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Co-released adrenaline markedly facilitates noradrenaline overflow through prejunctional β_2-adrenoceptors during swimming exercise

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Abstract

The effect of intravenously applied (−)-adrenaline, taken up by and released from sympathetic nerves, on swimming exercise-induced noradrenaline overflow in permanently cannulated adrenal demedullated rats was studied. Adrenaline (100 ng/min) was infused for 2 h, during which a plasma concentration of 500 pg/ml (approximately 2.5 nM) was reached. One hour later plasma adrenaline had returned to undetectable levels. During swimming, adrenaline was released into the plasma in concentrations up to 133 pg/ml and the noradrenaline concentration was markedly enhanced as well. The total catecholamine increase amounted to 178% of control (saline infusion) in the first 3 min of swimming and 165% for the whole 20 min. Cocaine (2.5 mg/kg plus 0.05 mg/kg/min), infused together with adrenaline and continued throughout the experiment, prevented the exercise-induced release of adrenaline and no increase in plasma noradrenaline concentration was observed. Yohimbine (0.25 mg/kg) strongly further enhanced the exercise-induced overflow of both noradrenaline and adrenaline. This further increase was completely blocked by the selective β_2-adrenoceptor antagonist ICI 118,551 ((±)-1-[(2,3-dihydro-7-methyl-1*H*-inden-4-yl)oxy]-3-[(1-methylethyl)amino]-2-butanol) (1.0 mg/kg). These results demonstrate that adrenaline can be taken up by sympathetic nerve endings through cocaine-sensitive uptake carriers and is released from these nerves during swimming exercise. Neuronally released adrenaline markedly enhances exercise-induced catecholamine overflow through activation of prejunctional β_2-adrenoceptors.

Keywords: Noradrenaline; Adrenaline; Catecholamine overflow; Exercise; β_2-Adrenoceptor, prejunctional; (Freely moving rat)

1. Introduction

Adrenaline released from the adrenal medulla is thought to be the endogenous activator of the prejunctional β_2-adrenoceptors facilitating noradrenaline release from postganglionic sympathetic nerve terminals. Circulating adrenaline may activate these receptors directly or act as a co-transmitter after being taken up in sympathetic nerve endings (Rand et al., 1979; Majewski et al., 1981). Despite their pronounced facilitatory capacity in the vasculature of freely moving rats, prejunctional β_2-adrenoceptors are not activated by released noradrenaline, even when inhibitory α_2-autoreceptors are blocked (Remie et al., 1988a,b).

Evidence supporting the facilitatory effect of co-released adrenaline has not only been found in vitro (Guimaraes et al., 1978; Majewski et al., 1980,1981; Valenta and Singer, 1991) but also in vivo. Exogenously applied or endogenously generated (through splanchnic nerve stimulation) adrenaline has been shown to enhance the release rate of noradrenaline in anaesthetized rabbits, after adrenaline levels had returned to normal (Majewski et al., 1982; Schmidt et al., 1984). More recently, using adrenodemedullated freely moving rats, we found that 1 h after adrenaline administration, when plasma levels had returned to undetectable levels, stimulation-evoked overflow of noradrenaline as well as adrenaline from the portal vein sympathetic nerves was strongly facilitated by prejunctional β_2-adrenoceptors (Coppes et al., 1993).

Little is known about the physiological relevance of

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this mechanism. During exercise, which is a physiological way of activating the sympathetic nervous system, increases in both plasma noradrenaline and adrenaline concentrations have been observed (Christensen and Galbo, 1983; Scheurink and Steffens, 1990). In humans, co-release of adrenaline was shown during supine bicycle exercise, by measuring catecholamine spill-over from individual organs to plasma, using radiotracer methodology (Esler et al., 1991). Small amounts of adrenaline were released from the heart when sympathetic nerve firing during exercise was increased 10- to 20-fold, but a facilitatory effect on noradrenaline release could not be shown.

In the present study we examined the possible role of adrenaline as a co-transmitter during swimming exercise and the extent to which adrenaline activates prejunctional β_2-adrenoceptors to facilitate neurotransmitter overflow. Adrenal demedullated Wistar rats were used to exclude interference by adrenaline released from the adrenal medulla.

