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Steroids 67 (2002) 119–126

Steroids

Effect of cellular differentiation on 11β-hydroxysteroid dehydrogenase activity in the intestine

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Received 21 September 2000; received in revised form 28 February 2001; accepted 2 April 2001

Abstract

11β-Hydroxysteroid dehydrogenase (11βHSD) converts endogenous glucocorticoids to their biologically inactive 11-dehydro derivatives and is therefore able to determine, at least in part, the biological action of glucocorticoids. Type 1 11βHSD has both oxidase and reductase activities interconverting corticosterone and 11-dehydrocorticosterone, whereas type 2 11βHSD has only oxidase activity converting corticosterone to 11-dehydrocorticosterone. Since 11βHSD expression is regulated during development and by hormones in a tissue-specific manner and since glucocorticoids play an important role in postnatal intestinal maturation, we investigated the role of corticosteroids and cytodifferentiation in the regulation of intestinal 11βHSD. Using rat intestinal organ cultures and epithelial cell lines derived from rat small intestine (IEC-6, IEC-18) and from human colon adenocarcinoma (Caco-2, HT-29), we analyzed the effect of corticosteroids and cytodifferentiation on 11βHSD. Screening of the clonal cell lines showed that Caco-2 cells expressed by far the greatest 11βHSD2 oxidase activity, lower activity was observed in HT-29 cells, and lowest activity was seen in IEC cells. Treatment with dexamethasone (50 nM) increased the activity of 11βHSD2 in IEC-6 cells (+59%) and HT-29 cells (+31%), whereas aldosterone (50 nM) stimulated 11βHSD2 in IEC-6 cells only (+31%). Caco-2 cells and IEC-18 cells did not respond to corticosteroids. Growth of IEC-6 cells on Matrigel, treatment of HT-29 cells with butyrate, and postconfluency of Caco-2 cells increased not only the markers of cytodifferentiation, such as alkaline phosphatase and sucrase, but also the activity of 11βHSD2 in all of these cell lines (IEC-6, +96%; HT-29, +139%; Caco-2, +95%). Addition of corticosteroids to these more differentiated cell cultures did not enhance 11βHSD2 activity. In intestinal organ cultures of suckling rat small intestine, dexamethasone and aldosterone stimulated 11βHSD by more than 300%. We conclude that corticosteroids markedly and differentially regulate intestinal 11βHSD2 and that cytodifferentiation of intestinal epithelial cells is associated with upregulation of 11βHSD2 activity that is independent of corticosteroids.

Keywords: IEC-6 cells; IEC-18 cells; HT-29 cells; Caco-2 cells; 11β-Hydroxysteroid dehydrogenase; Dexamethasone; Aldosterone

1. Introduction

11β-Hydroxysteroid dehydrogenase (11βHSD) is a microsomal enzyme that catalyzes the reversible conversion of active glucocorticoids (cortisol, corticosterone, respectively) to their hormonally inactive metabolites (cortisone, 11-dehydrocorticosterone, respectively). 11βHSD is thought to prevent the access of glucocorticoids to the mineralocorticoid receptors and to regulate glucocorticoid access to the glucocorticoid receptors [1]. Biochemical and molecular biological evidence indicates that there exists at least two distinct isozymes of 11βHSD that differ in terms of their requirements and affinity for substrate: a low affinity, NADP-dependent (11βHSD1) and a high affinity, NAD-dependent isozyme (11βHSD2). The NAD-dependent isoform is a strong dehydrogenase, whereas the type 1 isozyme acts predominantly as a reductase in vivo [1]. There is abundant evidence for the presence of 11βHSD in many tissues and organs including gastrointestinal tract, the distal part of which (ileum, caecum, colon) is a well characterized mineralocorticoid target tissue [2,3]. Using an immunohistochemical approach or enzyme assay, 11βHSD has been localized to the colon, caecum, and ileum, but not to the jejunum [4–7]. In colon, 11βHSD1 was found in nonepithelial cells of the lamina propria, whereas 11βHSD2 was localized to the epithelial surface and crypt cells [5] with higher expression in the surface colonocytes and progressive decrease along the crypt axis [8,9]. This distribution of 11βHSD follows the distribution of mineralocorticoid receptors and is quite opposite to the distribution of

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0039-128X/02/$ – see front matter
PII: S0039-128X(01)00143-X

glucocorticoid receptors along the crypt axis [10], i.e. the spatial distribution of 11βHSD seems to depend on the stage of colonocyte differentiation.

