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Interleukin-1β antagonizes phenobarbital induction of several major cytochromes P450 in adult rat hepatocytes in primary culture

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Abstract **We have investigated the effects of interleukin (IL)-1β and IL6 on expression and phenobarbital (PB) induction of ethoxyresorufin *O*-deethylase (EROD) and pentoxyresorufin *O*-deethylase (PROD) activities, as well as on mRNA levels of cytochromes P450 (CYP) 1A, 2B, 2C, 2E and 3A, in rat hepatocytes in primary culture. IL6 slightly antagonized PB-induced PROD activity. Strikingly, IL1β strongly inhibited basal EROD and PROD activities, and fully blocked their induction by PB in a dose-dependent fashion. Furthermore IL1β completely suppressed PB induction of all CYP mRNAs analyzed. Our results demonstrate that IL1β can suppress basal CYP activities, as well as PB-inducible expression of five CYP mRNAs in rat hepatocytes in primary culture.**

Key words: Adult rat hepatocyte; Cytochrome P450; Phenobarbital; Cytokine; Interleukin-1β; Interleukin-6

1. Introduction

Regulation of cytochrome P450 (CYP) gene expression is under complex regulatory control in mammalian liver. Not only do age, sex, and genetic polymorphisms influence CYP-mediated drug metabolism in the liver, but also health status as well as exposure to xenobiotics play critical roles. Indeed, expression of CYP genes from the four major families is inducible both in vivo and in vitro by a large variety of compounds, typified by polycyclic aromatic hydrocarbons (PAH) such as 3-methylcholanthrene (3-MC), barbiturates like phenobarbital (PB), glucocorticoids and fibrates.

It has recently become clear that infection and inflammation diseases generally depress CYP activity in rodents and humans [1–3]. We, and others, have demonstrated that individual cytokines can lower expression of several CYP genes when directly added to primary human and rodent hepatocytes or hepatoma cells in culture [4–6]. Therefore, decreased CYP activities in the liver can be attributed, at least in part, to the direct effect of cytokines on hepatocytes.

Recently, the relationship between cytokines and CYP inducers on modulating CYP expression has been put into question. 3-MC-mediated induction of CYP1A1 and CYP1A2 genes in primary rat hepatocyte cultures was shown to be suppressed, at the transcriptional level, in response to interleukin (IL)-1β, but not to IL6 [7]. It has also been reported that epidermal growth factor and transforming growth factor (TGF)-α blocked 3-MC-induced expression of CYP1A1, in mouse hepatocytes in culture [8]. We recently demonstrated that TGFβ1 was able to fully suppress 3-MC induction of the CYP1A1 and 1A2 genes in human hepatocytes in primary culture, whereas IL1β, tumor necrosis factor (TNF)-α and interferons (IFN) α and γ were only slightly active [9]. IL6 was shown to inhibit 3-MC induction of CYP1A1 in human hepatoma cells at the transcriptional level [10], and PB induction of CYP2B genes in rat hepatocytes in primary culture [11]. In addition, IL1β could inhibit CYP4A1 gene induction by fibrates and dexamethasone in fetal rat hepatocytes [12]. IFN inducers were able to counteract β-naphtoflavone-mediated ethoxyresorufin *O*-deethylase (EROD) induction in vivo [13]. Importantly, growth hormone (GH) was also able to antagonize CYP induction by PB in the rat in vivo and in vitro [14,15]. However, GH levels did not appear to be the primary determinants of the acute phase suppression of CYP gene expression in rat hepatocyte cultures, since the effect of cytokines was readily observed in absence of added GH [7]. Therefore, several categories of peptide hormones, presumably upon interaction with their specific membrane receptor(s) [5], are able to antagonize induction of CYP genes in response to distinct, structurally unrelated, inducing compounds. Moreover, TPA, an activator of protein kinase C, was shown to antagonize PB-mediated induction of selected CYP enzymatic activities [16], as well as 3-MC induction of CYP1A1 gene expression in rodent and human hepatocytes ([17], and unpublished observation in our laboratory). Taken together, these observations strongly suggest that signals initiated at the cell membrane can affect CYP induction, possibly through induction of kinase pathways. The goal of the present experiments was to extend investigations aimed at better understand, especially at the mRNA level, the interelations between 'classical' CYP inducers and several cytokines playing important roles during the inflammatory response.

