THE EFFECT OF PROLONGED SIMULATED NON-GRAVITATIONAL ENVIRONMENT ON MINERAL BALANCE IN THE ADULT MALE

VOLUME I

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Charles L. Donaldson, M.D., Stephen B. Hulley, M.D.,
Donald E. McMillan, M.D., Robert S. Hattner, M.D.,
and Jon H. Bayers, M.D.

SUMMARY AND CONCLUSIONS

1. Three healthy adult males were restricted to complete bed rest for periods of 30 to 36 weeks; freedom of movement in the horizontal plane was permitted.

2. Urinary calcium excretion was elevated throughout bed rest, averaging 67 mg/day above the baseline value of 193 mg/day. Mean peak excretion occurred during the 7th week and was 136 mg/day above the baseline value.

3. Mean calcium balance during bed rest ranged from -200 to -256 mg/day for the three subjects. This represents an estimated loss of 0.5 to 5.5 percent of the skeleton.

4. Gamma ray transmission scanning of the calcaneus showed much larger losses of mineral during bed rest. For the three subjects, decreased mass in the central portion of the bone ranged from 25 to 45 percent.

5. These data are taken to suggest that bone dissolution during bed rest may occur at different rates in different portions of the skeleton.

Summary and Conclusions

Weight bearing bones may be particularly at risk, and medulary regions diminish more readily than cortex.

6. Recovery of calcaneus mineral to values above the initial (3 month) level was observed 5-10 months following reambulation; disuse osteopenia therefore may be reversible.

7. Serum parathyroid hormone concentration increased during bed rest and achieved levels compatible with hyperparathyroidism in all three subjects. The data implicate this systemic factor in addition to local mechanical factors in the etiology of disuse osteopenia.

8. Urinary excretion of hydroxyproline and pyrophosphate increased during bed rest in comparison with ambulatory periods.

9. Mean urinary phosphorus during the entire bed rest period was 98 mg/day above the baseline value. Mean phosphorus balance for the three subjects during bed rest ranged from +4 to -68 mg/day.

10. Nitrogen balance became more negative during the first two months of bed rest. An absolute negative balance was not achieved during bed rest as a whole, but failure to measure gaseous nitrogen loss may artifactually increase the balance value. Weight and lower extremity limb girth decreased

during bed rest, suggesting that loss of lean body mass was occurring.

11. Sodium, potassium and magnesium balances were generally more negative during bed rest than during the baseline period.

12. During reambulation, balances of sodium, potassium, nitrogen, magnesium and phosphorus became strikingly positive and weight increased.

13. Measurement of dermal losses of calcium, sodium and potassium demonstrated a minor contribution of these electrolytes to overall balance.

14. Plasma volume and red blood cell volume were decreased during bed rest in comparison with the ambulatory periods.

15. No evidence for the occurrence of urinary calculi or calcium-containing crystals was noted during the study.

16. Clinical evaluation revealed no major morbidity during bed rest. During reambulation the soles of the feet were tender for 3-4 weeks and easy fatigability was subjectively noted for 4-6 months.

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INTRODUCTION

The importance of physical activity in maintaining skeletal mass is demonstrated by the loss of bone mineral which attends immobilization. This effect occurs locally when a single limb is fixed to promote fracture healing (39) and is generalized in quadriplegics (17,24,48). Metabolic studies have shown an increased rate of calcium excretion in patients with fractures (26) or paralysis (17,24,48), and also in normal individuals subjected to immobilization by bed rest in half-body casts (11,21). Other investigators have shown that bed rest per se results in loss of bone mineral, even when movements in the horizontal plane are not restricted (30,37,45), with a similar magnitude of loss (6).

Calcium loss during bed rest is estimated to be between 0.3 percent (21) and 1 percent (11) of the total body calcium per month. The possibility that bone mineral may be lost preferentially from weight bearing bones in this situation has not been investigated. Radiographic evidence for osteopenia has been observed after two (45) and twelve (17) weeks of bed rest, although these changes were not observed in another study of six weeks duration (11). These studies underscore the importance of gravitational forces, or of counter-gravitational muscular effort, on

factors controlling bone homeostasis.

