USAMRNL March 1967
PHYSIOLOGY DIVISION PROTOCOL

Project No. 3A104501B71R Research in Biomedical Sciences
Task No. 05: Environmental Medicine
Work Unit No. 82: Metabolic Effect of Altitude
Study No. 4: Endocrine Effects of Altitude

I. INTRODUCTION

It has been established that acute exposure of man to high altitude causes the onset of a distressing, incapacitating syndrome, termed "mountain sickness." (1, 2, 3) This syndrome is characterized by impaired physical and psychomotor performance. The transitory effects of high altitude exposure - the most debilitating symptoms - severe headache, nausea and vomiting, extreme fatigue and anorexia usually last up to 5 - 6 days in most subjects. These symptoms are variable in intensity in different people or in the same person at different times.

Various theories have been advanced in the attempt to explain the etiology of mountain sickness. Research has been concerned primarily with the evaluation of cardiopulmonary and acid-base alterations in man. Still, all of the reported, established alterations do not explain the phenomenon of acclimatization that occurs in most subjects within 5 - 7 days. Recently, in the attempt to discover the cause of mountain sickness and the mechanisms of adaptation, investigators have focused on endocrine function, shifts in body fluids, and electrolyte balance in man at high altitude. The following sections summarize the observed alterations of endocrine function, body fluids and electrolytes in man at high altitude. There is a wealth of literature of similar observations conducted on animals which is essentially consistent with the reports on man (4 - 14).

Body Fluids and Electrolytes in Man at High Altitude

When man ascends rapidly to high terrestrial locations, there is a continuous decrease in plasma volume, a decrease in extracellular fluid volume, and a rise in intracellular fluid volume. Total body water increases (15). Total body potassium obtained from body scanning for K^{40} is noted to be increased by one research group (16) but similar measurements made by USAMRNL show that total body potassium decreases (15). Excessive urinary excretion of sodium, potassium and chloride ions has been documented (15, 17). There is a significant rise in salivary sodium to potassium ratio (18) and urinary sodium to potassium ratio (17). In addition to electrolyte changes, urine volume increases at high altitude (19).

These observed effects of altitude on man resulting in body fluid shifts and electrolyte alterations suggest diminished aldosterone secretion by the adrenal glands. Very recently it has been reported that urinary aldosterone excretion does decrease at altitude, approaching zero within three days of high altitude exposure (19). These findings appear to be consistent with the observed pattern of urinary electrolyte excretion. Additional investigation is necessary in order to establish and correlate the electrolyte changes with aldosterone secretion at high altitude by means of balance studies with human volunteers. If aldosterone excretion is indeed diminished at high altitude in the face of sodium loss, diminished plasma and extracellular fluid volumes, the mechanism is unknown and is contrary to accepted physiological controls of aldosterone secretion.

Adrenocortical Function in Man at High Altitude

Various investigators using intermittent chamber studies in man (20, 21), or actual field studies at high altitude (17, 22, 23),

have observed a rise in the 24-hour urinary excretion of 17-ketosteroids and 17-hydroxycorticosteroids during acute exposure. One research group did note a difference in the excretion pattern of 17-hydroxy-corticosteroids and 17-ketosteroids. The latter decreased initially. Adrenal function has been studied in a group of high altitude natives, and is the same as a comparable group living at sea level (24).

These studies are generally consistent with the known physiological control of pituitary adrenal function during stress. The correlation of adrenocortical function to "mountain sickness" and the magnitude of environmental adrenocortical steroid secretion and ACTH release during stress require additional investigation. The accurate measurement of these hormones during acute altitude exposure can determine the desirability of a possible therapeutic means of controlling the symptoms of high altitude stress in man.

Glucose Tolerance in Man at High Altitudes

Studies in man (25, 26, 27, 28) have established that there is a lower fasting blood glucose level and greater utilization of glucose at high altitude. Whether the cause of enhanced glucose utilization is secondary to increased insulin secretion or the effect of adreno-corticosteroids on glucose metabolism, is not known, and should be determined.

II. OBJECTIVE

The objectives of the present proposal to study human subjects at high altitude are (1) to evaluate through balance studies, the nature of electrolyte alterations; (2) to determine the magnitude of the stress response at altitude by evaluation of pituitary and adrenocortical function; (3) to measure and correlate the aldosterone secretion and excretion in relationship to electrolyte changes; and (4) to investigate the mechanism for increased glucose utilization.

The purpose of the study will be to correlate these findings with the severity of mountain sickness symptoms at 14,100 feet.

III. JUSTIFICATION

Reports concerning the Indian Army (29) have furnished alarming evidence of incapacitating medical and personnel problems resulting from altitude sickness, when a military force attempts to function on mountain locations. An understanding of the physiological alterations during altitude acclimatization and the relationship of these to various mountain sickness symptoms might lead to successful methods of pre-selecting or pre-conditioning troops.

Knowledge of the endocrine function in man at high altitude would shed light on the adaptive changes that occur during the stress of hypoxia. Significantly patho-physiological alterations in hormone secretion can be dealt with therapeutically. Recent Army Research Office conferences (30, 31) have pointed to a military need for studies on physiology, pharmacology and performance in high terrestrial environments.

