# The effect of work level and dietary intake on sweat nitrogen losses in a hot climate

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1. Nitrogen intakes, and N output in urine, faeces and sweat have been measured in six young Tanzanian men who were accustomed to a hot climate. The measurements were done while the subjects were receiving first a normal and then a low-N diet; and when they were performing moderate physical work, and had undergone a period of acclimatization.

2. When the subjects were acclimatized and working on a normal diet, their sweat output increased, with a fall in its N concentration. Total sweat N loss increased from an average of 0.10 to 0.71 g/d.

3. The effect of the low-N diet was to decrease both the sweat N concentration, and the rate of increase of total N loss in sweat, as sweat volume increased.

4. It is estimated that maximum sweat N losses would not exceed 1 g/d on an adequate diet, or 0.5 g/d on a low-protein diet. Our results provide no basis for recommending extra protein allowances to cover sweat N losses for workers in tropical climates.

Increased sweating follows exposure to a hot environment, or an increase in work output, or both. Individuals doing heavy or moderately heavy work in tropical climates are liable to sweat considerably, and it has been suggested by Consolazio, Nelson, Matoush, Harding & Canham (1963) that if nitrogen losses in sweat are not compensated for in some way they could become dangerously high, for example, in tropical workers whose diets are low in protein.

Compensation could occur by a reduction in the N concentration in sweat with increasing volume of output. Such a reduction has been reported by Bass, Kleeman & Quinn (1953) in young American males who were in the process of becoming acclimatized to heat, and by Consolazio *et al.* (1963). The findings of Mitchell & Hamilton (1949), however, do not show such an effect. Workers in tropical climates are already acclimatized to heat to some extent, and this might affect the relationship between sweat volume and sweat N concentration. There is some information about N loss in people living in the tropics (summarized in Table 1) but none about the effects of increased sweat production.

# 'Table 1. Nitrogen content of whole body collections of sweat, diet 'normal' unless otherwise stated; all subjects male

	Range of total cutaneous N loss $(g/d)$						
Experiment	Minimal work	Heavy work					
Jamaican students*							
Hot climate		0.13-0.20					
English students†							
Cool climate	0.5-0.3	<del>_</del>					
Hot climate		0.2-0.2					
English seamen <del>t</del>							
Hot conditions		0.3-0.8					
Adult Africanst							
Hot climate	0.190.48						
American students							
Cool conditions§	0.06-0.14						
Warm conditions§	0.07-0.20						
o 5 g N intake/d, cool conditions	0.038-0.175						
24 0 g N intake/d, cool conditions	0.128-0.332	and the second					
96.0 g N intake/d, cool conditions	0.269-0.571						
* Ashworth & Harrower	(1967).						

† Collins, Eddy, Hibbs, Stock & Wheeler (1971).

1 Darke (1960).

§ Sirbu, Margen & Calloway (1967).

|| Calloway, Odell & Margen (1971).

Compensation for increasing sweat N losses might also be brought about by a reduction in urinary N output; but, as Ashworth & Harrower (1967) point out, sweat N losses are small in relation to urinary loss, and within the limits of accuracy of balance techniques it would be almost impossible to show such an effect if it occurred. There is some indication (see Table 1) that, when the dietary N content is low, sweat N concentration falls. There is, however, no information about the combined effects of moderate or heavy sweating and a low-N diet, or on the response of sweat N concentration to diet in acclimated subjects.

In this paper we report the effect of a combination of moderate physical work and a low N intake on the sweat, urinary and faecal output of N in young men who had spent all their lives in a hot climate, and who can therefore be presumed to be accustomed to it. The subjects of this experiment were in fact subjected to the additional heat load of exercise in a hot room in order to maintain and if possible enhance their state of acclimatization.

### EXPERIMENTAL

# Subjects and plan of experiment

The study was carried out in the Physiology Department of the Faculty of Medicine, University of Dar es Salaam, Tanzania, during April–June 1969 towards the end of the 'hot' season. The volunteer subjects were six male Tanzanian medical students, aged between 23 and 26 years.

