Effects of skin-cooling on hemodynamic, ventilatory, and arterial blood gas variables in cattle

Variables	Skin-cooling[a]	
	Before	After
Ts (°C)	38.6 ± 0.1	32.9 ± 0.5[b]
Tr (°C)	39.8 ± 0.0	39.7 ± 0.1
HR (min^{-1})	96 ± 3	92 ± 2[b]
Q (ml/min/kg)	133 ± 7	129 ± 4[b]
SV (ml/kg)	1.39 ± 0.06	1.34 ± 0.05
P_{pa} (mm Hg)	30 ± 2	32 ± 2
TPVR (mm Hg/ml/min/kg)	0.24 ± 0.02	0.26 ± 0.02[b]
Pao (mm Hg)	104 ± 3	112 ± 4[b]
TSVR (mm Hg/ml/min/kg)	0.80 ± 0.06	0.92 ± 0.05[b]
f (min)	85 ± 7	40 ± 4[b]
V_E (ml/min/kg, BTPS)	400 ± 30	220 ± 10[b]
V_T (ml/kg, BTPS)	5.0 ± 0.3	6.5 ± 0.8
PaO_2 (mm Hg)	68 ± 1	66 ± 1
$PaCO_2$ (mm Hg)	36 ± 1	37 ± 1
pHa	7.46 ± 0.01	7.46 ± 0.01

Ts, skin temperature; Tr, rectal temperature; TPVR, total pulmonary vascular resistance; TSVR, total systemic vascular resistance. [a] Mean values ± SEM are shown for 12 animals (except $n = 10$ for Pao and TSVR). [b] Before and after values differ significantly, $p < 0.05$ (determined by a paired t-test).

Results and Discussion. The cardiopulmonary effects of skin-cooling are shown in the Table. In the cooled area, skin temperature was lowered by nearly 6 °C. The decrease in skin temperature probably evoked a peripheral vasoconstriction that elevated systemic arterial pressure despite a reduction in cardiac output. The elevated systemic pressure might have caused the fall in heart rate via increased baroreceptor activity. The increase in total pulmonary resistance could have been related to pulmonary vasoconstriction or to an increased left atrial pressure. If this pulmonary effect of skin-cooling is exaggerated in a cold environment, it might account for the higher incidence of hypoxic pulmonary hypertension and brisket disease in cattle during cold weather at high altitudes[1].

After the skin was cooled, respiratory rate and minute volume were reduced by about 50%. Arterial blood gases were not altered. These results suggest that although cattle hyperventilate at an ambient temperature of 25 °C, the increased ventilation represents panting and alveolar gas exchange is not affected. Environmental temperature must apparently approach 40 °C before thermoregulatory ventilation influences alveolar ventilation and arterial blood gases[5]. Thus, it can be inferred that in cattle at moderate laboratory temperatures arterial blood gases reflect alveolar ventilation as determined by non-thermoregulatory mechanisms. If variations in environmental temperature from 15 to 25 °C affected alveolar ventilation, then much blood gas data collected from normoxic and hypoxic cattle might be subject to reinterpretation[4,6–9]. The thermoregulatory-induced disassociation between minute and alveolar ventilation might account for the lack of correlation between changes in minute ventilation and arterial blood gases observed in cattle exposed to simulated high altitude[7] and to carotid body excision[10]. It is obvious that attention must be paid to ambient temperature when minute ventilation is used as a variable in studies of ventilatory control in cattle. Minute ventilation would probably be more closely related to alveolar ventilation if cattle were studied at an ambient temperature somewhat lower than 25 °C.

Summary. Cooling the skin of cattle at an ambient temperature of 25 °C decreased cardiac output and increased systemic and pulmonary vascular resistances. Minute ventilation was reduced by about 50%. There was no change in alveolar ventilation as measured by arterial blood gases. These results indicate that thermoregulatory ventilation has significant cardiopulmonary effects in cattle at normal laboratory temperatures.