IV. EXPERIMENTAL DESIGN

A. The subjects will be ten U.S. Army volunteers (19 to 25 years old) who have signed a volunteer form indicating their knowledge of the scope of the procedures planned, including the intravenous administration of tracer quantities of radioactive steroid compounds and their willingness to serve as subjects. The radioactive concentration of these isotopes will be recorded in the subjects' Army Health Records. The subjects will be interviewed, examined and selected by a medical officer after review of their health records to exclude cardiopulmonary, renal or endocrine disorders. A medical officer is to be present during the entire study and is authorized to terminate the experiment at any time continuation would be detrimental to the health of the subject.

The study will be conducted for a total period of 21 days beginning 11 September 1967. The sea level test site is to be selected on the basis of available facilities and volunteers. It is imperative that the sea level site be located in a temperate climate. A hot, humid climate would make electrolyte balance studies inaccurate. In addition, heat exposure alters blood volume and this would interfere with studies on aldosterone secretion. Tentatively, Fort Lewis, Washington has been selected as a site meeting these specifications. Other possibilities include Fort Ord, California; Fort Devons, Massachusetts and Fort Wainwright, Alaska.

During the sea level test period, constituting days 1 to 14, the subjects will be started on a constant metabolic diet which will require eight days of equilibration of body electrolytes prior to initiating balance studies. This diet will be continued until the end of the study on day 21. On day 15 the subjects will be flown to Colorado Springs or Denver and transferred by Army vehicle to the Army mobile laboratory on Pikes Peak (14, 100 ft). The altitude test phase will be conducted there with living quarters provided for the subjects.

B. Clinical Study

1. Plan of study

a. Sea level test period - 09:00 hours day 1 to 09:00 hours day 15.

b. Travel period - day 15.

c. Altitude test period - 09:00 hours day 16 to 09:00 hours day 21.

2. Diet

A prepared liquid diet comprising daily consumption of 2,800 calories as 55% carbohydrate, 15% protein, and 30% fat. The daily electrolyte and mineral intake will be 100 mEq. of sodium, 80 mEq. of potassium, 1600 mg of calcium, and 1200 mg of phosphorus.

The diet and water content are to be constant and analysed. During the day of travel, the subjects will be maintained on this diet. On days 5 and 19, the subjects will fast from 21:00 hours until 14:00 hours the following day (glucose tolerance test). There will be no smoking during this interval.

3. Water intake

Distilled water will be used for drinking purposes. Daily water intake will be measured and recorded. A minimum of 1500 ml of water per subject per day is to be consumed.

4. Medications

a. Glucose - 100 grams in distilled water administered orally on days 6 and 20 at 09:00 hours

b. 4-C^{14} - cortisol (1μc) and 1, 2-H^{3} - aldosterone (2μc) - administered intravenously in 10 ml sterile solution of 10% ethanol in water at 09:00 hours on days 9 and 17.

c. Sodium chloride - 500 mg in gelatin capsules in the event of diminished dietary intake to maintain constant daily sodium intake.

d. Potassium replacement elixir (5 ml contains 10 mEq. of potassium as gluconate and citrate salts) - to maintain constant daily potassium intake if intake of food diminishes when anoretic.

e. Calcium gluconate tablets - 500 mg if food intake diminishes when anoretic to maintain constant daily intake of calcium.

f. Aspirin and Darvon - administered orally at the discretion of the medical officer for headache. The dosage and time of administration are to be recorded.

g. Intravenous infusions of isotonic sodium chloride and potassium will be administered if severe vomiting occurs in order to maintain electrolyte and water balance.

5. Collections

a. Urine

Total 24-hour urine collections will be obtained each subject starting at 09:00 hours each day. Each collection d will end on 09:00 hours the following day with the subjects ng at this time and adding this specimen to the previous day's ction. The days of collection are 8 through 14 and 16 through The urine will be stored in large plastic bottles and kept under geration at all times during collection. At the end of each our collection period, all urine collected will be frozen and will ept in this state until analysed at the USAMRNL. All specimen es will be labeled with name of subject, and the starting and ng dates of the 24-hour collection. Radioactivity labels will be ked to all 24-hour urine samples collected after the administra- of radioisotopes to the subjects.

b. Blood

(1) Plasma insulin and glucose

At 08:30 hours on days 6 and 20, 5 ml of nous blood will be drawn into fluoride-oxalate vacuum tubes. llowing oral glucose administration, similar samples will be ken at 15, 30, 45, 60, 90, 120, 240, and 300 minutes. The es will be labeled and the contents frozen. The subjects are to frain from smoking from the period of fasting until the final ood sample is drawn.

