Korean J. of Pharmacology
Vol. 26, No. 1, pp. 67-76, 1990

Influences of Hydrocortisone, DHEA, Estradiol and Testosterone on the Hepatic and Intestinal Polyamine Metabolism of Castrated Mice

Sang-Hyun Choi, Boe-Gwun Chun*, Nam-Hun Kim and Yeon-Sook Chun

Department of Pharmacology, Korea University College of Medicine, Seoul 110-522, Korea

ABSTRACT

Hydrocortisone 50 mg/kg (HC), dehydroepiandrosterone 250 mg/kg (DHEA), β-estradiol 5 mg/kg (E2), and testosterone 20 mg/kg (TS) were subcutaneously injected into the castrated ICR mice at noon for four days, and the animals were sacrificed at 10-12 A.M. of the fifth day.

The intestinal DAO activity was significantly decreased by HC, but it was rather increased by E2 and TS, respectively. And DHEA did not change the DAO activity. But the hepatic MAO activity was not affected by anyone of HC, DHEA, E2, and TS. Aminoguanidine 25 mg/kg produced the marked decrease of the intestinal DAO activity and the significant increases of the intestinal PT and SD contents, but it did not change the hepatic polyamine contents. HC and DHEA induced the significant increase of the intestinal PT content. E2 induced the marked increase of the hepatic PT content and the moderate increase of the intestinal PT content. TS little affected the polyamine contents of the liver and intestine.

These results suggest that the E2-induced increase of the hepatic PT content is rather ascribed to the greater enhancement of PT synthesis than the inhibition of polyamine catabolism, and that the HC-induced increase of the intestinal PT content is due partly to the inhibition of polyamine catabolism via DAO.

Key Words: Polyamine metabolism, Diamine oxidase, Hydrocortisone, Estradiol, DHEA, Testosterone

Abbreviations: HC, hydrocortisone; E2, estradiol; DHEA, dehydroepiandrosterone; TS, testosterone; PT, putrescine; SD, spermidine; SM, spermine; DAO, diamine oxidase; MAO, monoamine oxidase; ODC, ornithine decarboxylase; SAM-DC, S-adenosylmethionine decarboxylase

INTRODUCTION

The diamine putrescine and the polyamine spermidine and spermine have been postulated to be essential for growth and differentiation and to be capable of interacting with many metabolic processes and transmembrane signalling, although their mechanism of action at the molecular level remains largely to be understood (Williams-Ashman and Canellakis, 1979; Pegg and McCann, 1982; Tabor and Tabor, 1984; Pegg, 1986; Seiler, 1987; Caldarera *et al.*, 1990).

However, it has been recently shown that the changes in tissue polyamine contents might be thought essential in the processes of adaptation in responses to partial hepatectomy (Pösö and Jänne, 1976; Luk, 1986; Choi *et al.*, 1988), hepatotoxic CCl_4 (Pegg *et al.*, 1985), cytotoxic mucosal injury (Luk *et al.*, 1980), and jejunectomy, pancreaticobiliary diversion, and poststarvation refeeding (Luk and Baylin, 1984; Luk and Yang, 1987; Hösömi *et al.*, 1987a and 1987b; Luk and Yang, 1988).

In rabbit experiments, the degree of ischemic bowel disease varied with the intestinal DAO activity, the final catabolic enzyme of polyamine (Kusche *et al.*, 1981; Seiler *et al.*, 1985). α-Aminoguanidine, a DAO inhibitor (Okuyama and Kobayashi, 1961; Seiler *et al.*, 1985) shortened the survival time of the animals after vascular occlusion of the superior mesenteric artery (Kusche *et al.*, 1979), and the shortening of the survival time was reversed by ad-

* Correspondence Auther

ministration of a histamine-receptor blockade such as cimetidine (Kusche *et al.*, 1981). However, the intestinal DAO activity of newborn rat pups could be induced by hydrocortisone (Karp *et al.*, 1987), and the prenatal administration of glucocorticoid decreased the incidence of necrotizing enterocolitis (Bauer *et al.*, 1984).

17 β-Estradiol and testosterone have been shown to stimulate both ODC and SAM-DC activities in the uterus of immature rats (Kaye *et al.*, 1971) and in the prostate of castrated rats (Fjosne, *et al.*, 1988), respectively. These findings suggest to us that the effects of several steroid hormones on polyamine metabolism may be involved in the homeostatic processes of several visceral organs against the pathogenic or toxic insults. So, the influences of hydrocortisone, DHEA, estradiol, and testosterone on the hepatic and intestinal polyamine contents, the intestinal DAO activity, and the hepatic MAO activity were studied in castrated male ICR mice.

MATERIALS AND METHODS

Materials

Hydrocortisone acetate (HC), dehydroepiandrosterone (DHEA), estradiol cypionate (E2), and testosterone cypionate (TS) were purchased from Upjohn. α-Aminoguanidine sulfate, putrescine, spermidine, spermine, histidine, 4-hydroxy-3-methoxyphenylacetic acid (homovanilic acid), and horseradish peroxidase (type I) were purchased from Sigma. 1,8-diaminooctane, 4-fluoro-3-nitrobenzotrifluoride, dimethylsulfoxide, and 2-methylbutane were from Aldrich.

Methanol and acetonitrile were HPLC-grade. And other chemicals were analytical grade. Male ICR mice, weighing 17-20 g, were supplied from Korea Experimental Animal Lab. Company.