The results reported here demonstrate that IL1β but not IL6 can suppress the PB-mediated induction of five distinct CYP gene family/subfamily members, i.e. 1A1, 2B1/2, 2C6/7, 2E1 and 3A1/2 enzyme activities and/or mRNAs in primary rat hepatocyte cultures.

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Abbreviations: CYP, cytochrome P_{450}; DMSO, dimethylsulfoxide; EDTA, ethylenediaminetetraacetic acid; EROD, ethoxyresorufin *O*-deethylase; FCS, fetal calf serum; GH, growth hormone; IFN, interferon; IL, interleukin; 3-MC or MC, 3-methylcholanthrene; NO, nitric oxide; PB, phenobarbital; PBS, phosphate-buffered saline; PROD, pentoxyresorufin *O*-deethylase; TGF, transforming growth factor; TNF, tumor necrosis factor; TPA, 12-*O*-tetradecanoylphorbol 13-acetate.

2. Materials and methods

2.1. Recombinant cytokines and chemicals

Human recombinant IL1β and IL-6 were purchased from Genzyme (Cambridge, UK). Several independent batches of IL1β were used through the one year period covered by this study, and endotoxin

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contamination was below the detection limit, according to the manufacturer's data sheet. Nicotinamide, 7-ethoxyresorufin, pentoxyresorufin, 3-MC and PB were from Sigma (St. Louis, MO, USA).

2.2. Cell isolation and culture

Adult hepatocytes were obtained after perfusion of male rat liver (Sprague–Dawley; 150–200 g) with 0.025% collagenase solution (Boehringer-Mannheim) buffered with 0.1 M HEPES (pH 7.4), as previously reported [18].

Cell viability, estimated by Trypan blue exclusion, ranged between 80% and 90%. Hepatocytes were seeded on plastic dishes at a density of 63×10^3 per cm^2 in a standard medium, consisting of 75% (v/v) Minimum Essential Medium (MEM) and 25% (v/v) Medium 199, buffered with 0.22% (w/v) sodium bicarbonate and supplemented with 10 μg/ml bovine insulin, 1 mg/ml bovine serum albumin, 10% (v/v) fetal calf serum (FCS) and 1% (w/v) streptomycin. At the first medium renewal (4 h after seeding), FCS was withdrawn and hydrocortisone hemisuccinate and nicotinamide were added at concentrations of 1 μM and 10 mM, respectively. Nicotinamide is known to favor maintenance of differentiation of hepatocytes in primary culture [19]. Cytokines and inducers were added 24 h after seeding, at the second medium renewal. The medium was then changed every day.

The cytokines have previously been found to regulate basal cytochrome P_{450} expression in primary cultured human hepatocytes [4,9] and/or acute-phase protein expression in both rat and human hepatocytes in primary culture [20,21]. At concentrations used none of the cytokines induced morphological alterations. 3-MC was dissolved in DMSO and was added at the concentration of 5 μM which was found to be optimal for induction of EROD activity, although quite as strong an induction was observed for 0.5 μM. No further induction of EROD activity was obtained for 10 μM 3-MC. All culture conditions included 0.2% DMSO, a concentration having no influence on CYP expression.

2.3. Isolation of RNA and blot analysis

Hepatocyte monolayers were scraped in 0.1 M phosphate-buffered saline (PBS). Total RNA was prepared as previously described [22], dissolved in sterile water and stored at −80°C. For RNA blotting, 10 μg of each RNA sample were subjected to electrophoresis in a denaturing 6% (v/v) formaldehyde–1.2% (w/v) agarose gel, and transferred onto Hybond-N^+ nylon filters (Amersham, Arlington Heights, IL). Equal RNA sample loading and integrity were routinely ascertained by ethidium bromide staining of minigels, and filters following transfer. Prehybridization, hybridization and washes were performed according to Church and Gilbert [23]. cDNA probes were ^{32}P-labeled by random priming using a multiprime labeling kit (Amersham, Arlington Heights, IL). Filters were autoradiographed at −80°C. Hybridization signals were quantified by densitometry and integration of unsaturated signals. References for CYP1A1 (full-length rabbit cDNA (LM4) probe), 2B1/2B2 (full-length rat CYP2B1 cDNA probe), CYP2C6/2C7 (partial 1200 bp cDNA fragment from the 3' half of the rat mRNA), CYP2E1 (full-length rat cDNA probe), CYP3A1/3A2 (full-length rabbit 3A cDNA (LM3c) probe) are 24, 25, 26, 27 and 28. Chicken glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used as a control. The 18S probe was a human genomic DNA fragment.

