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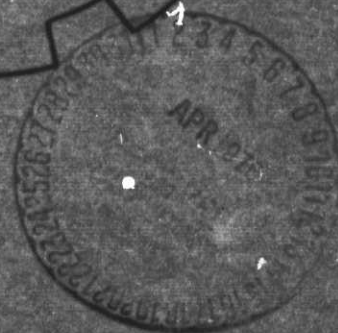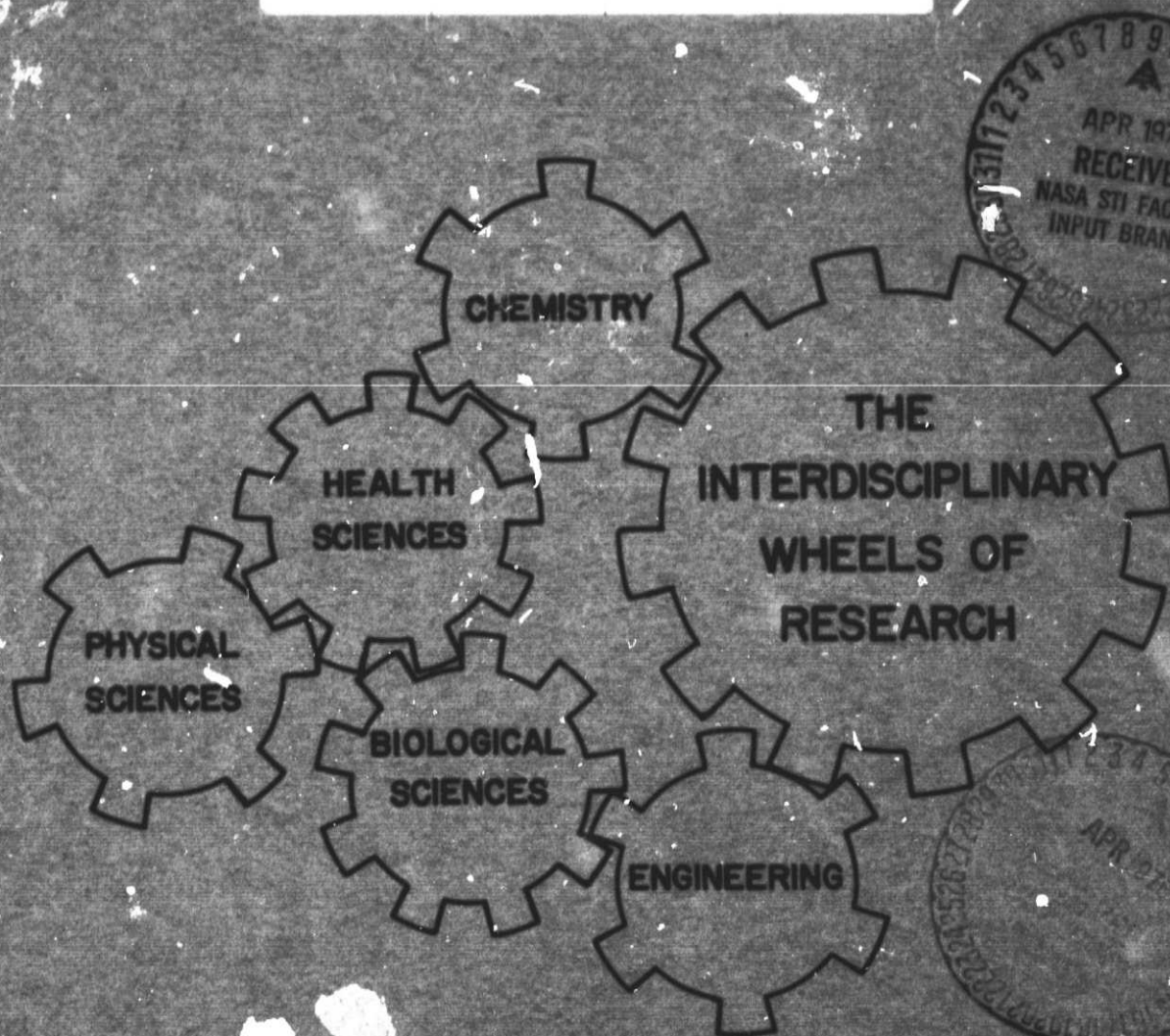
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UNIVERSITY OF MISSOURI-COLUMBIA
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The influence of the space environment on man was first noted in the Gemini missions which showed that weightlessness effected both cardiovascular and bone density changes. Left unanswered until Apollo, however, was the question of individual variation as well as the course and extent of these alterations. Data from Apollo showed several physiological responses to 0-G, the most important of which were decreased cardiovascular responsiveness, reduced red cell mass, and musculoskeletal deterioration. Vestibular related problems were also observed for the first time. In addition, a tendency to lose body weight was noted both through tissue loss as a result of decreased caloric intake, and intracellular water loss as a consequence of increased aldosterone production. The biomedical information obtained from Apollo permits a scientific description of physiological responses to 0-G, and a formulation of hypothesis of the course of the changes taken from Berry (1974).
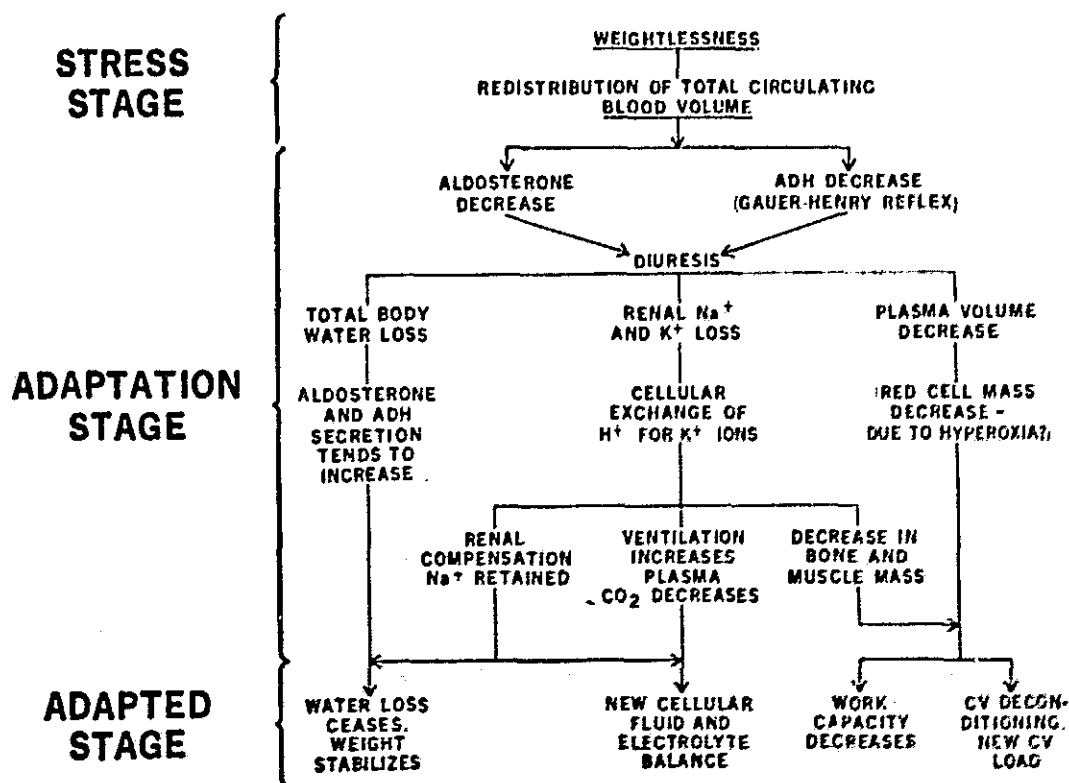

Fig. 4. Diagram of the hypothesized course of adaptation to weightlessness.