The recent advent of space flight has transformed consideration of the effects of a hypogravic environment on skeletal mass from the theoretical to the practical realm. Preliminary data from the short-term space expeditions have supported the prediction that weightlessness leads to loss of skeletal mineral (35,36,38). The magnitude of the effect is such that it could be an important consideration on space flights of longer duration. It was therefore thought necessary to gain further understanding of the pattern and degree of demineralization which would occur during prolonged space flight, and to devise effective prophylactic measures. This report describes the first phase of such an investigation.

Horizontal bed rest was employed as the most reasonable way to simulate a non-gravitational environment. Three normal males were studied during thirty to thirty-six weeks of bed rest, and during prior and subsequent ambulatory periods. Mineral balance information in this presentation (exhibit A) is correlated with data obtained by gamma ray transmission scanning of the calcaneus (exhibit B).

METHODS AND MATERIALS

Study Subjects

The study was carried out in the Metabolic Unit of the U.S. Public Health Service Hospital, San Francisco (appendix i). Three healthy male subjects ages 21-22 years volunteered to undergo prolonged bed rest and were judged suitable after medical and psychiatric screening.* All three subjects completed the planned period of continuous bed rest, thirty-six weeks for two and thirty weeks in the case of the third. In addition, the subjects were studied for 3-8 week periods before and after bed rest.**

During the initial ambulatory period, the subjects engaged in a relatively normal level of activity supplemented by thirty minutes twice daily of walking at three miles per hour up a 6° grade on a treadmill. The subjects were restricted to the hospital grounds except for occasional passes of several hours duration. During bed rest, the subjects were required to remain in bed or on a stretcher at all times, and were closely supervised. Freedom of horizontal movement was allowed in bed; the subjects were permitted to raise themselves on one elbow for eating and reading. They were not allowed to sit up or dangle their legs over the bed, and bowel movements and micturition were performed in the supine

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* One of the subjects was thought to desire a prolonged stabilizing environment. The other two were conscientious objectors who chose the study as an alternative to military service, one of whom had less than thirty weeks obligation to fulfill.

** During the second week of the ambulation period, subject G.B. left the hospital for three days due to a personal emergency. The balance values for this week have been omitted or extrapolated from those of contiguous weeks.

position.

Balance Diet

The palatable whole food diet was composed of seven daily menus (three meals and an evening snack), which were rotated randomly each week and which provided a relatively constant daily mineral intake. The diets are described in appendix ii, tables A-G. All foods, except for some staples, soft drinks, and meats, were purchased in common lots prior to the study to assure maximum constancy. Three lots of meat were obtained during the study due to limitations of freezer storage. Canned whole milk and frozen homogenized eggs were used, and fresh foods were avoided. Standard methods of cooking included steaming, baking, and broiling. Only distilled water was used for food preparation and drinking. Individual portions of food were weighed on a Mettler balance and the subjects were required to eat all of the food served to them. At the end of each meal the subjects cleaned their plates with a spatula, with bread, or by licking, and drank a small quantity of distilled water used to rinse their glassware. All dishes and utensils used in the study were washed in a dishwasher with a distilled water rinse. Tooth paste used by the subjects was free of mineral except silica.

Three duplicate analyses of the diet were obtained at spaced intervals during the study. For this purpose, an additional serving of each menu was weighed and prepared throughout three seven-day periods. The

meals were pooled daily, homogenized for seven minutes in a large Waring blender, and an aliquot was stored frozen for mineral analysis. The results of these analyses are compared to the calculated dietary contents in appendix ii, tables H and I. Calculated nutrient values of the seven menus are shown in table J. The values include the nutrients in one hexavitamin tablet daily, although this supplement was not initiated until eleven days before bed rest. The only other medications employed were dioctyl sodium sulfo-succinate (Colace), 0-300 mg/day, and occasionally acetyl salicylic acid, 600 mg, and pseudoephedrine, 60 mg (generally less than once weekly).