I. F. McMURTRY[11], J. T. REEVES, D. H. WILL and R. F. GROVER

Cardiovascular Pulmonary Research Laboratory, University of Colorado Medical Center, 4200 East Ninth Avenue, Denver (Colorado 80220, USA), and Department of Physiology and Biophysics, Colorado State University Fort Collins (Colorado 80521, USA), 15 April 1975.

[6] R. F. GROVER, J. T. REEVES, D. H. WILL and S. G. BLONT, JR. J. appl. Physiol. *18*, 567 (1963).
[7] D. H. WILL, C. S. CARD, G. D. VANDLANDINGHAM and A. F. ALEXANDER, Physiologist *10*, 347 (1967).
[8] D. H. WILL, J. L. HICKS, C. S. CARD, J. T. REEVES and A. F. ALEXANDER, J. appl. Physiol. *38*, 495 (1975).
[9] D. H. WILL, J. L. HICKS, C. S. CARD and A. F. ALEXANDER, J. appl. Physiol. *38*, 491 (1975).
[10] G. E. BISGARD and J. H. K. VOGEL, J. appl. Physiol. *31*, 431 (1971).
[11] Acknowledgments. We thank LINDA PANEPINTO, BEA KAPLAN and MARY MUNROE for technical assistance and DIANN SMITH for typing the manuscript. This work was supported by NIH grant No. HL 14985 and by a grant to Dr. WILL from the Colorado Heart Association.

Seasonal Variations of Cardiac Output in Rats

Data on the cardiac output of rats given in the literature range between 142 ml/kg/min[1] and 457 ml/kg/min[2]. This great variability was attributed to different techniques, or to differences in the age, strain and sex of the animals[3]. In our laboratory determination of cardiac output were performed in rats of the some strain and sex and nearly the same age over 4 years; inspite of the constant experimental conditions, values varied greatly. Trying to find an explanation for this phenomenon, we have detected seasonal variations in the cardiac output of rats.

Materials and methods. We used male Wistar rats (conventional animals; dealer P. Bäumler, Wolfratshausen) weighing 320 to 400 g which were housed at 23 °C and fed with Altromin pellets and tap water ad libitum. The

[1] J. KAPITOLA, F. KÖLBEL, M. SCHÜLLEROVÁ and O. SCHREIBEROVÁ, Physiologia bohemos. *22*, 137 (1973).
[2] Y. C. LIN, C. A. DAWSON and S. M. HORVATH, Comp. Biochem. Physiol. *33*, 901 (1970).
[3] M. VÍZEK and I. ALBRECHT, Physiologia bohemos. *22*, 573 (1973).

animals were anesthetized with 1.2 g/kg urethane i.p. The rectal temperature of the anesthetized animals was kept at 36.5 ± 0.5 °C with the aid of a heated operation table, a rectal thermocouple and an YSI telethermometer model 73 (Yellow Springs Instrument Co., Yellow Springs, Ohio). Arterial blood pressure was recorded from the left carotid artery with a Statham pressure transducer on a 'Helcoscriptor' (Hellige, Freiburg) and heart rate taken from the pulsatory changes of blood pressure. We determined cardiac output by the thermo-dilution method and calculated peripheral resistance as described previously[4]. Beginning in November 1971, however, an electronic calculator[5] (Herzzeitvolumen-Messgerät HZV BN 6560, August Fischer KG., Göttingen) was used. The comparison of the 2 methods for determination of cardiac output revealed no differences in the results.

After the preparation of the animals and the calibration of the apparatus, an adaptation time of about 30 min was necessary to reach a steady state. Thereafter all parameters were measured 3 times at intervals of 5 to 10 min. The values given in this paper are means based on the last of these 3 determinations. Pharmacological treatments were performed only after determination of the basic values.

Results and discussion. All values of cardiac output measured during the same month were compiled and the means of these values are given in Figure 1. Evidently, there are large differences in these values of cardiac output. The extremes were 245 ml/kg/min measured in June 1970 or 244 ml/kg/min in March 1972, on the one hand, and 415 ml/kg/min in November 1970, on the other hand. Thus, cardiac output of rats differed by about 70%. Considering the trace of the cardiac output, seasonal variations are evident. The low values were found in spring and summer, the high ones in autumn and winter. A further determination of cardiac output (not shown in Figure 1) was performed in October 1973. Again a high value was found (373 ml/kg/min) which is comparable with that measured in October 1970.