It is widely accepted that corticosteroids play an important role in intestinal development affecting the growth, differentiation [11], and regulation of intestinal transport functions [12]. The local regulation of corticosteroid metabolism via 11βHSD may therefore represent an important factor in intestinal development [13]. In addition, it has been reported recently that intestinal 11βHSD is stimulated in vitro in organ cultures of ileum and colon by corticosteroids [15] and that the ileal enzyme undergoes dramatic developmental changes that correlate with the developmental pattern of plasma concentrations of corticosterone [14]. However, the intestine is a very complex system composed not only of epithelial cells (enterocytes, colonocytes), but also of other elements, such as mesenchymal cells, muscle cells, and extracellular matrix molecules that form an integrated functional unit [16,17]. Due to this complex structure, the mechanism of corticosteroid action on intestinal 11βHSD is not obvious.

The aim of this study was, therefore, to test whether the effect of corticosteroids on intestinal 11βHSD activity represents a direct effect on the enterocytes and whether cytodifferentiation is able to modulate 11βHSD activity. We examined intestinal organ cultures [15] and four different cell lines: (i) the permanent epithelial cell lines derived from the rat jejunum and ileum (IEC-6 and IEC-18) that retain the characteristics of the immature crypt cells [18,19] and (ii) two cell lines derived from colonic adenocarcinomas that exhibit inductively (HT-29) or spontaneously (Caco-2) the differentiated phenotype of mature enterocytes [20].

2. Experimental

2.1. Materials

Dulbecco's modified Eagle's medium (DMEM) was obtained from Sevapharma a.s. (Prague, Czech Republic), fetal bovine serum (FBS) was acquired from Pansystems GmbH (Aidenbach, FRG), Petri dishes were purchased from Gama (České Budějovice, Czech Republic), and tissue culture flasks (25 cm^2) were obtained from Nunc (Roskilde, Denmark). Matrigel, a reconstituted basement membrane prepared from the Engelbreth-Holmswarm sarcoma, was purchased from Collaborative Research (Bedford, MA, USA), and 1,2,6,7-[^{3}H]corticosterone was acquired from Amersham International (Aylesbury, UK). Methanol was purchased from Merck (Darmstadt, FRG). All other chemicals were supplied by Sigma (St. Louis, MO, USA).

2.2. Cell cultures

Cells of four lines (IEC-6, IEC-18, Caco-2, and HT-29) were grown in monolayer cultures. Stock cultures were grown in plastic 25 cm^2 flasks and incubated at 37°C in 5% CO_2/95% air atmosphere. The culture medium consisted of DMEM supplemented with 40 μg/ml gentamycin (Lek, Ljubljana, Slovenia) and varying concentrations of FBS. Caco-2 and HT-29 cells were cultivated in the presence of 10% FBS, whereas IEC-6 and IEC-18 were cultured in the presence of 5% FBS and 0.1 U/ml insulin (Léčiva, Prague, Czech Republic) for optimal growth. For experiments, the stock cells were detached by treatment with trypsin-EDTA solution (Sigma, St. Louis, MO, USA) in PBS and seeded in 60 mm dishes at a density of 0.2×10^6 cells/dish. Four days before the experiments, aldosterone (50 nM) or dexamethasone (50 nM) was added to the cultures. After the cells were maintained in the medium for 4 days, the monolayers of cells were washed with fresh DMEM without steroids, and 11βHSD activity was measured. As the analysis of the medium did not demonstrate any detectable amount of corticosteroids, we did not strip the FBS with charcoal.

The effect of cell differentiation on 11βHSD was studied in Caco-2 cells in the postconfluent stage (17 days), in IEC-6 cells growing on Matrigel coated dishes, and in HT-29 cells incubated in the presence of butyrate (2 mM). In these experiments, IEC-6 cells were plated at much higher density (1.3×10^6 cells/dish) than in experiments with corticosteroids.

2.3. Intestinal organ cultures

Organ cultures were prepared as previously described [15]. Briefly, seven-day-old suckling rats were decapitated, and the ileum was removed aseptically, washed, opened longitudinally, and cut into small fragments. Fragments of the ileum were cultured in DMEM supplemented with gentamycin and 5% FBS in 60 mm Petri dishes at 37°C under a humidified atmosphere of 5% CO_2 and 95% O_2 for 72 h with or without dexamethasone (50 nM), aldosterone (50 nM), or butyrate (2 mM). 11βHSD activity was measured as mentioned later, and enzyme activity was expressed as the percentage of [^{3}H]corticosterone conversion per 24 h and per mg of dry weight.