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The plan of the experiment was as follows, the duration of each observation period being 3 d, after a run-up period of 3-4 d on a controlled diet:

Study period	Dietary N	Work level
I	Normal: 10–13 g/d	Habitual activity only
	Acclimatiz	ation period of 10 d
2	Normal: 10–13 g/d	Habitual activity, plus 2 h exercise daily
3	Low: 4-6 g/d	As study 2

The exercise consisted of rhythmic stepping, using a step 23 cm high, at an average environmental temperature of  $32^{\circ}$  dry bulb,  $28^{\circ}$  wet bulb. Each subject spent a total of 2 h stepping, in bouts of 15 min work followed by 15 min rest. No restriction was put on the subjects' ordinary activity, but they were following an experimental schedule which kept them in the laboratory ( $26^{\circ}$  dry bulb) or in their hostel (about  $28-30^{\circ}$  dry bulb) for most of the day. Activity diaries kept during this time show that most of their habitual activity can be graded as sitting, standing and performing light work, and walking. Outside shade conditions were about  $32^{\circ}$  dry bulb,  $27^{\circ}$  wet bulb. Energy expenditures were calculated from these diaries, using factors obtained by calibrating the subjects during habitual activities by means of a Douglas bag and Haldane gas analysis apparatus.

#### Diets

The subjects' diets were prepared in the medical students' hostel by a nutritionist and a cook. During each study, and the preceding run-up period, each subject adhered to a constant menu with the same amount of food and drink each day. Cooked foods were prepared from fixed recipes, all cooking was done in distilled water, and all foods given to the subjects were weighed to the nearest 5 g. Fluids were measured to the nearest 1 ml. Although some foods and drinks contained negligible amounts of N, studies were also being done (to be reported elsewhere) of sodium, potassium and iron losses, so that it was necessary to control the intake of everything except glassdistilled water.

At the same time as each subject's food was weighed out, a duplicate was weighed, and each day's duplicate intake was homogenized, mixed thoroughly, and sampled for later analysis.

Studies 1 and 2 were preceded by a 3 d 'run-up' period during which the subjects ate their constant diet, but did not collect excreta. Study 3 was preceded by a 5 d run-up.

The diets during studies 1 and 2 were based on menus of the subjects' own choice. In study 2 the basic menu remained the same, but the subjects increased their calorie intake by about 10% in response to their increased work output. In study 3 these diets were modified by the removal of eggs, meat and milk and the addition of more carbohydrate-rich foods. Energy intakes fell slightly during study 3, partly because of the unfamiliar diet and partly because the subjects had increased their intakes somewhat in excess of needs during study 2.

The subjects had access to distilled water for drinking at all times and had a daily

allowance of beer, Coca-cola and orange squash, which they could take at any time during the 24 h and which was increased slightly in studies 2 and 3 to facilitate the intake of extra fluid.

## Urine and faecal collections

For faecal collections each subject was provided with his own plastic bucket and seat. The samples were weighed, and homogenized in the bucket with 200 ml of distilled water. The homogenate was stored in a deep-freeze. Faecal losses were related to the balance period by use of carmine markers. A pill containing carmine dye was taken by each subject at the start and again at the end of the 72 h study, and faeces was collected between the time of the first appearance of dye and its final disappearance.

Urine was collected under supervision, and volumes were measured, from each subject before retiring at night and again before breakfast. To these samples the subject added any urine passed during the 24 h period.

## Sweat: whole body 24 h collection

07.00 hours. The subject was weighed nude and then stood in a large polyethylene bag which was suspended from an overhead metal ring. He was sprayed down with about 5 l distilled water and then dried and reweighed. He then dressed in cotton pants, shirt, trousers and socks, all previously washed in distilled water. Excess sweat produced during the day was absorbed by a small, clean towel carried in a polyethylene bag.

*Exercise period.* When the subject was about to do step climbing he undressed and placed his clothing inside a large polyethylene bag. He then put on shorts, socks and plimsolls washed in distilled water. During the exercise period he kept himself dry with a towel washed in distilled water. At the end of the exercise period he undressed, was weighed, and the socks, shorts and towel were added to his previously used clothes. He then put on a new set of clothing.

22.00 hours. The subject was washed down, dried and weighed as before. Cotton pyjamas washed in distilled water were supplied for overnight wear and these were brought along to the hospital in a polyethylene bag to be added to the washings at 10.00 hours the next morning.

The absorbent clothing and towels added to the washings of the subject were spin-dried and the excess fluid was returned to the polyethylene bag. The volume of the washing fluid was recorded and a 200 ml portion taken into a polyethylene bottle and stored in a deep-freeze.