Body Measurements

Daily weights were obtained in the supine position, using an Acme in-bed scale. Limb and trunk girths were determined periodically, using a metal tape measure that encircled the part while subjects lay supine in bed. One end of the tape measure was fixed, the other weighted by 2.0 kg of lead shot to provide reproducible tension in successive measurements. Skin fold thickness was measured with calipers. The sites of measurement are indicated in the tables.

Blood and Extracellular Fluid Volumes

Red cell mass was measured using ^{51}Cr tagged autologous cells. Plasma volume was measured using ^{131}I labelled serum albumin. Extracellular fluid volume was measured using ^{82}Br. Technics employed in these measurements were the same as described in previous studies at this Unit (27).

Serum, Stool, and Urine Collections

Thirty-five ml of blood were drawn in the fasting state on the first day of each 7-day period. After standing for 1-2 hours at room temperature the serum was separated and stored frozen at -22° C. Individual urines were pooled and refrigerated during each 24-hour period beginning and ending at 7:30 a.m.; individual urine specimens were rinsed from the urinal into a polyethylene bottle with distilled water after determining the volume. A 5 percent aliquot of each was acidified with concentrated HCl to 1 percent by volume and was stored at 4° C until closeout of the 7-day period. An aliquot of the pooled 7-day urine aliquots was then stored at -22° C for subsequent mineral analysis. In addition, pyrophosphate was determined weekly on a fresh unacidified 24-hour specimen, and a second morning urine was examined weekly for microscopic crystals.

A stool marker (100 mg brilliant blue) was administered by mouth at the beginning of each 7-day period. The stools for that period were

collected from the first appearance of the marker in the stool until the appearance of the subsequent marker. Stools were collected in bedpans lined with mylar film and were subsequently transferred with distilled water rinsing into unused epoxy-lined 1-gallon paint canisters and refrigerated. Upon completion of the 7-day collections the stools were further diluted with distilled water and 300 ml glacial acetic acid to a final weight approximately three times the initial weight. They were homogenized for thirty minutes on a paint shaker, and an aliquot was stored at -22° C for subsequent ashing and mineral analysis.

Sweat Collections

Forty-eight hour analyses of sweat mineral content were performed in the following manner. At the beginning each subject was thoroughly washed with an 0.1 percent acetic acid solution to remove residual sweat mineral. They were then dressed in distilled water-rinsed pajamas and similarly treated linen was placed on their beds. All washing and skin care was discontinued for the subsequent 48 hours. At this time the entire body was washed again with an 0.1 percent acetic acid solution and wrung out. 2.5 L of the collected 22 L were evaporated to dryness by gentle boiling and reconstituted in 30 ml of 1 normal HCl for mineral analyses. A control set of linen and pajamas were treated in a similar fashion each week to allow correction for contained mineral residue.

Sweat determinations were performed weekly during the first half of the study, and monthly thereafter, the results being extrapolated to provide continuous data. This extrapolation was felt justified by

the relative constancy of the measured sweat mineral content, and by the minor contribution of these losses to overall mineral balance.

Ashing of Stool and Diet

Three methods were employed in stool and diet preparation. Muffle furnace ashing was used to prepare material for calcium, magnesium, sodium, and potassium analysis. Sulfuric acid digestion was the only method found acceptable for nitrogen analysis; solutions prepared in this manner were also analyzed for phosphorus. Calcium and magnesium analyses using nitric acid digestion were done to confirm muffle furnace data. When the results did not agree within 5 percent, additional determinations were performed.

For the muffle furnace ashing, a weighed aliquot of diet or stool homogenate (approximately 20 gm) was ashed in a covered crucible at 575° C for 72 hours in a muffle furnace. The residual was reconstituted with 2 ml concentrated HCl, 2 ml concentrated NH_4OH, and 1 ml H_2O, followed by three washes of 5 ml of 0.6 normal HCl. Recovery of added calcium was 98.8 ± 3.1 percent (S.D.) and that of added phosphorus 98.4 ± 2.7 percent.