2.4. Determination of cell proliferation and differentiation

Cell proliferation was estimated by determining the total number of cells per dish. The cells were trypsinized, diluted with PBS, and cell number was counted using a Bürker chamber. The cells were homogenized at 4°C in a glass homogenizer using a grinding tube and ultrasonicator. Cell differentiation was followed by measuring the increase in sucrase and alkaline phosphatase activity as described previously [13]. The amounts of glucose liberated from sucrose and nitrophenyl liberated from p-nitrophenyl-phosphate were determined spectrophotometrically. The enzyme activities were expressed as mU per mg of protein (one unit being 1 μM of substrate hydrolyzed per minute).

2.5. 11βHSD activity assay

11βHSD oxidase activity was determined by measuring the rate of conversion of corticosterone to 11-dehydrocorticosterone. On the day of experiment, the medium was aspirated from the dish, and the cells were preincubated in fresh DMEM without steroids or butyrate for 4 h. 11βHSD2 activity was assayed using 6.25 nM [^{3}H]corticosterone whereas 11βHSD1 activity was measured using 6.25 nM [^{3}H]corticosterone and 1 μM corticosterone. Under these conditions, the radiometric conversion assay provided a measure of net conversion of corticosterone by intact cells in the presence of substrate concentrations that approximated K_m values for intestinal 11βHSD1 and 11βHSD2 [5,21]. 11βHSD reductase activity was measured using 6.25 nM [^{3}H]11-dehydrocorticosterone and 1 μM 11-dehydrocorticosterone. [^{3}H]11-dehydrocorticosterone was generated by incubating guinea-pig colon with [^{3}H]corticosterone followed by Sep-Pak extraction and HPLC purification. Preliminary studies demonstrated that the investigated cell lines possessed different levels of 11βHSD expression, and therefore, the cell monolayers were incubated for respective times in order to be within the linear portion of the conversion curves. The reaction was stopped by cooling and aspiration of the medium. Aliquots of the media were extracted using C18 reversed-phase Sep-Pak column (Waters, Milford, MA), dried under nitrogen, and analyzed using HPLC as described previously [15]. ^{3}H-labelled steroids were monitored by on-line radioactive detection using a radioisotope detector with a solid cell (Beckman Type 171, Fullerton, CA). The activity of 11βHSD was expressed as pmol of 11-dehydrocorticosterone produced per hour and per 10^6 cells.

2.6. HPLC/MS analysis

The method used was similar to our previously described method for the analysis of corticosterone metabolites [22]. Briefly, the apparatus used was a HP 1100 LC/MSD system (Hewlett-Packard, Palo Alto, CA, USA) consisting of a degasser, a binary pump, an autosampler, a thermostated column compartment, and mass selective detector (MSD). Chromatographic separation was carried out on the Zorbax Eclipse XDB-C18 column (150 × 2.1 mm I.D., 5 μm, Rockland Technologies [Hewlett-Packard]). A 5 μl sample (dissolved in 1% acetic acid in methanol) was injected. Elution was achieved by a linear gradient between mobile phase A (methanol–water–acetic acid 40:60:1, v/v/v) and B (methanol). The gradient started from 20% B to 100% B at 55 min, and then, the column was eluted with 100% B for 5 min. Equilibration before the next run was achieved by 10 min of washing with 20% B. The flow-rate was 0.25 ml/min for the first 55 min, and then, the flow-rate was 0.4 ml/min; column temperature was held at 25°C.

2.6.1. Mass spectrometry

Atmospheric pressure ionization-electrospray ionization (API-ESI) positive mode mass spectrometry was used. Operating conditions were as follows: drying gas (N_2), 6 l/min; drying gas temperature, 350°C; nebulizator pressure, 20 psi (138 kPa); capillary voltage, 4500 V; ions were observed at mass range m/z 200–500; the fragmentor was set at 80 V.

2.7. Data analysis

Data are presented as the mean ± SEM. Statistical analysis was performed using the unpaired Student's *t* test. $P < 0.05$ was considered statistically significant.