# Controlled hyperthermia test and sweat samples

The controlled hyperthermia test bed (Fox, 1967) was used to assess the degree of heat acclimatization at intervals throughout the investigation. As indices of acclimatization, the deep body temperature at which sweating was initiated and the total sweat produced were recorded for a standard period of controlled hyperthermia. Unfortunately it was not possible to use the sensitive method of determining sweat onset by applying starch-iodine papers; the temperature at which the first sweat sample was

obtained by aspiration from the suit was used instead. The subject was washed down by spraying with distilled water. After weighing on a Spido man-balance, he was then dressed in a polyethylene inner suit, dressed in the outer ventilated suit attached to the heating system of the bed and allowed to rest comfortably for 30 min (air to suit  $30^{\circ}$ ). Temperatures in the mouth and external auditory meati were recorded continuously with thermistors and temperature meters. After body temperature readings had reached equilibrium, body temperature was raised at first slowly (air to suit  $45^{\circ}$ ) until the temperature of sweat initiation was reached, and then rapidly (air to suit  $55^{\circ}$ ) until the target body temperature of  $38^{\circ}$  was attained. Hyperthermia was then maintained at  $38^{\circ}$  for a further 30 min, and during this time sweat was continuously sucked by vacuum from the inner polyethylene suit and collected for analysis. At the end of the experiment, the subject was cooled by blowing cool air through the suit, washed with distilled water after the inner suit was removed and finally reweighed. The sweat collected from the suit was analysed for N.

#### Calculation of cutaneous and respiratory losses

The subjects were weighed daily to the nearest r g on a Spido man-balance. Their intakes of food (F) and drink (D) were known, also their urinary (U) and faecal (S) output, and their change in weight  $(\Delta W)$ . Total cutaneous and respiratory weight loss (C/R) over each 24 h period was calculated as

$$C/R = F + D - (U + S + \Delta W).$$

Respiratory losses were estimated as averaging 300 g/d. The weight of epidermal debris was ignored, and total sweat output was thus estimated as (C/R-300).

## Analytical methods

Urine, sweat and sweat-washing samples were stored in a deep-freeze under toluene. Food and faeces were homogenized, sampled and frozen. For analysis, food and faeces were thawed, remixed and dried; sweat-washing samples were filtered. N in all samples was determined by an autoanalyser, after Kjeldahl digestion using a sodium sulphate and selenium catalyst.

The energy content of the food samples was also determined with a ballistic bomb calorimeter (Miller & Payne, 1959).

#### RESULTS

#### State of acclimatization

The total loss of sweat and amount of sweat collected during the controlled hyperthermia test (see Table 2) increased significantly in studies 2 and 3 compared with study 1, showing that some degree of increased acclimatization had been achieved by the step-climbing routine. The failure to find a reduction in the temperature threshold for initiation of sweating, which is an invariable occurrence with this acclimatization procedure (cf. Collins, Eddy, Hibbs, Stock & Wheeler, 1971), is ascribable to the insensitive method used.

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## Table 2. Results from the controlled hyperthermia test on six male adults

(Mean values and standard deviations)

Sweat collected (ml) $203 \pm 101$ $314^{*} \pm 163$ $201^{*} \pm 1$	3
Total sweat output (ml) $668 \pm 284$ $888 * * \pm 335$ $810 * * \pm 2$ Temperature at which sweating was initiated (°C) $37 \cdot 0 \pm 0 \cdot 26$ $36 \cdot 9 \pm 0 \cdot 11$ $36 \cdot 9 \pm 0$ Total nitrogen output (mg) $64 \cdot 8 \pm 26 \cdot 1$ $82 \cdot 8^* \pm 40 \cdot 9$ $62 \cdot 9^* \pm 2$ Average sweat N concentration (mg/l) $360 \pm 123$ $289^* \pm 81 \cdot 1$ $223^* \pm 3$	137 266 0·12 27·4 34·9

Significance of mean value for studies 2 and 3, compared with study 1: \* P < 0.05; \*\* P < 0.01.

#### Balance studies

The N and energy intake of each man during each 3 d study period is shown in Table 3, together with N losses in urine, sweat and faeces. The difference between intake and the sum of these losses is described as 'balance', but no account has been taken of the growth of hair and the nails or of epidermal tissue lost in the form of cell debris. The extent of hair and nail loss has been estimated as 0.024 g/d (Sirbu, Margen & Calloway, 1967).

On average (as shown in Table 3), the subjects were in approximate N balance during study 1, with a slight positive balance in study 2, which can be related to the subjects' increased work level and food intake. In study 3, all subjects were in negative N balance.

Table 4 shows the volume of urine and sweat produced, with fluid intakes; from Tables 3 and 4 it can be seen that the subjects all increased their fluid intake, and their sweat production, in response to the exercise in studies 2 and 3. Urinary N output remained constant in study 2 in spite of a small increase in intake, whereas sweat N output in study 2 increased by about 20%.