Although this hypothesis has begun to be tested in the Skylab mission, many questions remain unanswered. The mechanisms concerning the adaptation of the cardiovascular system is unknown. Hormonal and nervous changes responding to circulating bloot .ume, shifts in blood volume, or intrinsic changes in cardiac muscle may singly or in various combinations underly the observed alterations. The 0-G adaptation syndrome, and the 1-G re-adaptation syndrome which may require treatment by counter measures, cannot be properly treated until mechanisms underlying these changes have been elucidated. Much of the experimentation concerning physiological responses to extraterrestial flight stress and adaptations has been conducted in human subjects, the astronauts. Only limited research has been done on laboratory animals under controlled conditions. Data concerning the human response obtained using this experimental approach has served the immediate purpose of demonstrating that man can withstand prolonged periods in a 0-G environment. However, little or nothing is known about long lasting effects, nor is there any substansive information concerning mechanisms involved in the short term changes. Our initial effort in this regard has been a review of literature in order to assess the state of the art. The conclusion reached from this review is that whereas some simulations can be used in recognizing physiological responses in man, particularly in readaptation.to a 1-G environment, little is known about mechanisms relevant to the changes. These remain to be investigated in animals, where invasive techniques and organ removal are possible, and where exposures to actual environments of 0-G followed by 1-G are also possible. Only experimentation with Sky-lab and/or shuttle capabilities are realistic in the immediate future, and the efforts in

our laboratory will continue to press for an on-board mammalian experiment in which many animals can be used and in which several integrated parameters can be studied (for example: cardiovascular, renal system, intestinal function, temperature regulation, and bioenergetics). These animal experiments will begin to answer questions raised by the human trials.

We propose to continue our investigations in concert with other laboratory ef.orts and focusing on problem areas relating to cardio-vascular and renal physiology, intestinal function and temperature regulation. In brief, our experiments are designed to parallel mans exposure to extraterrestial environments and to be supportive of manned flight through the assessment of alterations which occur in response to 0-G. Mechanisms responsible for both the changes already observer and adaptive homeostatic responses will be investigated in the areas mentioned above.

Although the U.S. effort through NASA has decreased to some extent, our laboratory continus to investigate problem areas we believe to be supportive of prolonged manned space flight. These investigations, carried out in concert with laboratories of other consortium members, have focused in problem areas noted in previous reports. Briefly, these areas include the following.

1. Renal function and its relationship redistribution to shifts of body fluid compartments (renin-angiotensin-aldosterone system).

2. The relationship of catecholamines to thermoregulation.

3. Gastrointestinal function examined directly through quantitative assessment of alterations in the flux of specific metabolites such as glucose.

Funding constraints, both in terms of support of extramural laboratories and flight missions have prompted investigations in yet another area, simulated 0-G. These studies have been r dertaken in an effort to assess mechanisms underlying reported changes in space, thereby predicting and preventing or treating the untoward effects of 0-G on man in space.

The Simulation of Zero Gravity

Continued exploration of space in missions of increasingly longer duration have encouraged and will continue to argue for ground based research designed to predict the effects of 0-G, and minimize its insult to man. In many areas of investigation, the scientist can design laboratory experiments where the variable of interest can be altered while other parameters are held fixed. Unlike these experiments, gravity is a unique variable which may neither be eliminated nor reduced in any laboratory confined to the earth's surface. Although these alterations in gravity cannot be achieved on the earth's surface, various methods have been developed to simulate zero or subnormal gravity.

Many of the effects of gravity and its absence on biological systems are predictable on the basis of physical law concerning mass, density, fluids, etc. On the basis of these laws, experiments can be designed in which one or more of the predictable passive effects of the interactions between gravity and a biological system are compensated for or negated. Current techniques may provide virtual elimination of gravity tissue interaction relative to one physical property, but be of little value in approximating 0-G relative to other physical parameters. The experimental design often poses intrinsic properties unrelated to those of primary interest. Furthermore, since only an aspect of weightlessness is simulated, the influences of concomitant changes in other systems of the experimental subject may confuse the results. The various techniques employed and their advantages and disadvantages are reviewed by Wunder et al. (1968), and will only be briefly listed here.

In simulating specific anticipated effects a direct reduction of mechanical force in specific areas has been employed. This has involved such procedures as immobilization involving denervation, tenotomy, and plaster casts. A reduction in the forces required to oppose gravity has also been employed in various ways to simulate the effects of weightlessness. These techniques have involved bed rest, fluid immersion, and tumbling devices, and have yielded much pertinent and useful information. Indirect methods involving simulated high G forces have also be used. These studies have involved both extrapolation from data obtained at hyper G and examination of responses to 1 G after prolonged exposure to hyper G. Finally, opposing accelerations have been employed in a limited way to simulate 0-G, and involve such techniques as free fall and combined linear and radial acceleration, i.e., parabolic trajectories.

All of these techniques for the simulating of weightlessness suffer from deficiencies which must be considered in the evaluation of experimental data. Artifacts, introduced as a consequence of the method of simulation, must be carefully separated from data relating to effects of gravity. A few of the more recent publications in the area of simulated weightlessness are given below as an example of the effort currently being expended in this area. A 30-day experiment in Modelling the Physiological Effects of Weightlessness (Gernin and Kakurin, 1972) is representative of the Russian effort in this area. This study examined the effects of simulated weightlessness on such diverse parameters as cardiovascular system function and bone demineralization. Their observations had shown that initial adaptation to weightlessness cause

a more intensive redistribution of fluid compartments than ordinary hypodynamia in the horizontal position. Cosmonauts had reported sensations of blood rushing to the head, puffiness and reddening of the face and skin. Taking these observations into account, a more adequate model of 0-G was selected utilizing a tilted body position in which the head is somewhat below the position of the legs. This position is termed the orthostatic 4° incline. Our purpose is not to review the data obtained from this large and complex study, but rather to point out the model employed. These workers note criticisms of models employed in the simulation of weightlessness, but call attention to the fact that predictions made on the basis of model studies have been correct and, therefore, valuable. In particular, models have predicted orthostatic instability, a decrease in physical performance, impairments in the mechanisms enduring the maintenance of vertical posture, impairments of coordination of walking movements, and atrophy of antigravity muscles.