3. Results

Using corticosterone as a substrate, all four cell lines were able to convert corticosterone to 11-dehydrocorticosterone. The conversion of corticosterone to 11-dehydrocorticosterone increased progressively during the incubation (IEC-6, IEC-18: 24 h period; HT-29, Caco-2: 8 h period) but the conversion of 6.25 nM corticosterone was much higher (~60–80% in HT-29 and Caco-2 cells, ~20% in IEC cell lines) than the conversion of 1.006 μM corticosterone (< 1%). When the cells were incubated with 11-dehydrocorticosterone (1.006 μM) there was not observed any conversion to corticosterone. These experiments indicated that the cell lines used in our experiments expressed predominantly type 2 oxidase activity and therefore only the low concentration of corticosterone was used in further 11βHSD assays.

Intact Caco-2 cells in the preconfluent stage metabolized corticosterone most avidly (0.954 ± 0.078 [6] pmol/h/10^6 cells), whereas HT-29 (0.362 ± 0.018 [6] pmol/h/10^6 cells) and especially IEC-6 (0.087 ± 0.006 [26] pmol/h/10^6 cells) and IEC 18 (0.044 ± 0.003 [12] pmol/h/10^6 cells) exhibited much lower activity. When IEC-6, IEC-18, and HT-29 cells were incubated with [^{3}H]corticosterone, they produced exclusively one metabolite, which was identified as [^{3}H]11-dehydrocorticosterone. In contrast, Caco-2 cells metabolized [^{3}H]corticosterone not only to [^{3}H]11-dehydrocorticosterone, but also to two other radioactive products - [^{3}H]20-dihydrocorticosterone and the unknown product (product X). This latter product was analyzed by HPLC/MS system (Fig. 1). When we compared this analysis with chromatographic analysis of steroid standards (Fig. 1A) and mass spectra, the presence of corticosterone was clearly demonstrated (Fig. 1B). The mass spectrum of the unknown peak at retention time 14.4, which corresponds to the product X of the radioactive system, is shown in Fig. 1C. Comparison of this spectrum with our previous results [22] demonstrates the steroid structure of peak X. We obtained similar mass spectra (315, 333, 373, 389, 297, 279 m/z) for two steroids (5β-pregnan-3β,11β,21-triol-20-one and 5α-pregnan-3β,