2.4. Ethoxyresorufin O-deethylase and pentoxyresorufin O-deethylase activity assays

EROD and PROD activities, mainly supported by CYP1A and CYP2B subfamilies, respectively, were measured essentially according to Burke and Mayer [29,30], with slight modifications for living hepatocyte monolayers [31]. Briefly, the medium was removed after 12, 24, 72 and 96 h of treatment, and cells were washed with 100 mM PBS and then incubated with ethoxyresorufin or pentoxyresorufin (50 μM) and salicylamide (1.5 mM) in PBS at 37°C. Kinetic reading with a spectrofluorimeter was performed over 15 min. Fluorescence values were converted to pmoles with a calibration curve of resorufin fluorescence and expressed per mg of total protein and per min. PROD activity levels in our cultures agreed well with results from in vivo and other in vitro studies [32,33], although they were on the lower side in some experiments.

2.5. Statistical analysis

Comparisons were performed with the Student's t-test following ANOVA. In Fig. 1 and Table 1 (upper part), the Student's t-test was used to perform analysis on each experiment by comparing, for each condition, 8 repeated determinations of EROD and PROD activities on the same cell population. In Table 1 (lower part), the analysis compares the means of 3–5 independent experiments, performed with cell preparations obtained over a one year period.

The densitometry analysis of RNA blotting/hybridization experiments was performed with hybridization signals obtained from 7 independent cell preparations for IL1β and 4 independent cell preparations for IL-6, without normalization to another message, since hybridization to the 18S probe demonstrated similar loading levels. As anticipated, variations were observed between absolute values of the signals, since the RNA preparations and hybridization experiments were performed independently. The effect of IL1β on CYP mRNA levels was analyzed with Wilcoxon's test using densitometric values. Significance was set at a limit of less than 5%. The specific sets of comparisons are described in the legends to figures.

3. Results

We set out to investigate the effects of IL1β and IL6 on phenobarbital induction of CYP-dependent enzyme activities

Table 1
Effects of IL1β and IL6 on induced EROD and PROD activities (dose–response)

Condition	Activity (U/ml)	PROD (pmol/mg protein/min)	EROD (pmol/mg protein/min)
Control		0.44 ± 0.19	19.33 ± 1.87
IL1β	100	0.14 ± 0.10***	7.30 ± 1.46***
IL6	50	0.53 ± 0.13	19.53 ± 1.15
PB		1.91 ± 0.23***	104.40 ± 11.48***
PB + IL1β	1	1.89 ± 0.48	75.83 ± 15.33§
	10	1.29 ± 0.27§§	55.84 ± 9.52§§
	100	0.58 ± 0.20§§	30.50 ± 3.92§§
	500	0.23 ± 0.21§§	17.55 ± 2.68§§
PB + IL6	250	2.25 ± 0.47	119.14 ± 16.63
	50	1.66 ± 0.29	83.66 ± 7.83§§
Control		0.52 ± 0.13	14.62 ± 2.85
IL1β	100	0.15 ± 0.05**	6.57 ± 2.90**
IL6	50	0.53 ± 0.13	17.36 ± 4.61
PB		1.67 ± 0.29***	79.17 ± 18.62***
PB + IL1β	100	0.42 ± 0.22^{aaa}	23.59 ± 9.01^{aa,*}
PB + IL6	50	1.35 ± 0.24^{a,**}	67.06 ± 15.50**
MC		1.24 ± 0.05**	71.18 ± 6.80**
MC + IL1β	100	0.72 ± 0.09^{bb}	46.23 ± 2.14^{b,***}
MC + IL6	50	1.27 ± 0.03*	61.50 ± 1.32**