(2) Plasma ACTH and cortisol

On days 9, 10, 13, 16, 17 and 18, venous lood will be taken at 08:45 hours. Thirty ml of blood will be rawn into a syringe containing heparin and transferred to entrifuge tubes and centrifuged. The plasma will be transferred o screw-cap glass tubes labeled "cortisol" and frozen. Forty ml

venous blood will be collected through sterile plastic tubing directly into a large centrifuge tube containing heparin. The centrifuge tube will remain immersed in an ice-water bath during the collection, then immediately centrifuged at 12 degrees centigrade. The plasma will be transferred to screw-cap tubes labeled "ACTH" and frozen. All tubes will be labeled with name of subject, time and date.

3) Fifteen ml of venous blood will be taken at 15:00 hours on days 8, 9, 10, 13, 14, 16, 17, 18, 19, and 20. Following centrifugation, the serum will be transferred to labeled test tubes and frozen.

c. Feces

Feces will be collected in plastic bags on days 8, 9, 10, 11, 12, 13, 14, 16, 17, 18, 19 and 20.

d. Vomitus

Any vomitus will be collected in plastic bags for analysis.

C. Measurements

1. Electrolytes

Concentrations of sodium and potassium in the diet, venous blood, stools and urine will be determined by AutoAnalyzer or flame photometry. Calcium will be measured by atomic absorption spectrophotometry. Chloride and creatinine will be measured by the Auto-Analyzer. The Fiske-Subbarow method will be used for phosphate concentrations.

2. Nitrogen

Urinary nitrogen content will be determined by the Auto-Analyzer. The macro-Kjeldahl technique will be used for nitrogen in the diet and in the feces.

3. Fat

Dietary and stool fat will be determined by routine analytical methods.

4. Glucose and insulin

Plasma glucose will be measured by AutoAnalyzer.
ma insulin will be determined by Captain J. Anderson, Metabolic
sion of the USAMRNL employing the method of Morgan and Lazarow

5. Steroids

a. Plasma cortisol by the Peterson modification (33)
thod of Silber and Porter (34) using the Beckman DU Spectro-
tometer.

b. Total 24-hour urinary 17-ketosteroids and 17-ketogenic
eroids by the Sobel (35) modification of the Norymberski method.
as-liquid chromatography will be used to determine individual
-ketosteroids.

c. Twenty-four hour urinary aldosterone excretion
y a modification of the double isotope technique of Kliman and Peterson
6) using 1,2-H^3-aldosterone, 1,2-H^3-tetrahydroaldosterone, and
cetic-1-C^{14} anhydride.

d. Twenty-four hour urinary cortisol excretion by the
method of Erlich (37), using 1,2-H^3-cortisol and acetic-1-C^{14} anhydride.

6. Plasma adrenocorticotropic hormone

ACTH levels in plasma will be determined by the method
of Vernikos-Danellis (38) by means of bioassay in hypophysectomized
rats.

7. Plasma and urine osmolality

Osmolality will be determined by means of the Fiske
osmometer.

D. Radioactive Steroids as Tracers in Human Subjects

1. Purification

4-C^{14}-cortisol (specific activity SA 15-30 mc/millimole)

H^3-aldosterone (SA 35 curies/millimole) obtained commercially
w England Nuclear Corporation will be tested for purity by
ography on three separate systems. The purified isotopes will
olved in absolute ethanol, and sterilized rendering them pyrogen-
Millipore filtration. The isotopes will be kept as a 10% solution
hol in sterile water in a sterile, multi-dose, stoppered vial.
ill be administered intravenously to the subjects by a medical

2. Dosage considerations and calculations

It has been determined that over 90% of injected radio-
ly labeled cortisol is excreted by human subjects via the kidney
h the first 48 hours (39, 40). Furthermore, no radioactivity
e detected in the body fluids in four days (39). The biological
life of radioactive cortisol in the human bloodstream is 60 - 80
tes). Similarly, over 90% of radioactive aldosterone injected
human subjects is excreted in the urine within 48 hours (42). The
tion of the tritium and carbon-14 labels in the steroid nucleus is
h that the labels remain an integral part of the compound in the
y and are excreted intact as steroid metabolites without degradation.
). This factor has enabled numerous investigators to employ these
topically labeled steroids in clinical research without the hazards
critical organ concentration, the random labeling of body water by
itium, or the expiration of carbon-14 carbon dioxide in human subjects.

Based on knowledge gained from reports in the literature, one
an make the safe assumption that the effective half-life (T 1/2) of
hese isotopes in man is one day (an over-assumption). The total
body burden would be calculated as follows: (44)

1. 4-C^{14}-cortisol. 1 μc injected into a 70 kg man assuming
tota dy distribution with an effective T 1/2 of one day.

$$D\beta = 73.8 \times C \times E\beta \times T \text{ rads}$$

$\bar{E}\beta \approx 0.050$ Mev. for C^{14}

$C \approx 1.0/70,000$

$T \approx 1$ day

$D\beta \approx 73.8 \times 1.0/70,000 \times 0.050 \times 1$ rads

$D\beta \approx 5.27 \times 10^{-5}$ rads

2. 1, 2-H^3- aldosterone. Two μc injected into a 70 kg man assuming total body distribution with an effective T 1/2 of one day.