Individual values for urine and sweat output are shown in Figs. 1 and 2. (In these figures, results from studies 1 and 2 are grouped together, as a 'high-intake' group.) The relationship between volume (V) and N concentration (N) in both urine and sweat is a logarithmic one, such that  $N = b/V^a$ , or  $\log N = \log b - a \log V$ .

The main characteristic of this relationship is that if the value of the constant a is approximately equal to 1, then the product NV will remain constant, implying that increases in volume will be completely compensated for by reductions in N concentration. Values for the constants a and b found in the different studies were:

		Urine		Sweat						
Study	a	b	r	a	b	<b>r</b>				
1 and 2 3	0.94 1.10	3·817 3'945	-0.88 -0.82	o∙46 o•77	4·054 4·824	-0.55 -0.54				

## Hyperthermia sweat

Table 2 shows the N concentration of sweat samples collected from subjects in the hyperthermia bed during the run-up periods for all three studies. In all subjects, the N concentration in sweat fell significantly during study 2 and then again during study 3.

Table 3. Nitrogen and energy intake, and nitrogen output of male adults during the three study periods

	(1/4)	menditure	by diary)	2447	2468	2823	2427	2334	2110	) 2435	2594	3145	2498	2520	2505	2098	) 2642	2584	3123	3079	2442	2462	2075	) 2636						
	Rneem (	Diatory F.	intake (	2751	2509	2756	2926	2758	2489	2698 (11.3 MJ	2751	2765	3229	2832	2586	2432	[M 711) 9972	2817	2422	2662	2590	2433	1912	2575 (10 <sup>.</sup> 8 M]						
(	-	ſ	Faeces	0.040 0	0.037	0.032	0.036	0.036	0.026	0.035	0.053	o.036	610.0	0.030	: L90.0	0.041	0.038	0.057	0.043	0.020	0.022	0.030	0.031	0.037						
	g/kg d)	Output	Sweat	910.0	600.0	110.0	٥.00	£10.0	٥-00	010.0	0.020	0.012	0.015	800.0	010.0	800.0	210.0	200.0	200.0	200.0	200.0	900. <b>0</b>	900.0	900.0						
	Mean (9		Urine	0.186	6£1.0	0.163	0.185	0.186	0.140	0.168	181.0	0'141	0.155	661.0	0.209	0.156	0/1.0	990.0	020.0	920.0	<b>0.0</b> 73	<u>560-0</u>	9 <u>5</u> 0.0	69 <b>0.0</b>						
	gen		Intake	0.268	0. 177	o.189	0.225	0.253	o.186	912.0	0.274	0.189	0.211	0.241	0.283	0.218	0.236	0.087	090.0	0.073	<i>LL</i> 0.0	o.o78	<i>LL</i> 0.0	920.0						
gen		Balanco	(g/d)	L.1 +	- 1.3	I.I –	- 0:2	6.0+	4 0.7	1.0+	+1.3	<b>г.о</b> +	+1.4	8. <b>0</b> +	+1.5	+0.8	<b>0</b> , I +	- 2.7	-4-0	- I.4	<u>с.</u> 1 –	-3.2	6.0 -	-2.2						
Nitro		ſ	Balance	+ 5.1	6.8 –	13:4	9.0-	+2.7	+2.1	+ 0.3	1.4+	+0.3	+4'3	+2.5	+4:4	+2.5	+3.0	0.4	1.21	1.4-1	- 2.0	2.6 -	-2.6	-6.7						
	(g) p		Faeces	6.4	7-7	6.5	6·8	5.7	4.5 2	6.3	8.6	7-3	4:0	7-8	6.5	1.4	6.8	6.3	0.6	6.5	4.8	6.2	5:4	6.8						
	during 3	Output	Swcat	2.2	6.1	2.3	1.4	2.0	1.2	8·1	3.3	2:4	9.0 3	1.5 I	9·1	<b>1.4</b>	2.5	1.1	1.1	9.I	0.1	6.0	1.1	1.1						
	Total		Urine	2.62	30.5	32.9	34.8	2.62	24.2	30.2	2.62	28.9	31.4	34.4	33.5	1.72	30.8	10 <sup>.8</sup>	14.5	4.II	6.£1	1.51	8.6	9.21						
	Ĺ	~	-	~	~	~	-		Intake	42.7	36.2	38-3	42.4	9.68	32.0	38.5	44.3	38.9	42.7	46.2	45.4	37-8	42.5	14.2	12.5	14.8	14-7	12.5	13-7	13.7
		Body-wt	on ist day (kg)	1.23	68.2	67.3	62-7	52.2	57-4	1.09	53.8	68.4	67.3	63-8	53.5	6.45	60.8	53.9	68.6	9.29	63.4	53.0	58.6	6.09						
			Subject	JS	FK	CM	EM	GU	FM	Mean	Js	FK	CM	EM	GU	FM	Mean	Js	FK	CM	EM	GU	FM	Mean						
		F40	period	I,							и							ŝ												