Treatments of animals

Ten male mice were kept to a cage and allowed acclimated to a 12 hr light (7 AM to 7 PM) and 12 hr dark cycle for one week before being studied. The mice were subjected to bilateral castration at 11 A.M. under light diethyl ether anesthesia (Waynforth, 1980), and the sham-operated mice underwent similar surgical procedure. One hour after that, the administration of steroid hormones was started. Mice were subcutaneously injected with HC 50 mg/kg or E2 5 mg/kg in cotton seed oil of 0.18% benzyl alcohol, and with DHEA 250 mg/kg or TS 20 mg/kg. α-Aminoguanidine sulfate was intraperitoneally injected in 0.85% NaCl solution.

Polyamine HPLC analysis

Apparatus: The high performance liquid chromatography (HPLC) system was consisted of a Gilson HPLC pump, a Rheodyne 7125 injection valve, a ERC ODS-1161 column (3μm; 6$\times$100mm), a Knauer variable UV/VIS spectrometer, and a Linear dual-channel chart recorder.

HPLC analysis: The extraction process was based upon that of Choi *et al.* (1989). Mouse livers and intestines excised after decapitation were first freezed on dry ice powder and then stored in freezers. Within 7 days after their sampling, both tissues were homogenized with a teflon homogenizer in 4 volumes of 0.4 M perchloric acid containing 2mM disodium EDTA and diaminooctane (50-100μg) as an internal standard.

One ml of the homogenate was spun at 15,000$\times$g for 10 min and 100μl of the supernatant was evaporated to dryness with streams of nitrogen gas at room temperature. And the 4-fluoro-3-nitrobenzotrifluoride (FNBT) derivatization of polyamines and the HPLC analysis condition were those originally described by Spragg and Hutchings (1983).

The N-2'-nitro-4'-trifluoromethylphenyl (NTP) polyamine derivatives in the 20μl of methanol extract were quantitatively analyzed on a isocratic HPLC system equipped with an ODS column, using diaminooctane as an internal standard (Spragg and Hutchings, 1983; Choi *et al.*, 1989). The recovery rates of NTP-polyamine derivatives were over 94.4%, and the calibration curves of them were consistently linear over a range of 50 picomole to 10 nanomole with the variations of less than 5% between identical samples, and the detection limit was less than 10 picomole on column with a S/N ratio of 5:1.

Assays of monoamine and diamine oxidases

After decapitation, the mouse liver and small intestine were quickly excised and extensively washed with ice-cold 0.85% NaCl-saline, and then the tissue (200-250mg) was homogenized with 4 volumes of 0.1M sodium potassium phosphate buffer, pH 7.8, using a teflon homogenizer, and the homogenate was spun at 10,000$\times$g for 15 min at 2°C. The precipitate of the liver homogenate and the supernatant of the intestinal homogenate were used for the assays of liver MAO and intestinal DAO activities, respectively, according to the method described originally by Snyder and Hendley (1968).

The liver precipitate obtained above was resuspended with 1.0 ml of 0.1 M sodium potassium phosphate buffer, pH 7.8. The incubation mixture was made up to contain in a final 3 ml of 0.1 M sodium potassium phosphate buffer, pH 7.8: 0.1 ml of the phosphate suspension of liver precipitate or the supernatant of intestinal homogenate, 40μg of horseradish peroxidase, 160μg of homovanillic acid, and 100μg of tyramine or putrescine as a MAO or DAO substrate, respecgively. The mixture reaction was proceeded for 60 min at 37°C, and then stopped by chilling the tube in ice-cold water. After centrifugation at 15,000×g for 10 min at 4°C, the fluorescence intensity of the supernatant was measured in an Aminco-Bowman spectrophotofluorometer at an excitation wave length of 325nm and emission of 415nm. The results were expressed as H_2O_2 nanomole formed/hr per mg of protein in the initial phosphate homogenate. The protein content was determined by the method of Lowry *et al.* (1951).

RESULTS

Effects of steroid hormones and aminoguanidine on intestinal DAO and liver MAO activities

As shown in Fig. 1, treatments of castrated male mice with 100mg/kg of aminoguanidine, a specific DAO inhibitor (Okuyama and Kobayashi, 1961; Seiler *et al.*, 1985), did not affect the hepatic MAO activity, but the intestinal DAO activity was markedly decreased at 6 and 24 hrs after the treatment to 41.8% and 41.4% of the control values, respectively. Injection once a day for 4 days with HC (50mg/kg), DHEA (250mg/kg), E2 (5mg/kg), and TS (20mg/kg) did not change the hepatic MAO activity of castrated mice (Fig. 2).

However, the intestinal DAO activity of castrated mice was markedly decreased from 17.23 to 10.71 nanomole H_2O_2/mg protein/hr by HC, but the DAO activity was moderately increased by E2 and TS to 128.8% and 133.8% of the control values (Fig. 2). DHEA did little affect both MAO and DAO activities.

