

after restraint except 3 isoproterenol-treated rats of the high fat diet group.

It has been demonstrated that physical exertion protects rats against the cardiotoxic action of catecholamines, through a net uptake of myocardial potassium and a decrease in sodium⁴. It is possible that this mechanism was invoked by stress in our experiments, and may explain the similar survival rate of stressed rats with and without myocardial damage⁵.

Résumé. Une nécrose myocardique a été produite chez le rat mâle adulte avec de l'isoprotérénol. Aucune différence de survie significative n'a été constatée entre les individus traités et les non traités lorsque les uns et les

autres ont été soumis à l'hypoxémie, la nage forcée dans l'eau froide, ou la restriction par attache.

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Lederle Laboratories Division, American Cyanamid Company, Pearl River (N.Y. 10965, USA), August 11, 1966.

⁴ E. BAJUSZ, *Arzneimittel-Forsch.* 14, 1115 (1964).

⁵ This work was conducted in the Department of Chemical Pharmacology. We wish to thank Dr. D. A. BURSKE for his interest and suggestions.

Spermine and Spermidine Distribution During Wheat Growth

Very little information has been reported on the occurrence, distribution and metabolism of polyamines in higher plants¹. MORUZZI and CALDARERA found spermine and spermidine in wheat germ². On the other hand, BERTOSI et al.³ and BAGNI⁴ showed that spermine 10⁻⁴M and spermidine 10⁻⁵M are growth-promoting factors for *Helianthus tuberosus* explants in vitro.

In this research we have studied the distribution of spermine and spermidine in wheat plant, variety 'Hard Red Winter', grown regularly in the field, and have followed the changes during growth. We have used, for polyamine determination, the method developed by RAINA⁵ with paper electrophoresis separation using sulphosalicylic acid buffer 0.065M at pH 3.5 and stained with amido black. Spermine and spermidine occur in all parts of the plant examined, except in the root and anther.

These polyamines (see Table) are present in unfertilized ovules and rapidly increase, given as γ /unit, after ferti-

zation. When the caryopsis is formed, but still in the milk stage, these polyamines occur not only in the embryo, but also, and in greater relative content, in the remaining parts of caryopsis. In the embryo of mature caryopsis (seed), spermine and spermidine increase especially in respect to the remaining part of caryopsis.

The determination on 170 mg of fresh weight of pure pollen grain has shown that spermidine is present in appreciable quantities while spermine appears only in traces. No significant changes of polyamine contents were noted in leaves before and after the fertilization of ovules.

¹ H. TABOR and C. W. TABOR, *Pharmac. Rev.* 16, 245 (1964).

² G. MORUZZI and C. M. CALDARERA, *Archs Biochem. Biophys.* 105, 209 (1964).

³ F. BERTOSI, N. BAGNI, G. MORUZZI, and C. M. CALDARERA, *Experientia* 21, 80 (1965).

⁴ N. BAGNI, *Experientia* 22, 732 (1966).

⁵ A. RAINA, *Acta physiol. scand.* 60, Suppl. 218 (1963).

Spermine and spermidine distribution in wheat plant

	Ovules unferti- lized	Ovules fertilized		Milk stage caryopsis		Mature caryopsis (seeds)		Pollen grains	Leaves		Culms	Plants 1 month old
		After 6 days	After 20 days	Em- bryos	Remain- ing parts	Em- bryos	Remain- ing parts		1 month before fertili- zation	1 month after fertili- zation		
Spermine												
γ/g fresh weight	25.0	21.0	12.4	67.7	4.9	399.8	7.2	traces	5.3	5.6	traces	16.0
γ/g dry weight	210.0	105.0	53.2	310.0	17.2	444.7	8.0	traces	16.2	16.7	traces	110.8
γ/unit	0.029	0.143	0.208	0.075	0.177	0.381	0.220					5.0
Spermidine												
γ/g fresh weight	87.5	33.0	51.0	125.0	8.0	853.9	18.1	27.6	8.8	9.1	traces	21.3
γ/g dry weight	735.0	165.0	218.0	574.0	27.6	949.8	20.1	212.1	26.5	27.1	traces	146.6
γ/unit	0.100	0.224	0.854	0.139	0.309	0.815	0.555					6.7
Spermidine/ Spermine	3.50	1.57	4.09	1.85	1.61	2.14	2.50		1.66	1.62		1.34
% dry weight	11.9	18.9	23.3	21.8	29.0	86.3	87.2	13.0	32.7	33.5	28.0	14.4

To obtain a more profound understanding of polyamine changes, we planted wheat seeds in boxes and grew them in the greenhouse. In whole 1-month-old plants without roots, before earing, spermine increases, given as γ /unit of seed, about 8 times and spermidine about 5 times. The increase of these polyamines, observed in plants before earing, suggest that leaves are one site of biosynthesis. Besides, the data show that the spermine and spermidine content is greater in organs of higher biosynthesis of the wheat plant.

