive esters [35]. In fact, based on our finding [16] that the sulfotransferase inhibitor pentachlorophenol strongly reduces group II adduct formation *in vivo*, we earlier postulated independently an important role for ethyl α-hydroxylation and sulfonation in TAM genotoxicity [16].

Humans are exposed constantly to numerous xenobiotics that may alter profoundly activities of drug-metabolizing enzymes; thus, TAM metabolism in patients undergoing TAM therapy may be subject to modulation causing increased or decreased TAM genotoxicity. The effects of CYP inducers on TAM–DNA adduct formation *in vivo* have not been studied thus far. In the current investigation, we have addressed this question and also determined the possible influence of TAM on the inductive properties of classic CYP inducers.

MATERIALS AND METHODS

Materials

TAM (99.7% purity), PB, BNF, PCN, ethylmorphine, ethoxyresorufin, methoxyresorufin, resorufin, $NADP^+$, glucose-6-phosphate, glucose-6-phosphate dehydrogenase, sucrose, bovine serum albumin, and corn oil were purchased from the Sigma Chemical Co. (St. Louis, MO). Benzphetamine hydrochloride was a gift from the Upjohn Co. (Kalamazoo, MI). Materials for ^{32}P-postlabeling and other chemicals have been reported previously [15, 16, 37].

Animals and Treatment

Female ICR mice (25 g) obtained from Harlan Sprague–Dawley (Houston, TX) were supplied with Purina 5001 diet and tap water *ad lib*. Animals were acclimatized for 1 week before each treatment. Each group consisted of at least 3 animals. With the exception of PB, which was dissolved in saline, all compounds were dissolved in TO. The mice were randomized into 9 groups of 3 or 4 animals each. Each of these groups received the following regimens i.p. once daily between 9:00 and 11:00 a.m. for 4 days: (i) TO (8 μL/g body weight) only as vehicle control; (ii) TAM (120 μmol/kg); (iii) saline (8 μL/g body weight) followed 1 hr later by TAM; (iv) PB (215 μmol/kg) in saline; (v) PB followed 1 hr later by TAM; (vi) BNF (146 μmol/kg); (vii) a mixture of BNF and TAM; (viii) PCN (140 μmol/kg); and (ix) a mixture of PCN and TAM. All animals were killed 24 hr after the last treatment. Livers were dissected and minced, and a 0.5-g portion of each preparation was frozen immediately and stored at −80° until DNA extraction. The remaining material was used without freezing for microsome preparations.

DNA Isolation and ^{32}P-Postlabeling

DNA was isolated by a modified solvent extraction procedure [38], and its purity and concentration were established spectrophotometrically from absorbances at 230, 260, and 280 nm. DNA was analyzed by the dinucleotide/monophosphate version of the ^{32}P-postlabeling assay [37], as modified for TAM adducts [16]. Autoradiography and quantitative analysis followed previously published procedures [15, 16, 39]. Adduct levels were estimated as RAL values [37, 39].

Microsomal Enzymes

Tissue was homogenized in ice-cold 0.25 M sucrose, 0.01 M Tris–HCl buffer, pH 7.4. Microsomes were isolated by a calcium chloride sedimentation method [40]. Liver microsomal CYP concentrations were determined by the method of Omura and Sato [41]. Protein was assayed by the protein-dye binding method of Bradford [42]. EROD activity was determined essentially by the method of Pohl and Fouts [43], as described in previous publications [44, 45]. MROD assay was identical to that of EROD, with ethoxyresorufin being replaced with methoxyresorufin [45, 46]. BZD and EMD activities were determined as described previously [47, 48]. For each of the enzyme assays, pilot experiments were performed to study the effect of protein concentration on enzyme activity, and experiments to study the effect of CYP inducers and TAM on the drug-metabolizing enzymes were performed under conditions wherein the enzyme activity was proportional to the amount of protein.

Statistical Analysis

For TAM–DNA adducts, mean RAL values of adducts for individual tissue preparations from each treatment group were analyzed by one-way ANOVA and Fisher's multiple comparison test. CYP parameters were analyzed for statistically significant differences by Student's unpaired *t*-test, assuming a significance level of $P < 0.05$. The ability of TAM to elicit synergistic induction of CYP enzyme activities in mice when given along with an inducer was tested. For this purpose, the activity of each enzyme in TO controls was subtracted from that in inducer or TAM groups, and the sum of the resulting values was divided by the corresponding inducible activity in animals given a combination of TAM and inducer.