Efforts in this country, in the simulation of the 0-G environment, have been reviewed by Murray and McCally (1969) and are typified by Jacobson et al. (1974), a more recent study. The latter experiment, a bed rest model predicts the effect of hyper-G encountered upon return to earth by astronauts, and evaluates the effect of a cutaway G-suit in preventing visual impairment and syncope which occurs upon exposure to hyper-G following adaptation to weightlessness. Although the authors comment that the bed rest model chosen is not a perfect analog of weightlessness, their data strongly suggest a problem which will be encountered by the astronaut upon return to earth, and experiments with bed rest models can offer a partial solution.

In short, a variety of models exist for this simulation of weightlessness, each suffering from one or more disadvantages or deficiences. Nonetheless, the similarity of alterations observed under the influence of 0-G and in the models would suggest that terrestrial experiments do indeed make possible the simulation of weightlessness. Such simulation not only enables the recognition of physiological responses to weightlessness, but also enables the evaluation of different agents and regiments to prevent and treat certain disorders. We feel that simulation studies have yielded and will continue to yield valuable information supportive of NASA's manned space effort. In addition, such studies employing small mammals will prove of added benefit by enabling the investigator to search the mechanisms underlying changes which occur during prolonged weightlessness through the use of invasive techniques not possible with human subjects. Knowledge of mechanisms will point to agents or counter measures necessary in dealing with changes occuring in man's adaptation to weightlessness.

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RENAL FUNCTION

Scintigraphic Studies

Introduction

Kidney function, i.e., glomerular filtration and tubular secretion, have been examined in the golden hamster (Mesocricetus auratus) as a function of body temperature. Tempel and Musacchia (1975) investigated the corticomedullary solute gradient in the kidney of hypothermic and hibernating hamsters. The presence of a gradient from cortex to medulla was used to infer the presence of glomerular filtration while its absence suggested the elimination of filtration. Data from this study suggested the absence of filtration in the artificially hypothermic hamster (rectal temperature, T_{re} 7°C) and marked reduction in the hibernator. Rewarming hypothermic animals by placing them in an environment at an ambient temperature (T_a) 22°C showed the return of filtration two hours after reaching T_{re} 37°C. Further, data suggested function had not returned in the animal at T_{re} 18°C.

Studies are currently in progress utilizing the more direct technique of scintigraphic imaging of the kidneys. Gamma emitting radiopharmaceuticals that are selectively taken up by the kidney are injected intravenously into animals that are positioned below a Nuclear of Chicago H.D. Anger scintillation camera and periodic recordings are taken. The data is both displayed on a cathode ray tube, and stored in computer core. The latter data may then be analyzed to quantify kidney function in terms of whole body activity. ^{131}I-orthoiodohippurate and [99m]TC-Sn-diethylene triamine-pentoacetic acid (DTPA) have been employed to investigate tubular secretion and glomerular filtration, respectively.

These studies were taken both to confirm and extend the previous
studies involving assessment of the renal solute gradient. The more
direct approach afforded by this method has the following advantages:
(1) a more accurate assessment of the correlation of temperature and
function is now possible. Kidney function is visualized directly with
no lag present so that dynamic studies are now possible; (2) the two
major components of urine formation can be separated, i.e., filtration
and secretion; and (3) this extremely sensitive method enables the in-
vestigator to ascertain when, in relation to temperature, urine forma-
tion becomes functionally significant by noting the appearance of the
label and hence, urine in the bladder.

Results and Discussion

The scintiphoto work has supported earlier conclusions of the lack
or marked reduction of glomerular filtration in the profoundly hypother-
mic hamster (T_{re} 7°C), as seen in Figure 2, a representative example.
Several additional findings possible with the new technique are noteworthy.

1. Perfusion of the kidneys is evident in the hypothermic animal
(T_{re} 7°C), although filtration is absent. This is evident from studies
employing two different labelled compounds: ^{99m}TC-Sn-DTPA, and ^{99m}TC-
labelled erythrocytes.

2. At these very low core temperatures secretion by the tubular
cells appears to be less depressed than is glomerular filtration. This
finding is suggested by the concentration of ^{131}I-orthoiodohippurate by the
kidneys at T_{re} 7°C.

3. Filtration begins to occur at a significant rate in hypothermic animals which have rewarmed to rectal temperatures in the range of 12 to 14°C. This is in contrast to earlier interpretation of solute gradient data which suggested little or no function at T_{re} 18°C. The scintigraphic technique enables a dynamic evaluation of renal function not afforded by an examination of solutes. Although filtration begins between T_{re} 12-14°C, it is undetectable by the gradient technique due to the time required to re-establish the solute gradient.

4. Finally, the scintigraphic technique enables the investigator to determine the temperature at which the processes of filtration and secretion contribute to actual urine formation through observation of bladder activity. Our studies have shown that urine begins to appear in the bladder as soon as a rectal temperature of approximately 22°C has been reached.

5. The technology described and the results can be utilized in studies of mammals exposed to extraterrestrial environments, for example, the 0-G adaptation envisioned in a prolonged space experiments using mammals, and in support of man related problems suggested by Skylab and other experiments, the cortico-medullary method and the radio chemical/scintigraphic techniques can be employed.

The scintigraphic techniques provide an approach to the study of dynamic change which may be measured after an animal is returned from prolonged space flight to an earth side laboratory.

RENAL FUNCTION

Mechanism Studies

Introduction

These investigations have prompted studies designed to elucidate the mechanisms underlying the reported changes in renal function at reduced body temperatures. The large dependence of filtration on physical factors has led to studies of such parameters as flow to the kidney, blood pressure, and volumes of such fluid compartments as red blood cells and plasma. The size of the hamster has prevented direct estimates of rates of renal perfusion or the use of electromagnetic flowmeters. However, numbers and disposition of the animal have made possible quantitation of renal blood flow by the technique of Sapirstein (19). This method employs the radiotracer ^{86}Rb which is injected intravenously. This γ emitter is then deposited in the tissues in direct proportion to the extent of its perfusion. Removal of the tissue or organ and subsequent counting enables an estimate of perfusion of an organ in terms of percent of total body flow. This may better be converted to actual flow using the Fick method to determine cardiac output. Blood pressures are being measured using standard techniques as are red blood cell and plasma volumes. The latter have involved use of ^{51}Cr labelled red cells and ^{125}Iodinated serum albumin for red cell and plasma volumes, respectively.