Effects of steroid hormones and aminoguanidine on the liver and intestinal polyamine contents

Like to the MAO activity, the hepatic polyamine contents were not changed by aminoguanidine (Fig. 3). But the intestinal putrescine content was significantly increased 6 hr and 24 hr after aminoguanidine injection by 49.2% and 69.7%, respectively. And the intestinal spermidine content was also significantly increased 24 hr after the injection by

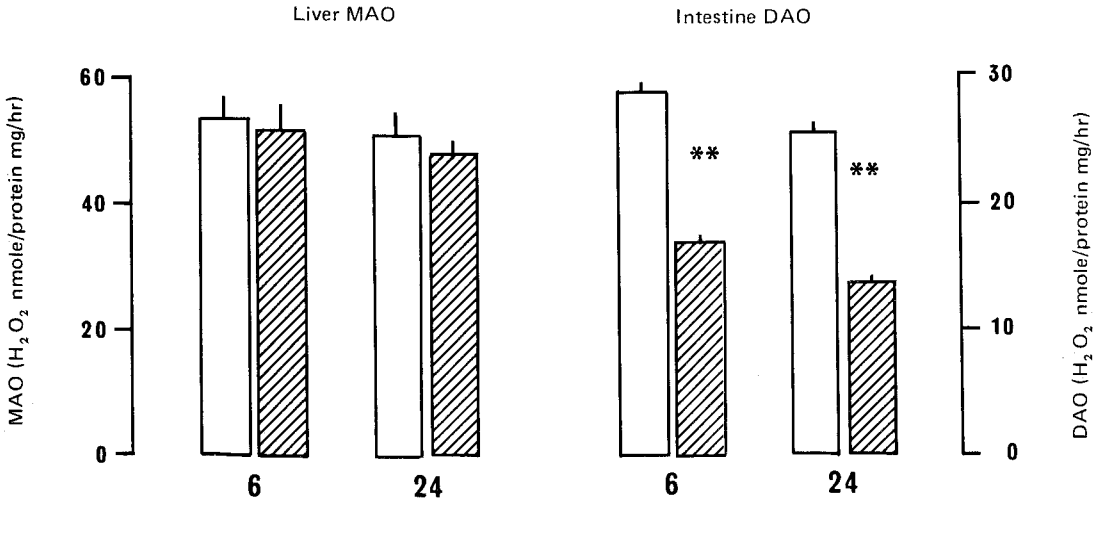

Fig. 1. Influence of aminoguanidine on the activities of the liver MAO and the intestine DAO. Each column and bar represents the mean and standard error of 8 data.
** indicates $p<0.02$.
□: control - isotonic saline, ▨: aminoguanidine

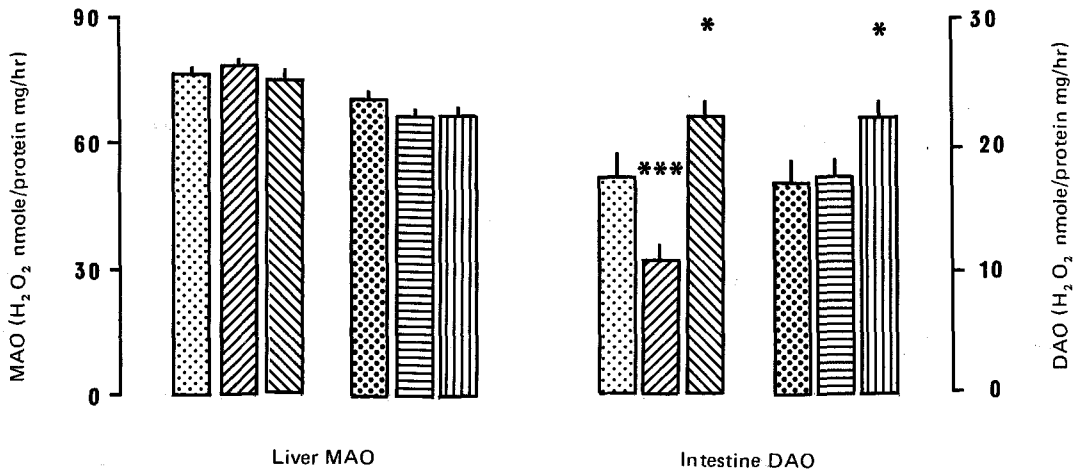

Fig. 2. Influences of hydrocortisone, estradiol, DHEA and testosterone on the activities of the liver MAO and the intestine DAO. * and *** indicate $p<0.05$ and $p<0.01$, respectively.