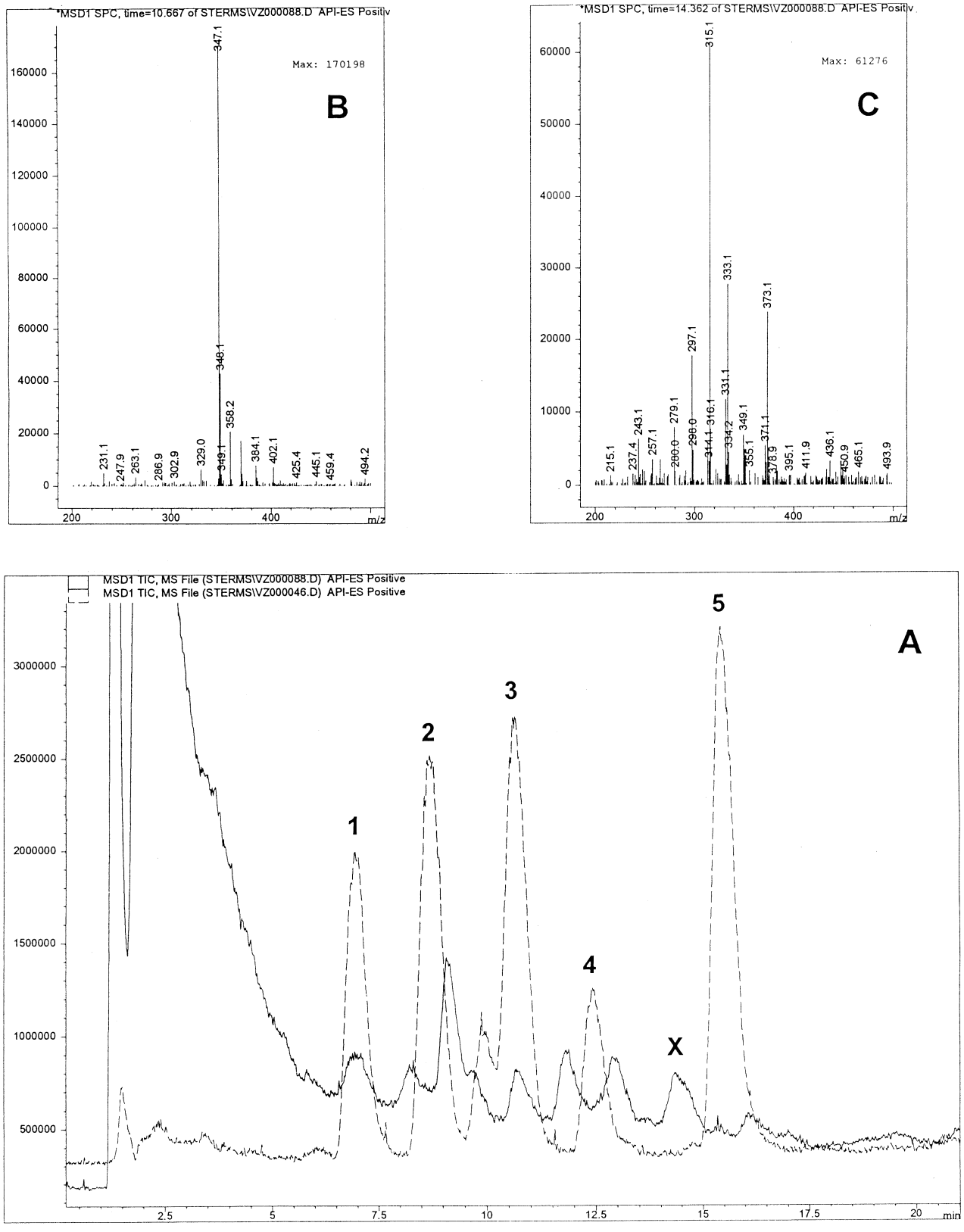

Fig. 1. HPLC/MS analysis of corticosterone metabolism in Caco-2 cells. **A:** Total ion current (TIC) profile of standards (dashed lines) and corticosterone metabolites produced by Caco-2 cells (full line); **B:** mass spectra of corticosterone and **C:** mass spectra of the unknown product X. 1, 11-dehydrocorticosterone; 2, 11-dehydro-20-dihydrocorticosterone; 3, corticosterone; 4, 20-dihydrocorticosterone; 5, deoxycorticosterone; X, unknown product.

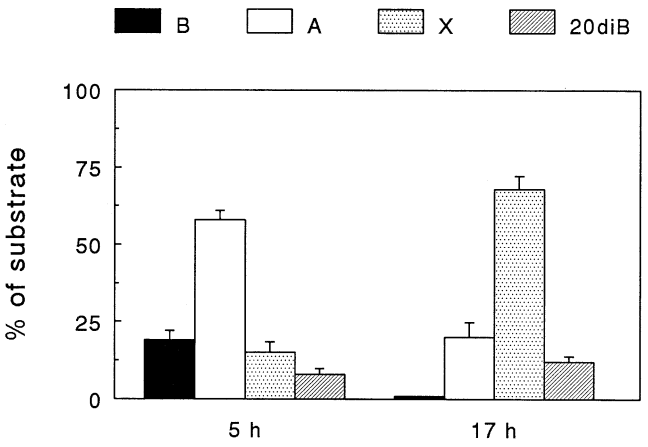

Fig. 2. Distribution of radioactivity from corticosterone after 5 and 17 h of incubation of the Caco-2 dishes with [^{3}H]corticosterone. B, corticosterone; A, 11-dehydrocorticosterone; X, unknown product; 20diB, 20-dihydrocorticosterone. All data are presented as the mean ± SEM.

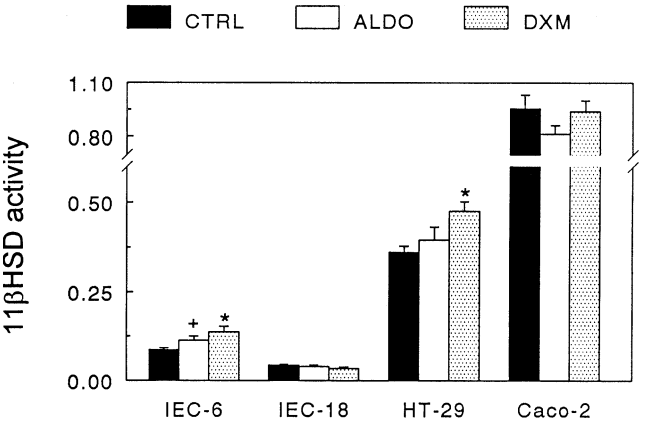

Fig. 3. 11βHSD activity in intact cells of various cell lines incubated in the presence and absence of corticosteroids. The cells were assayed after the cultures were between 80 and 100% confluence. Results are expressed as pmol of 11-dehydrocorticosterone produced per hour and per 10^6 cells and are presented as the mean ± SEM. $^+P < 0.05$ or $^*P < 0.01$ compared to controls.