Primary hepatocyte cultures were treated with IL1β (1 to 500 U/ml) or IL6 (up to 250 U/ml, only results with doses of 50 and 250 U/ml are shown) in combination with 3-MC (5 μM) or PB (2 mM) for 96 h, from 24 h after seeding. In the upper part, the dose–response analysis was performed on cells from a single animal. Each value represents the mean ± S.E.M. of 8 samples. For EROD and PROD activities, ANOVA analysis for all culture conditions on the same animal has been performed ($P = 0.0001$) before performing the Student's t-test. Statistically different from control, untreated cells (Student's t-test, $^{***}P < 0.001$, $^{**}P < 0.01$ and $^{*}P < 0.05$). Statistically different from PB-treated cells (Student's t-test, $^{\S\S}P < 0.001$ and $^{\S}P < 0.01$). In the lower part, each value represents the average of 5 experiments (5 animals) ± S.E.M., except for 3-MC alone or in combination with IL1β (3 animals) and 3-MC + IL6 (2 animals). In each experiment measurements were repeated at least 8 times. Control refers to cells kept in culture for the same time as treated cells. Statistically different from control, untreated cells (Student's t-test following ANOVA ($P < 0.05$), $^{***}P < 0.001$, $^{**}P < 0.01$ and $^{*}P < 0.05$). Statistically different from PB-treated cells (Student's t-test, $^{aaa}P < 0.001$, $^{aa}P < 0.01$ and $^{a}P < 0.05$). Statistically different from 3-MC-treated cells (Student's t-test, $^{bb}P < 0.01$ and $^{b}P < 0.05$).