Results and Discussion

Figure 2 summarizes the findings concerning organ flow in control and hypothermic hamsters. It is noteworthy that flow to the kidneys of the hypothermic hamster is decreased by greater than 50%. Blood pres-

DISTRIBUTION OF CARDIAC OUTPUT IN NORMOTHER...
AND 18 HR. HYPOTHERMIC C... DEN HAMSTERS
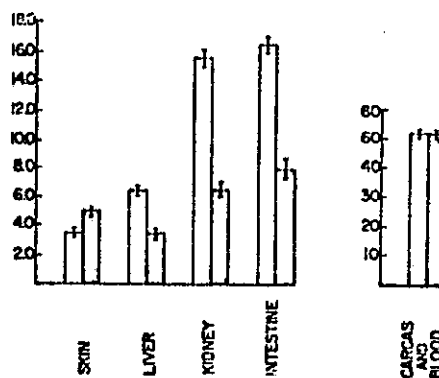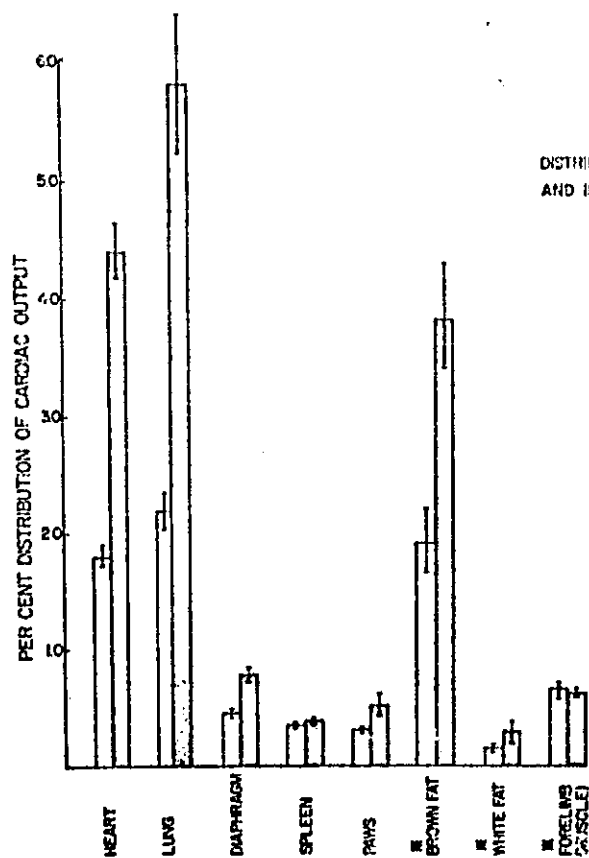
HYPOTHERMIC (T_{re} 7 C) N=17
CONTROL   (T_{re} 37 C) N=12
K   = %/gram
± SEM

sure data seen in Figure 4 also explain in great part the reduction in glomerular filtration seen in the profoundly hypothermic animal. The T_{re} 7°C animal demonstrates a mean pressure of approximately 60 mmHg, roughly a 60% decrease from a control value of approximately 120 mmHg. Figure 4, which shows the correlation of temperature and pressure, suggests that mean aortic pressure must increase to approximately 80 mmHg.

Data on red cell and plasma volumes is not yet summarized, but it appears to support the earlier interpretation of a reduction in plasma volume in hypothermia inferred from an increased hematocrit.

These results have explained in large measure the findings of markedly reduced glomerular filtration in the profoundly hypothermic animal, and have provided a more accurate picture of the correlation of temperature and function in rewarming studies. Studies are currently in progress to compare and contrast these data with data on the same parameters from hibernators. It is felt that such a comparison will yield much useful and interesting information concerning responses in the hibernator which are truely adaptive, and not merely cold suppressed.

Our findings have also stimulated an interest in assessing the role, if any, in the depressed metabolic state. There is suggestive evidence for its involvement, although a literature search has yielded information concerning this system in only one study. Brown et al. (1971) noted high urinary potassium concentration, and increased vascular resistance in the dormant brown bear; however, no significant increase in peripheral renin concentration was found. Nevertheless, the following evidence would argue for a role for this system in a true hibernator. Chaffee et al. (1963) found the histology of kidney from cold-acclimated

hamsters to reflect changes observed in renovascular hypertension.
Zimny and Levy (1969) in a study of 13-lined ground squirrels observed
an increase in the juxtaglomerular granulation index, suggestive of in-
creased renin secretion in hibernation. Increased activity of the adre-
nal cortex, the source of mineralocorticoids, has also been noted during
hibernation by such investigators as Engel et al. (1957), Hoffman (1968),
and Soumalainen (1960). The lack of information concerning the role of
this system in both the control and hibernating states has prompted
efforts to develop assay procedures for renin. The bioassay procedure
frequently employed has not proved sufficiently accurate and reproducible.
We are currently working on modifying a commercially available radioimmuno-
assay for use in our experimental animal. We also feel that the eluci-
dation of the workings of this system in control animals will yield use-
ful information which when combined with studies on simulated weightless-
ness, will have bearing on the elucidation of mechanisms underlying fluid
compartment shifts in astronauts in space.

The development and utilization of these methods with small mammals
such as the hamster or rat should be considered essential to the planning
of mammalian experiments in future Skylab, Shuttle or other space explora-
tion programs. Although some of the methods may not lend themselves to
actual inflight experimentation, they are more important in determining
the nature and mechanisms involved in pathophysiologic changes which occurred
during exposure to the extraterrestial environment. Such changes, and
their ultimate effects in physiologic and biochemical homeostasis can be
assessed after animals are returned from the space environment. Thus, we
are recommending that in planning an experiment in which numbers of mammals

(amenable to statistical analysis) will be used, then selection of methods herein employed with hamsters will have been tested and readily adaptable for rats, hamsters or other species.

METABOLIC AND CIRCULATORY ALTERATIONS IN
HEAT ACCLIMATED HAMSTERS

Background and Rationale

Investigations of heat acclimation in the golden hamster indicate a depression of in vitro tissue metabolism as well as whole animal oxygen consumption (Cassuto and Chaffee, 1966; Cassuto, 1968). Such metabolic alterations are of obvious advantage in maintaining heat balance at 35°C. More recent studies involving the hamster (Jones and Musacchia, 1973, 1974, 1975) indicated increases in myocardial and renal catecholamine concentrations and concomitant decreases in utilization of NE in these tissues with heat acclimation. Such decreases in sympathetic neurotransmitter utilization (considered to reflect decreased sympathetic nerve activity) may be in response to the overall reductions in metabolism and organ weight (Cassuto and Chaffee, 1966; Cassuto, 1968). However, interpretation of functional advantages of altered sympathetic activity with heat exposure is difficult based on the present understanding of heat acclimation in rodents. Indeed, the quantitative assessment of temperature regulation and cardiovascular function in heat acclimated hamsters has not been reported. Adaptive characteristics of the heat acclimated state might involve cardiovascular changes as displayed by altered vasodilation of the heat exposed rat (Rand et al., 1965). Expansion of vascular volume might also be expected in long term acclimation to hot environments. The facilitation of heat dissipation from core to periphery is likely to involve shifts in volume

distribution as indicated with heat conservation of the cold acclimated rat (Jansky and Hart, 1968).

The hamster is specialized for survival in cold temperatures as evidenced by hibernation. However, the hamster is also capable of adapting to high temperatures (Cassuto and Chaffee, 1966; Jones and Musacchia, 1973, 1974). Since decreases in heat production with heat acclimation will reach minimal levels, changes in heat dissipation mechanisms would appear to be of more adaptive significance in meeting environmental demands of high temperatures.

A study has been initiated to focus on heat dissipation and cardiovascular adaptive changes of heat acclimated hamster. Metabolic measurements are being made to define the acclimated state in terms of previous studies (Cassuto and Chaffee, 1966; Cassuto, 1968). Avenues of heat dissipation changes in blood flow distribution and vascular volumes are also being examined.

Methods

Male golden hamsters from our closed colony at approximately 3 months of age, weighing 120-140 g are being acclimated to either 34°C or 22°C. After approximately 6-9 weeks in the hot environment animals are removed and the various measurements made.