From May 1970 to April 1971, cardiovascular experiments were performed most frequently. Values of cardiac output, heart rate, stroke volume, blood pressure, and peripheral resistance, registered in this period, are compiled in Figure 2. As can be seen, changes of cardiac output were accompanied by coincident changes of heart rate and stroke volume. However, the stroke volume was much more implicated in the seasonal changes of cardiac

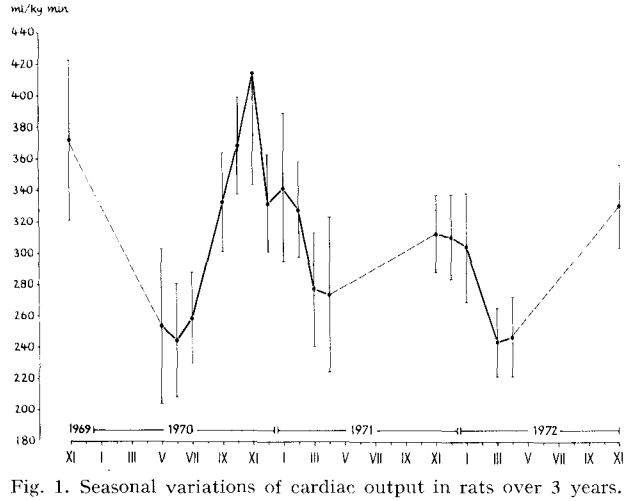

Fig. 1. Seasonal variations of cardiac output in rats over 3 years. Values are means ± confidence limits for p 0.05 ($\bar{x} \pm t \cdot (s\bar{x})$) of 9–13 animals each.