$D\beta \approx 73.8 \times C \times \bar{E}\beta \times T$ rads

$\bar{E}\beta \approx 0.006$ Mev. for H^3.

$C \approx 2.0/70,000$

$T \approx 1$ day

$D\beta \approx 72.3 \times 2.0/70,000 \times 0.006 \times 1$ rads

$D\beta \approx 1.26 \times 10^{-5}$ rads

During the control period 1.0 μc of 4-C^{14}-cortisol and 2.0 μc of 1, 2-H^3-aldosterone will be injected simultaneously (6.53×10^{-5} rads). This will be repeated once at high altitude the following week. The total radiation dose received by each individual will be no more than 1.3×10^{-4} rads which is considerably below the limits generally agreed upon for an internal emitter - approximately 0.1 rem per week (5 rem per year).

3. Monitoring radioactivity

During the experimental studies, after the administration of radioisotopes all excreta will be collected until levels of radioactivity are equal to background levels. All radioactive waste will be disposed of by the Radioisotope Section of the USAMRNL as outlined in "Procedures for Use of Radioactive Material" (See Appendix E Application for Renewal and Amendment to AEC Byproduct Material License No. 5-46-13 (A66) dated June 1966). Periodic wipes will be

taken of all areas for possible contamination and counted by means of a liquid scintillation counter.

All rules, regulations and limitations set forth by Army AEC and local authorities, including those embodied in AR 70-25; AR 40-37; Title 10, Part 20, Code of Federal Regulations "Standards for Protection Against Radiation"; and Handbook 69 of the National Bureau of Standards will be complied with.

Enclosed and attached to this protocol is the Voluntary Consent Statement relating to the intravenous administration of radioisotopically labeled steroid compounds as tracers. (See Appendix I)

4. Determination of the Secretory Rates of Cortisol and Aldosterone

a. Cortisol secretion rate.

The cortisol secretion rate will be determined by the method of Roginsky, et al. (45) from the combined 48-hour urine collection following isotope injection

b. Aldosterone secretion rate

This will be determined by the method of Kelly, et al. (46) using the double isotope derivative method.

V. ADMINISTRATION

A. The study will be the responsibility of the Physiology Division of USAMRNL.

B. The personnel and their responsibilities will be assigned as follows:

1. Captain A. H. Janoski, MC: Project leader; responsible medical officer.

2. John P. Hannon, Ph. D.: Project Co-leader.

3. J. L. Shields, Ph. D.: Project Co-leader.

4. Captain R. P. Carson, MC: Medical Officer
5. Captain B. Whitten, MSC: Administrative officer
6. George J. Klian, Ph. D.: Technical supervisor
7. Major C. G. Liddle, V. C.: Radiation officer
8. One NCOIC: Subject control and dietary supervisor
9. Four enlisted military technicians
10. Two civilian technicians

C. Continuous physician coverage during the study will be the responsibility of Captains A.H. Janoski and R. P. Carson of the U. S. Army Medical Corps.

D. Cost

1.	Equipment	800.00
2.	Chemicals, including solvents	$4,000.00
3.	Per diem for subjects	210.00
4.	Travel for subjects	1,500.00
5.	Diet for subjects	500.00
6.	Air freight (samples collected during study)	700.00
7.	Class A Funds	600.00
8.	Travel for project personnel (7 investigators, 5 enlisted men, 2 civilians). Includes one round trip to Seattle	2,100.00
9.	Rental truck for equipment (30 days)	900.00
10.	GSA vehicles (2 for 21 days)	400.00
11.	Lab rats for bioassay (250 rats)	800.00
12.	Per diem for investigators	2,400.00
13.	Per diem for enlisted men	
	a. Sea level site - 14 days (government quarters available, government mess not available).	560.00

14.	Per diem for civilian technicians (21 days)	$672.00
15.	Three investigators' per diem for three-day site survey	144.00
16.	Three investigators' travel to Seattle for site survey (3 round trips)	450.00
17.	Additional per diem for 3 investigators for four days at sea level site to set up study	192.00
18.	Additional per diem for 4 enlisted men for four days at sea level site to set up study (government quarters available; government mess not available)	128.00
19.	Miscellaneous expendable items	$1,200.00
	Total	$18,816.00

E. Miscellaneous

Since the final approval by the AEC for the use of isotopes in human studies rests on the approval of the protocol and definite site selection for sea level studies, it is requested that final action on this protocol be no later than 5 June 1967. If the protocol is approved at this time, application would then be made to the AEC for action on the license amendment. Furthermore, final action on the protocol at this date would enable the investigators to prepare the isotopes for human administration and to obtain the necessary chemical supplies for the field study.

F. Additional Information

See Appendix I and Appendix II

A. H. JANOSKI
Captain, MC

APPENDIX I

VOLUNTARY CONSENT STATEMENT

Military _____ Military Patient _____ Civilian _____ Civilian Patient _____

I, _____, having the capacity to consent, voluntarily and without force or duress consent to participate in research involving the use of tracer amounts of radioisotopes. I have been informed of, and understand, the nature, duration, and purpose of the experiment, the method and means by which it is to be conducted, the inconveniences and hazards to be expected, and the effects upon my health and person which may possibly come from participation in the experiment.