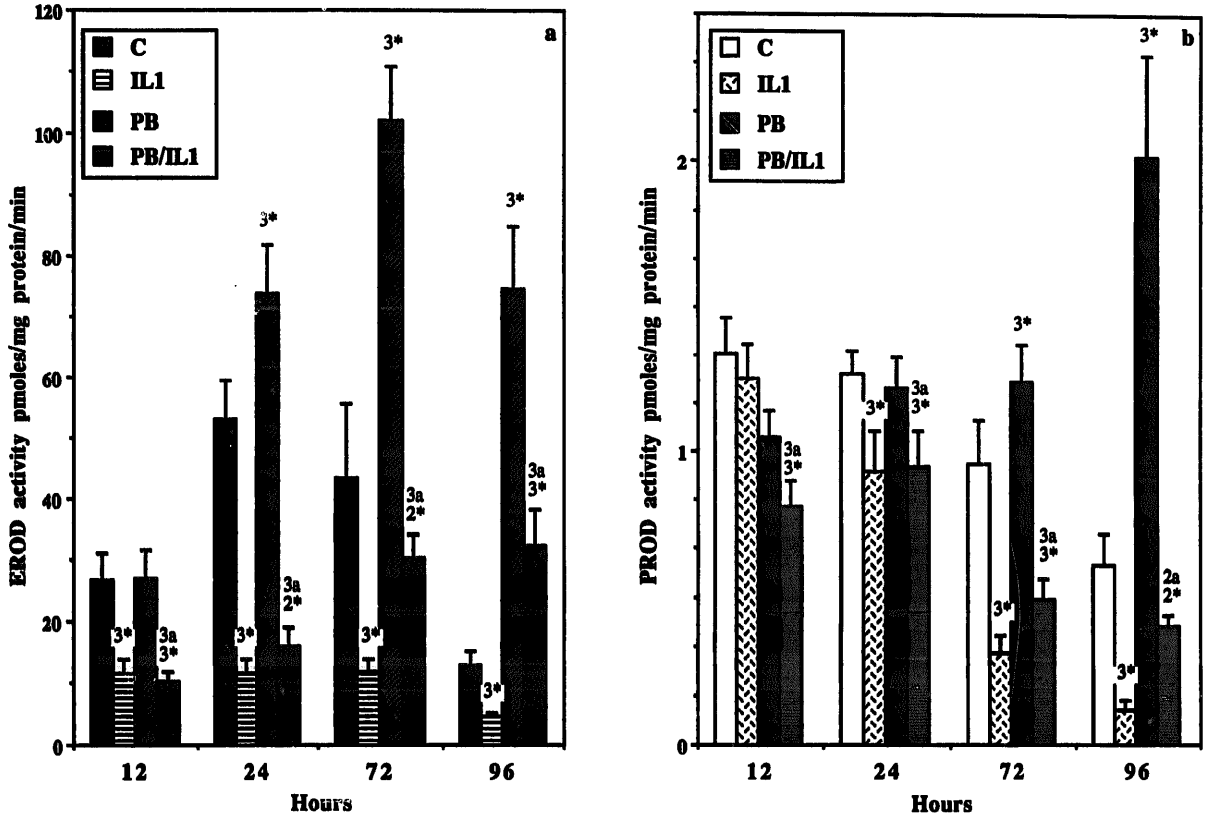

Fig. 1. Time-course analysis of IL1β effects on PB-induced EROD (2a) and PROD (2b) activities. Primary rat hepatocytes were treated with IL1β (100 U/ml) in combination with PB (2 mM) for 12, 24, 72 and 96 h, from 24 h after seeding. Control refers to cells kept in culture for the same time as treated cells. IL1 refers to IL1β. Each value represents the mean ± S.E.M. of 8 samples. (3*, 2*, *)Statistically different from the corresponding control, untreated cells (Student's *t*-test following ANOVA ($P < 0.05$), (3*)$P < 0.001$, (2*)$P < 0.01$, and (*)$P < 0.05$). (3a, 2a, a)Statistically different from PB-treated cells (Student's *t*-test; (3a)$P < 0.001$, (2a)$P < 0.01$ and (a)$P < 0.05$).

in primary rat hepatocyte cultures. Cytokines were chosen according to known effects on either CYP expression or acute phase protein production by human and rodent hepatocyte primary cultures [4,9,20,21]. No apparent cytotoxicity was observed in response to the cytokines alone or in combination with the inducers.

3.1. Effect of IL1β and IL6 on basal and induced PROD and EROD activities

The dose-response experiment in Table 1 (upper part) shows that PB induced PROD activity by at least 4-fold ($P < 0.001$) and, as reported in several previous studies [34–36], EROD activity by a factor of at least 5 ($P < 0.001$). IL1β was able to depress both basal and PB-induced activities in a dose-dependent manner. Complete inhibition of induction was obtained for 500 U IL1β/ml ($P < 0.001$), while 1 U IL1β/ml induced a partial block of only EROD activity (27%; $P < 0.01$). In this dose–response experiment, IL6 (50 U/ml) was able to partially block the PB-induced EROD activity by an average of 25% ($P < 0.001$) and had no effect on PB-induced PROD activity. Table 1 (lower part) shows results from five additional independent experiments at optimal exposure times. PB induced EROD and PROD activities by 5.4- and 3.2-fold ($P < 0.001$), respectively. IL1β produced drops in basal EROD (55%) and PROD (71%) activities, whereas IL6 did not have any significant effect. IL1β blocked PB induction of EROD ($P < 0.01$) and PROD activities ($P < 0.001$) by 70% and up to 100%, respectively. Through all analyzed animals, IL6 did not have any significant effect on EROD induction, although it produced a 20% decrease of PROD activity ($P < 0.05$).

3.2. Effect of IL1β on PB-induced PROD and EROD activities: time-course analysis

In order to determine the kinetics of IL1β-mediated depression of PROD and EROD activities under both basal and PB-treated conditions, we conducted a time-course analysis. Fig. 1a shows that PB induction of EROD activity was detected only after 24 h. It was largest after 96 h (4.2-fold; $P < 0.001$), time at which basal activity had dropped by 52%, as compared to its value at 12 h. IL1β (100 U/ml) was able to induce large decreases (57–78%; $P < 0.001$) in basal EROD activity, and this effect was already observed as soon as at 12 h of culture. IL1β fully blocked PB induction at 24 and 72 h, but only by 57% ($P < 0.001$) at 96 h. However, a higher concentration of IL1β (500 U/ml) fully prevented any induction at 96 h (see above).

Fig. 1b shows that PB induction of PROD activity was only observable after 72 h of treatment (1.3- to 3.3-fold; $P < 0.001$). At these later time-points, full blockades in induction were

