Medical Research Division,
Biomedical Laboratories,
Edgewood Arsenal,
Aberdeen Proving Ground, Maryland 21010,
U.S.A.

T. L. YAKSH
H. I. YAMAMURA
L. A. FEDELE

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Resistance of intestinal α-adrenoceptors to cold storage

It has been suggested that storage of rabbit intestine at 6–8° for 24 to 72 h leads to selective impairment of α-adrenoceptor function (Lum, Kermani & Heilman, 1966; Lum, Heilman & Gaunt, 1967; Salimi, Kermani & others, 1970).

In the present experiments the α-agonist (−)-phenylephrine and the β-agonist (−)-isoprenaline have been compared as inhibitors of the spontaneous isometric contractions in fresh and cold-stored segments of rabbit intestine, in the presence and absence of the selective antagonists, phentolamine and propranolol. The bathing solution used in the experiments was a modified Krebs solution with the following composition (mM):—NaCl 119·6, KCl 4·95, $CaCl_2$ 2·45, $MgSO_4$ 1·2, NaH_2PO_4 1, sucrose 10, glucose 10·9, $NaHCO_3$ 25.

A complete dose-response curve for either agonist was determined on a fresh jejunal segment. One of the two antagonists was then added to the bathing solution and after an equilibration period of 30 min the dose-response curve determination for the agonist was repeated. A similar procedure was subsequently followed with an adjacent segment which was stored at 6–8° for 24 h. The concentrations of antagonists which we used were those used in the experiments of Lum & others (1966). The potencies of the agonists have been expressed as pD_2 values (Ariens & van Rossum, 1957) where the maximal response was arbitrarily defined as complete extinction of at least one spontaneous contraction. The mean pD_2 values so obtained are shown in Table 1.

In fresh tissues, isoprenaline yielded log dose-response curves flatter than those produced by phenylephrine, the onset of its inhibitory action was slower and its effects lasted longer; however, its potency was similar to that of phenylephrine. These findings are in general agreement with those of van Rossum & Mujic (1965) and of Bowman & Hall (1970), and with the hypothesis of a dual adrenoceptor mechanism subserving inhibition in rabbit jejunum (Ahlquist, 1948; Ahlquist & Levy, 1959; Levy 1959; Furchgott, 1960).

Storage of tissues at 6–8° for 24–48 h did not affect the rate of spontaneous movement (15 min^{-1} for tissues equilibrated at 37°). In cold-stored tissues, the inhibitory potencies of both isoprenaline and phenylephrine were increased; there was less variability in tissue response; log dose-response curves were flatter; the onset and duration of the effects were indistinguishable from those observed in fresh tissues.

Phentolamine proved to be somewhat more potent an antagonist of phenylephrine in cold-stored tissues than in freshly excised tissues; thus the pA_2 (molar) estimates calculated from the results shown in Table 1 were 8·2 and 9·2 for fresh and cold-

Table 1. *Mean pD_2 values for phenylephrine and isoprenaline in the absence and presence of antagonists*

Agonist	Tissue condition	Mean pD_2 values ± s.e. No antagonist	Phentolamine 100 ng ml^{-1}	Propranolol 200 ng ml^{-1}
(−)-Phenylephrine	Fresh	7·16 ± 0·29 (23)	5·35 ± 0·93 (7)*	7·97 ± 0·49 (15)
	Cold	7·97 ± 0·26 (15)	5·17 ± 0·18 (7)*	7·49 ± 0·24 (7)**
(−)-Isoprenaline	Fresh	7·66 ± 0·19 (35)	7·3 ± 0·47 (6)**	6·87 ± 0·19 (27)*
	Cold	8·07 ± 0·18 (25)	7·8 ± 0·46 (4)**	7·0 ± 0·17 (22)*

( ) No of experiments.

* indicates that the value differs significantly (at $P = 0·05$) from appropriate control value on on the basis of an unpaired Student's *t* test.

** indicates that there was a significant difference (at $P = 0·05$) between these and the control values when a paired *t*-test was applied to values obtained on each preparation.

stored tissues respectively. Propranolol was a less potent antagonist of isoprenaline and was approximately equipotent in fresh and cold-stored tissues. The pA_2 (molar) estimates for propranolol/isoprenaline were 6·8 and 7·1 in fresh and cold-stored tissue respectively.

Some antagonism of isoprenaline by phentolamine and of phenylephrine by propranolol (the latter in cold-stored tissues only) was indicated when "within" tissue comparisons of pD_2 values were undertaken. Such findings suggest some overlap of the properties of α- and β-adrenoceptors in rabbit intestine, or more probably, that there may be some lack of specificity in the actions of the agonists and/or antagonists used. The results clearly indicate that cold storage of rabbit intestine under the present conditions does not lead to selective impairment of intestinal α-adrenoceptors. Possibly, the difference in the present results and those of Lum & others (1966, 1967) may relate to their use, and our omission of atropine from the bathing medium.

Department of Physiology,
Monash University,
Clayton, Victoria, Australia

SAN-SAN-WAI
JOCELYN N. PENNEFATHER

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