2. Materials and methods

2.1. Animal care and surgery

Male normotensive Wistar rats, weighing 300–375 g, were used. All animals were housed in individual cages and were permitted tap water and standard laboratory chow ad libitum. Lights were on from 7.00 to 19.00 h.

All rats underwent surgical bilateral adrenodemedullation under halothane anaesthesia. A 2-week recovery period was allowed for adrenocortical regeneration. During the recovery period the adrenodemedullated rats had free access to 0.9% NaCl in addition to normal drinking water, but they hardly used saline. Adrenodemedullation was verified histologically after the termination of the experiments. After the 2-week recovery period, the rats were prepared for surgery under halothane anaesthesia. All rats were provided with two silicon cannulas inserted just proximal to the entrance of the right atrium, one through the right jugular vein for sampling of blood (0.94 mm OD, 0.51 mm ID) and a second in the left jugular vein (0.64 mm OD, 0.28 mm ID) for infusion of drugs. Because of the small size, the infusion cannula did not prevent venous return from the head of the animals. The cannulas were run subcutaneously to emerge at the crown of the head, where they were fixed on the skull with acrylic glue and four stainless steel screws (Steffens, 1969). Experiments were started approximately 7 days after implantation of the cannulas, when the animals had completely recovered their preoperative weight. Using these techniques we were able to apply drugs and to take blood samples simultaneously without disturbing the animal.

2.2. Exercise

Exercise was performed in a Plexiglas swimming pool which was provided with a resting platform at its upper side (Benthem et al., 1993). Lowering the platform to the bottom of the pool forced the rat to swim against a counter current (0.22 m/s). The temperature of the water was maintained at 32 ± 1°C. After the 20-min swimming period, the resting platform was raised, enabling the rat to leave the water. An infra-red heating lamp was switched on to prevent the rat from cooling. To avoid emotional (novelty) stress the rats were accustomed to the experimental set-up in six training sessions.

2.3. Blood sampling and determination of catecholamines

Blood samples of 200 μl were collected according to the method described by Remie and Zaagsma (1986) and put into small ice-cooled cups containing some EDTA crystals. Red blood cells were spun down immediately (Beckmann Microfuge). After each blood sample a transfusion of 200 μl of citrated donor blood (0.6% citrate) was given. Donor blood was obtained from adrenodemedullated donor rats.

After extraction plasma catecholamines were determined using high-performance liquid chromatography with electrochemical detection as described by Remie et al. (1988a) and modified by Coppes et al. (1993). Briefly, the analytical column was an Alltech Adsorbosphere Catecholamines C18 3μ cartridge (100×4.6 mm) and the mobile phase was composed of ultra pure water/methanol (97.0:3.0 v/v), citric acid 0.78%, NaH_2PO_4 0.68%, octanesulphonic acid 0.02%, and 0.01% of EDTA and NaCl. The pH of the mobile phase was adjusted to 3.8 with citric acid and the solution was filtered (Schleicher and Schüll, 0.2 μm) and degassed by sonication at low pressure.

An ESA Coulochem 5100 A electrochemical detector with a high-sensitivity analytical cell (5011) was used for the detection of catecholamines. The guard potential was set to +450 mV, the first detector potential was set to −350 mV and the second detector to +350 mV. Correction for incomplete recovery was performed, using 3,4-dihydroxybenzylamine as an internal standard.