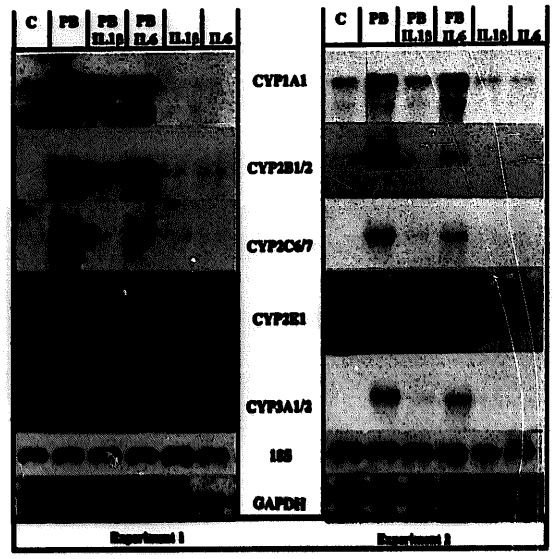

Fig. 2. Effects of IL1β and IL6 on PB-induced CYP1A1, CYP2B1/2B2, CYP2C6/2C7, CYP2E1 and CYP3A1/3A2 mRNA. Primary rat hepatocyte cultures from 7 animals were treated with IL1β (100 U/ml), IL6 (50 U/ml) and PB (2 mM). When cytokines were combined with PB, their addition was concomitant. Total RNA was prepared after 96 h of treatment and CYP mRNA expression was analyzed by RNA blot. Filters were hybridized with ^{32}P-labeled cDNA probes and autoradiographed. C: Control refers to cells kept in culture for the same time as treated cells. Hybridization of the same filters with 18S and GAPDH DNA probes was used as a control. Results from 2 experiments are shown in parts a and b.

obtained in response to IL1β ($P < 0.001$). In addition, IL1β decreased basal PROD activity after 24 h (25%, $P < 0.001$) to a minimum after 96 h (80%; $P < 0.001$).

Earlier time-points and a concentration of 250 U/ml of IL6 have also been surveyed. The effect of this cytokine on EROD and PROD activities was similar for both 72 and 96 h time-points regardless of the dose (data not shown).

3.3. Depression of PB-induced levels of CYP1A, -2B, -2C, -2E and -3A mRNAs by IL1β

In order to determine if IL1β was able to also affect accumulation of CYP mRNAs contributing to EROD and PROD activities, as well as that of other CYP mRNAs, we performed hybridization experiments with specific cDNA probes for CYP1A1, -2B1/2B2, -2C6/2C7, -2E1 and -3A1/3A2. As a whole, seven experiments were performed, allowing us to apply a statistical test (Wilcoxon), following densitometry and integration of unsaturated hybridization signals. Autoradiograms from two independent hybridization experiments from two separate cell populations are shown in Fig. 2.

A strong induction of all CYP mRNA signals was observed in response to PB. In addition, a faint signal corresponding to what might be CYP1A2 mRNA (smaller-sized transcript) was observed in the PB sample upon hybridization with the CYP1A1 cDNA probe. However, since this band co-migrated with the 18S ribosomal RNA band, it is also possible that it represents an artefact due to trapping of some CYP1A1 mRNA highly abundant in these lanes.

After densitometric analysis of autoradiograms from the 7 experiments, the Wilcoxon test was applied to determine the significance level of IL1β effects. IL1β could significantly, albeit slightly, decrease basal CYP2C6/7 (26%; $P < 0.05$) and CYP2E1 (34%; $P < 0.05$) mRNA levels through all of the seven experiments, although this was not visually apparent in Fig. 2. The effect of IL1β on basal accumulation levels of CYP1A1 and CYP2B1/2B2 was not significant. This cytokine produced, however, about 55% reduction of the PB-induced CYP1A1 (induction factor of 7.6-fold) and CYP2B1/2B2 (induction factor of 3.4-fold) mRNA levels ($P < 0.05$). PB-induced CYP2C6/2C7 and CYP3A1/3A2 mRNAs (induction factors of 2.6- and 2.1-fold, respectively) were suppressed by 63% and 58% by IL1β, respectively ($P < 0.05$). CYP2E1 mRNA level was induced by 2.4-fold in response to PB, and this induction was reduced by 44% by IL1β ($P < 0.05$).

Significance levels of the effect of IL6 on CYP mRNA accumulation could not be accurately determined due to the limited number [4] of independent experiments, even though apparent decreases of the PB response were generally observed. GAPDH mRNA levels did not show substantial variations under the various conditions. Finally, hybridization to an 18S ribosomal DNA probe demonstrated equivalent RNA loading.