11β,21-triol-20-one), but these compounds had another retention time (see our previous publication) [22].

We conclude, therefore, that the new product seems to be a steroid of unknown structure. The distribution of all three products depended significantly on the length of incubation (Fig. 2). The DMEM medium extracts prepared from Caco-2 cells incubated for 5 h with [^{3}H]corticosterone contained 58 ± 3% [11] of radioactivity in 11-dehydrocorticosterone and much less in the unknown product X (15 ± 3%). After 17 h of incubation, however, only 20 ± 4% [18] of radioactivity was localized into 11-dehydrocorticosterone and 68 ± 4% [18] was incorporated into product X. These results indicate that 11-dehydrocorticosterone is a substrate for the synthesis of product X.

To study the effect of corticosteroid hormones on 11βHSD, the intestinal cell lines were treated for 48 h with 50 nM dexamethasone or aldosterone. The treatment regime produced a decrease in proliferation that was assessed by counting the total number of cells per dish (Table 1). Both aldosterone and dexamethasone arrested the growth of IEC-6 and IEC-18 cells, but not that of colonic HT-29 cells and Caco-2 cells in the preconfluent stage. The effect of corticosteroids on 11βHSD activity was assessed by incubating the cells with [^{3}H]corticosterone and measuring the percentage of conversion. As shown in Fig. 3, 11βHSD activity in HT-29 and IEC-6 cells was significantly increased following treatment with dexamethasone, whereas aldosterone induced 11βHSD activity only in IEC-6 cells. This stimulatory effect of aldosterone on IEC-6 cells may reflect the moderate affinity of glucocorticoid receptors to aldosterone [23]. The activity of 11βHSD was not changed in IEC-18 and Caco-2 cells.

To study the effect of cell differentiation on 11βHSD activity we cultured the cells under different experimental conditions that showed antiproliferative, but differentiation-inducing effects (growth on intestinal matrix, butyrate, or postconfluency). As shown in Tables 1 and 2, the decrease in proliferation and a more differentiated phenotype of these cells compared to their untreated (IEC-6, HT-29) or preconfluent (Caco-2) controls was attained. Data summarized in Fig. 4 demonstrate that 11βHSD activity was significantly higher in the differentiated cells than in their undifferentiated counterparts. Growth of IEC-6 cells on Matrigel, which inhibits proliferation and stimulates cellular maturation [24], doubled the activity of 11βHSD (+ 96%). Incubation of HT-29 cells in the presence of butyrate, which has antiproliferative and differentiation-stimulating effects [25], increased the activity by 139%. Also, the comparison of Caco-2 cells in preconfluent and postconfluent stages indicated that cell differentiation was followed by an increase in 11βHSD activity (+ 95%). Neither dexamethasone nor aldosterone stimulated the activity of 11βHSD in the differentiated IEC-6, HT-29, and Caco-2 cells (data not shown).

To examine the effect of corticosteroids and butyrate on immature intestine, organ cultures of the ileum were treated

Table 1
Effect of corticosteroids, Matrigel, and butyrate on proliferation of IEC-6, IEC-18, HT-29, and Caco-2 cells

Cell line	Aldosterone	Dexamethasone	Butyrate or Matrigel
IEC-6	77 ± 5 (6)**	64 ± 2 (6)**	70 ± 3 (14)**
IEC-18	81 ± 6 (8)*	82 ± 7 (8)*	n.d.
HT-29	91 ± 9 (6)	87 ± 8 (7)	53 ± 5 (6)**
Caco-2 preconfluent	104 ± 5 (6)	105 ± 5 (6)	n.d.

Values represent the mean ± SEM; numbers of determinations are in parentheses; n.d., not determined. All results are given as a percentage of the control values (untreated dishes). In some experiments, IEC-6 cells were grown on Matrigel and HT-29 cells were cultured in the presence of butyrate. For further details, see Materials and Methods. $^*P<0.05$, $^{**}P<0.01$.