Control groups -

▨: cotton seed oil of 0.18% benzyl alcohol,

▨: cotton seed oil

Experimental groups -

▨: hydrocortisone, ▨: estradiol,

▨: DHEA, ▨: testosterone

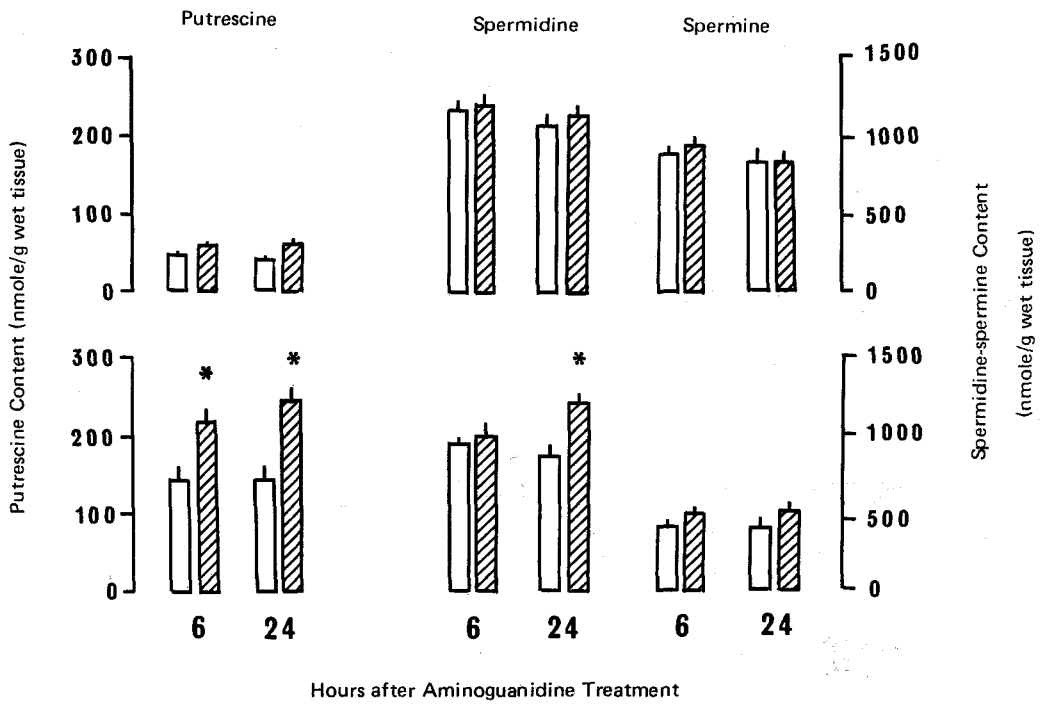

Fig. 3. Influence of aminoguanidine on the contents of putrescine, spermidine and spermine in the liver (upper figure) and in the intestine (lower figure).

□: control - isotonic saline, ▨: aminoguanidine

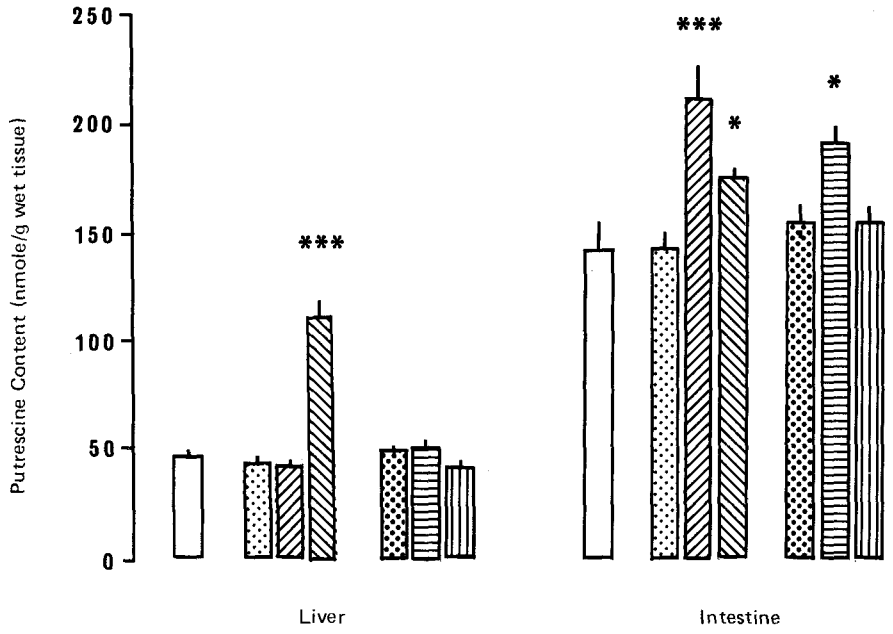

Fig. 4. Influences of hydrocortisone, estradiol, DHEA and testosterone on the putrescine contents of the liver and the intestine.
□: normal group
Control groups -
▨: cotton seed oil of 0.18% benzyl alcohol,
▩: cotton seed oil
Experimental groups -
▨: hydrocortisone, ▧: estradiol,
▤: DHEA, ▥: testosterone

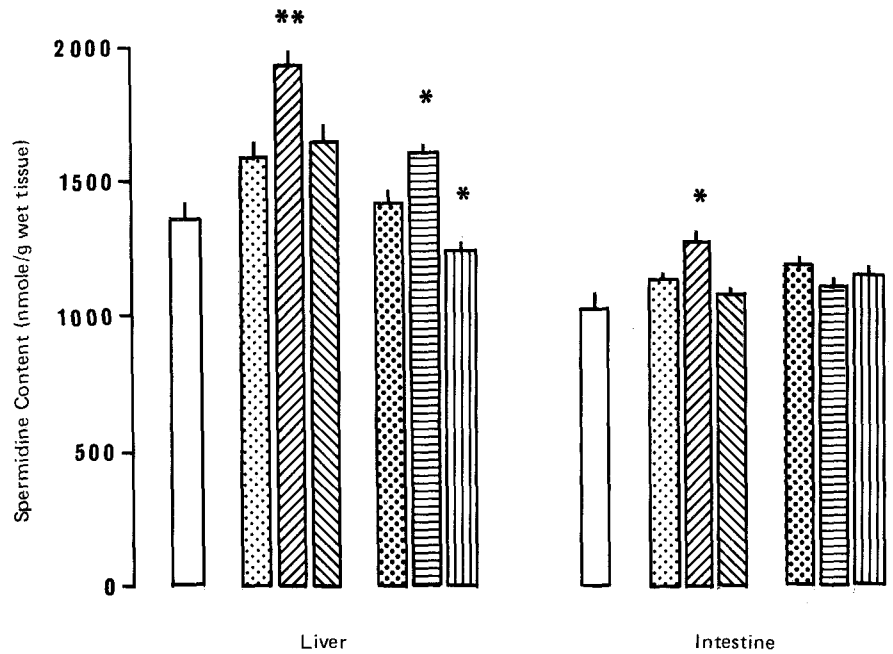

Fig. 5. Influences of hydrocortisone, estradiol, DHEA and testosterone on the spermidine contents of the liver and the intestine.