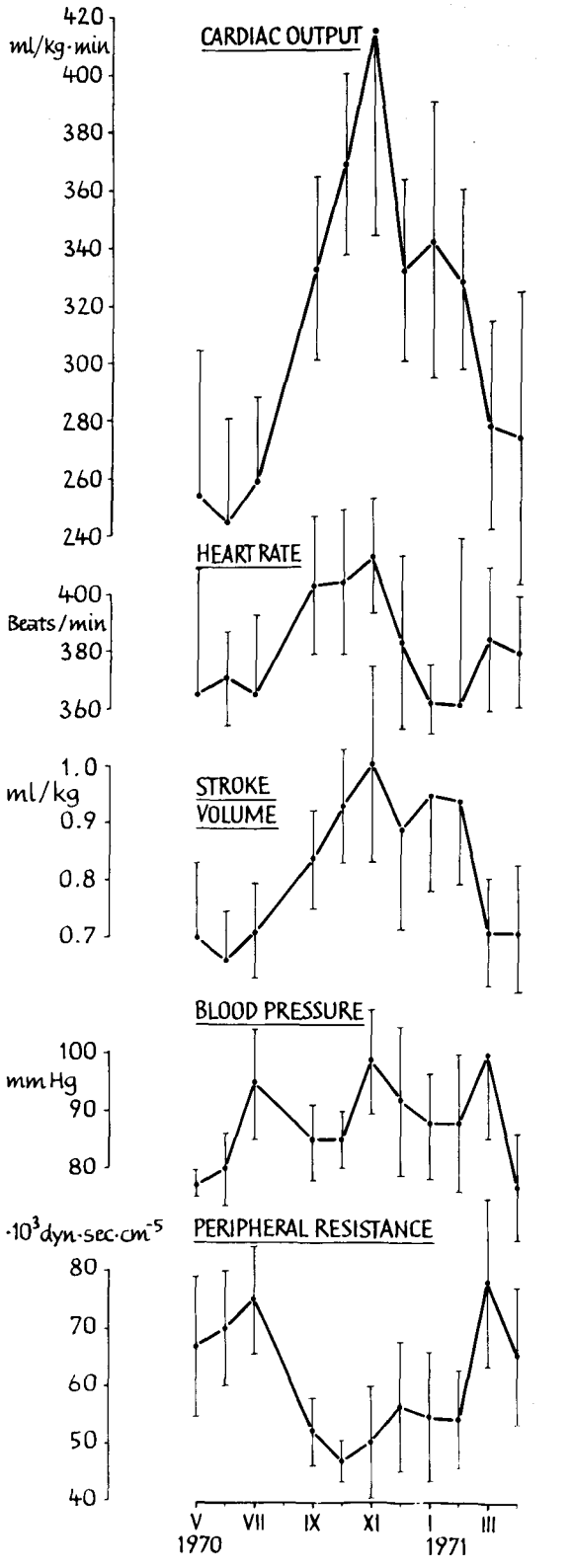

Fig. 2. Seasonal variations of cardiovascular parameters in rats throughout 1 year. Values are means ± confidence limits for p 0.05 ($\bar{x} \pm t \cdot (s\bar{x})$) of 9 animals each.

[4] O. Strubelt, G. Back, E. Uhl and G. Zetler, Naunyn-Schmiedeberg's Arch. Pharmak. *271*, 346 (1971).

[5] H. Slama and J. Piiper, Z. Kreisl.-Forsch. *53*, 322 (1964).

output than the heart rate. For example, between June and November 1970, cardiac output increased by 69%, stroke volume by 53%, but heart rate by only 12%. Arterial blood pressure, on the other hand, did not coincide with the seasonal variations of cardiac output (Figure 2). Thus, the changes of cardiac output were compensated by reversal changes of peripheral resistance.

What is the cause of these seasonal variations? Cardiac output is mainly regulated by the blood demands of peripheral tissues[6]. Exercise and motility on the one hand, thermal regulation on the other hand are the most important factors demanding a higher body-blood supply. Both can be excluded as causes of the variations in cardiac output seen in our experiments. Firstly, the cardiovascular investigations were done in anesthetized rats which were not able to move. Secondly, the rats were housed at a constant environmental temperature of 23 °C before the experiments, and body temperature was kept at 36.5 °C during the experiments.

We suppose that the thyroid gland is involved in seasonal variations of cardiac output. It was shown[7] that the thyroid gland of rats is at a higher level of activity during the winter than during the summer season. These changes occurred even though the animals were maintained under conditions of constant temperature (22 to 25 °C) and light. Seasonal variations in thyroid function (higher serum thyroxine levels during the winter period than during the summer period) were also seen in new-born children[8]. Thyroid hormones increase the metabolic rate and moreover have a direct positive inotropic and chronotropic effect on the heart[9–11]. Both actions are capable of increasing cardiac output.

Since cardiac output, heart rate, stroke volume, and peripheral resistance of rats are subject to seasonal variations, results obtained at different times of the year cannot be compared. Therefore, cardiovascular research in rats always requires simultaneous control experiments.

Summary. Cardiac output of rats shows seasonal variations with low values in spring and summer and high ones in autumn and winter. The stroke volume was much more implicated in these changes than the heart rate. The seasonal changes of cardiac output are probably due to changes of thyroid function.

G. BACK and O. STRUBELT

Abteilung für Toxikologie der Medizinischen Hochschule Lübeck, Ratzeburger Allee 160, D-2400 Lübeck (German Federal Republic, BRD), 20 June 1975.

[6] A. C. GUYTON, New Engl. Med. J. *277*, 805 (1967).
[7] R. A. HOFFMANN and M. X. ZARROW, Acta endocr., Copenh. *27*, 77 (1958).
[8] P. ROGOWSKI, K. SIERSBAEK-NIELSEN and J. MØLHOM HANSEN, J. clin. Endocr. *39*, 919 (1974).
[9] M. BEZNÁK, Can. J. Biochem. *40*, 1647 (1962).
[10] R. A. BUCCINO, J. F. SPANN, P. E. POOL, E. H. SONNENBLICK and E. BRAUNWALD, J. clin. Invest. *46*, 1669 (1967).
[11] G. S. LEVEY, Am. J. Med. *50*, 413 (1971).