RESULTS

DNA Adducts

Exposure of mice to TAM, but not PB, BNF, or TO gave rise to the formation of 12 chromatographically distinct DNA adducts (Fig. 1). PCN administration did not result in DNA adduct formation (data not shown). We previously reported that i.p. administration of TAM to mice results in the formation of two distinct groups of DNA adducts belonging, respectively, to polar group I (spots 1, 2, 3, and 4) and non-polar group II (spots 5, 6, 8, 9, 10, and 12), with relative amounts of group I and group II adducts strongly depending upon the route of TAM administration [15, 16]. Animals pretreated with PB prior to administration of TAM showed diminished intensities of the major TAM adducts compared with those in animals given TAM only (Fig. 1). Quantitative analysis showed that group I adduct levels were not greatly affected overall by inducer pretreatment, with certain exceptions, as revealed by statistical analysis (Table 1). On the other hand, levels of several group II adducts were depleted significantly in the PB group compared with animals given TAM alone (Table 1), with spots 5, 6, 8, 9, 10, and 12 being reduced by 80.9, 50.6, 56.9, 60.7, 66.8, and 49.2%, respectively. Total adduct levels for group II (Subtotal II) and groups I and II combined (Total) were reduced by 66.7 and 64.4%, respectively.

Similar to animals pretreated with PB + TAM, animals given a combination of BNF and TAM displayed reduced group II adduct levels versus those that received TAM alone (Fig. 1). As illustrated in Table 1, spots 5, 6, 8, 9, 10, and 12 were reduced by 63.4, 52.7, 44.5, 48.3, 61.7, and 63.0%, respectively, versus the TAM samples. Group II (Subtotal II) and groups I plus II (Total) adduct levels were diminished by 57.5 and 54.7%, respectively.

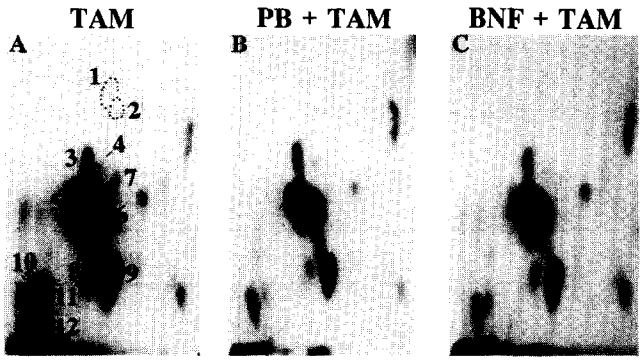

FIG. 1. Representative autoradiograms of ^{32}P-postlabeled liver TAM–DNA adducts from mice exposed to TAM (A), PB + TAM (B), or BNF + TAM (C). Female ICR mice were i.p. administered TAM dissolved in TO either alone or in combination with PB or BNF once daily for 4 days. All animals were killed 24 hr after the last treatment, and their liver DNAs were analyzed for TAM adducts by ^{32}P-postlabeling. For details, consult Materials and Methods. Adducts 7, 11, and an unmarked adduct located to the right of spot 6 were not consistently recovered and, therefore, were excluded from calculations used for Table 1. Several weak unmarked spots were also seen in vehicle controls and in animals given CYP inducers only and thus were not TAM related. Autoradiography was conducted for 16 hr at 23° using Kodak XAR-5 X-ray films with Du Pont Lightning Plus intensifying screens.

Exposure of animals to a mixture of PCN and TAM, also, significantly reduced levels of group II adducts (maps not shown in Fig. 1) compared with the TAM only groups: spots 5, 6, 8, 9, 10, and 12 were lowered by 85.3, 71.3, 46.1, 59.6, 73.9, and 27.9%, respectively. Subtotal II and Total were reduced by 74.3 and 71.1%, respectively (Table 1).