	Flui	d intako Study	e (ml)	Urine	volum Study	e (ml)	Sweat volume (ml) Study				
Subject	ī	2	3	I	2	3	ī	2	3		
JS	7755	8320	8970	3405	2510	539 <b>0</b>	6742	9247	7079		
$\mathbf{FK}$	7890	9845	10490	6195	4395	7490	3906	8571	5666		
CM	9160	11035	10135	4305	3380	6185	7442	11099	7477		
$\mathbf{E}\mathbf{M}$	6650	6955	7355	5648	4105	3355	5105	6548	7852		
GN	6885	7340	8645	4710	488 <b>0</b>	5035	5225	4713	5982		
$\mathbf{F}\mathbf{M}$	7905	7870	8640	5232	4770	5115	5957	6155	7522		
Mean	7708	8561	9039	4916	4007	5428	5730	7625	693 <b>0</b>		

Table 4. Total fluid intake and urine and sweat volumes of male adults during each 3 d study period

When account had been taken of the water content of food and faeces, and of the metabolic production of water, the mean calculated water balance was approximately -200 g/d in each period.



Fig. 1. Relationship between urine volume and urine nitrogen concentration in men receiving normal (studies 1 and 2) and low-N diets (study 3).  $\times$ , studies 1 and 2;  $\bigcirc$ , study 3. Fig. 2. Relationship between sweat volume and sweat nitrogen concentration in men receiving normal (studies 1 and 2) and low-N diets (study 3).  $\times$ , studies 1 and 2;  $\bigcirc$ , study 3.

#### DISCUSSION

### Urinary and faecal N output

Although our subjects were in negative N balance in study 3 they were not on a protein-free diet, and it might be expected that their urinary and faecal N output would have been somewhat higher than estimates of minimum endogenous losses. This was so; the average urinary N output of our subjects on the 6th-9th days of the low-N diet was 0.069 g/kg per d, whereas Young & Scrimshaw (1968) found a value of

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0.041 g/kg per d in young adults on the 6th day of a protein-free diet. The faecal loss in our subjects was hardly affected at all, whereas in adults on a protein-free diet it may fall to 0.02 g/kg per d (World Health Organization, 1965).

Our results show an inverse relationship between urinary N concentration and urine volume, which has the effect of keeping total urinary N output almost constant, at a given level of intake.

# Sweat losses

As shown by Fig. 2 and the values for the constants a and b found in the different studies (see p. 548), there is an inverse relationship between sweat volume and N concentration, such that a large increase in sweat volume is accompanied by a reduction in N concentration and a small increase in total N loss in sweat. This is also shown by the values for sweat N loss in the hyperthermia test bed (Table 2). In study 2 the 24 h sweat output increased, partly because of an increase in energy expenditure (see Table 3) and partly because of the acclimatization procedure. Over the 3 d of study 2 there was a small increase in total sweat N loss. The increased sweating in the hyperthermia tests in study 2, however, was not due to increased activity but solely to short-term acclimatization Thus it appears that increased sweat volume, whether due to acclimatization or to increased work, results in a reduction in sweat N concentration.

The effect of a low N intake is to reduce sweat N concentrations; this is shown by the analysis both of sweat washings and of pure sweat samples from the hyperthermia test. Thus total sweat N losses were less in period 3 than in period 1, although sweat volume was considerably higher. The rate of increase of sweat N loss, as sweat volume increased, was also reduced.

From the constants a and b (p. 548), it can be calculated that in our subjects a daily sweat output of 1 l would contain on average 0.46 g N, and that this total output would increase to 0.9 g in 5 l (or 0.015 g/kg for a 60 kg man). On the low-N diet, 1 l of sweat contained an average 0.32 g N, and 5 l would contain 0.45 g (0.0075 g/kg for a 60 kg man). It seems unlikely from these values that sweat N output in acclimatized subjects would exceed 1 g/d on a good diet, or 0.5 g/d on a low-protein diet. These estimates of maximum sweat loss are of the order of one-sixth to one-tenth of the total endogenous loss by other routes.

In view of the difference in magnitude between urinary and sweat losses, it is hardly surprising that our results provide no evidence for a compensatory reduction in urinary N output, after an increase in sweat N loss.

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