Sulfuric acid digestion was performed as follows: An aliquot of the homogenates (approximately 4 gm) was placed in a tared 100 ml volumetric flask and weighed. Twenty ml of concentrated sulfuric acid

and 2 selenized granules were added. The solution was boiled for approximately 2 hours, resulting in a fine suspension of brownish material. Recovery of added urea nitrogen was 97.8 ± 2.1 percent (S.D.) and that of phosphorus 100.4 ± 2.4 percent.

Nitric acid digestion was carried out in a similar manner. A quantity of homogenate (approximately 2 gm) was placed in a tared 100 ml volumetric flask and weighed. Ten ml of 90 percent HNO_3 and 4 boiling beads were added. Boiling for approximately five minutes at maximal heat on a Kjedahl burner resulted in a clear solution of about 1 ml volume. The solution was diluted to 100 ml with distilled water and analyzed for calcium and magnesium. Recovery of added calcium was 100.4 ± 2.4 percent (S.D.).

Laboratory Determinations

Calcium was determined by atomic absorption spectrophotometry on an automated Perkin-Elmer Model 303, using 0.25 percent lanthanum as an electron flux stabilizer and Harleco calcium standard solution C (19,50). Magnesium was also analyzed by atomic absorption spectrophotometry, using magnesium chloride in water as a standard (19,50). Phosphorus was analyzed on a Technicon Autoanalyzer by the standard adaptation of the Fiske and Subbarow technic (14). KH_2PO_4 standard were made up in 0.02 N HCl except for stool and diet solutions which contained sulfuric acid and were compared with standards which had been

brought to the same pH with H_2SO_4.

Total nitrogen was determined on a Technicon Autoanalyzer using the standard method (13), except that 0.2 percent perchloric acid was employed in the digestant; urea standards in 0.1 N H_2SO_4 were used. Although recovery of added urea by this method was satisfactory, recovery of creatinine nitrogen was 62.3 $\pm$ 2.1 percent (S.D.)*. Urinary nitrogen excretion was corrected for incomplete recovery of creatinine nitrogen by using this factor and the measured excretion of creatinine (a correction of approximately 2 percent). Urine and serum creatinine concentrations ([Cr]) were determined on a Technicon Autoanalyzer by the standard modification of the Folin Wu method (16), employing Technicon standards. Creatinine** (C_{Cr}) was calculated from the relationship C_{Cr} = urine [Cr] x urine volume/serum [Cr].

Sodium and potassium were analyzed by lithium internal standard flame photometry on a Technicon Autoanalyzer (appendix iii). Hydroxyproline was determined by the method if Kivirikko and Prockop (31), using hydroxyproline in distilled water as the standard. Pyrophosphate was analyzed on a fresh 24-hour urine sample once weekly by a modification of the method of Fleisch and Besaz (15) as described in appendix iv.

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* These findings are similar to those previously reported (1).

** clearance

Each of the above methods was found to yield satisfactory results in initial recovery studies. Commercial standard solutions* were included in all subsequent runs for quality control. In addition, all calcium, phosphorus, hydroxyproline and pyrophosphate determinations were carried out in duplicate, as were urinary nitrogen assays.

Assay of the serum concentration of parathyroid hormone ([PTH]) was carried out by Dr. Eric Reiss at the Michael Reese Hospital in Chicago, Illinois. This immunoassay employs cockerel antibody to bovine PTH (41,42). The results are expressed in relation to an arbitrary selected standard hyperparathyroid serum assigned a potency of 1000 µl equivalents/ml. This amount of standard serum corresponds to 115 mµg of bovine PTH

Data Presentation

Metabolic observations for each activity period were carried out for a similar, but not identical, duration in each of the three subjects. Accordingly, while all available data is presented on figures and tables depicting individuals, only the portions common to all three subjects could be shown for those data presentations which employ averages. Complete balance data were obtained on all three subjects during the last 3 weeks of baseline ambulation, the first 30 weeks of bed rest, and the first 3 weeks of reambulation. Complete urinary and serum data were

* Commercial standards employed were: Versatol, Versatol A, Hyland Urine, and Brook serum.

obtained for the last 2-4 weeks of baseline ambulation, the first 30 weeks of bed rest, and the first 4 weeks of reambulation. When the averages of data presented in these two ways are compared, minor numerical differences are sometimes present.