Specifically, I agree to receive (intravenously / orally) a small quantity of _____ containing _____ microcuries of _____. I also agree to furnish urine and stool samples for the period following until no detectable radioactivity is present and submit to measurements of expired gases if Carbon-14 has been received.

I understand that I may at any time during the course of the experiment revoke my consent and withdraw from the experiment without prejudice.

I do not at this time have any physical diseases, except for the following _____ _____, or mental disease, to the best of my knowledge.

_____ _____

DATE SIGNATURE

_____

SIGNATURE OF WITNESS

APPROVAL

I have personally ascertained that the quality of the foregoing consent is sufficient to permit the volunteer to participate in the experiment.

_____ _____

ATTENDING PHYSICIAN PROJECT LEADER

APPENDIX II

SUBJECT STATEMENT

Date _____

I voluntarily agree to participate as a subject in the experiment to be conducted on high altitude. I am aware that I may withdraw from the experiment at any time without prejudice or penalty of any kind. It has been explained to me that constant medical supervision will be maintained and that neither the exposure to high altitude nor the experimental techniques used in this study are unduly hazardous. I realize that in some subjects the high altitude may cause any or all of the following symptoms: dryness of the mouth and nose, excitement, blurring of vision, dizziness, tiredness, tremor, lack of appetite, mild cramps, thirst, confusion, a sense of well-being, sleepiness, muscular aches, ringing in my ears, nausea, runny nose, headache, hunger, sleeplessness, coughing, rapid heart beat, chest pains, fatigue, constipation, fever, muscular stiffness, stomach ache, itching or sneezing.

The nature and purpose of the experiment have been explained to me and I sign this statement fully understanding the project, any hazards connected with it, and my rights.

_____
(Name)

BIBLIOGRAPHY

1. C. W. Harris and J. E. Hansen. Electrocardiographic changes during exposure to high altitude. Amer. J. of Cardiology 18: 183-190, 1966.

2. Stickney, J. C. and E. J. Van Liere. Acclimatization to low oxygen tension. Physiology Reviews 33: 13, 1953.

3. Harris, C. W., J. L. Shields and J. P. Hannon. Acute altitude sickness in females. Aerospace Med. 37: 1163, 1966.

4. Sundstrom, E. S. and G. Michaels. The adrenal cortex in adaptation to altitude, climate and cancer. Memoirs of the University of California, Vol. 12, 1942. University of California Press, Berkeley, California.

5. Langley, L. L. and R. W. Clarke. The reaction of the adrenal cortex to low atmosphere pressure. Yale Journal of Biology and Medicine 14: 529, 1941-1942.

6. Dohan, F. C. Effects of low atmosphere pressure on the adrenals, thymus and testes of rats. Proc. Soc. Exper. Biol. and Med. 49: 404, 1942.

7. Darrow, D. C. and E. L. Sarason. Some effects of low atmosphere pressure on rats. Journal of Clinical Investigations 23: 11, 1944.

8. Hailman, H. F. The effect of preventing acapnia on adrenal cortical hypertrophy under conditions of decreased baronietric pressure. Endocrinology 34: 187, 1944.

9. Demopoulos, H. B., B. Highman, P. D. Altland, M. A. Gerving and G. Kaley. Effects of high altitude on granular juxtaglomerular cells and their possible role in erythropoietin production. American Journal of Pathology 46: 497, 1965.

10. Thorn, G. W., M. Clinton, Jr., S. Farber and H. W. Edmonds. Studies on Altitude Tolerance, I. Bulletin of Johns Hopkins Hospital 79: 59, 1946.

11. Thorn, G. W., M. Clinton, Jr., B. M. Davis and R. A. Lewis Effect of adrenal cortical hormone therapy on altitude tolerance. Endocrinology 36: 381, 1945.

12. Lewis, R. A., G. W. Thorn, G. F. Koepf and S. S. Dorrance. The role of the adrenal cortex in acute anoxia. Journal of Clinical Investigation 21: 33, 1942.

13. Reeves, J. L. Influence of intermittent exposure to simulated altitude on plasma and tissue electrolytes in rats. USAF Aerospace Medicine Center Pamphlet 61-37, February, 1961.

14. Marks, B. H., A. N. Bhattacharya and J. Vernikos-Danellis. Effect of hypoxia on secretion of ACTH in the rat. American Journal of Physiology 208: 1021, 1965.

15. Unpublished data. Physiology Division of the United States Army Medical Research and Nutrition Laboratory, Fitzsimons General Hospital, Denver, Colorado.

16. Ayres, P. J., R. C. Hunter and E. S. Williams. Aldosterone excretion and potassium retention in subjects living at high altitude. Nature 191: 78, 1961.

17. Pugh, L. G. Physical and medical aspects of the Himalayan Scientific and Mountaineering Expedition, 1960-1961. British Medical Journal 2: 621, 1962.

