Thrombin exhibits a pattern of release and metabolism of 5-HT different from that of reserpine (Figures 1 and 2)[12]. In the presence as well as in the absence of Ca^{++}, thrombin causes a rapid loss of 5-HT from isolated blood platelets. The liberation of the amine is much more rapid than with reserpine, being maximal within 15 min (according to preliminary experiments already after 2 min). As found by previous authors with another buffer (NaCl-Tris)[13], the thrombin-induced 5-HT liberation is more marked in the presence of Ca^{++}. Thus with thrombin + $CaCl_2$ the platelets lose about 95% of the original 5-HT and simultaneously coarse platelet aggregates are formed. On incubation with thrombin alone, about 75% of the 5-HT leaves the platelets; aggregation is in general very fine and often cannot be seen macroscopically. After 15 min, practically all the 5-HT liberated from the platelets is identified in the incubation fluid and no appreciable amounts of metabolites are detectable with the spectrophotofluorimetric method. After 1 and 2 h, about 2 and 10–15% respectively of the liberated 5-HT can be recovered as 5-hydroxytryptophol and 1–2% as 5-hydroxyindoleacetic acid. Both these metabolites have been identified by paper chromatography[2,3]. Their formation explains the slight decrease in 5-HT in the incubation medium between 15 min and 2 h.

The experiments show that after thrombin the 5-HT leaves the platelets very rapidly without being metabolized initially. If the liberated 5-HT remains in contact with the thrombocytes, however, part of it is secondarily transformed into 5-hydroxytryptophol and to a minor extent into 5-hydroxyindoleacetic acid. Both these metabolites derive from 5-hydroxyindoleacetaldehyde, the product of oxidative deamination of 5-HT by monoamine oxidase. In the absence of major amounts of erythrocytes, which probably contain aldehyde oxidase, the 5-hydroxyindoleacetaldehyde is mainly converted to 5-hydroxytryptophol[3]. These findings demonstrate therefore that monoamine oxidase is probably still active in platelets aggregated by thrombin. They furthermore indicate the possibility that 5-hydroxytryptophol formation might also occur in vivo, e.g. in white thrombi which contain only few erythrocytes.

Zusammenfassung. In isolierten Blutplättchen von Kaninchen bewirkt Thrombin rasche und hochgradige Freisetzung von unverändertem 5-Hydroxytryptamin, welches durch weitere Inkubation sekundär teilweise zu 5-Hydroxytryptophol und 5-Hydroxyindolessigsäure umgewandelt wird. Nach Reserpin scheint hingegen ein Teil des 5-Hydroxytryptamins bereits bei seiner Freisetzung zu diesen Metaboliten abgebaut zu werden.

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Inhibition of Acute Effects of Hydralazine by an Adrenergic β-Receptor Blocking Agent

Some of the acute effects of hydralazine such as reduction of peripheral resistance, tachycardia and coronary dilatation, resemble the effects that would result from stimulation of adrenergic β-receptors. We, therefore, attempted to inhibit these effects of hydralazine by administration of an adrenergic β-receptor blocking agent, pronethalol.

Blood pressure and heart rate were measured in the unanaesthetized trained dog, in the pentobarbital-anaesthetized[1] dog and in the allobarbital-urethane anaesthetized[2] cat. Blood pressure, heart rate, total coronary flow and oxygen saturation in arterial and coronary venous blood were measured in the allobarbital-urethane-anaesthetized[2] cat. For these experiments the open-chest, right heart bypass preparation with fixed cardiac output was used[3]. Oxygen saturation was measured by a photometric method. Doses of the substances used are given in Tables I and II.

In the unanaesthetized dog hydralazine reduced blood pressure and increased heart rate. Pronethalol, in a dose which has no pronounced influence on blood pressure or heart rate, inhibited these effects (Table I). In the anaesthetized dog and cat, hydralazine produced no tachycardia. The blood pressure fall was more pronounced in the anaesthetized than in the unanaesthetized dog. Administration of the β-blocker in the anaesthetized dog, did not inhibit, but enhanced the hypotensive effect of hydralazine.

Coronary flow was markedly increased by hydralazine and, as oxygen consumption remained unchanged, oxygen extraction decreased correspondingly. In the doses used, pronethalol had no pronounced effect on the parameters measured. Pronethalol abolished the effect of hydralazine on coronary flow. In addition, oxygen consumption, which was not influenced by either compound given singly, was markedly reduced. Therefore, oxygen extraction remained low in spite of the return of coronary flow towards normal levels.

The effect of pronethalol is apparently different in the anaesthetized and in the unanaesthetized dog. An increase in heart rate in the unanaesthetized and a decrease in heart rate in the anaesthetized dog were seen. The blood pressure and heart rate effects of hydralazine were inhibited in the unanaesthetized dog by pronethalol, while the hypotensive effect was enhanced in the anaesthetized dog and cat.