Table 2
Effect of culture treatment on alkaline phosphatase and sucrase activity

Cell line	Treatment	Alkaline phosphatase	Sucrase
IEC-6	Controls	1.5 ± 0.3 (9)*	>0.05[a]
	Matrigel	3.8 ± 0.4 (8)	>0.05[a]
HT-29	Controls	0.3 ± 0.1 (8)	n.d.
	Butyrate	5.1 ± 0.5 (8)*	n.d.
Caco-2	Preconfluent stage	24.5 ± 1.8 (13)	4.3 ± 0.7 (8)
	Postconfluent stage	128.2 ± 12.3 (14)*	31.6 ± 5.9 (12)*

Values represent the mean ± SEM; numbers of determinations are in parentheses; [a]sucrase activity in IEC-6 cells was not detected under the experimental conditions of this study; n.d., not determined. Values are expressed as mU/mg prot. The IEC-6 cells were grown on plastic (controls) or on basement membrane extract (Matrigel), HT-29 cells were incubated in the absence (controls) or presence of butyrate, and Caco-2 cells were studied in the preconfluent (between 80 and 100% confluence) and postconfluent stages. For further details, see Materials and Methods. *$P <$ 0.01 compared to controls.

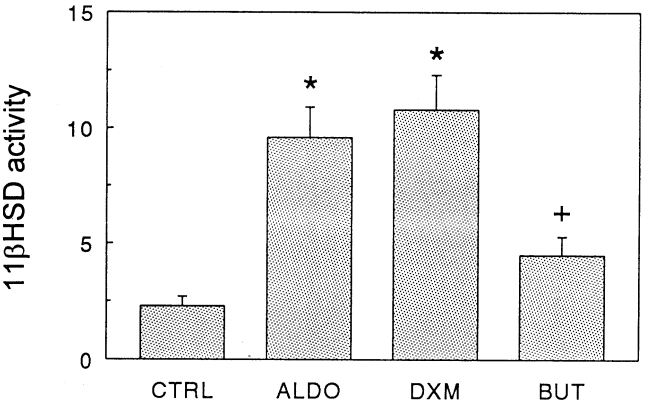

Fig. 5. Effect of corticosteroids and butyrate on 11βHSD activity in the organ culture of rat ileum. Pieces of ileum were incubated in the absence or presence of aldosterone, dexamethasone, or butyrate as described in Experimental. Activity is expressed as the percentage of [^{3}H]corticosterone conversion per 24 h and per mg of dry weight and presented as the mean ± SEM. $^+P < 0.05$ or *$P < 0.01$ compared to controls.

with dexamethasone, aldosterone, or butyrate, and 11βHSD activity was evaluated. The data summarized in Fig. 5 show that corticosteroids exerted a striking effect on 11βHSD activity. Treatment with dexamethasone or aldosterone increased 11βHSD activity by 370% and 317%, respectively, whereas butyrate stimulated 11βHSD activity only by 96%.

4. Discussion

In accordance with previous findings in the intestine [5,14], these results showed that intestinal cell lines expressed 11βHSD even if the activity of this enzyme was largely different among the cell lines. Caco-2 cells possessed the highest level of glucocorticoid metabolism, whereas HT-29 and especially IEC cells had much lower activity. These findings reflect the situation in the intact intestine, where the highest level of 11βHSD activity was localized to the mineralocorticoid-target intestinal segment, i.e. the colon, and the activity was lower in the ileum and absent in the upper part of the small intestine [4,7]. The findings, that the conversion of corticosterone operated well in the nanomolar range whereas it was nearly absent in the micromolar range of substrate concentrations and that 11-reductase activity was absent, suggest the presence of