2.4. Experiments

At least 1 h before the start of the experiment the rats were placed on the resting platform and were connected to two polyethylene tubes, one for blood sampling and one for infusion of drugs. When the rats were totally accustomed, a first basal blood sample was taken ($t = -190$ min). Adrenaline (100 ng/min +

0.05% ascorbic acid) or saline (containing 0.05% ascorbic acid) was infused for 2 h (1.2 ml/h) starting at $t = -180$ min, after a second basal blood sample had been taken. In some experiments cocaine was given as a bolus injection (2.5 mg/kg) immediately followed by an infusion (0.05 mg/kg/min) which lasted throughout the experiment, starting at the onset of the adrenaline or saline infusion. In this case, cocaine and adrenaline (or saline) were infused together for 2 h, after which the infusion syringe was changed to one containing cocaine only. Yohimbine and ICI 118,551, when used, were injected at $t = -10$ min at 0.25 mg/kg and 1 mg/kg, respectively. Additional blood samples were taken at $t = -120$, -60, -30, -10 and $t = -1$ min. To measure plasma adrenaline levels during infusion the infusion was interrupted for approximately 30 s at $t = -120$ min and a blood sample was taken. At $t = 0$ the resting platform was lowered. During swimming blood samples were taken within 10 s at $t = 1$, 3, 5, 10, 15, 20 min (just before raising the platform) and at $t = 25$ min.

2.5. Data analysis

The increase in plasma catecholamines during exercise is expressed as area under the curve (AUC) both during the first 3 min of swimming and during the whole period of 20 min, and was compared to control (saline infusion in the absence of drugs) which was averaged and set at 100% (see Fig. 1).

The results are expressed as means ± S.E.M. Wilcoxon's matched pair signed rank test was used to compare data obtained within an experiment. The Mann-Whitney *U*-test was applied to determine differences between data obtained in separate experiments. *P* values of less than 0.05 were regarded as being significant.

2.6. Drugs

(−)-Adrenaline bitartrate (Sigma, St. Louis, MO, USA); cocaine hydrochloride (ACF, Maarssen, Netherlands); yohimbine hydrochloride (17-hydroxyyohimban-16-carboxylic acid methyl ester hydrochloride) (Brocacef, Maarssen, Netherlands); ICI 118,551 hydrochloride ((±)-1-[(2,3-dihydro-7-methyl-1*H*-inden-4-yl)oxy]-3-[(1-methylethyl)amino]-2-butanol hydrochloride) was a kind gift from ICI (Macclesfield, UK). All drugs were dissolved in sterile saline.

3. Results

3.1. Basal overflow

Plasma adrenaline levels of the adrenal demedullated rats were below the limit of detection (10 pg/ml), both during infusion of saline and during subsequent swimming exercise. Medullary tissue was completely absent in the recovered adrenal, showing that adrenodemedullation was complete. Following the infusion or injection of drugs the rats were at rest on the platform until it was lowered, except during cocaine infusion when the rats were slightly active.

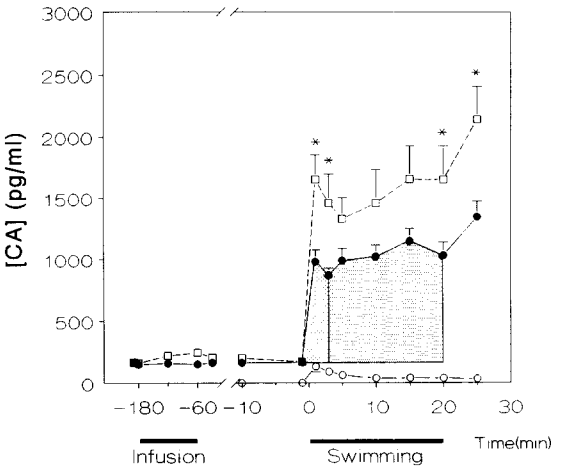

Fig. 1. Plasma catecholamine (CA) levels during saline (0.02 ml/min; $n = 9$) and adrenaline (100 ng/min; $n = 8$) infusion and during subsequent swimming exercise. Closed circles and continuous lines: noradrenaline during and after saline infusion. Open circles and continuous lines: adrenaline after adrenaline infusion. Open squares and dashed lines: noradrenaline during and after adrenaline infusion. Values are the mean ± S.E.M of *n* experiments. S.E.M. values were sometimes smaller than the size of the symbol representing the mean. Dotted areas represent areas under the curve for 0–3 min and 3–20 min of swimming. * Significantly different from saline infusion: $P < 0.05$.