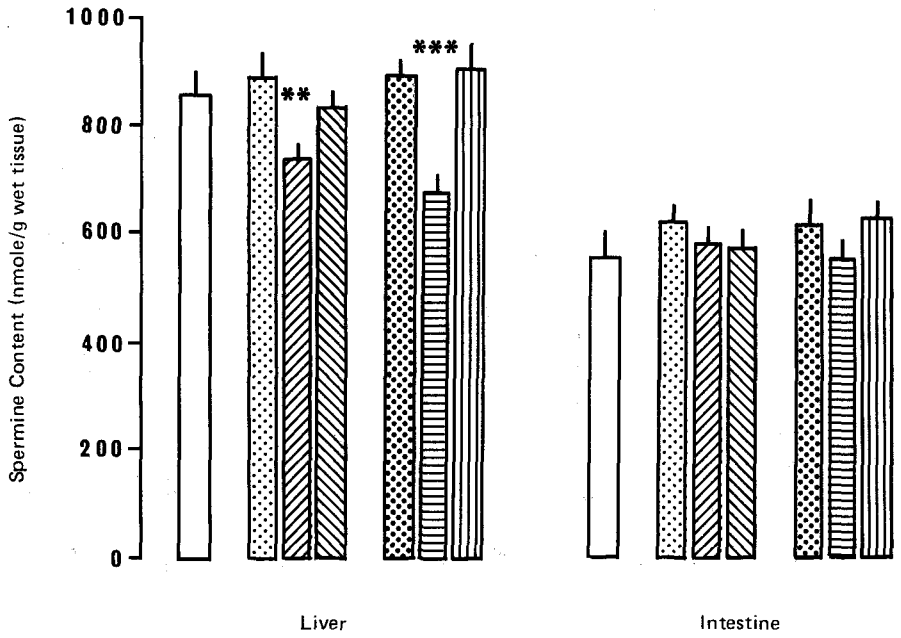

Fig. 6. Influences of hydrocortisone, estradiol, DHEA and testosterone on the spermine contents of the liver and the intestine.

37.9%. However, the intestinal spermine content was not changed by aminoguanidine (Fig. 3).

The treatment of mice with HC (50 mg/kg), DHEA (250 mg/kg), or E2 (5 mg/kg) once a day for 4 days, significantly increased the intestinal putrescine content by 49.0%, 23.5%, or 23.9%, respectively, but TS did not affect the content (Fig. 6). And the hepatic putrescine content was not changed by HC, DHEA, or TS, but markedly increased by E2 up to 245.7% of the control value (Fig. 4). It is noteworthy that the intestinal putrescine content was significantly increased by HC and the hepatic content was markedly increased by only E2. But the hepatic spermidine content of castrated mice was moderately increased by HC to 126.9% of the control value and little affected by other steroid hormones (Fig. 5). Similarly, the intestinal spermidine content was moderately increased by only HC and little changed by other steroids (Fig. 5). However, the hepatic spermine content of castrated mice was significantly decreased by DHEA to 76.4% and was moderately decreased by HC to 82.9%, comparing with the control value of each (Fig. 6). E2 and TS did not affect the hepatic spermine content. But the intestinal spermine content of castrated mice was not significantly changed by all of the steroids (Fig. 6).

DISCUSSION

There is a reported association between administration of prenatal glucocorticoids and a decreased incidence of necrotizing enterocolitis in human infants (Bauer *et al.*, 1984). In animal experiments, the degree of ischemic bowel disease correlates negatively with intestinal DAO activity (Kusche *et al.*, 1979; 1981), suggested that one important factor in gut maturity that relates to the risk of necrotizing enterocolitis may be histamine-DAO activity.

However, the enhanced DAO activity in rapidly growing conditions may represent a means by which the excess of polyamine can be controlled (Perin *et al.*, 1983; 1986), and the increased ODC activity with mucosal proliferation has been observed, regenerating mucosa after cytotoxic insults (Luk *et al.*, 1980), and the adaption conditions of several organs such as the intestinal hyperplasia showed in response to poststarvation refeeding (Luk and Yang, 1987; 1988; Ulrich-Baker *et al.*, 1988) and jejunectomy or pancreatico-biliary diversion (Luk and Yang, 1987; Hösömi *et al.*, 1987a; 1987b). Furthermore, the indices of mucosal proliferation, including new DNA

synthesis, crypt cell proliferation, and crypt cell labelling index, were highly correlated with the time course of the increase in ODC activity (Luk and Baylin, 1984). And ODC activity increases rapidly and markedly in regenerating liver after partial hepatectomy (Luk, 1986; Choi *et al.*, 1989) and participates in the cAMP-dependent activation of the hepatic drug-metabolizing enzyme by several xenobiotic drugs (Russell and Haddox, 1979).