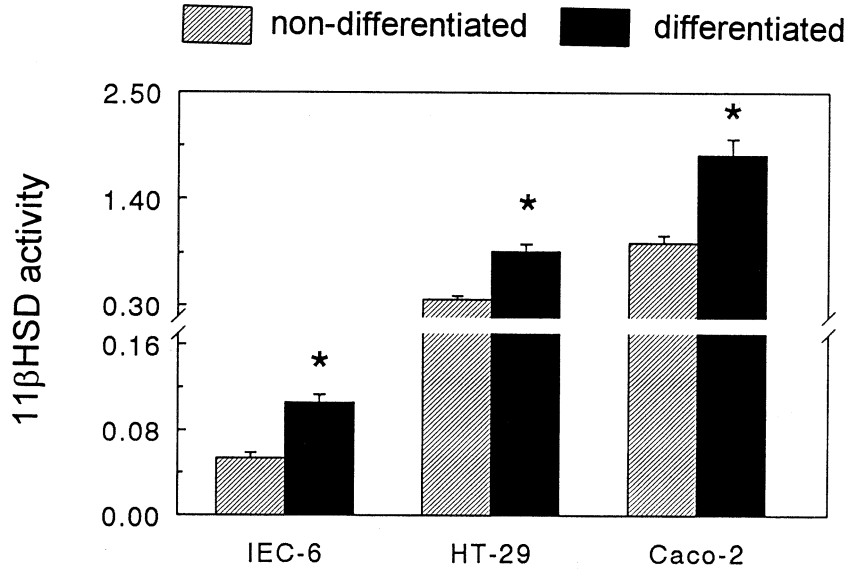

Fig. 4. Effect of differentiation of intestinal cells on 11βHSD activity. IEC-6 cells were grown on plastic (cross-hatched bars) or a Matrigel-coated surface (solid bars). HT-29 cells were grown in the absence (cross-hatched bars) or presence (solid bars) of butyrate. Caco-2 cells were analyzed in the preconfluent (cross-hatched bars) and postconfluent (solid bars) stages. Values are expressed as pmol of 11-dehydrocorticosterone produced per hour and per 10^6 cells and are presented as the mean ± SEM, *$P < 0.01$ compared to controls.

11βHSD2 in the studied cell lines. This is similar to other human colonic cell lines and the intestine. Recent studies of colonic cell lines T84 and SW-620 have shown that only 11βHSD2 is expressed in these cells [21,25]. Similarly, 11βHSD2 isoform is present only in the intestinal epithelium, whereas 11βHSD1 isoform is localized exclusively to the non-epithelial cells of the lamina propria [5,26].

The present study demonstrated that corticosteroids induced a much smaller response in the intestinal cell lines than in the intestinal organ cultures. Our failure to induce 11βHSD may be due to a lack of corticosteroid responsiveness of some genes in the culture of immortalized cell lines or due to the absence of the mesenchymal compartment. Previous studies have shown that glucocorticoids are able to influence epithelial cells indirectly via the mesenchymal cells and extracellular matrix [16]. In addition, the presence of different isoforms of 11βHSD in the lamina propria and in the intestinal epithelium [5,8,26] leaves open the possibility that the increase of corticosterone conversion in organ cultures reflected the effect of corticosteroids on 11βHSD1 of the lamina propria. This isoform was reported to be stimulated by dexamethasone in various cell types including fibroblasts [27–30], however, the substrate concentration of our assay was appropriate for 11βHSD2 measurement but at least 150 fold too low for 11βHSD1 assay. Additional studies will be required to resolve this issue.

The small stimulatory effect of corticosteroids on IEC-6 and HT-29 cells was associated with the stimulation of cell differentiation because we did not observe any stimulatory effect of corticosteroids on cultures of IEC-6, HT-29, and Caco-2 cells in higher stages of differentiation. Nevertheless, we report for the first time that enterocyte differentiation, induced either spontaneously (Caco-2 cells) or by appropriate manipulations (HT-29, IEC-6), increases the expression of 11βHSD2 activity independent of corticosteroids. In view of these findings and the increased immunoreactivity of 11βHSD2 positive T84 cells cultured in the presence of sodium butyrate [31], our current findings raise the possibility that intestinal 11βHSD2 activity parallels differentiation. As higher expression of 11βHSD2 in the intestine is more localized to surface enterocytes than to less differentiated crypt cells [8,9], it is obvious that the differentiation process must play a role not only in the distribution of 11βHSD2 along the crypt axis, but also in functional processes regulated by corticosteroids. Indeed, the expression of 11βHSD2 displays coexpression along the crypt axis with mineralocorticoid receptors, which are expressed predominantly in differentiated surface cells whereas it is quite opposite to the distribution of glucocorticoid receptors [10].

In conclusion, the present data suggest that the ability of intestinal cells to metabolize glucocorticoids via 11βHSD2 is a feature of the differentiated enterocytes and that upregulation of 11βHSD2 by cytodifferentiation is independent of corticosteroids. The findings support the hypothesis that regulation of intestinal 11βHSD activity might play a role in the functional differentiation of intestinal epithelium.

Acknowledgments

We wish to thank Mrs. I. Mezteková and R. Somolová for technical assistance. This research was funded by grants from the GA ČR (grant # 306/99/0210) and from the Academy of Sciences (grant # A7011713).

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