Biochemical Pharmacology, Vol. 29, p. 3017.

0006-2952/80/1101-3017 $02.00/0

SHORT COMMUNICATIONS

In vitro inhibition of the absorption of oestradiol-17β in rat intestine by acetylsalicylic acid

(*Received* 4 *February* 1980; *accepted* 9 *May* 1980)

It is now established that orally administered oestradiol-17β and ethinyl oestradiol are absorbed from the intestinal lumen after metabolic conversion in the intestinal wall to oestrone-3-glucuronide and ethinyloestradiol-3-glucuronide, respectively [1–3]. This gut wall metabolism plays a significant role in reducing the oral bioavailability of these compounds [3].

Other factors that may affect the oral bioavailability of oestrogens via an effect on intestinal absorption include the simultanuous presence of other drugs and the effect of dietary constituents. Although attention has been paid in recent years to drug–drug interactions in oestrogen metabolism, much of the interest has centred on the effect of other drugs on the plasma clearance rates and hepatic metabolism [4–6] of oestrogens, such as occurs when oral contraceptives are used. Little or no information is available on the possible effects of drugs at the intestinal level.

We report here the effect of the commonly used analgesic, acctylsalicylic acid (a.s.a.), on the *in vitro* absorption of oestradiol from rat intestine.

Intestinal sacs 3.5 cm in length were prepared from the duodenal segments of female albino rats (Wistar stain) weighing 200–230 g, according to the method of Wilson and Wiseman [7]. The mucosal medium consisted of 2 ml 0.1M phosphate buffer, pH 7.0, containing 11 mM KCl, 13.4 mM NaCl, $CaCl_2$, 20 mM glucose, 3.35 pM [2,4,6,7-^{3}H]oestradiol-17β (sp. act. 100Ci/mmole; Radiochemical Centre, Amersham, U.K.) and 3.0 or 30 mM acetylsalicylic acid. The serosal medium consisted of 2 ml of the above solution in an incubation tube but without oestradiol and a.s.a. Control sacs were set up simultaneously as above but without a.s.a. Filled sacs were incubated at 30° for 3 hr in a metabolic incubator. Aliquots (20 μl) of serosal medium were taken at 15, 30, 60, 120 and 180 min and mixed with 10 ml Instagel (Packard Instruments Co.) and the radioactivity determined in a Packard 'Tricarb' liquid scintillation spectrometer.

In each sample the unconjugated oestradiol and its unconjugated metabolites in the serosal medium were extracted twice with 5 vol. diethyl ether. The combined ether extracts were dried under a stream of nitrogen and chromatographed on thin-layer plates of silica gel using the solvent system benzene–ethanol (9:1 v/v).

The conjugated oestrogens in the serosal medium were hydrolysed in 2 N hydrochloric acid (100°, 30 min) before ether extraction and chromatography as described above.

The results were expressed as net transport [2], i.e. radioactivity transported from mucosal to serosal medium. The results in Fig. 1 therefore showed that at a concentration of 30 mM a.s.a. net transport of oestradiol was significantly inhibited compared to controls ($P < 0.05$ at 30 min, $P < 0.001$ at 60, 120 and 180 min, paired *t*-test). Analysis of the radioactive fractions separated by thin-layer chromatography showed that significant differences between experimental (30 mM a.s.a.) and control samples were only associated with the fraction of the hydrolysed conjugate chromatographing with the mobility of oestrone. Radioactivity in this fraction was reduced by 44 per cent compared to controls at 180 min.

Thus it is apparent that in the process of absorption, the oxidation of oestradiol to oestrone or the conjugation of oestrone was inhibited by acetylsalicylic acid.

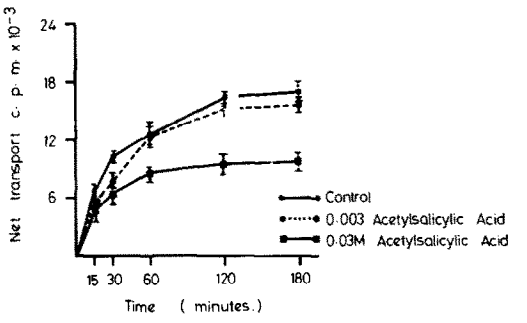

Fig. 1. Effect of acetylsalicylic acid on the time course of the transport of [2,4,6,7-^{3}H]oestradiol-17β *in vitro* by rat intestinal sacs. Each point represents the mean ± S.E. of six determinations.

An oral dose of 500 mg a.s.a. (e.g. aspirin) in an adult would result in an intestinal concentration of a.s.a. greater than 30 mM if an intestinal fluid volume of 1–2 l. is assumed [8]. Therefore the observed inhibitory effect of a.s.a. on oestradiol absorption may be of clinical significance for women taking oestradiol-containing oral medications, for example in oral contraceptive and oestrogen replacement therapy.

Acknowledgements—This study was supported by a grant from the World Health Organisation Expanded Programme of Research, Development and Research Training in Human Reproduction. We thank Professors B. Kwaku Adadevoh and D. A. Olatunbosun for their interest and encouragement of this work.

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REFERENCES

1. B. P. Lisboa, I. Drosse and H. Breuer, *Z. physiol. Chem.* **342**, 106 (1965).
2. F. R. Smith, D. F. Topley and J. E. Ross, *Biochim. biophys. Acta* **69**, 68 (1963).
3. D. J. Back, M. W. Bates, A. M. Breckenridge, F. E. Crawford, M. L. Orme, P. H. Rowe, G. Smiles and E. Smith, *J. Steroid Biochem.* **9**, 823 (1978).
4. H. M. Bolt, M. Bolt and H. Kappus, *Acta Endocrin.* **85**, 189 (1977).
5. H. M. Bolt, H. Kappus and M. Bolt. *Eur. J. clin. Pharmac.* **8**, 301 (1975).
6. R. M. Welch, W. Levin and A. H. Conney, *J. Pharmac. exp. Ther.* **160**, 171 (1978).
7. T. H. Wilson and G. Wiseman, *J. Physiol.* **123**, 116 (1954).
8. R. Passmore and M. H. Draper, in *Biochemical Disorders in Human Disease* (Eds. R. H. S. Thompson and I. D. P. Wooton), p. 5. Churchill, London (1970).