RESULTS

Calcium Metabolism

During bed rest mean urinary calcium excretion rose from an average baseline value of 193 mg/day to a maximum of 329 mg/day in the 7th week (figure 1). The value subsequently fell, but did not return to the mean baseline level until reambulation was instituted. On the average, the increment in urinary calcium excretion during bed rest was 67 mg/day.

Calcium balance data are shown in figures 2-7 and tables 1-4. Mean calcium balances for the three subjects during the entire bed rest period were -256, -200 and -208 mg/day. During the first 3 weeks of reambulation the mean calcium balance was only slightly less negative, averaging -158 mg/day. For the two subjects whose observations extended into subsequent weeks, a further trend toward a positive balance was seen. For each subject, a more negative mean calcium balance was observed during bed rest than during the baseline ambulatory period. Fecal calcium excretion increased during bed rest in two of the three subjects. Calcium lost by way of the integument amounted to 2.2 percent of the total calcium output and was not consistently altered by bed rest. The total net losses of calcium for the three subjects during bed rest were 42.0, 64.5 and 52.4 gm.

Mean serum calcium concentration did not change greatly during bed rest when compared with the prior ambulatory period (figure 8, table 5).

There was an early tendency towards an increase, followed by a slight depression below baseline values. A more marked decrease was seen during reambulation.

Phosphorus Metabolism

Mean urinary phosphorus excretion was higher throughout the period of bed rest than during the baseline period in all three subjects; the average increment was 98 mg/day (figure 9, table 6). During reambulation, the values fell abruptly to a mean level 105 mg/day below baseline. Phosphorus balance data are shown in figures 10-15 and tables 6-9. Bed rest did not induce consistent changes in fecal phosphorus excretion. Mean phosphorus balances for the three subjects during the bed rest period were -64, +8 and -37 mg/day. Total net changes of body phosphorus during the entire period of bed rest were -17.1, +0.8 and -10.3 gm. There was no appreciable change in mean phosphorus concentration during bed rest; a slight decrease was seen during reambulation (figure 8, table 10). Phosphorus lost by way of the integument was too low for measurement.

Parathyroid Hormone

During the baseline period ([PTH]) values were well within the normal range of 10-60 µl equivalents of standard serum per ml (figure 16, table 11). During the first 19 weeks of bed rest [PTH] rose steadily to a value four-fold higher than the mean baseline level, and well into

the hyperparathyroid range. Twenty-four hours after reambulating [PTH] had fallen sharply, and normal levels were achieved two weeks later.

Hydroxyproline and Pyrophosphate

Urinary hydroxyproline excretion was consistently higher throughout the bed rest period than during the baseline ambulatory period; the mean increment was 9 percent (figure 17, table 12). During reambulation hydroxyproline excretion fell to an average value 6 percent below the mean baseline level. The changes in urinary pyrophosphate excretion were similar in nature, but more pronounced (figure 18, table 13). The mean bed rest value is 36 percent above, and that during reambulation 12% below, the mean baseline value.

Nitrogen Metabolism

Mean urinary nitrogen excretion was increased, in comparison with the baseline ambulatory period, during the first 10 weeks of bed rest (figure 19). The value subsequently stabilized near the baseline level until reambulation, when a fall was generally observed. The three subjects showed a slight decrease in the mean nitrogen balance when the whole bed rest period is compared with the baseline ambulatory period (from +1.0 to +0.6 gm/day) (figures 20-22, tables 14-17). Nitrogen excreted by C.S. during the first week of reambulation was anomalously high for unknown reasons. If this value is omitted, all subjects showed

a marked increase in nitrogen balance during reambulation.