During adrenaline infusion (100 ng/min), measurements of the plasma concentration of adrenaline did not yield information about the content of adrenaline in the circulation, since the substance was infused in close proximity to the sampling cannula where blood samples were taken (just before the heart). After interrupting the infusion for approximately 30 s, plasma adrenaline levels were measured to be about 500 pg/ml. However, 50 min after cessation of the 2 h infusion the adrenaline levels at $t = -10$ had returned to zero (undetectable levels) in all experiments.

Infusion of saline (0.02 ml/min), cocaine (2.5 mg/kg + 0.05 mg/kg/min) and cocaine plus adrenaline for 120 min did not significantly influence the basal plasma concentration of noradrenaline prior to swimming (Figs. 1 and 2). During adrenaline infusion in the absence of cocaine, the plasma noradrenaline concentration was increased significantly from 160 ± 18 pg/ml at −180 min to 243 ± 34 pg/ml at −120 min and to 261 ± 35 pg/ml at −60 min ($P < 0.01$, $n = 22$, collective data from all experiments in the absence of cocaine). Following yohimbine treatment (0.25 mg/kg) at $t = -10$ min, the plasma noradrena-

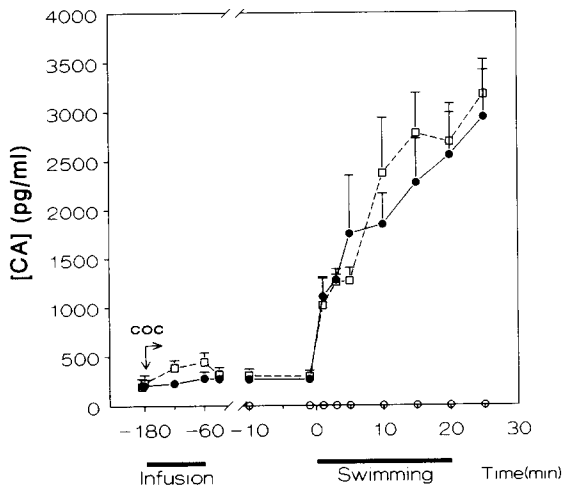

Fig. 2. Plasma catecholamine (CA) levels during cocaine (COC, 2.5 mg/kg + 0.05 mg/kg/min; from $t = -180$ until $t = 20$ min; $n = 4$) and cocaine + adrenaline (100 ng/min; from $t = -180$ until $t = -60$ min; $n = 4$) infusion and during subsequent swimming exercise. Closed circles and continuous lines: noradrenaline during cocaine infusion. Open circles and continuous lines: adrenaline after adrenaline and during cocaine infusion. Open squares and dashed lines: noradrenaline during cocaine infusion and during and after adrenaline infusion. Values are the mean ± S.E.M of n experiments. S.E.M. values were sometimes smaller than the size of the symbol representing the mean.

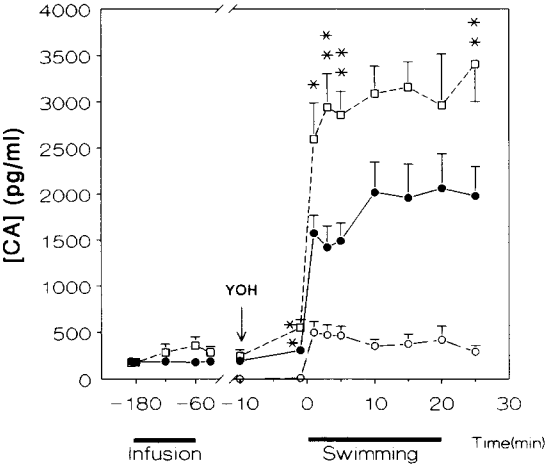

Fig. 3. Plasma catecholamine (CA) levels during saline infusion plus yohimbine injection (YOH, 0.25 mg/kg; $t = -10$ min; $n = 6$) and adrenaline (100 ng/min) infusion plus yohimbine injection ($n = 7$) and during subsequent swimming. Closed circles and continuous lines: noradrenaline during and after saline infusion and yohimbine injection (arrow). Open circles and continuous lines: adrenaline after adrenaline infusion and yohimbine injection. Open squares and dashed lines: noradrenaline during and after adrenaline infusion and after yohimbine injection. Values are the mean ± S.E.M of n experiments. S.E.M. values were sometimes smaller than the size of the symbol representing the mean. *Significantly different from saline infusion plus yohimbine injection: $^{*}P < 0.05$, $^{**}P < 0.01$.