However, hydrocortisone or dexamethasone increased the ODC activity in freshly isolated rat hepatocytes (Lumeng, 1979), regenerating rat livers (Thrower and Ord, 1974), or in rat brains (Anderson and Schanberg, 1975); the SAM-DC and spermidine synthetase of mouse mammary glands (Oka *et al.*, 1982), and the DAO activity in newborn rat intestines (Karp *et al.*, 1987). E2 stimulated both ODC (Cohen, *et al.*, 1970; Kaye, *et al.*, 1971) and SAM-DC (Kaye *et al.*, 1971). And 5'-methylthioadenosine phosphorylase activity in the rat uterus was also increased by E2 (Nicolette *et al.*, 1980), and that emzyme in the ventral prostate of rats was stimulated by TS (Danzin *et al.*, 1979; Nicolette *et al.*, 1980). Both ODC and SAM-DC activities were markedly decreased in response to castration, but their decreased activities were recovered to control values by dihydrotestosterone (Russell and Haddox, 1979; Fjosne *et al.*, 1988). DHEA is more abundantly synthesized than other steroids and circulates at very high levels (greater than 10 times of cortisol level), but little is known about its physiological activities, and the receptor for it has not been discovered (Norman and Litwack, 1987). However, it is apparantly important in fetal development, in supplying cells with a precursor for androgen and estrogen synthesis, and for certain tissue-protective functions which are as yet not understood (Norman and Litwack, 1987).

Barrett-Connor *et al.* (1986) suggested that the plasma level of DHEA-sulfate might confer protection against death, particularly from cardiovascular diseases. While, in an experiment which was designed to assist in clarifying the role of DAO in the polyamine transformations, there was a linear relationship between the putreanine content of mouse liver and the time after intraperitoneal injection of spermidine (Seiler *et al.*, 1983).

So, in the present study, the influences of HC, DHEA, E2, and TS on the intestinal DAO activity and the polyamine contents in the liver and the intestine were investigated in castrated male mice, referring their effects on the liver MAO activity. Injection once a day for 4 days with HC, DHEA, E2, or TS did not changed the liver MAO activity. While, the intestinal DAO activity was markedly decreased by HC but rather increased by E2 and TS, respectively. HC and DHEA significantly increased the intestinal putrescine content and the liver spermidine content but moderately decreased the liver spermine content. And E2 significantly increased the liver PT content but did not show any significant effect on the other polyamine contents measured in this study.

Those findings may be in agreement with Rojansky *et al.* (1979) and seem to be supported by the results in this study showing that after treatment with aminoguanidine, a specific DAO inhibitor (Okuyama and Kobayashi, 1961; Seiler *et al.*, 1985), the intestinal DAO was selectively inhibited and the intestinal putrescine and spermidine contents were increased, similar to other previous papers (Seiler *et al.*, 1983; 1985).

And the HC-induced increase of the intestinal putrescine and liver spermidine contents can be considered in connection with several studies (Lumeng, 1979; Oka *et al.*, 1982).

But the decrease-induced by HC of the intestinal DAO activity is opposed to the result of Karp *et al.* (1987).

The observed difference seems to be attributable to the newborn rat used by Karp *et al.* (1987); in their study, HC increased the intestinal DAO activity when the injection of HC was begun on 4, 6, or 8 days of life, but did not, really, increase when its injection was started on 10 days of life. In the present study, the intestinal DAO activity was moderately increased by E2 and TS, but the hepatic MAO activity was not changed.

The results may be, in consideration of the 1,000 fold increase of plasma DAO activity in pregnant women (Russell and Durie, 1978), suggesting that E2 can induce the catabolism of polyamines.

But E2 significantly increased the hepatic putrescine content, and, to lesser extent, increased the intestinal content. However, the other polyamine contents of the liver or intestine were little affected by E2. These are consistent with previous reports (Cohen *et al.*, 1970; Kaye *et al.*, 1971), demonstrating that E2 may enhance more greatly the ODC activity than the DAO activity and those effect can be more dominant in the liver than in the intestine.

On the other hand, although TS moderately increased the intestinal DAO activity, TS did not produce any meaningful change of the polyamine contents in the liver and intestine of castrated male mice, suggesting that TS does much less affect the polyamine metabolism of the liver and intestine than that of the prostate and kidney, in which both ODC

and SAM-DC activities are markedly dependent upon androgenicity (Isomaa *et al.*, 1983; Seely and Pegg., 1983; Fjosne *et al.*, 1988).

In summary, the results obtained in the present study suggest that E2 may enhance more greatly putrescine synthesis in the liver, comparing with its effect on the catabolism of polyamine, that HC may, unlike E2, increase in intestinal putrescine content due partly to the inhibition of polyamine catabolism via DAO, and that the modes of the polyamine metabolism in the liver and intestine, in response to steroid hormones such as HC, E2, and TS, are distinguishably different from each other.

REFERENCES

Anderson TR and Schanberg SM: *Effect of thyroxine and cortisol on brain ornithine decarboxylase activity and swimming behavior in developing rat. Biochem Pharmacol 24:495-501, 1975*

Barrett-Connor E, Khaw K-T and Yen SSC: *A prospective study of dehydroepiandrosterone sulfate, mortality, and cardiovascular disease. N Eng J Med 315:1519-1524, 1986*

Bauer CR, Morrison JC, Poole WK, Korones SB, Boehm JJ, Rigatto H and Zachman RD: *A decreased incidence of necrotizing enterocolitis after prenatal glucocorticoid therapy. Pediatrics 73:682-688, 1984*