Metabolic rate is determined by placing the animal in an air tight plexiglass chamber and measuring the change in percent oxygen content of the effluent air. By measuring the air flow rate and correcting to standard temperature pressure, oxygen consumption ($\dot{V}O_2$) can be measured, using a Beckman paramagnetic oxygen analyzer, model G-2,

according to the method of Depocas and Hart (1957). During the course of the $\dot{V}O_2$ run, Evaporative Water Loss (EWL) is measured by determining the increase in weight of glass tubes containing indicating drierite which are placed at the exit port of the chamber. Respiratory frequency is determined visually with a stop watch during the same time that $\dot{V}O_2$ and EWL are being measured.

Deep body temperature measurements (T_{re}) are made with Wesco rapid-recording thermometers (Schultheis Company, N.Y.C.) inserted about 2 cm into the rectum. Skin temperatures (Ts) are measured using a Yellow Springs Instrument telethermometer recording thermometer and a calibrated thermistor probe attached to the dorsal posterior section of the hamster's back approximately 2-3 cm from the base of the tail.

Measurement of cardiac distribution are based on the technique described by Sapirstien (1958) employing the tracer [86]RbCl. Venous injection of this radiolabel is taken up by the capillary beds of all tissues except the brain at a rate such that each tissue retains a constant amount for a given period. Since the tracer behaves as if there were zero venous drainage, tissue concentration of the isotope reflect the percent of cardiac output received.

Plasma and red cell volumes were measured simultaneously to determine whole body hematocrit (WBH) in acclimated animals. Both techniques employed isotopic dilution principles using ^{51}Cr tagged red cells and ^{125}I labeled serum albumen according to standard techniques (ICSH, 1973).

It is reasonable to state that each of the methods and techniques used herein with the hamster are adaptable for use with other small rodent

species which may be used in space related experiments.

Results

Results of $\dot{V}O_2$, measurements indicate that oxygen consumption is drastically reduced with heat exposure as the 34°C group $\dot{V}O_2$ was 49% of the 22°C (control) group. In contrast EWL is elevated almost 2 fold in heat exposed animals (.966 ± .043 ml/kg hr^{-1}) compared to control animals (.519 ± .096 ml/kg hr^{-1}). Differences in respiratory frequency between the two groups are not significant. Thus, long term acclimation of the hamster to 34°C results in a adaptive decrease in $\dot{V}O_2$ while increasing EWL.

Measurements of core temperatures indicate that with heat exposure hamsters are slightly hyperthermic (Tre = 37.40°C, P < .001) compared to their temperatures at 22°C (Tre = 36.85°C). In contrast, alterations in skin temperature are markedly different with heat acclimation. Control values averaged 32.35 ± .17°C while the heat acclimated temperatures were 36.60 ± .10°C (P < .001). The core to shell gradient is therefore, reduced from 4.5°C to 0.81°C. These changes suggest that in the heat exposed animal the core temperature is expanded to encompass the shell area and thus more heat is dissipated.

Alterations in the distribution of cardiac output suggest that organs in the splancnic bed of the heat exposed group, i.e., liver, kidney and intestine, receive a lower percentage of total blood flow than in the control group. Fractional distribution to the liver is decreased by 11%, the kidney by 27% and the intestine by 16% in the heat exposed animals compared to the distribution in the controls.

Brown fat tissue also appears to receive a decreased fraction of cardiac output in heat acclimated animals. In contrast, the carcass of the heat acclimated animals receives an increased fraction of total flow in that 13% more of the ^{86}Rb went to the carcass of the 34°C exposed group than went to the same tissue in the controls. Thus, these results suggest that with heat exposure of the hamster blood flow is decreased in the splancnic beds and increased in the periphery.

Alterations in vascular volume with heat acclimation indicate a 10% decrease in red cell volume (P < .001) and a slight decrease in plasma volume. Although the change in red cell volume is significant, comparison of the whole body hematocrit of the two groups is not different (.476 ± .004 for 22°C vs. .461 ± .009 for 34°C). However, a similar comparison of large vessel hematocrit indicates a slight (3.7%) but significant reduction (P = .0075) with heat acclimation.

Discussion

The drastic reduction in whole-body metabolism with heat acclimation is of major significance in the animal's ability to maintain heat balance. The extent to which evaporative water loss is an important avenue of heat dissipation for the hamster is determined by comparison of calories of heat dissipated (EWL x latent heat of vaporization at .57 Kcal/g H_2O) against calories of heat produced. The calculated fraction of heat production removed by EWL is 4.0% in the 22°C group and 15% in the 34°C group. Thus, the 50% reduction of heat production in heat acclimation is complimented by an almost 4 fold increase in the fraction of heat production dissipated by evaporative mechanisms. How-

ever, the majority of metabolic heat in both groups must be dissipated by other mechanism, namely radiation and convection.

Temperature measurements of heat acclimated animals indicate that the core temperature has greatly expanded to encompass most of the shell area. In greatly reducing the core to surface gradient by increasing skin temperature, a positive gradient from skin to air is maintained which assures convective and radiative heat loss. If skin temperatures were not elevated above ambient, a net heat gain from the environment to the animal would likely result in a pathological elevation of T_{re}.

Mechanisms which may account for the heat acclimated hamsters' ability to maintain elevated T_s is likely to involve redistribution of blood flow or expansion of vascular volume or both. Experiments of fractional distribution of cardiac output indicate that at 34°C there is a decrease in blood flow to the liver, kidney and intestine but an increase in flow to the carcass and presumably the periphery. These data would suggest that there is displacement of flow from splanchnic area to the periphery.

The 11% decrease in RCV of the heat acclimated hamsters could be attributed to the overall decrease in metabolism with a reduced demand for oxygen delivery to tissues. Combined reductions of red cell and plasma volume (4%) constitute a 7.4% reduction in total circulating volume with 34°C acclimation. Whole-body hematocrit decreased 3.2% in heat exposure but the change was not significant ($P > .05$). However, large vessel hematocrit was significantly reduced (3.7%, $P < .01$). This is interpreted as reflecting a combination of the reduced RCV and

the greater hematocrit of large vessels due to the Fahraeus-Lindquist effect.

Such reductions in vascular volumes will reduce the circulating volume available for fractional distribution to various organs. Thus, the reduced blood flow to the liver, kidney and intestine, as suggested for the heat acclimated animal, would be decreased to an even greater degree than is apparent by the fractional distribution experiments.

Reductions in whole-body and tissue metabolism combined with decreases in vascular volumes, and the redistribution of blood flow away from splanchnic areas suggest that adaptive thermoregulatory features of the hamster to high ambient temperatures consists primarily of massive reductions in metabolic activity. Although there is an elevation in T_{re}, the magnitude of the reduction in metabolism combined with previous reports of decreased sympathetic function (Jones and Musacchia, 1974) and decreased tissue metabolism (Chayoth and Cassuto, 1972) would suggest a depressed metabolic state.

Summary

1) Oxygen consumption is greatly reduced by heat acclimation (50% of control value) in the hamster.