line concentration was significantly elevated at $t = -1$ min, but after adrenaline infusion it was significantly more enhanced than after saline infusion (Table 1, Fig. 3). When ICI 118,551 (1.0 mg/kg) was injected together with yohimbine, plasma noradrenaline concentrations still increased significantly both after saline infusion and after adrenaline infusion (Table 1). However, after adrenaline infusion and yohimbine plus ICI 118,551 injection, plasma noradrenaline levels were not significantly different compared to the levels reached after saline infusion and yohimbine alone or yohimbine plus ICI 118,551 (Table 1).

3.2. Exercise

Fig. 1 shows the effect of swimming exercise on plasma noradrenaline and adrenaline levels after saline infusion and after adrenaline infusion. Adrenaline could not be detected during exercise after saline infusion. However, plasma adrenaline was detected during swimming, reaching levels as high as 133 ± 45 pg/ml ($t = 1$ min) after adrenaline infusion. Noradrenaline levels were significantly more increased during swimming (at 1, 3, 20 min) after adrenaline infusion than after saline infusion. When the rats were back on the

Table 1
Basal plasma noradrenaline concentrations in rats previously infused with saline or adrenaline, before ($t = -10$ min) and after ($t = -1$ min) the injection of yohimbine and/or ICI 118,551

Group (infusion)	Injection	Plasma noradrenaline concentration (pg/ml)		
		$t = -10$ min	$t = -1$ min	n
Saline (0.02 ml/min)	Saline (1 ml/kg)	163 ± 14	165 ± 16	9
Adrenaline (100 ng/min)	Saline (1 ml/kg)	202 ± 28	171 ± 20	8
Saline (0.02 ml/min)	Yohimbine (0.25 mg/kg)	193 ± 46	307 ± 57 [a]	6
Adrenaline (100 ng/min)	Yohimbine (0.25 mg/kg)	244 ± 68	549 ± 87 [a,b]	7
Saline (0.02 ml/min)	Yohimbine (0.25 mg/kg) + ICI 118,551 (1 mg/ml)	175 ± 11	299 ± 18 [a]	6
Adrenaline (100 ng/min)	Yohimbine (0.25 mg/ml) + ICI 118,551 (1 mg/kg)	192 ± 21	275 ± 24 [a,c]	7

Data are expressed as means ± S.E.M.
[a] Significant differences ($P < 0.05$) from the appropriate values before injection at $t = -10$ min. [b] Significant difference between saline- and adrenaline-pretreated groups ($P < 0.05$). [c] Significant difference between ICI 118,551-treated and ICI 118,551-nontreated groups after injection of ICI 118,551 plus yohimbine and yohimbine alone following adrenaline infusion ($P < 0.05$).

platform they started to groom intensively, which was accompanied by a rise in plasma noradrenaline levels at $t = 25$ min both with and without adrenaline preloading (Fig. 1).

The total AUC (noradrenaline + adrenaline) for the first 3 min of swimming, after adrenaline infusion, increased significantly compared to control (saline infusion) (Fig. 5, upper panel). Furthermore, the total AUC for the whole period of swimming increased significantly compared to control (Fig. 5, lower panel).

During cocaine infusion, exercise-induced noradrenaline overflow significantly increased, over the whole period of swimming, compared to that after saline infusion (Figs. 2 and 5, lower panel). After cocaine plus adrenaline no further increase in plasma noradrenaline concentration occurred either during the first 3 min of swimming or during the whole period (Figs. 2 and 5). Adrenaline could not be detected in the plasma (< 10 pg/ml).