Magnesium Metabolism

Mean urinary magnesium excretion followed a similar pattern, an early increase during bed rest followed by stabilization near the baseline level, and a decrease during reambulation (figure 23). Magnesium balance averaged -14 mg/day during both the baseline and the bed rest periods (figures 24-29, tables 18-21). An increase in balance to -3 mg/day was seen during reambulation. Serum magnesium concentration appeared to be slightly lower throughout bed rest than during the ambulatory periods (figure 30, table 22).

Sodium and Potassium

Mean urinary sodium excretion was increased during bed rest and decreased during reambulation, in comparison with the baseline period (figure 31). Sodium balance changed in a similar manner, the average values before, during and after bed rest being +3.3, -1.8 and +18.0 mEq/day, respectively (figures 32-34, tables 23-26). Sodium losses in the feces were small, averaging 2.1 mEq/day. Losses via the integument were more important, and usually decreased during bed rest. Serum sodium concentration did not appear to be affected by bed rest (figure 35, table 27).

Mean urinary potassium excretion was increased during the first 14 weeks of bed rest, then stabilized near the baseline level (figure 31).

Reambulation was accompanied by a sharp fall in all three subjects. Potassium balance changed in a similar manner, the average values before, during and after bed rest being +3.4, zero and +12.4 mEq/day respectively (figures 36-38, tables 28-31). Potassium losses in stool and sweat were small, and neither these nor serum potassium concentration appeared to be affected by bed rest (figure 35, table 32).

Weight, Fluid Balance and Creatinine Clearance

Weight loss was observed during the baseline period in subjects R.R. and G.B., and during the first 13 weeks of bed rest in subjects R.R. and C.S. (figure 39, table 33). During reambulation weight increased in all three individuals.

Fluid intake was higher during bed rest than in the ambulatory periods (table 34). Hydration was encouraged while in bed to maintain a high urine volume (table 35). Figures 40-42 depict fluid balance; intake is plotted downward on the ordinate, and urine output upward from the intake value. Insensible loss of fluid during bed rest was lower than in the ambulatory periods. "Insensible loss" as graphed is not corrected for changes in body weight or fecal content of water, since these factors were not very large.

Creatinine clearance was unchanged during bed rest in comparison with the baseline period (figure 43, table 36). The mean value increased

slightly during reambulation.

Blood and Extracellular Fluid Volumes

Blood volume was decreased an average of 19 percent during bed rest in comparison with the baseline period (figure 44, table 37). This change, which was seen in all three subjects, had completely reversed two weeks following ambulation. Independent determinations of red blood cell volume and plasma volume showed that both were reduced during bed rest (by 13 percent and 22 percent respectively) and both returned to the baseline level during reambulation (tables 38,39).

Extracellular fluid volume 'bromine space) was not determined during the baseline period (figure 45, table 40). Values obtained during and following bed rest appeared to be stable.

Body Measurements

Thoracic and abdominal girths were not altered by bed rest (tables 41,42). Measurements of upper and lower arm circumferences were also stable (tables 43,44). All three subjects showed a reduction in leg girth at the thigh, the mean decrease being 6 percent by the 29th week. Leg girth at the calf was reduced 16 percent in the same time interval. After 4 weeks of ambulation, the value of the thigh was still 3 percent, and at the calf 8 percent below baseline (tables 45,46). Skin fold thickness was not consistently altered by bed rest

(tables 47-49).

Clinical Evaluation

No significant morbidity resulted from the study. Two subjects began to have hard stools early in the course of bed rest and had transient rectal bleeding. Neither showed pathology on sigmoidoscopy, and with Colace administration stools became softer and bleeding never recurred. After the early weeks of bed rest, bowel movements tended to follow the same patterns as during ambulation. Flatus was not a problem on the diet and under the conditions of the study, and subjectively there was no change of flatulence during bed rest.

Emotional stress became evident periodically, manifest particularly in interpersonal relationships with the staff and one another. However, lethargy was a more striking consequence of bed rest, and all subjects agreed that the confinement during ambulatory periods proved more taxing than did prolonged bed rest (appendix vi).

