Short communications

Further studies on the effect of ageing on β-adrenoceptor activity of rat aorta

J. H. FLEISCH

Laboratory of Chemical Pharmacology, National Heart and Lung Institute, Bethesda, Maryland 20014, USA

β-Adrenoceptor activity of rat thoracic aorta decreased with the increasing age of the animal. The ability of aortas to relax to isoprenaline was completely lost when rats were 3–6 months old, depending on the source and strain of animals. $NaNO_2$, however, completely relaxed aortic tissue from animals of all ages tested.

In a previous study, β-adrenoceptor activity of rabbit and rat aorta was shown to decrease with increasing age (Fleisch, Maling & Brodie, 1970); that is, the ability of thoracic aortas to be relaxed by isoprenaline was lost when New Zealand rabbits reached 2 years of age. In addition, isoprenaline-induced relaxation of NIH Sprague-Dawley rat aorta was also shown to decrease with increasing age; aortas from 78- to 90-day-old rats retained only 20% of the activity demonstrated in 41- to 60-day-old animals. By contrast, sodium nitrite completely relaxed aortic strips not only from 78- to 90-day-old rats but from older rabbits and rats as well. Therefore, the loss in capacity to relax to isoprenaline was due to a defect in the β-adrenoceptor system. This study shows that the time at which β-adrenoceptor activity of rat thoracic aorta is lost depends on the source and strain of the animals.

Methods.—Sprague-Dawley rats of either sex were obtained from Hormone Assay, Inc., Chicago, and Zivic-Miller, Allison Park; Wistar rats (CFN) from Carworth Farms, New City; and Osborn-Mendel rats from NIH. Helically cut thoracic aortic strips prepared by the method of Furchgott & Bhadrakom (1953) were suspended in 10 ml organ baths containing Krebs bicarbonate solution of the following composition in mmol/litre: KCl, 4·6; $CaCl_2 \cdot 2H_2O$, 2·5; KH_2PO_4, 1·2; $MgSO_4 \cdot 7H_2O$, 1·2; NaCl, 118·2; $NaHCO_3$, 24·8; and dextrose 10·0. The baths were aerated with 95% O_2 and 5% CO_2. Isometric contractions in g tension were measured by means of a Grass FTO3 force-displacement transducer connected to a Sanborn polygraph. β-Adrenoceptor activity of rat thoracic aorta was determined by the magnitude of the isoprenaline-induced relaxation of 5-hydroxytryptamine contracted tissues. All experiments were performed after most of the α-adrenoceptor sites had been blocked by 0·3 μg/ml phentolamine methanesulphonate. The following drugs were used: (−)-isoprenaline (+)-bitartrate dihydrate (Winthrop Labs), $NaNO_2$ (reagent grade, J. T. Baker), 5-hydroxytryptamine, creatinine sulphate (Sigma) and phentolamine methanesulphonate (gift of Ciba Co.).

Results.—With aortas from all groups of animals, the magnitude of the isoprenaline-induced relaxation decreased with increasing age (Fig. 1) whereas that due to $NaNO_2$ was unchanged. In contrast to our previous study, however, β-adrenoceptor activity of aortas from the present group of animals was lost only in rats 6 months old or older. The difference is best seen upon comparison of isoprenaline-induced relaxations of aortas from the NIH rats used in the previous study with those obtained from Hormone Assay (Fig. 1). Figure 1 also shows that 0·1 μg/ml of isoprenaline induced a 90% relaxation in these aortas, whereas this concentration of isoprenaline produced only 75% of maximal relaxation in aortas used in the previous study. However, $NaNO_2$ also was more active in aortas from this study, causing a complete relaxation in all tissues at a concentration of 10 μg/ml compared to 30–100 μg/ml needed in the previous experiments.

Discussion.—β-Adrenoceptor activity of rat thoracic aorta clearly decreases with increasing age, although the age at which the decrease occurs varies. This study also suggests that the decrease probably begins earlier than was previously suspected. The youngest animals used in this study were between 10 and 20 days younger than those used before. Of further interest was the finding that the relaxation due to $NaNO_2$ was the same in all aortas examined, being independent of the age of the animal. This differed somewhat from our earlier observation in that the $NaNO_2$