Yohimbine injection alone significantly increased exercise-induced noradrenaline overflow after saline infusion both during the first 3 min as well as during the whole period of swimming (Figs. 3 and 5). After adrenaline infusion and yohimbine treatment, the total AUC during exercise was strongly elevated during the first 3 min of swimming and during the whole period (significantly higher than after saline and yohimbine: $P < 0.01$ and $P < 0.05$, respectively) (Figs. 3 and 5). At individual time points, noradrenaline levels were signif-

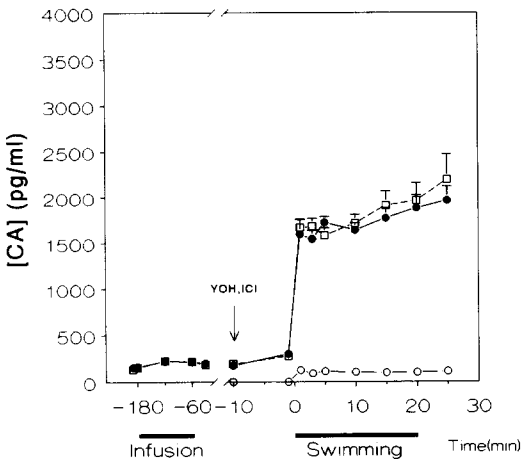

Fig. 4. Plasma catecholamine (CA) levels during saline infusion plus yohimbine (YOH, 0.25 mg/kg) and ICI 118,551 (ICI, 1.0 mg/kg; $n = 6$) injection and adrenaline infusion plus yohimbine and ICI 118,551 injection ($n = 7$) and during subsequent swimming. Closed circles and continuous lines: noradrenaline during and after saline infusion and yohimbine and ICI 118,551 injections (arrow). Open circles and continuous lines: adrenaline after adrenaline infusion and yohimbine and ICI 118,551 injections. Open squares and dashed lines: noradrenaline during and after adrenaline infusion and yohimbine and ICI 118,551 injections. Values are the mean ± S.E.M of n experiments. S.E.M. values were sometimes smaller than the size of the symbol representing the mean.