18. Williams, E. S. Salivary electrolyte composition at high altitude. Clinical Science 21: 37, 1961.

19. Williams, E. S. Electrolyte regulation during the adaptation of humans to life at high altitude. Proc. Royal Society, Series B: 266, 1966.

20. Pincus, G., and H. Hoagland. Steroid excretion and the stress of flying. Journal of Aviation Medicine 14: 173, 1943.

21. Clinton, Jr., M., G. W. Thorn and V. D. Davenport. Studies on Altitude Tolerance. II. Bulletin of Johns Hopkins Hospital 79: 70, 1946.

22. Timeras, P. S., W. Pace and C. A. Hwang. Plasma and urine 17-hydroxycorticosteroid levels in man during acclimatization to high altitude. Fed. Proc. 16: 340, 1957.

23. Hornbein, T. F. Adrenal cortical response to chronic hypoxia. Journal of Applied Physiology 17: 246, 1962.

24. Moncloa, F. and E. Pretell. Cortisol secretion rate, ACTH and methopyrapone tests in high altitude natives. Journal of Clin. Endo. and Metab. 24: 915, 1964.

25. Forbes, W. H. Blood sugar and glucose tolerance at high altitude. American Journal of Physiology 116: 309, 1936.

26. Picon-Reategui, E. Studies in the metabolism of carbohydrates at sea level and at high altitude. Metabolism 11: 1148, 1962.

27. Picon-Reategui, E. Intravenous glucose tolerance test at sea level and at high altitude. Journal of Clin. Endo. and Metab. 23: 1256, 1963.

28. Calderon, R. and L. A. Llerema. Carbohydrate metabolism in people living in chronic hypoxia. Diabetes 14: 100, 1965.

29. Evans, W. O. and J. E. Hansen. Troop performance in high altitude. Army: February, 1966

30. Army Research Office Symposium on High Altitude March 1963.

31. Army Research Office Symposium on High Altitude September 1963.

32. Morgan, C. R. and A. Lazarow. Immunoassay of insulin using a two-antibody system. Proc. Society Exp. Biology and Medicine 110: 29, 1962.

33. Peterson, R. E., A. Karrer and S. L. Guerra. Evaluation of Silber-Porter procedure for determination of plasma hydrocortisol. Anal. Chemistry 29: 144: 1957.

34. Silber, R. H. and C. C. Porter. The determination of 17, 21-dihydroxy-20-ketosteroids in urine and plasma. Jour. of Biol. Chemistry 210: 923, 1954.

35. Sobel, C., O. J. Golub, R. J. Henry, S. L. Jacobs and G. K. Basu. Study of the Norymberski methods for determination of 17-ketogenic steroids (17-hydroxycorticosteroids) in urine. Jour. of Clinical Endocrinology and Metabolism 18: 208, 1958.

36. Kliman, B. and R. E. Peterson. Double isotope derivative assay of aldosterone in biological extracts. Jour. of Biol. Chemistry 235: 1639, 1960.

37. Erlich, E. N. Reciprocal variations in urinary cortisol and aldosterone in response to increased salt intake. Jour. of Clinical Endocrinology and Metabolism 26: 1160, 1966.

38. Vernikos-Danellis, J. and E. Anderson. Changes in adrenal corticosterone concentration in rats: Method of bioassay for ACTH. Endocrinology 79: 624, 1966.

39. Peterson, R. E., J. B. Wyngaarden, S. L. Guerra, B. B. Brodie and J. J. Bunim. The physiological disposition and metabolic fate of hydrocortisone in man. J. of Clin. Investigation 34: 1779, 1955.

40. Cope, C. L. and E. G. Black. The behavior of 14^{C}-cortisol and estimation of cortisol production rate in man. Clinical Science 17: 147, 1958.

41. Kumagai, L. F., E. L. Simons, H. Brown and C. D. West. The transformation of radioactive cortisol in normal humans. Steroids Supplement II: 119, 1965.

42. Kelly, W. G. and J. Laragh. In: Advances in Metabolic Disorders. Vol. I, 1964, New York, Academic Press.

43. Kowarski, A., J. Finkelstein, B. Lopas and C. J. Migeon. The in vivo stability of the tritium label in 1, 2-H^{3}-d-aldosterone when used for measurement of aldosterone secretion rate by the double isotope dilution technique. Steroids 3: 95, 1964.

44. Quimby, E. H. and S. Feitelberg. Radioactive isotopes in medicine and biology. Lea and Febiger, Philadelphia 1963.

45. Roginsky, M., J. Shaver and N. P. Christy. A study of adrenocortical function in acromegaly. Jour. of Clinical Endocrinology and Metabolism 26: 1101, 1966.

46. Kelly, W. G., L. Bandi, J. N. Shoolery and S. Lieberman. Isolation and characterization of aldosterone metabolites from human urine; two metabolites bearing a bicyclic structure. Biochemistry 1: 172, 1962.