2) The per cent of heat production dissipated by evaporation in heat-acclimated hamsters is 400% greater than that of non-acclimated hamsters.

3) Red cell volume is decreased by 10% in heat-acclimated hamsters but there is no change in plasma volume.

4) Radiative and convective heat loss are maintained in heat accli-
   mated hamsters by means of increased skin temperatures.

5) Mechanisms of elevated skin temperatures with heat acclimation
   are suggestive to involve a shunting of blood flow from the core
   to the periphery.

INTESTINAL TRANSPORT OF SUGARS IN
THE HEAT STRESSED HAMSTER AND RAT

In the earlier part of this study, we have been concerned with the effect of chronic exposure to 34°C on intestinal transport of sugar _in vitro_ by the hamster. Our earlier findings may be summaried as follows: Glucose uptake per gram intestinal tissue in an _in vitro_ everted gut sac preparation is unchanged from control values following exposure to the animals to T_a 34°C for one to eight weeks. Serosal transfer is increased after eight weeks heat exposure. Small intestinal mass is reduced in heat stressed hamsters with two weeks to eight weeks exposure compared to both pair fed and _ad lib_. controls at 22°C.

The purpose of the present study was two-fold. In part one we examined the ability of the _in vitro_ hamster intestine to transport a non-metabolizable sugar 3-O methyl glucose. We attempted to quantitate alterations in sugar transport kinetics by the determination of the K_m and V_{max} for 3-O methyl glucose using radioisotopic techniques. Secondly, we extended out study to a second model, the rat, to determine if resistance of intestinal transport in the hamster to heat stress might not be species specific.

Materials and Methods

Hamster: Male golden hamsters weighing 125-145 grams taken from our closed colony were individually caged and randomly divided into two groups. One group was housed in a Hotpack walk-in chamber at T_a 34°C for six to eight weeks. The control group was housed in animal quarters at 22°C.

Food (Wayne Lab Blox) and water were available ad libitum for both groups. Lighting was on a 12:12 cycle. At the end of the exposure periods animals were fasted for 24 hours prior to sacrifice.

The kinetics of ^{14}C 3-0 methyl glucose transport in the hamster was determined in intestinal tissue slices by the method of Crane and Mandelstom (1966) as modified by Olsen and Rosenberg (1970). Extracellular water was determined by ^{14}C mannitol and by ^{14}C inulin. Radioactivity was measured on a Beckman liquid scintillation counter LS250.

Rat: Male Sprague-Dawley rats weighing 140-180 grams were individually caged and randomly divided into two groups following an initial acclimation of one to two weeks in the animal quarters. One group was maintained in a Hotpack walk-in environmental chamber at 34°C for two to three weeks with food (Wayne Lab Blox) and water available ad libitum. Control animals at 22°C were pair fed by a single daily evening feeding to maintain the same growth rate as the heat exposed animals. Water was available ad libitum. Lighting was on a 12:12 cycle. Animals in both groups were fasted the final 24 hours of the exposure period. Animals fed ad libitum were also used for gut weight determinations and a growth rate study.

Analytical Methods: In vitro mucosal uptake and serosal transfer of glucose were measured in everted gut sacs in the rat by methods described by Wurth and Musacchia (1973). Three sacs were made from the jejunum of each animal. Glucose concentrations were determined by a modified othro-toluidine method ("Trucose", American Monitor) on a Technicon auto-analyzer. Inulin was used as an indicator for alterations in serosal fluid volume and appropriate corrections were made in serosal glucose

concentration. Inulin analyses were made on a Technicon autoanalyzer. Tissue metabolism was calculated by determining the difference between the initial quantity of glucose in the mucosal plus serosal fluids and the final concentrations in those volumes plus tissue glucose. Tissue glucose was determined by homogenizing the tissue at the end of incubation period in 5% TCA and analyzing an aliquot of the supernatant.

Table 1. Effect of Chronic Exposure to 34°C on In Vitro Transport of 3-0 Methyl Glucose

	Heat Exposure 34°C	Control 22°C	Statistical Significance
[Intracellular] [Extracellular]	5.56 ± .185 (18)	4.92 ± .183 (16)	P < .025
Tissue weight	.0538 ± .002	.0792 ± .003	P < .001

Number of flasks ( ) Incubation time is 30 minutes in 5 mM 3-0 methyl glucose and tracer isotope

Table 2.   Effect of Chronic Exposure to 34°C on Tissue Characteristics

   In Vitro

	Heat Exposed 34°C	Control 22°C	Statistical Significance
Tissue water %	79.08 ± .058 (8)	78.95 ± .167 (7)	N.S.
Extracellular Space (%)	11.79 ± 1.26 (10)	9.06 ± 1.50 (10)	N.S.
Tissue weight (gms)	.049 ± .002	.080 ± .005	P < .001

Number of flasks ( ) Incubation time is 30 minutes in 5 mM 3-0 methyl glucose plus tracer inulin

Table 3. Effect of Chronic Exposure to 34°C on the Kinetics of 3-0
         Methyl Glucose Transport

	Heat Exposed 34°C	Control 22°C
K_m (mM)	16.79 (6)	23.32 (6)
V_{max} (µm/ml tissue water/15 min)	52.16	68.35

Number of animals ( )$_{14}$ Incubation time is 15 minutes. Extracellular
space determined with ^{14}C mannitol

Results

The ability of hamster gut tissue to concentrate 3-0 methyl glucose is increased in heat stressed hamsters _in vitro_ (Table 1) although parameters such as tissue water and extracellular space remain unchanged (Table 2). The kinetics of 3-0 methyl glucose transport is also altered with heat stress (Table 3).

Data from the rat study suggests that rats respond to chronic heat exposure in a manner similar to hamsters. _In vitro_ uptake of glucose per sac is depressed at three jejunal locations (Figure 1) but there is no change when mucosal uptake is calculated on a wet weight basis (Table 4). However, serosal transfer per gram wet weight is decreased at all three locations (Table 4). Increased serosal transfer in the presence of unchanged mucosal uptake could be the result of decreased diffusional barriers or depressed utilization of the substrate. Figure 2 shows that there is depression of glucose metabolized by tissues from animals with chronic heat exposure. As in the hamster, total gut tissue is decreased following heat exposure suggesting that total capacity for transport is also depressed in the rat (Figure 3).

Conclusions

Both the hamster and rat show altered sugar transport following chronic heat stress. The ability of the tissue to take up the natural metabolite, glucose per gram tissue, appears to remain constant with chronic heat exposure although serosal transfer values increase. In comparison with the hamster, in the rat alterations of serosal transfer appear earlier. Both animals show depression of total gut

mass with heat exposure suggesting an over all depression of transport capacity.

Study of transport on the non metabolizable 3-0 methyl glucose suggests there is alteration of sugar transport in the hamster exposed to heat. Both concentrating ability and reduced K_m suggest that transport in heat exposed animals is increased for this species.