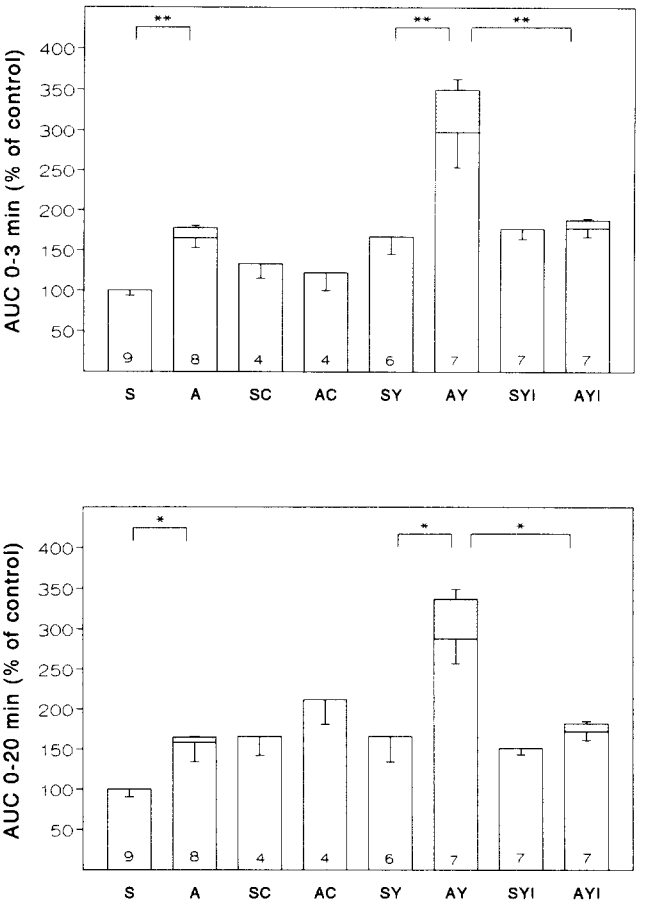

Fig. 5. Plasma catecholamine levels (expressed as area under the curve (AUC)) during swimming exercise. Control experiments (swimming-induced noradrenaline increase after saline infusion) were set at 100%. Upper panel: AUC for 0–3 min of swimming. Lower panel: AUC for 0–20 min of swimming. Bars represent the total AUC for catecholamines, with the open portion indicating noradrenaline levels, and the dotted portion indicating adrenaline levels. Saline infusion (S), adrenaline infusion (A), cocaine plus saline infusion (SC), cocaine plus adrenaline infusion (AC), yohimbine injection after saline (SY) or adrenaline (AY) infusion, ICI 118,551 and yohimbine injection after saline (SYI) or adrenaline (AYI) infusion. $^{*}P < 0.05$, $^{**}P < 0.01$.

icantly higher than after saline and yohimbine at 1, 3, 5, and 25 min (Fig. 3). Plasma adrenaline overflow increased after yohimbine to 496 ± 118 pg/ml at $t = 1$ min, which was significantly higher than after adrenaline infusion alone ($P < 0.05$) (cf. Fig. 1).

Yohimbine given together with ICI 118,551 significantly increased exercise-induced noradrenaline overflow after saline infusion (0–3 min and 0–20 min, Figs. 4 and 5); however, these values were not significantly different from the data with yohimbine alone (cf. Fig. 5). Yohimbine plus ICI 118,551 injection after adrenaline infusion did not increase the AUC during exercise any further (0–3 min and 0–20 min, not significantly different from corresponding saline infusion). Adrenaline, however, was still released up to levels of 125 ± 23 pg/ml at $t = 1$ min, which was considerably

less than that in experiments without ICI 118,551 (496 ± 118 pg/ml at $t = 1$ min) (Fig. 3).

4. Discussion

The objective of the present study was to investigate the role of adrenaline as a possible co-transmitter taken up by, stored in and released from sympathetic nerve terminals, and its ability to facilitate evoked noradrenaline overflow through prejunctional β_2-adrenoceptors. Swimming exercise was used as a physiological activator of the sympathetic nervous system. The rats used were adrenal demedullated to prevent release of adrenaline from the adrenal medulla during exercise.

During infusion of adrenaline (100 ng/min) for 2 h, resulting in plasma levels of about 500 pg/ml, i.e. about 2.5 nM, the plasma noradrenaline concentration increased 1.5-fold. This increase is probably due to direct stimulation of facilitatory prejunctional β_2-adrenoceptors since during saline infusion plasma noradrenaline concentrations remained at baseline levels. Adrenaline may also activate prejunctional inhibitory α_2-adrenoceptors (Majewski et al., 1981,1985). Despite the fact that prejunctional α_2-adrenoceptors have a higher capacity to inhibit noradrenaline overflow than β_2-adrenoceptors to facilitate it (Starke, 1987; Remie et al., 1988b, Coppes et al., 1993), the β_2-adrenoceptor-mediated facilitation during adrenaline infusion was not overruled by α_2-adrenoceptor-mediated inhibition. This can be explained by the fact that the animals were at rest during adrenaline infusion, which is accompanied by a low activity of the sympathetic nerves. Under these conditions, facilitatory β_2-adrenoceptors are more operative than at higher levels of neural activity (Misu and Kubo, 1986).

The half-life of adrenaline in plasma is less than 30 s (Ferreira and Vane, 1967). Once taken up in sympathetic nerves the half-life increases to about 4 h and adrenaline can be released up to 24 h after its administration (Majewski et al., 1981). Adrenaline may therefore be far more effective in activating prejunctional β_2-adrenoceptors after being released from sympathetic nerve endings. In a previous study it was shown that co-released adrenaline can strongly facilitate electrically evoked noradrenaline overflow in the portal vein of freely moving rats (Coppes et al., 1993). To investigate whether adrenaline also acts as a co-transmitter, stimulating prejunctional β_2-adrenoceptors, during a more physiological activation of the sympathetic nervous system, a 20-min period of swimming exercise was applied in the present study.