3. Discussion

The present study shows that IL1β but not IL6 was able to decrease basal EROD and PROD activities in rat hepatocytes in primary culture. The effect of cytokines on basal mRNA accumulation levels was not always accurately measurable in the absence of inducer treatment, due to a very low expression level. In a previous study, we found that IL1β, IL6 and TNFα suppressed basal CYP mRNA levels in cultured human hepatocytes [4]. IL1β could significantly, although modestly, lower basal mRNA accumulation levels of CYP2C and CYP2E1 throughout the 7 experiments conducted in the present study. As previously described in various other model systems [34,35,37–40], PB was able to induce EROD activity, as well as CYP1A1 mRNA level. We also observed induction in hepatocytes cultured in Williams' E medium (data not shown). In addition, IL1β was able to strongly antagonize this induction as well as PB-induced PROD activity and CYP2B1/2, -2C6/7, -2E1 and -3A1/2 mRNAs through 7 independent experiments. The effects of IL1β and IL6 on the 3-MC-mediated induction of EROD and PROD activities agree with previous work [7]. No significant effect of TNFα, IFNα or IL4 on basal or PB-induced EROD and PROD activities was observed (data not shown). However, IFNγ was able to depress basal as well as PB-induced EROD activity by an average of 15% ($P < 0.01$) (data not shown).

In the present study, we observed that IL1β, but not IL6, was able to fully antagonize 3-MC-mediated induction of EROD activity. These observations were in perfect agreement with results from Barker et al. [7] who showed, in addition, that the antagonism of IL1β over induction took place at the transcriptional level. We made the unexpected observation that 3-MC was also able to reproducibly induce PROD activity, although no such inductive effect was observed for CYP2B mRNA (data not shown). Further investigations are needed to explain this effect which was also blocked by IL1β.

Although IL6 was able to slightly suppress PB-induced EROD level in the dose–response experiment (Table 1, upper

part), this cytokine depressed only PB-induced PROD activity (20%; $P < 0.05$) over a set of 5 experiments (Table 1, lower part). Nevertheless, we did not observe the total blockade of CYP2B-associated enzyme activity level reported by others using the similar dose range of IL6 in Fisher 344 rat-derived hepatocytes cultured on matrigel [11,41]. In addition, no effect of IL6 on CYP2B mRNAs could be proven to be significant, although a tendency towards decrease was generally seen. However, similar results with regard to the effect of IL1β on enzyme activity levels were obtained in this study [41] and in ours.

The mechanism(s) by which IL1β down-regulates basal and induced CYP expression remain(s) unclear. However, another peptide hormone, namely growth hormone, has been shown to be a strong suppressor of CYP gene induction by PB in vivo in the rat and in rat hepatocytes in primary culture, presumably upon interaction with its membrane receptor [14,15]. Activation of AP1 and/or NFkB, two major transduction signal effectors known to be activated by IL1β in various systems, remains to be analyzed [42,43].

A potential inhibitory pathway of CYP gene expression could involve an increase in nitric oxide (NO) synthase. Indeed, it has been recently demonstrated that rat and human CYP1A expression was inhibited by NO in recombinant V79 Chinese hamster cell lines and in rat hepatocytes. In addition, inhibition of NO synthesis, in the presence of cytokines and endotoxin, led to restoration of CYP1A1 activity [44]. Other studies showed induction of NO synthase following treatment with a combination of different cytokines (IL1β, TNFα and IFNγ), which acted synergistically on hepatocytes and other cell types [45,46]. Administration of lipopolysaccharide (LPS) diminished PB-mediated PROD induction in vivo in the rat, and PB induction was partially restored when a specific NO synthase inhibitor was given to animals concomitantly with LPS [47]. In addition, glucocorticoids, at a concentration similar to that found in our cell culture medium, have been shown to inhibit NO synthase induction ellicited by a combination of IL1β, TNFα, IFNγ and LPS in primary cultures of rat hepatocytes [48]. This observation suggests that NO synthase activity might not be the main determinant of the effect of IL1β on PB-mediated induction CYP genes. Moreover, preliminary data from our laboratory suggest that IL1β suppresses PB induction of CYP genes as effectively in presence of an inhibitor of NO synthase (*N*-methyl-L-arginine) as in its absence (data not shown).

In summary, our results support the idea that cytokines, and especially IL1β, are major regulators of phase I detoxication enzymes. Their effects on phase II enzymes remain to be investigated. The observation that cytokines can antagonize CYP induction suggests that the overall hepatic detoxication activity results from a balance of major inductive and suppressive mechanisms.

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