Effect of Furosemide on the Permeability to Cl of the Isolated Skin of *Leptodactylus ocellatus*

Furosemide is known to be a potent inhibitor of active chloride transport in the epithelia. Evidence of this effect was provided by the abolition of the potential difference (PD) (positive inside) in the thick ascending limb of Henle's loop perfused with chloride-containing solutions[1], and by the decrease in short circuit current (SCC) in the frog cornea which only transports Cl^-[2]. More recently, furosemide was found to decrease Cl^- influx in selected, low voltage, short-circuited skins of the European *Rana temporaria*[3]. Since in this preparation Cl^- influx slightly excedes Cl^- efflux, the results were thought to represent a specific inhibitory effect on active Cl^- transport.

The possibility of additional effect of furosemide on passive Cl movements is still controversial and largely supported by indirect evidence: 1. According to BURG et al.[1] furosemide inhibits passive Cl^- influx into the lumen of the perfused ascending limb more than can be accounted for by changes in PD; 2. EIGLER et al.[4] reported that furosemide increases the PD of the toad skin with little change in the SCC which was considered to be indirect evidence of decreased permeability to anions. Contrasting with these findings, LOTE[3] reported that furosemide has no effect on passive Cl^- efflux in the skin of *Rana temporaria*. It is possible that this discrepancy is due to low permeability to Cl^- of the preparations used in those studies.

The aim of this study was to investigate the effect of furosemide on passive Cl fluxes in a preparation which is highly permeable to Cl. For this purpose the isolated skin of the South American frog (*Leptodactylus ocellatus*) was chosen because its active and passive Cl fluxes are 10 times faster than those of *Rana temporaria*, and passive Cl effluxes 10 times faster than passive Na fluxes[5,6]. In this respect it compares favorably with the thick ascending limb where the ratio between the 2 passive fluxes is only 0.5[7]. Also the possibility of eliminating electrical gradients by short-circuiting the isolated skin makes it possible to detect permeability changes without any correction for PD changes.

The results indicate that, in addition to its known inhibitory effect on active Cl transport, furosemide also decreases passive Cl fluxes in the isolated skin of *Leptodactylus ocellatus*. Extension of this effect to the thin ascending limb of Henle's loop may bear some relationship to the natriuretic properties of the drug.

Methods. The isolated abdominal skins of *Leptodactylus ocellatus* were used in all experiments. After dissection the skins were mounted in lucite chambers as described by USSING and ZERAHN[8] covering 3.14 cm^2 of skin. Both chambers were filled with 5 ml of Ringer solution (NaCl 115.5 m*M*; $NaHCO_3$ 2.4 m*M*; KCl 2.0 m*M* and Ca gluconate 1.0 m*M*) and bubbled with air. PD was measured through agar-Ringer bridges and calomel electrodes with a Keithley 200 B electrometer and the SCC with the external circuit described by USSING and ZERHAN[8]. Tissue conductance was calculated as SCC/PD.

[1] M. BURG, L. STONER, J. CARDINAL and N. GREEN, Am. J. Physiol. *225*, 119 (1973).
[2] O. A. CANDIA, Biochim. biophys. Acta *298*, 1011 (1973).
[3] C. J. LOTE, J. Physiol., Lond. *239*, 27P (1974).
[4] J. EIGLER, H. CARL and H. H. EDEL, Klin. Wschr. *44*, 417 (1966).
[5] J. FISCHBERG, J. A. ZADUNAISKY and F. W. DE FISCH, Am. J. Physiol. *213*, 963 (1967).
[6] J. A. ZADUNAISKY, O. A. CANDIA and D. J. CHIARANDINI, J. gen. Physiol. *47*, 393 (1963).
[7] M. B. BURG and N. GREEN, Am. J. Physiol. *224*, 659 (1973).
[8] H. H. USSING and K. ZERAHN, Acta physiol. scand. *23*, 110 (1951).