After saline infusion, plasma noradrenaline levels were clearly increased during swimming. However, 1 h after the adrenaline infusion, when plasma levels of adrenaline had returned to undetectable levels, exercise-induced noradrenaline overflow was clearly enhanced compared to that in control experiments (saline infusion). Adrenaline was released as well, especially in the first minute of swimming, after which plasma adrenaline levels gradually decreased. This confirmed the findings of Moura et al. (1990) who showed in guinea pig vas deferens preloaded with catecholamines that recently incorporated amines are stored in varicosities close to the surface of the organ and are more easily released than the endogenous noradrenaline pools. Plasma concentrations of noradrenaline after adrenaline infusion were increased over the whole period of swimming (Fig. 5).

When cocaine was co-infused with adrenaline, the release of adrenaline as well as the facilitation of the noradrenaline overflow during exercise was prevented, indicating the neuronal origin of the released adrenaline. After yohimbine administration, both the basal noradrenaline concentration and the swimming exercise-induced overflow of noradrenaline were increased, similar to findings with the portal vein of the freely moving rat (Remie and Zaagsma, 1986; Remie et al., 1988b; Coppes et al., 1993). After adrenaline preloading, yohimbine further increased basal as well as exercise-induced noradrenaline levels (up to 3158 pg/ml) significantly more than in the saline-infused animals (up to 2062 pg/ml). Moreover, adrenaline overflow was markedly elevated (up to 496 pg/ml) compared to that in adrenaline-infused animals not receiving yohimbine. These data confirm that, when prejunctional α_2-autoreceptors are blocked, co-released adrenaline is capable of facilitating noradrenaline and adrenaline overflow to a much larger extent. These results are comparable with previous results for the portal vein of the freely moving rat where, after preloading with adrenaline, the stimulation-evoked noradrenaline overflow in the presence of yohimbine was also strongly elevated (Coppes et al., 1993). It might be argued that the actions of yohimbine are due to an increased sympathetic firing rate as the result of a central action (Laubie and Schmitt, 1988). However, Szabo et al. (1989) showed that yohimbine even at a higher dose (1.0 mg/kg + 0.1 mg/kg/h) increased basal plasma noradrenaline levels without any increase of renal sympathetic nerve activity in anaesthetized rabbits.

As in a previous study in the portal vein of unrestrained rats (Coppes et al., 1993), the selective β_2-adrenoceptor antagonist ICI 118,551, given together with yohimbine, completely blocked the facilitation of exercise-induced noradrenaline and adrenaline overflow after adrenaline infusion. ICI 118,551 also blocked the increase in basal noradrenaline above the level observed with yohimbine alone after adrenaline infu-

sion. These results demonstrate the involvement of prejunctional β_2-adrenoceptors.

During infusion, plasma levels of adrenaline reached about 500 pg/ml (circa 2.5 nM), which corresponds with a relatively low level of stress since Popper et al. (1977) have reported that physical immobilization stress of rats increases plasma adrenaline levels to 2400 pg/ml. In humans, infusion of 15 ng/kg/min adrenaline for 6 h amplified the blood pressure response to sympathetic stimulation by both cold stress and isometric exercise up to 18 h after the cessation of the infusion (Blankestijn et al., 1991). Furthermore, the stimulation-evoked tritium release from isolated, [^{3}H]noradrenaline-loaded renal arteries of 4-week-old prehypertensive spontaneously hypertensive rats, measured approximately 2 h after swimming exercise, was significantly inhibited by the β-adrenoceptor antagonist carteolol (10 and 100 nM), in contrast to the release from arteries of animals not undergoing swimming (Misu et al., 1990). These data indicate that not only exogenously applied but also endogenous adrenal-derived adrenaline, released during exercise, may have a tonic facilitatory effect on evoked noradrenaline release from sympathetic nerves.

In conclusion, this study has shown that exogenously applied adrenaline, in a dose producing physiological plasma levels, is taken up into sympathetic nerves through cocaine-sensitive uptake carriers and is released, together with noradrenaline, during swimming exercise. Co-released adrenaline markedly facilitates the exercise-induced noradrenaline and adrenaline overflow through prejunctional β_2-adrenoceptors. This facilitation is further enhanced when inhibitory α_2-adrenoceptors are blocked.

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