Microsomal Enzymes

Administration of TAM to mice led to a 1.3-fold induction in the liver microsomal activity of BZD (Table 2); however, total CYP levels or activities of other microsomal enzymes were not altered significantly. PB administration elevated total CYP content as well as individual microsomal enzyme activities. The total CYP content was increased 2.8-fold, and the activities of BZD, EROD, MROD, and EMD were enhanced 3.9-, 5.2-, 8.1-, and 1.9-fold, respectively, over controls. Animals pretreated with PB prior to TAM administration exhibited a 3.3-fold increase in BZD activity versus TAM only. A comparison of data from the PB + TAM group with the PB group revealed no significant change in any of the parameters, except that MROD activities were reduced markedly by the combined treatment.

BNF significantly induced EROD and MROD activities compared with vehicle controls, by factors of 10.1 and 17.2, respectively (Table 2). Other CYP parameters were not affected significantly. Animals that were exposed to a combination of BNF and TAM showed significant induction of total CYP content (1.9-fold), EROD activity (15.6-fold), and MROD activity (34.9-fold) over the corresponding values in the TAM only samples (Table 2). Comparison of the BNF + TAM and the BNF only groups showed that the former displayed 2.1- and 1.7-fold increases, respectively, in EROD and MROD activities, respectively, over the latter. Interestingly, as calculated from the data of Table 2, BNF + TAM samples displayed a 2.2- and 1.8-fold synergistic induction, respectively, of EROD and MROD activities versus the added inductive effects of BNF and TAM, i.e. when the chemicals were given separately (Table 2).

PCN administration resulted in marked induction of total CYP content and activities of BZD and EMD. The inducers enhanced total CYP content by a factor of 2.8 and activities of BZD, EROD, and EMD 2.3, 2.0, and 2.5 times, respectively, over vehicle controls (Table 2). In response to a combination of PCN and TAM, total CYP content was increased 3.4-fold, and activities of EROD and EMD were enhanced 3.3- and 2.8-fold, respectively, over the TAM samples. The other enzymes were not affected significantly. When data from the PCN + TAM group were compared against the PCN only group, the EROD and EMD activities in the former displayed 2.3- and 1.2-fold induction, respectively (Table 2), while the other enzyme activities remained unaltered. Furthermore, combined administration of PCN and TAM synergistically induced EROD (2.6-fold)

TABLE 1. Effect of CYP inducers on mouse hepatic TAM–DNA adduct formation*

Adduct	TAM (1)	PB + TAM (2)	BNF + TAM (3)	PCN + TAM (4)	Source†	P‡
1	5.3 ± 0.5	4.3 ± 0.3	4.8 ± 0.8	3.2 ± 0.7	1 vs 4	0.11
2	10.7 ± 0.8	6.0 ± 0.0	7.8 ± 1.5	11.6 ± 2.3	2 vs 1, 4	0.06
3	100.6 ± 12.2	101.7 ± 10.4	131.5 ± 37.1	88.1 ± 18.5		0.56
4	30.6 ± 1.5	21.7 ± 0.3	21.0 ± 3.1	53.6 ± 18.6	4 vs 2, 3	0.07
Subtotal I	147.2 ± 13.1	133.7 ± 9.8	165.1 ± 42.0	156.5 ± 26.2		0.86
5	1563.6 ± 160.4	299.3 ± 97.0	571.5 ± 131.5	230.5 ± 37.5	1 vs 2, 3, 4	0.0002
6	1045.9 ± 85.1	516.3 ± 162.6	494.5 ± 88.1	299.8 ± 53.2	1 vs 2, 3, 4	0.0002
8	97.4 ± 9.8	42.0 ± 10.3	54.0 ± 8.7	52.5 ± 4.8	1 vs 2, 3, 4	0.0003
9	480.0 ± 54.0	188.7 ± 50.6	248.3 ± 39.8	194.1 ± 30.3	1 vs 2, 3, 4	0.0016
10	226.7 ± 29.02	75.3 ± 22.6	86.8 ± 18.1	59.1 ± 9.9	1 vs 2, 3, 4	0.0006
12	84.6 ± 12.8	43.0 ± 9.3	31.3 ± 9.8	61.0 ± 10.6	1 vs 2, 3	0.058
Subtotal II	3498.2 ± 338.3	1164.6 ± 345.2	1486.4 ± 288.2	897.0 ± 130.8	1 vs 2, 3, 4	0.0001
Total	3645.4 ± 333.5	1298.3 ± 335.3	1651.5 ± 309.0	1053.5 ± 132.3	1 vs 2, 3, 4	0.0001

* Data represent RAL × 10^{10} values (± SEM), N = 3–5 individual animals.
† Source obtained from Fisher's multiple comparison test.
‡ *P* values obtained from ANOVA.

and EMD (1.3-fold) activities versus the added inductive effects of the chemicals given separately.