In terms of a potentially utilized subject for NASA space laboratories, where intestinal function is of interest, the rat appears to be a promising model. These are the first such experiments reported for rats, the basis of our previous measures of intestinal responses to stress have been with studies of the hamster. Additional studies with the rat as a potential subject for flight experiments are recommended.
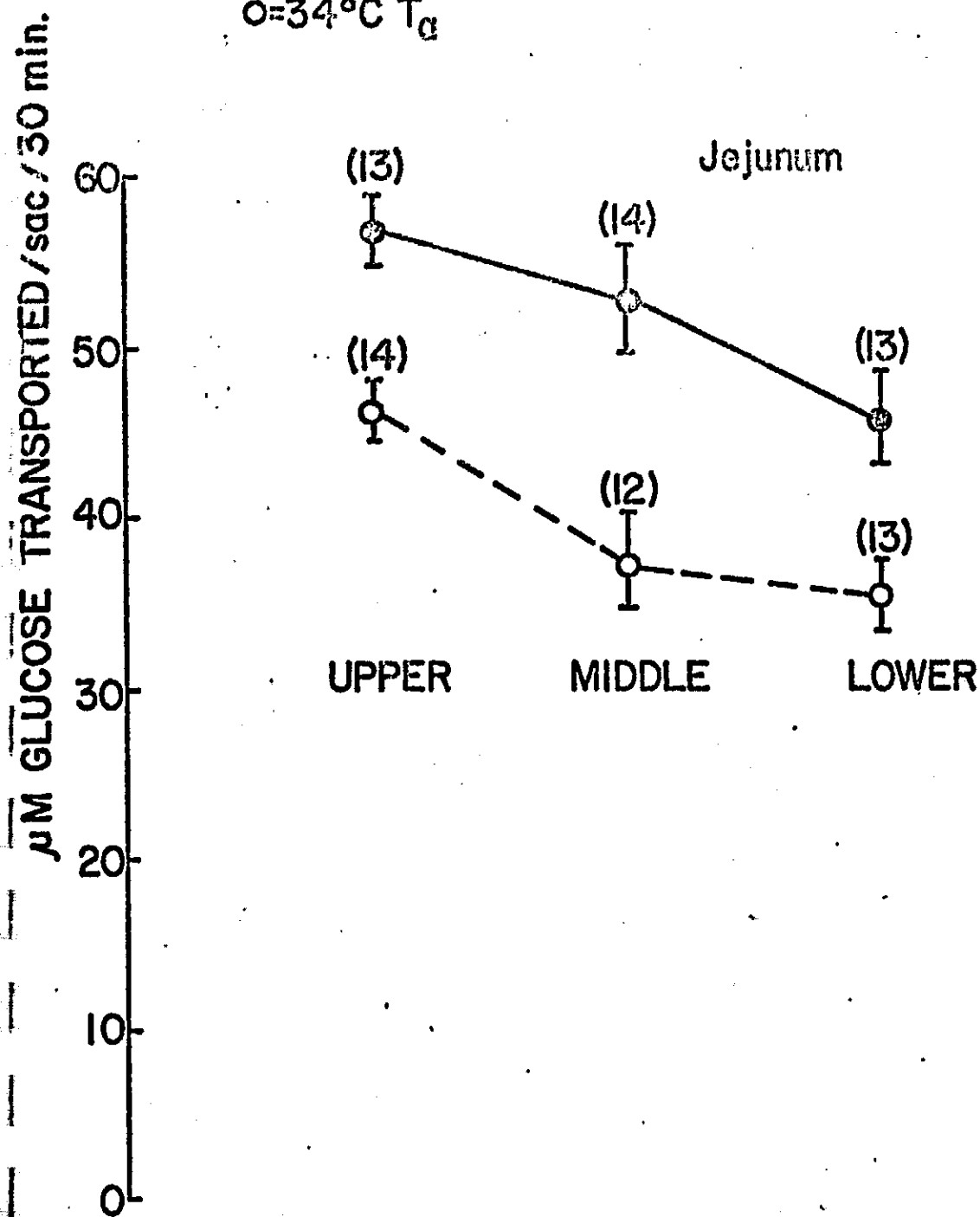
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INTESTINAL TRANSPORT OF GLUCOSE IN THE RAT
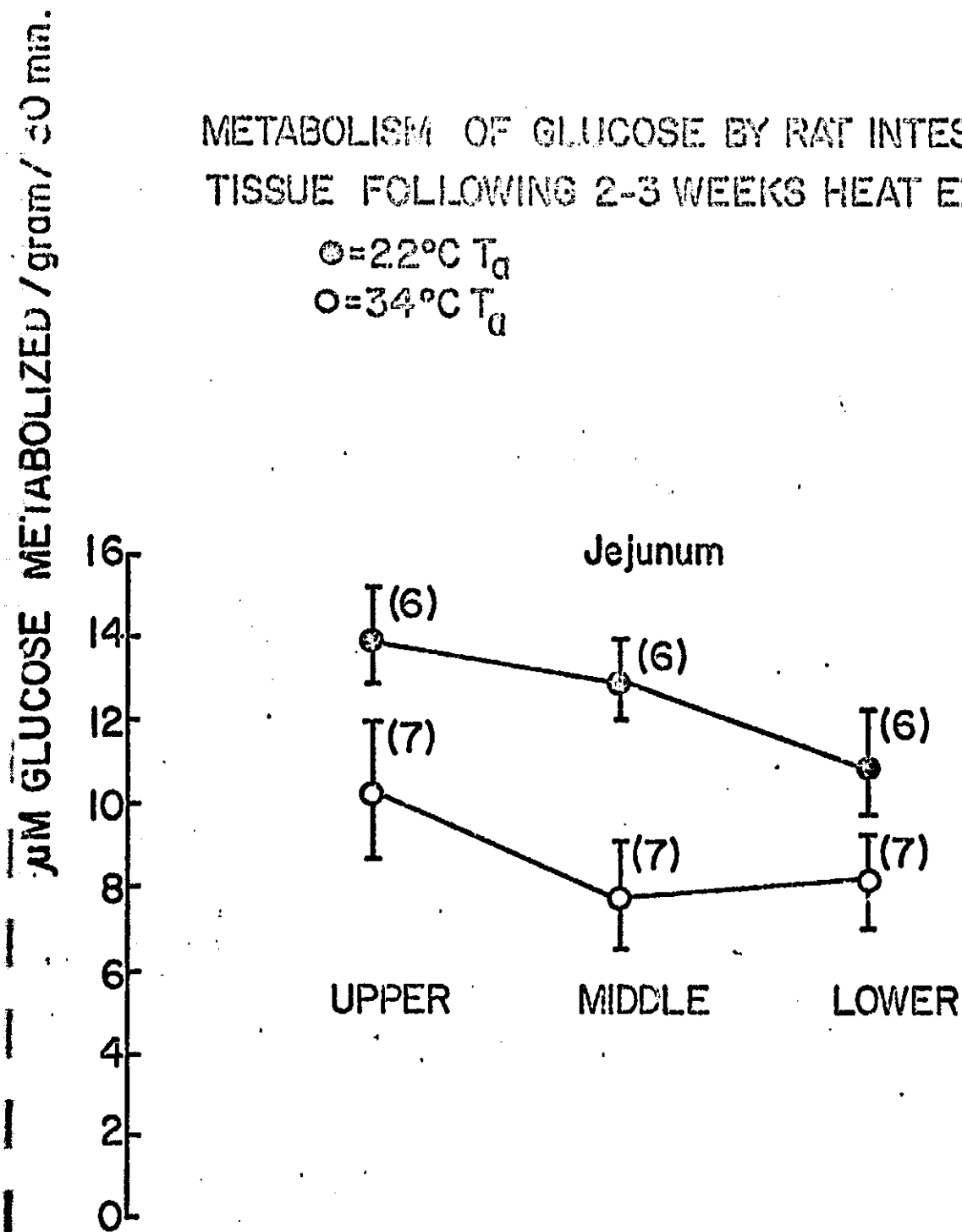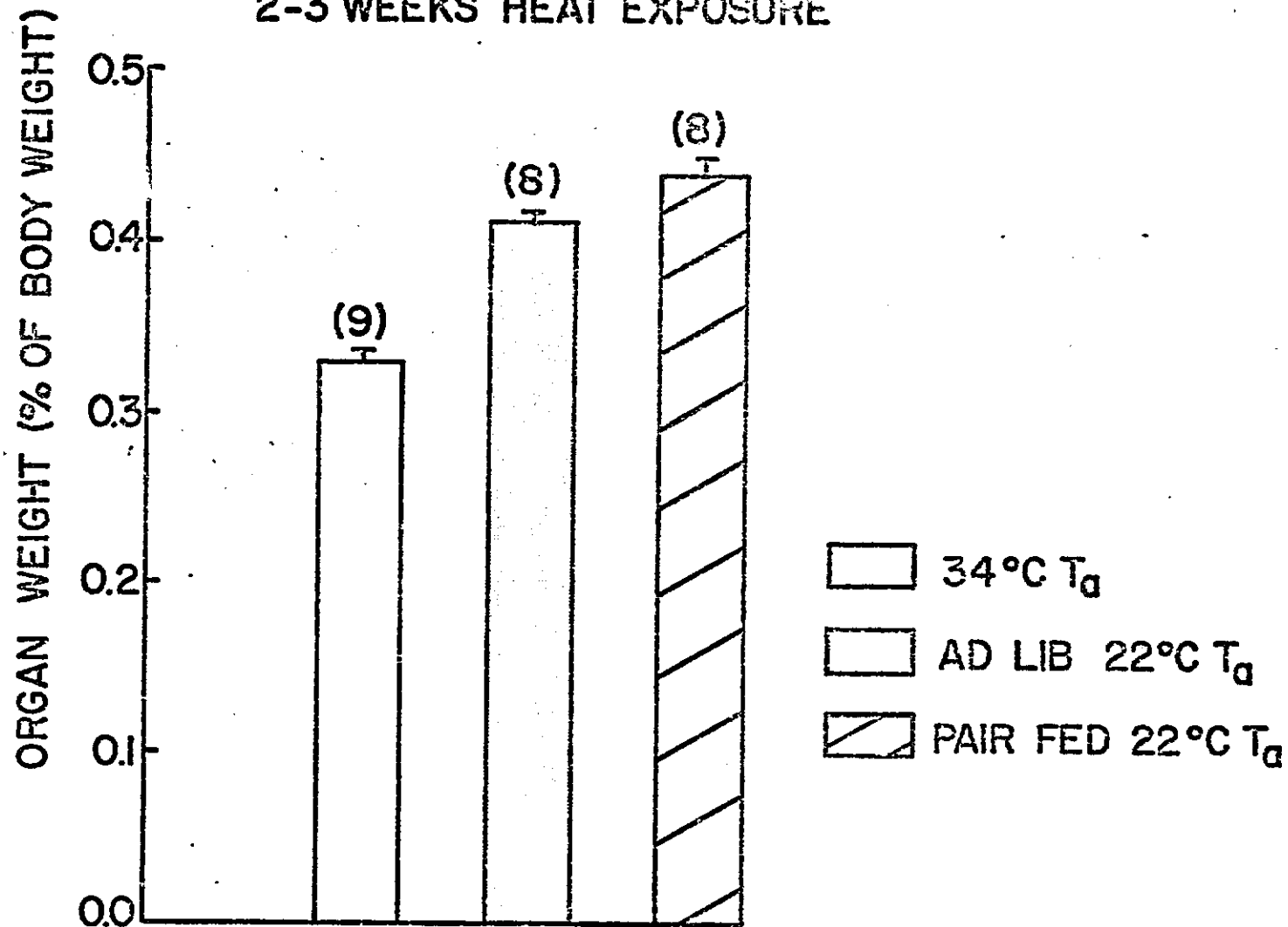FOLLOWING 2-3 WEEKS HEAT EXPOSURE