The relationship of spermine and spermidine to nucleic acid biosynthesis⁶, amino acid rate of incorporation⁷ and 1 C unit metabolism⁸, shows the importance of their presence during wheat plant development.

Riassunto. Spermina e spermidina sono presenti in tutte le parti della pianta di frumento eccetto radici ed antere. Sono particolarmente abbondanti negli ovuli dove aumen-

tano dopo la fecondazione. I risultati inoltre indicano che le foglie sono uno dei principali luoghi di biosintesi di queste sostanze.

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⁷ R. G. MARTIN and B. N. AMES, *Proc. natn. Acad. Sci. USA* 48, 2171 (1962).

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The Influence of the Experimental Conditions on the Renal Response to Angiotensin in the Rat

Although there is increasing evidence that the renin-angiotensin system is important in the control of sodium metabolism and arterial pressure, its exact role is far from clear. Studies on the effect of angiotensin on renal function have led to contradictory findings. Thus all previous investigators¹⁻⁴ have found that in the rat infusion of angiotensin increases urinary sodium excretion, and in this species angiotensin is thought to have the role of a sodium-excreting hormone. Since the rat is the common model for the study of hypertension as well as for micro-puncture studies of sodium transport mechanisms, the action of angiotensin in this species is of critical importance. We have found that the effect of angiotensin on urinary sodium excretion in the rat is dependent on the experimental conditions. The same dose of angiotensin in the same animal is consistently natriuretic under one set of circumstances and antinatriuretic under another. This observation is of importance for the correct interpretation of the role of angiotensin, which in the rat appears to be that of a sodium-retaining hormone.

The experiments were performed on 7 young black-hooded rats weighing between 180 and 220 g. Under light ether anaesthesia catheters were inserted into a tail vein and, through a small suprapubic incision and cystotomy, into the bladder. The operative preparation took approximately 30 min, and the animals were then allowed to recover in special restraining cages.

The response to angiotensin was determined in the same animal at varying times after surgery, with and without anaesthesia, and in states of sodium loading and depletion. The protocol of each angiotensin infusion was identical under these different experimental conditions. For 1 h before and throughout each experiment the animal was infused continuously by means of a constant infusion pump at a rate of 0.37 ml/min. The infusate was half-strength Hartmann's solution, giving a sodium infusion rate of 24 μ Eq/min in all experiments except those performed during sodium depletion, when it was 2.5% dextrose. Urine was sampled at 10 min intervals and

sodium concentration estimated by flame photometry. Following 2 stable control periods, angiotensin II (Ciba) dissolved in one of the above infusates was administered in doses of 0.00005–0.005 μ g/kg/min for 12–32 min, the urine passed during the initial 2 min of the angiotensin infusion being discarded. In the following description, changes in urinary sodium excretion during angiotensin infusion less than 10% of control are considered insignificant.

Infusion of angiotensin in doses of 0.00005–0.005 μ g/kg/min 2–6 h after operation, when the animal was fully conscious, had no consistent effect on urinary sodium excretion (Table). In separate experiments on 3 animals in whom anaesthesia after operation was maintained with i.v. nembutal 15–25 mg/kg, angiotensin in these doses never reduced sodium excretion and sometimes increased it.

After the acute post-operative infusion of angiotensin, the animal was placed on a high sodium intake by infusing approximately 0.5 mEq of sodium daily as quarter-strength Hartmann's solution and fed standard rat chow. In 3 animals the response to angiotensin was studied after only 1 or 2 days of post-operative sodium loading. Infusion of angiotensin at this time again had no consistent effect on sodium excretion. After 3–8 days of chronic sodium loading the response to angiotensin was again investigated. Doses of 0.0005–0.005 μ g/kg/min now consistently reduced urinary sodium excretion in all 7 animals, and in 5 out of the 7 doses of 0.00005 μ g/kg/min also reduced it (Table). Immediately after this study, the animal was anaesthetized with i.v. nembutal, 15–25 mg/kg, and the angiotensin infusion repeated in identical fashion. Doses of angiotensin which had been antinatriuretic in conscious animals immediately before anaesthesia, failed to affect

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