During reambulation, orthostatic symptoms were fleeting and very minor. Tenderness of the feet was noted immediately upon ambulating, and the two subjects who tried to walk as much as possible on the first day out of bed after 36 weeks developed edema and petechiae of their ankles and marked tenderness of the soles of their feet. Their gait became so impaired as to be grotesque, and ambulation was severely

limited for the next two weeks. X-rays of their feet revealed no fractures or visible periosteal changes, but their bones appeared demineralized although no attempt was made to quantitate this in view of the bone mass studies being conducted simultaneously (exhibit B). Finally, aided by the buoyancy of water in a hydrotherapy pool, they were able to bear weight progressively, their need for wheelchairs diminished, and their symptoms gradually disappeared. Gait was almost completely normal in 4 weeks, although easy fatigability was noted for 4-6 months.

DISCUSSION

Calcium and Phosphorus Metabolism

In this study, three healthy adult males were restricted to bed rest for a period of 30-36 weeks. All three subjects showed significant losses of bone calcium, as judged by negative mean balances ranging from -200 to -256 mg/day. Both urinary and fecal calcium excretions were increased during bed rest when compared to the baseline ambulatory period, except in the case of one subject who showed a slight decrease in the fecal value. Mean urinary calcium reached a maximum during the 7th week which was 70 percent above the mean baseline value; it then returned to a plateau at an intermediate level. The degree of negative calcium balance did not change throughout the bed rest period. With reinstitution of ambulation, urinary and fecal calcium excretions dropped in all subjects, although in each case the balance remained slightly negative during the 3-5 weeks under observation.

The negative calcium balance during the baseline period of the two subjects is unexplained. The data might reflect accommodation by the subjects to a prestudy diet higher in calcium, with failure to equilibrate during the baseline period of 6-8 weeks.* Less likely, a relative vitamin D deficiency might have existed, since this vitamin was not present in the canned milk and supplementary vitamins were not begun

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* Dietary history confirmed this suggestion in patient G.B. whose intake prior to admission was estimated to be 1700 mg calcium/day. Patient C.S. had an estimated intake of 1150 mg calcium/day.

until 11 days before the beginning of bed rest. Inaccuracy in fecal calcium analysis is rendered very unlikely by the fact that duplicate analyses by two different technics were employed throughout, and by the observed recovery of 98.8 ± 3.1 percent (S.D.) of added calcium.

All three subjects manifested an increase in urinary phosphorus excretion during bed rest in comparison with the baseline period. Changes in fecal phosphorus excretion were variable. Mean phosphorus balance was much more negative during bed rest than during the ambulatory period in only one subject; the other two showed little change. Dissolution of bone would be expected to yield 43 mg of phosphorus per 100 mg of calcium (44). The magnitude of phosphorus loss relative to that of calcium during bed rest was well below the value which would be expected from simple bone loss in two of the subjects. A methodologic explanation for this disparity was carefully sought but none was found. The measured phosphorus balance agreed well with the theoretical balance of this mineral* in one previous bed rest study (11), but not in another (21).

Estimation of mineral losses through the integument has only occasionally been attempted in the past, with disparate results (20,28). Since sweat mineral loss might reasonably be expected to vary with the

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* Theoretical phosphorus balance can be calculated from the balances of calcium and nitrogen (29).

degree of physical activity, it was important to determine the magnitude of mineral excretion by this route in the current study. Calcium recovered from skin averaged 2.2 percent of the total calcium output, and no phosphorus was detected, as reported previously (29). No consistent change in mineral loss by this route was observed when bed rest values were compared with those during ambulatory periods. These data confirm the lower previous estimate of sweat calcium content (20), and support the validity of ignoring dermal mineral losses in studies of this type, in view of the technical difficulty in obtaining sweat collections.

The absolute loss of calcium observed in these three subjects during the entire bed rest period was 42.0, 52.4 and 64.5 gm. If balance values during bed rest are expressed as comparisons with the baseline values (11), the losses were 6.8, 8.0 and 68.3 gm. The total body calcium of these subjects is estimated at 1250 gm (10). The loss observed during 30-36 weeks of bed rest therefore represents 0.5 to 5.5 percent of the body calcium store.