Caldarera CM, Tantini B, Pignatti C and Clo C: *A possible involvement of polyamines in the regulation of transmembrane signalling events. In The Biology and Chemistry of Polyamines (ed. SH Goldemberg and ID Algranati), IRL Press, pp 19-28, 1990*

Choi S-H, Kim H-G, Park H-I and Chun B-G: *A simple, sensitive, and specific HPLC analysis of tissue polyamines using FNBT derivatization: Its application on the study of polyamine metabolism in regenerating rat liver. Kor J Pharmacol 24:233-240, 1988*

Choi S-H, Lee H-J, Chun B-G and Chun Y-S: *Influences of adrenoceptor blockades and nifedipine on Polyamine metabolism in regenerating rat liver. Kor J Pharmacol 25: 199-207, 1989*

Cohen S, O'Malley BW and Stastny M: *Estrogenic induction of ornithine decarboxylase in vivo and in vitro. Science (NY) 170:336-338, 1970*

Fjosne HE, Strand H, Ostensen MA and Sunde A: *Ornithine decarboxylase and S-adenosylmethionine decarboxylase activity in the accessory sex organs of intact, castrated, and androgen-stimulated castrated rats. Prostate 12:309-320, 1988*

Hösömi M, Lirussi F, Stace NH, Vaja S, Murphy GM and Dowling RH: *Mucosal polyamine profile in normal and adapting (hypo and hyperplastic) intestine: effects of DFMO treatment. Gut 28(S1):103-107, 1987a*

Hösömi M, Stace NH, Lirussi F, Smith SM, Murphy GM and Dowling RH: *Role of polyamines in intestinal adaptation in the rat. Eur J Clin Invest 17:375-385, 1987b*

Isomaa VV, Pajunen AE, Bardin CW and Janne OA: *Ornithine decarboxylase in mouse kidney. Purification, characterization, and radioimmunological determination of the enzyme protein. J Biol Chem 258:6735-6740, 1983*

Karp WB, Robertson AF and Kanto WP Jr: *The effect of hydrocortisone, thyroxine, and phenobarbital on diamine oxidase activity in new born rat intestine. Pediatr Res 21:368-370, 1987*

Kaye AM, Icekson I and Linder HR: *Stimulation by estrogens of ornithine and S-adenosylmethionine decarboxylases in the immature rat uterus. Biochim Biophys Acta 252:150-159, 1971*

Kusche J, Lorenz W, Stahlknecht CD, Richter H, Hesterberg R, Schmal A, Hinterlag E, Weber D and Ohmann C: *Intestinal diamine oxidase and histamine release in rabbit mesenteric ischemia. Gasteroenterology 80:980-987, 1981*

Kusche J, Stahlknecht CD, Lorenz W, Reichert G and Dietz W: *Comparison of alterations in the histamine-diamine oxidase system during acute intestinal ischemia in pigs, dogs, and rabbits: evidence for a uniform pathophysiological mechanism. Agents Actions 9:49-52, 1979*

Lowry OH, Rosebrough NJ, Farr AL and Randall RJ: *Protein measurement with the folin phenol reagent. J Biol Chem 193:265-275, 1951*

Luk GD: *Essential role of polyamine metabolism in hepatic regeneration: Inhibition of deoxyribonucleic acid and protein synthesis and tissue regeneration by difluoromethylornithine in the rat. Gastroenterology 90:1261-1267, 1986*

Luk GD and Baylin SB: *Inhibition of intestinal epithelial DNA synthesis and adaptive hyperplasia after jejunectomy in the rat by suppression of polyamine biosynthesis. J Clin Invest 74:688-704, 1984*

Luk GD and Yang P: *Polyamines in intestinal and pancreatic adaptation. Gut 28(S1):95-101, 1987*

Luk GD and Yang P: *Distribution of polyamines and their biosynthetic enzymes in intestinal adaptation. Am J Physiol 254:G194-G200, 1988*

Lumeng L: *Hormonal control of ornithine decarboxylase in isolated liver cells and the effect of ethanol oxidation. Biochim Biophys Acta 587-556-566, 1979*

Manni A, Wright C, Feil P, Baranao L, Demers L, Garcia M and Rochefort H: *Autocrine stimulation by estradiol-regulated growth factors of rat hormone-responsive mammary cancer: interaction with the polyamine pathway. Cancer Res 46:1594-1598, 1986*

Nicolette JA, Wrobel NC and Perro AJ: *The stimulatory effect of testosterone propionate and 17-β* estradiol on 5'-methylthioadenosine phosphorylase activity in rat target tissues. Biochim Biophys Acta 627-190-198, 1980

Norman AW and Litwack G: *Adrenal corticoids. In Hormones (ed. AW Norman and G Litwack), Academic Press, chap. 10, pp 397-448, 1987*

Oka T, Perry JW and Rerada N: *The regulatory function of spermidine in hormonal control of the development of mouse mammary gland in culture. Fed Proc 41:3073-3077, 1982*

Okuyama T and Kobayashi Y: *Determination of diamine oxidase activity by liquid scintillation counting. Arch Biochem Biophys 95:242-250, 1961*

Pegg AE and McCann PP: *Polyamine metabolism and function. Am J Physiol 243:C212-C221, 1982*

Perin A, Sessa A and Desiderio MA: *Polyamine levels and diamine oxidase activity in hypertrophic heart of spontaneously hypertensive rats and of rats treated with isoproterenol. Biochim Biophys Acta 755:344-351, 1983*

Perin A, Sessa A and Desiderio MA: *Response of tissue diamine oxidase activity to polyamine administration. Biochem J 235:119-123, 1986*