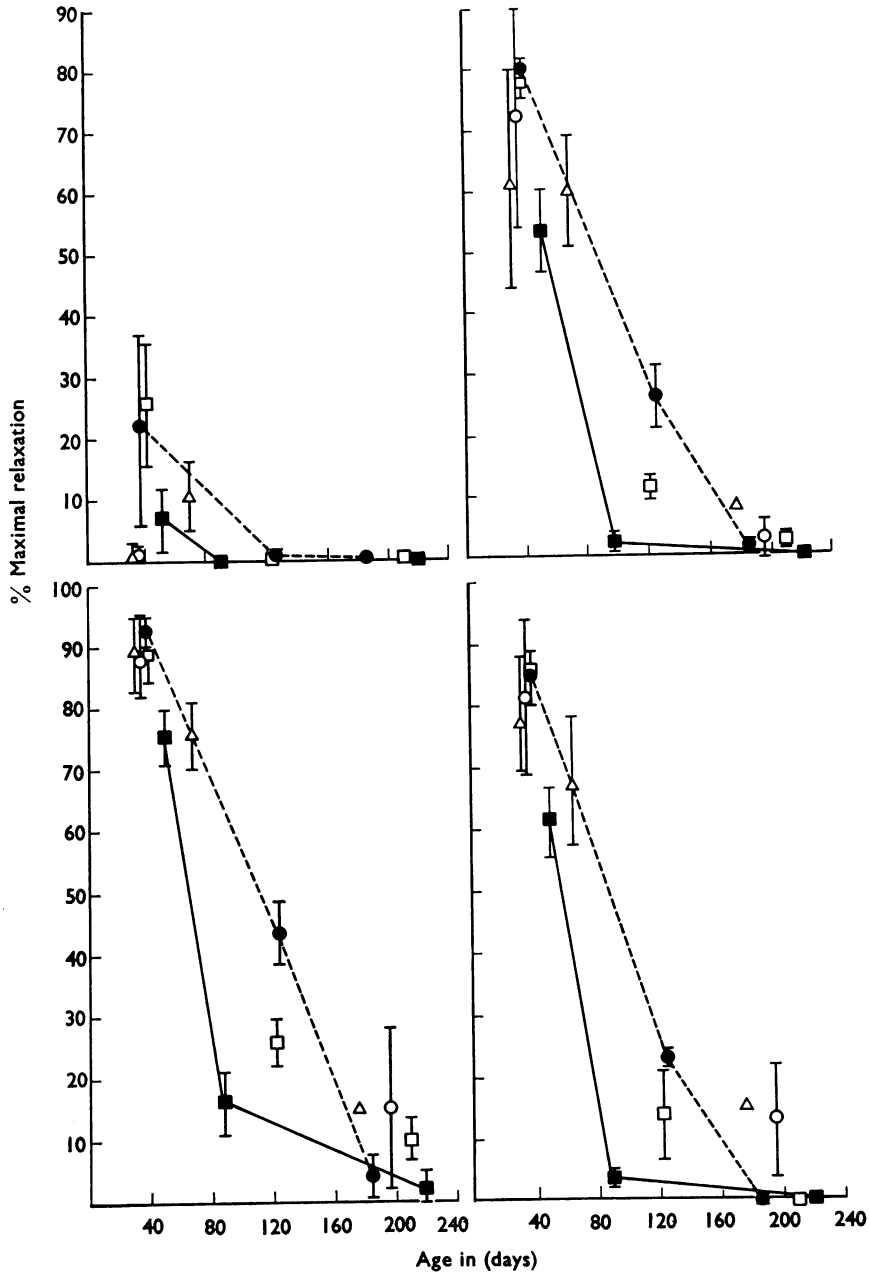

FIG. 1. Dose-response curves for the relaxant effect of isoprenaline (free base) in helically cut thoracic aortic strips from twenty-eight NIH Sprague-Dawley rats (■——■), fifteen Hormone Assay Sprague-Dawley rats (●– – –●), eleven Zivic-Miller Sprague-Dawley rats (□), ten Carworth Wistar (CFN) rats (○) and eight Osborn-Mendel rats (△). The strips were previously made to contract with 5-hydroxytryptamine after α-adrenoceptor sites were blocked with phentolamine. Upper left, 0·001 μg/ml isoprenaline; upper right, 0·01 μg/ml isoprenaline; lower left, 0·1 μg/ml isoprenaline; lower right, 1 μg/ml isoprenaline. Each point represents the mean per cent of the maximal relaxation $\pm$ S.E.

dose-response curves were slightly shifted to the right in aortas from older animals even though $NaNO_2$ still produced the same maximal response. It is difficult to explain the increased responsiveness to isoprenaline of young rat aortas from this study to an increase in β-adrenoceptor activity, since an increase in the ability of the tissues to relax to $NaNO_2$ stimulation was also observed. Nevertheless, these experiments indicate that tissues from very young animals might produce quantitatively different responses to those from animals just slightly older and adds to the thesis that animals should be selected for studies based on their age and not on their weight.

Finally, we believe that factors other than the age of the animal may control β-adrenoceptor activity of aortic tissue. The variability in the response to isoprenaline is greater than that to noradrenaline, 5-hydroxytryptamine, KCl and $NaNO_2$. It seems plausible that variables, such as genetic make-up, environment, diet, seasonal variation, various circadian rhythms, and certain pathological states, influence the overall receptor activity of a tissue. These effects would be especially manifested in tissues where the number of receptors does not provide a clear margin of safety, that is, only enough receptors are present to produce the overt response with no 'spare receptors' available. These factors might explain the relatively large standard errors usually seen in experiments designed to determine β-adrenoceptor activity.

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