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INVESTIGATORS PRINCIPAL ASSOCIATE: Janoski, A. H. Capt.; Hannon, J. P. PhD; Shields, J.L.
TEL
TYPE: PhD

21. TECHNOLOGY UTILIZATION

22. COORDINATION

23. KEYWORDS: Altitude; Adaptation, Physiological; Endocrine; Biochemistry; Stress; Physiology; Adrenal Function

24.
(U) Technical Objective: (Study No. 4 - Endocrine Effects of Altitude). Research under this work unit will be directed toward evaluation of endocrine changes in human volunteers produced by the stress of high altitude. In particular, there will be investigation into electrolyte alterations, pituitary and adrenocortical function aldosterone secretion in relation to electrolyte changes and glucose metabolism during acute exposure of man to a hypoxic enrironment. Determination of the alterations in endocrine funtion during altitude exposure will contribute to understanding the basic physiologic mechanisms producing mountain sickness and acclimatization.

25.(U) Approach; Biochemical and radioisotope techniques will be utilized to measure the alterations in endocrine function in human volunteers. Investigations will be conducted at sea level and high altitude with the volunteers serving as their own controls. Emphasis will be placed on the acute stress of continuous exposure to an altitude of 14,100 feet for six days. A constant diet of known chemical composition will be maintained during the sea level and altitude test periods. Studies will be concerned with electrolyte and water balance; steroid excretion and production; pituitary functions; and glucose metabolism.

26.(U) Progress: Initial report.

27. COMMUNICATIONS SECURITY ☐ a. COMSEC OR COMSEC RELATED ☐ b. NOT RELATED	28.	29. OSD CODE	30. BUDGET CODE

31. MISSION OBJECTIVE	32. PARTICIPATION

33. REQUESTING AGENCY	34. SPECIAL EQUIPMENT

35. EST. FUNDS (In thousands)	36.
CPY01	

DD FORM 1498 (Items 1 to 36 identical to NASA Form 1122) REPLACES DD FORM 613 &

Typed Copy

29 August 1966

JANOSKI, Alfonso H., Captain, Medical Corps, Physiology Division,
U. S. Army Medical Research & Nutrition Laboratory

Date and Place of Birth:	3 November 1935 Wilkes-Barre, Pa.
Wife:	Jane Stewart
Children:	Stephen Gregory
Education:	BA, Seton Hall University South Orange, N. J. 1957 MD, Columbia University, N.Y. 1961
Experience:	Internship, Bellevue Hospital, Columbia (First) Medical Division 1st & 2nd year Medical Residencies, Bellevue Hospital Visiting Fellow in Medicine (endocrinology), Columbia Presbyterian Medical Center, New York 1964-66
Publications:	Two
Hobbies:	Photography, hunting, fishing
Member of:	Harvey Society of New York American Association for Advancement of Science New York County Medical Society

Form AEC-313 a (10-61) Page 3

UNITED STATES ATOMIC ENERGY COMMISSION

APPLICATION FOR BYPRODUCT MATERIAL LICENSE

SUPPLEMENT A—HUMAN USE

Form approved Budget Bureau No. 38-R0301

This page may be completed by the physician's preceptor (if any) in the medical use of radioisotopes. When the information is not furnished by the preceptor, the name and present address of this preceptor (if any) should be shown in item 12 below.

9. (a) USING PHYSICIAN'S NAME	(b) NAME AND ADDRESS OF ATTICANT (If different from P(a))
Alfonso H. Janoski Capt MC USAMRNL Denver, Colo. 80240	

10. CLINICAL TRAINING AND EXPERIENCE OF PHYSICIAN WHO WILL USE BYPRODUCT MATERIAL

(A) ISOTOPE	(B) CONDITION(S) DIAGNOSED OR TREATED	(C) NUMBER OF CASES	(D) TYPE OF PARTICIPATION FOR ALL CASES IN COLUMN B (circle applicable numbers of items in accordance with key set forth below)
I-131	Diagnosis of thyroid function	50	(1) 2 (3) (4)
	Treatment of hyperthyroidism	15	(1) 2 (3) (4)
	Treatment of thyroid cancer	4	(1) 2 (3) (4)
	Treatment of cardiac conditions		1 2 3 4
	Brain tumor localization	5	1 2 3 (4)
	Blood determinations		1 2 3 4
	Kidney function		1 2 3 4
	Others:		1 2 3 4
P-32 Soluble	Treatment of polycythemia and leukemia	10	(1) 2 3 (4)
	Brain tumor localization		1 2 3 4
	Treatment of bone metastases	2	1 2 3 (4)
	Others:		1 2 3 4
			1 2 3 4
P-32 $CrPO_4$	Treatment of prostatic cancer		1 2 3 4
	Treatment of cervical cancer		1 2 3 4
	Treatment of pleural effusions and/or ascites		1 2 3 4
	Others:		1 2 3 4
			1 2 3 4
Au-198 Colloid	Treatment of prostatic cancer		1 2 3 4
	Treatment of cervical cancer		1 2 3 4
	Treatment of pleural effusions and/or ascites	2	1 2 3 (4)
	Others:		1 2 3 4
			1 2 3 4
Cr-51	Blood determinations	25	(1) 2 (3) (4)
	Others:		1 2 3 4
			1 2 3 4
Other Isotopes	Tritium labelled & Carbon-14 labelled Steroids - Research	6	(1) (2) 3 (4)
			1 2 3 4
			1 2 3 4