⊘ = 22°C T_a
O = 34°C T_a

Jejunum

(13) (14) (13)

(14) (12) (13)

UPPER   MIDDLE   LOWER

μM GLUCOSE TRANSPORTED/sac/30 min.

60
50
40
30
20
10
0

METABOLISM OF GLUCOSE BY RAT INTESTINAL TISSUE FOLLOWING 2-3 WEEKS HEAT EXPOSURE

$\odot = 22°C\ T_a$
$\bigcirc = 34°C\ T_a$

RESPONSE OF RAT INTESTINAL WEIGHT TO 2-3 WEEKS HEAT EXPOSURE

ORGAN WEIGHT (% OF BODY WEIGHT)

Legend:
- 34°C T_a
- AD LIB 22°C T_a
- PAIR FED 22°C T_a

INTESTINAL TRANSPORT OF GLUCOSE IN VITRO IN RATS CHRONICALLY EXPOSED TO 34°C T_A

	JEJUNAL LOCATION		
	UPPER	MIDDLE	LOWER
HEAT EXPOSED			
MUCOSAL UPTAKE	154.10 ± 7.83 (13)	119.80 ± 11.51 (12)	104.73 ± 6.10 (13)
SEROSAL TRANSFER	63.22 ± 6.65 (13)*	43.74 ± 5.81 (12)*	33.36 ± 4.32 (12)*
CONTROLS			
MUCOSAL UPTAKE	136.70 ± 6.31 (13)	118.61 ± 7.46 (14)	96.92 ± 5.72 (13)
SEROSAL TRANSFER	42.52 ± 6.11 (13)	25.25 ± 3.92 (14)	21.60 ± 3.09 (13)

MEANS ± SE. uM/ML/GM WW. NUMBER OF SACS ( ). *$P < .05$.

Personnel and facilities in this laboratory in the Dalton Research Center are relatively unchanged although a new technician, Mr. R. Rick, has joined us. Staff members are identified with each project, these include Dr. George Tempel, an Assistant Professor in the Department of Physiology, Ms. Mecca Carpenter and Mr. Stephen Jones, both Ph.D. candidates, and Ms. Janet Burnett, a technician. In addition, Dr. Wynn Volkert, Associate Professor of Radiology, has been associated with these projects for several years.

We have continued our research projects in tissue intermediary metabolism with Dr. Cecil Entenman and currently have two papers accepted for publication in Comparative Biochemistry and Physiology. Two additional manuscripts are in preparation.

Our funds are being expended and are committed in accordance with provisions set forth in our proposal.