The effect of hydralazine on coronary flow was antagonized by pronethalol. The effect on oxygen consumption may have been antagonized, if we assume that the

[1] Nembutal, Abbott; 25 mg/kg i.v.
[2] Dial, CIBA; 35 mg/kg i.p. + 35 mg/kg s.c.
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Table I. Influence of pronethalol on the effects of hydralazine in the dog and cat

Control values BP	HR		Hydralazine mg/kg i.v. BP	HR		Pronethalol mg/kg i.v. BP	HR	
Dog, unanaesthetized trained			1.0			1.0		
133 ± 5	104 ± 9	(13)	116 ± 5	237 ± 14	(13)	—	—	
138 ± 7	95 ± 17	(6)	115 ± 7	223 ± 23	(6)	137 ± 8	162 ± 11	(6)
128 ± 9	98 ± 6	(8)	—	—		135 ± 6	122 ± 5	(8)
Dog, pentobarbital sodium anaesthesia 25 mg/kg i.v.			1.0			1.0		
144 ± 2	163 ± 10	(3)	102 ± 10	155 ± 5	(3)	75 ± 6	133 ± 9	(3)
112 ± 6	168 ± 2	(3)	—	—		100 ± 23	154 ± 9	(3)
Cat, allobarbital-urethane anaesthesia[a]			0.3			1.0		
154 ± 13	183 ± 15	(4)	121 ± 11	192 ± 26	(4)	—	—	
145 ± 11	178 ± 11	(7)	116 ± 6	180 ± 15	(7)	99 ± 6	137 ± 11	(7)
139 ± 8	194 ± 9	(7)	—	—		115 ± 6	145 ± 8	(7)

[a] Dial CIBA, 35 mg/kg s.c. + 35 mg/kg i.p. BP = blood pressure in mmHg. HR = heart rate in beats/min. The order of injection of the compounds studies is given in the Table from left to right. Values given are means ± SE of the maximal effects. Number of experiments given in brackets.

Table II. Influence of pronethalol on the effects of hydralazine in the cat

BP	HR	CF	O_2-Ext.	O_2-Cons.	BP	HR	CF	O_2-Ext.	O_2-Cons.	BP	HR	CF	O_2-Ext.	O_2-Cons.
Control values					Hydralazine 3 mg					Pronethalol 10 mg				
107 ± 12 (4)	198 ± 24	122 ± 15	79 ± 1	13.8 ± 1.8	81 ± 7 (4)	219 ± 26	300 ± 93	38 ± 6	15.4 ± 3.4	83 ± 7 (4)	179 ± 17	141 ± 24	45 ± 6	8.8 ± 0.9
114 ± 5 (4)	174 ± 15	106 ± 11	60 ± 5	11.0 ± 1.1	–	–	–	–	–	113 ± 6 (4)	157 ± 16	97 ± 5	54 ± 1	9.1 ± 1.3

BP = blood pressure in mmHg. HR = heart rate in beats/min. CF = total coronary flow in ml/100 g heart weight · min. O_2-Ext. = myocardial oxygen extraction in % of maximal saturation. O_2-Cons. = myocardial oxygen consumption in ml/100 g · min. Other values as in Table I.

oxygen consumption under the influence of hydralazine is relatively high in view of the reduced cardiac work load.

It seems unlikely that the effect of hydralazine on blood pressure and peripheral resistance is exclusively the result of a direct stimulation of adrenergic β-receptors. β-receptor blockade (isoproterenol antagonism) is demonstrable in isolated organ preparations; and, in the same dose range, in the anaesthetized and unanaesthetized animal. Antagonism of the hypotensive effect of hydralazine was, however, seen only in the unanaesthetized animal. In chronic experiments in renal hypertensive rats, the hypotensive effect of hydralazine after repeated injections could be inhibited by simultaneous administration of pronethalol[4].

It has been shown that pronethalol inhibits the uptake of catecholamines into tissue stores[5]. Therefore, the pressor effect of endogenous catecholamines may be potentiated through a cocaine-like mechanism. It has been postulated that hydralazine as well as other hypotensive agents causes an increase in sympathetic discharge by some reflex mechanism[6]. This effect may be more pronounced in the unanaesthetized animal. In this case, the inhibition of the effects of hydralazine on blood pressure could be explained by a cocaine-like mechanism of action; and the inhibition of its actions on the heart by direct blockade of β-receptors.

A central site of action as a component of the mechanism of action of pronethalol cannot, however, be excluded. This possibility is suggested by the differences in the results in anaesthetized and unanaesthetized animals.

Zusammenfassung. Die durch Hydralazin verursachte Blutdrucksenkung und Tachycardie am wachen Hund wurde durch Pronethalol antagonisiert. Am narkotisierten Tier änderte Hydralazin die Herzfrequenz nicht, sondern führte zu einer Drucksenkung, die durch Pronethalol verstärkt wurde. Hydralazin steigerte an der narkotisierten Katze den Coronardurchfluss. Dieser Effekt wurde durch Pronethalol aufgehoben. In den verwendeten Dosen hatte Pronethalol auf die gemessenen Grössen keine Eigenwirkung.

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