Key to above numbers (column D)

Active Participation and Discussion in the:

1. Examination of patients to determine suitability for radioisotope diagnosis and/or treatment and recommendations on dosage to be prescribed.
2. Collaboration in calibration and administration of dosages including related measurements and plotting of data.
3. Active period of training and experience of sufficient duration to permit followup of patients through treatment and posttreatment period including reevaluation as to effectiveness and complications.
4. Study and discussion of case histories to establish most efficacious diagnostic and/or therapeutic techniques for this radioisotope use.

11. TOTAL NUMBER OF HOURS OF PARTICIPATION IN CLINICAL TRAINING 40 Hours

12. THE TRAINING AND EXPERIENCE INDICATED ABOVE WAS OBTAINED UNDER THE SUPERVISION OR GUIDANCE OF

Dr Sergei Feitelberg - Mt. Sinai Hosp. N.Y.; Dr Sidney Werner - Columbia-Presbyterian Med Center N.Y.; Dr Nicholas P. Christy - Roosevelt Hospital

(Name of physician (preceptor)) AT (Institution) (Signature)

7. TRAINING AND EXPERIENCE OF EACH INDIVIDUAL NAMED IN ITEM 4 *(Use supplemental sheets if necessary)*

8. TYPE OF TRAINING	WHERE TRAINED	DURATION OF TRAINING	ON THE JOB (Circle answer)	FORMAL COURSE (Circle answer)
a. Principles and practices of radiation protection	Columbia Univ College of Physicians & Surgeons	2 yrs	(Yes) No	(Yes) No
b. Radioactivity measurement standardization and monitoring techniques and instruments	Mt Sinai Hosp (Radiophysics Dept)	1 yr	(Yes) No	(Yes) No
c. Mathematics and calculations basic to the use and measurement of radioactivity	"		(Yes) No	(Yes) No
d. Biological effects of radiation	"		Yes (No)	(Yes) No

9. EXPERIENCE WITH RADIATION. *(Actual use of radioisotopes or equivalent experience)*

ISOTOPE	MAXIMUM AMOUNT	WHERE EXPERIENCE WAS GAINED	DURATION OF EXPERIENCE	TYPE OF USE
H^3	5 Mc	Columbia Univ College of Physicians & Surgeons	2 yrs	Purification, Human metabolism, Isolation & Synthesis
C^{14}	10 Mc	"	"	"

10. RADIATION DETECTION INSTRUMENTS. *(Use supplemental sheets if necessary)*

TYPE OF INSTRUMENTS (Include make and model number of each)	NUMBER AVAILABLE	RADIATION DETECTED	SENSITIVITY RANGE (mr/hr)	WINDOW THICKNESS (mg/cm²)	USE (Monitoring, surveying, measuring)

11. METHOD, FREQUENCY, AND STANDARDS USED IN CALIBRATING INSTRUMENTS USED ABOVE

12. FILM BADGES, DOSIMETERS, AND BIO-ASSAY PROCEDURES USED *(For film badges, size, type, method of calibrating and processing, or name of supplier)*

INFORMATION TO BE SUBMITTED ON ADDITIONAL SHEETS

13. FACILITIES AND EQUIPMENT. Describe laboratory facilities and remote handling equipment, storage containers, shielding, fume hoods, etc. Explanatory sketch of facility is attached. *(Circle answer)* Yes No

14. RADIATION PROTECTION PROGRAM. Describe the radiation protection program including control measures. If application covers sealed sources, submit leak testing procedures where applicable, name, training, and experience of person to perform leak tests, and arrangements for performing initial radiation survey, servicing, maintenance and repair of the source.

15. WASTE DISPOSAL. If a commercial waste disposal service is employed, specify name of company. Otherwise, submit detailed description of methods which will be used for disposing of radioactive wastes and estimates of the type and amount of activity involved.

CERTIFICATE (This item must be completed by applicant)

16. THE APPLICANT AND ANY OFFICIAL EXECUTING THIS CERTIFICATE ON BEHALF OF THE APPLICANT NAMED IN ITEM 1, CERTIFY THAT THIS APPLICATION IS PREPARED IN CONFORMITY WITH TITLE 10, CODE OF FEDERAL REGULATIONS, PART 30, AND THAT ALL INFORMATION CONTAINED HEREIN, INCLUDING ANY SUPPLEMENTS ATTACHED HERETO, IS TRUE AND CORRECT TO THE BEST OF OUR KNOWLEDGE AND BELIEF.

Alfonso H. Janeski Capt MC
Applicant named in item 1

Date _____ By: _____

Title of certifying official

WARNING.—18 U. S. C., Section 1001; Act of June 25, 1948, 62 Stat. 749, makes it a criminal offense to make a willfully false statement or representation to any department or agency of the United States as to any matter within its jurisdiction.