Pösö H and Jänne J: *Inhibition of ornithine decarboxylase activity and spermidine accumulation in regenerating rat liver. Biochem Biophys Res Commun 69:885-892, 1976*

Rojansky N, Neufeld E and Chayen R: *Excretion of polyamines by the pregnant rat following ibhibition of diamine oxidase. Biochim Biophys Acta 586:1-9, 1979*

Russell D and Snyder SH: *Amine synthesis in rapidly growing tissue: Ornithine decarboxylase activity in regenerating rat liver, chick embryo, and various tumors. Proc Nat Acad Sci, USA, 60:1420-1427, 1968*

Russell DH and Haddox MK: *Cyclic AMP-mediated induction of ornithine decarboxylase in normal and neoplastic growth. In Adv. Enzyme Regul. vol 7 (ed. G Weber), Pergamon Press, pp 61-87, 1979*

Russell DH and Durie BGM: *Polyamines as biochemical markers of normal and malignant growth. Prog Cancer Res Ther 8:121-165, 1978*

Seely JE and Pegg AE: *Changes in mouse kidney ornithine decarboxylase activity are brought about by changes in the amount of enzyme protein as measured by radioimmunoassay. J Biol Chem 258:2496-2500, 1983*

Seiler N, Knodgen B, Bink G, Sarhan S and Bolkenius F: *Diamine oxidase and polyamine catabolism. In Adv in Polyamine Res, vol 4 (ed. U Bachrach, A Kaye and R Chayen), Raven Press, pp 135-154, 1983*

Seiler N, Bolkenius FN and Knodgen B: *The influence of catabolic reactions on polyamine excretion. Biochem J 225:219-226, 1985*

Seiler N: *Functions of polyamine acetylation. Can J Physiol Pharmacol 65:2024-2035, 1987*

Spragg BP and Hutchings AD: *High-performance liquid chromatographic determination of putrescine, spermidine and spermine after derivatization with 4-fluoro-3-nitrobenzotrifluoride. J Chromatogr 258: 289-291, 1983*

Snyder SH and Hendley ED: *A simple and sensitive fluorescence assay for monoamine oxidase and diamine oxidase. J Pharmacol Exp Ther 163:386-392, 1968*

Thrower S and Ord MG: *Hormonal control of liver regeneration. Bichem J 144:361-369, 1974*

Ulrich-Baker MG, Wang P, Fitzpatrick L and Johnson LR: *Amiloride inhibits rat mucosal ornithine decarboxylase activity and DNA synthesis. Am J Physiol 254:G408-G415, 1988*

Waynforth HB: *Orchiectomy (castration). In Experimental and Surgical technique in the rat., Academic Press, pp 160-161, 1980*

Williams-Ashman HG and Canellakis ZN: *Polyamines in mammalian biology and medicine. Perspect Biol Med 22:421-453, 1979*

=국문초록=

Hydrocortisone, DHEA, Estradiol 및 Testosterone에 의하여 나타나는 마우스-간 및 소장 Polyamine 대사의 변동에 관한 연구

고려대학교 의과대학 약리학교실

최상현 • 전보권 • 김남헌 • 천연숙

웅성-마우스의 고환을 diethyl ether 마취하에서 제거하고, 수종의 steroid 홀몬을 각각 매일 1회씩 4일간 피하주사하여, 간 및 소장의 polyamine 함량과 소장의 diamine oxidase (DAO) 활성도에 미치는 그들의 영향을 검색하였다.

1. Hydrocortisone succinate 50 mg/kg (HC) 및 dehydroepiandrosterone 250 mg/kg (DHEA)에 의하여, 소장의 putrescine (PT)은 유의하게 증가되었으나, spermidine (SD) 및 spermine (SM)은 별 영향을 받지 않았고, 간의 SD은 다소 증가되고, SM은 다소 감소 되었으나, PT은 별 변동을 보이지 않았다.
2. Estradiol cypionate 5 mg/kg (E2)에 의하여, 간의 PT은 현저히 증가되었으나, 소장의 PT은 다소 증가되었고, 그외 소장 및 간의 SD와 SM의 변동은 보이지 않았다.
   Testosterone cypionate 5 mg/kg (TS)에 의하여는 간의 SD이 다소 감소되었을 뿐 별 변동이 없었다.
3. 소장의 DAO 활성도는 HC에 의하여 현저히 감소되었으나, E2 및 TS에 의하여는 유의하게 증가되었고, DHEA에 의하여는 별 영향을 받지않았다. 그러나 간의 monoamine oxidase 활성도는 HC, E2, DHEA, 및 TS에 의하여 영향을 받지 않았다.
4. Aminoguanidine 25 mg/kg로 소장의 DAO 활성도가 현저히 감소되었으나, 간 MAO 활성도는 영향을 받지 않았고, 소장의 PT 및 SD은 유의하게 증가되었으나, 간의 polyamine은 별 변동을 보이지 않았다.

이상의 결과로 미루어 볼때, 간 및 소장의 polyamine 대사—특히 PT 함량의 변동이 각각 E2 및 HC에 의하여 특이적으로 조절되는 바, E2에 의한 간 PT 함량의 증가는 주로 생성촉진 작용에 연유되며, HC에 의한 소장 PT 함량의 증가는 주로 polyamine의 이화성 대사를 억제함에 기인될 수 있는 것으로 사료된다.