DISCUSSION

The present study has examined the effect of CYP inducers on the formation of ^{32}P-postlabeled TAM–DNA adducts and the activities of CYP-dependent drug-metabolizing enzymes *in vivo*. The selective depletion of group II but not group I adducts by coadministration of TAM with classic CYP inducers was consistent with our previous hypothesis of two distinct pathways of TAM metabolism leading to genotoxic metabolites [16]. The marked and selective reduction of group II adduct levels by each of the CYP inducers suggested that CYP enzymes were involved in the detoxication of TAM to non-genotoxic metabolites *in vivo*. The observation that no significant differences were observed in the extent of reduction of TAM group II adducts between the different inducers (Table 1) indicated that TAM group II adduct formation was inhibited to a similar extent irrespective of the inducer used. As microsomes from animals treated with each of these inducers have been shown to efficiently N-demethylate TAM [25, 30], our results support the hypothesis that this reaction was involved in detoxication of TAM to non-genotoxic metabolites.

Mani *et al.* [24] and White *et al.* [25] have shown in *in vitro* experiments that CYP3A forms are primarily involved in the covalent binding of TAM to microsomal proteins. However, our results cannot be explained solely on the basis of the CYP3A effect, since PB and BNF, which do not markedly induce CYP3A forms [49], also elicited reduction

TABLE 2. Effect of TAM and CYP inducers on microsomal enzymes

Assay	TO	TAM*	PB	PB + TAM	BNF	BNF + TAM	PCN	PCN + TAM
Total P450	0.80	0.71	2.25†	2.30‡	1.11	1.38‡	2.22†	2.74‡
	(0.09)	(0.15)	(0.12)	(0.58)	(0.28)	(0.40)	(0.06)	(0.05)
BZD	12.53	16.23†	48.72†	53.69‡	11.10	18.49	28.32†	26.60
	(0.69)	(0.42)	(0.84)	(4.24)	(1.26)	(3.06)	(1.37)	(7.87)
EROD	0.20	0.28	1.04†‡	0.94	2.03†	4.36‡§‖	0.40†	0.92‡§‖
	(0.08)	(0.03)	(0.21)	(0.19)	(0.50)	(1.93)	(0.10)	(0.08)
MROD	0.13	0.11	1.05†	0.48§	2.24†	3.84§‖	0.37	0.50
	(0.03)	(0.015)	(0.21)	(0.15)	(0.60)	(1.35)	(0.20)	(0.12)
EMD	12.24	12.49	22.99†	28.68	20.20	21.30	30.48†	35.70‡§‖
	(0.89)	(0.89)	(4.54)	(2.80)	(1.16)	(2.30)	(1.18)	(1.75)

All assays were performed in duplicate, and the values presented in the table are means of activities from 3–5 individual animals. SD is given in parentheses. CYP content and enzyme activities represent nmol/mg protein and nmol/min/mg protein, respectively.

* Because there were no significant differences in enzyme activities in animals given TAM only versus animals that were pretreated with saline prior to TAM, data from the TAM group were used for assessing statistical significance.

†–‖ Values were statistically significant (by Student's *t*-test) at $P < 0.05$.

† Comparison of TAM- or inducer-treated groups.

‡ Comparison of inducer + TAM groups with TAM only group.

§ Comparison of inducer + TAM groups with corresponding inducer groups.

‖ Inducer + TAM versus sum of inductive effects of the drugs given separately (see Materials and Methods).

of group II TAM DNA adducts *in vivo*. Thus, it appears that metabolic intermediates that are responsible for DNA binding *in vivo* are different from those involved in protein binding *in vitro*. In fact, White *et al.* [25] have proposed that the reactive metabolites involved in protein and DNA binding could be different, since mouse liver microsomes are more efficient in the metabolic activation of TAM to protein-bound products, whereas *in vivo* administration of TAM to rats leads to higher levels of DNA adducts than in mice. The differences in the reactive metabolites of TAM responsible for DNA and protein binding would be in analogy to the metabolic activation of other carcinogens, e.g. 4-aminobiphenyl, that undergo N-hydroxylation, followed by oxidation to a nitroso derivative that binds to cysteine residues of hemoglobin [50]. On the other hand, metabolic activation of this aromatic amine to DNA-reactive species involves N-hydroxylation followed by sulfuric acid esterification [50]. Similarly, safrole undergoes α-hydroxylation and subsequent sulfonation forming a DNA- or protein-reactive metabolite, while on the other hand, the methylenedioxy group of safrole can also be oxidized to yield protein-binding metabolites [51].

We recently reported that 4-OH-TAM gives rise to the major group I adduct *in vitro* upon incubation with DNA and rat liver microsomes in the presence of cumene hydroperoxide [21]. In contrast, incubation of TAM itself with microsomes and cumene hydroperoxide leads to the formation of the major group II adduct. These reactions are not supported by NADPH, suggesting that metabolic activation of TAM leading to both groups of adducts may involve CYP functioning as a peroxidase [52]. The reason as to why in the present work group I adducts were not affected by CYP inducers could be that the metabolic conversion of TAM to 4-OH-TAM, the proximate metabolite of group I adducts [16], is catalyzed by constitutive CYPs [25, 30], and therefore was resistant to CYP inducers. Other possibilities are that 4-OH-TAM may have been partially or completely resistant to detoxication by CYP-catalyzed N-demethylation.

While levels of phase II enzymes such as UDP-glucuronyl transferases (UDPGT) are also elevated by the CYP inducers used here [49, 53–55], it is unlikely that these enzymes played a major role in the reduction of group II adducts for the following reasons:

1. 4-OH-TAM, the precursor of group I adducts, is glucuronidated [56], but group I adducts were not affected by the CYP inducers.
2. Batt *et al.* [55] have shown that exposure of mice to PB, BNF, or PCN leads to stimulation of UDPGT activities, but the potency of PB is much greater than that of BNF or PCN. However, in our experiments each of the inducers elicited a comparable reduction of group II TAM adducts.
3. As regards the phase II enzymes, sulfotransferases are not induced by CYP inducers [53, 54], and glutathione *S*-transferases are induced only 2- to 3-fold [49].
4. Nuwaysir *et al.* [57] have shown recently that TAM itself induces a specific isozyme of UDPGT, i.e. UGT2B1, which is the major PB-inducible form, without affecting levels of other UDPGT isozymes. However, in our study the combination of PB with TAM did not lead to a greater depletion of group II adducts compared with other inducers.

The induction by TAM of BZD activity (Table 2), which is reflective of catalysis by CYP2B1/B2 [58], suggests that TAM selectively induced CYP enzymes of the CYP2B family, consistent with the results of White *et al.* [59] and Nuwaysir *et al.* [60] that TAM induces CYP2B1 enzymes in rodent liver microsomes. The significant induction by BNF of EROD and MROD activities (Table 2), which reflect CYP1A1 [61] and CYP1A2 [62] dependent activities, was in accord with findings of other investigators that BNF and polycyclic aromatic hydrocarbons induce enzymes of the CYP1A family [49, 58]. However, the mechanisms underlying the synergistic induction of EROD and MROD activities in animals given TAM + BNF are not understood. Since TAM is estrogenic to mice [5], it is possible that TAM enhances BNF-induced transcription of the *CYP1A1* gene by a hormone-mediated process. In fact, TAM has been shown recently to modulate xenobiotic-metabolizing enzymes, at least in part, by perturbing the hormonal milieu [60]. The elevation by PCN of EMD activity (Table 2), which is catalyzed preferentially by CYP3A [63], was probably due to induction of CYP3A isoforms [49, 58, 63, 64]. The significantly greater enhancement of EROD and EMD activities in the PCN + TAM versus PCN samples could again have been due to the estrogenic action of TAM.

In conclusion, classic CYP inducers substantially inhibited total TAM–DNA adduct formation in female mouse liver. Since a growing list of medications taken by humans as well as certain nutrients are potent inducers of the CYP enzymes, combined administration of CYP inducers and TAM may be efficacious in reducing genotoxic risk in humans treated with this antiestrogen.

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