Previous bed rest studies have indicated bone loss of similar (21) and rather larger (11) magnitudes. In these studies, half-body casts assured lower extremity immobilization. In the current study, freedom of movement in the horizontal plane was permitted for practical reasons and to more closely simulate an astronaut's environment. A second difference in experimental design is that subjects were permitted

out of bed for up to thirty minutes per day in the two previous studies, but not in the current one. In spite of differences in design, the similarity of data obtained in these bed rest investigations is notable, and it may be concluded that bone mineral loss during 30-36 weeks of bed rest is less than 6 percent of total body calcium.

In a companion study of the same three subjects, serial measurements of calcaneus mineral content were obtained, employing a modification of Cameron's technic for gamma ray transmission scanning (exhibit B). Due to technical difficulties in developing the method, bone scans were not obtained during the baseline period. Scans were obtained from the third thru ninth month of bed rest at approximately monthly intervals, and showed a highly significant decrease in the mineral content of the calcaneus . On the average, the total loss of mineral from the central portion of this bone ranged from 25-45 percent of the initial value measured in the third month of bed rest. The lowest region of the calcaneus, which represents predominantly cortical bone, was less severely affected than the central region which has a higher proportion of medullary bone. During reambulation, mineral content was gradually restored until 5-10 months later when it exceeded the initial value. This observation is important since a previous study indicated that disuse osteopenia was not completely reversed after 1-14 years of normal activity (39). The very large losses of calcaneus mineral during prolonged bed rest contrast with the relatively small losses of skeletal

mineral observed by balance technics. This disproportion appears to demonstrate three important features of bone loss during bed rest: (A) The entire calcaneus is affected more than the total skeleton, probably because of its weight bearing function. (B) Medullary loss appears to be greater than cortical loss, probably because the turnover rate is more rapid (18). (C) The loss appears to be reversible, mineral being regained at a rate similar to that at which it was lost.

The Mechanism of Disuse Osteopenia

The mechanism for the loss of bone mineral during bed rest has been the subject of two recent reviews (6,23). Interest in the problem stems from 1892 when Wolff suggested that disuse osteopenia is due to mechanical factors: either to absence of pressure transmitted to bone, or to absence of muscular tension applied to bone by muscle, or to both. The bulk of published evidence suggests that the absence of pressure forces on bone is primarily responsible for disuse osteopenia. Quiet standing for three or more hours per day appears to reverse the changes in mineral metabolism induced by bed rest, while vigorous supine exercise for four hours per day is ineffective (30). Forces on bone during standing may generate piezoelectric potentials to maintain homeostasis (4).

Local factors causing net resorption of bone might be expected to raise serum calcium, depressing parathyroid hormone production [PTH]. Heaney has supported this speculation that PTH is depressed, by demon-

strating rising serum and urine calciums, and declining gastro-intestional absorption of calcium in paralyzed adults (24). Direct determination of serum [PTH] in the current study demonstrated rising, rather than falling, levels. This change was appreciable after one week of bed rest and by the 20th week concentrations compatible with hyperparathyroidism had been achieved. An abrupt decline to near normal [PTH] values was observed 24 hours after reambulation.

The cause of the elevation of PTH is not clear. The level of PTH secretion is inversely related to the serum calcium concentration ([Ca]) (5,43). Observed values for [Ca] changed very little during bed rest, and actually showed a transient rise during the first few weeks at a time when [PTH] was already increasing. The only time a distinct decrease in [Ca] occurred was during reambulation, and this was accompanied by a sharply falling [PTH].

Secretion of PTH may be responsive to the serum level of ionized calcium, rather than that of total calcium (2). Although changes in these entities are usually parallel, this might not be the case under the conditions of this study. Measurements of ionized calcium were not carried out.

The level of PTH secretion may also be inversely related to serum magnesium concentration (8,47). Decreased levels of [Mg] were seen

throughout bed rest, and higher levels occurred during reambulation, but the changes were very slight.

Whatever the cause of PTH elevation, certain effects would be expected to occur:

a. The anticipated elevation in [Ca] was not observed during most of bed rest, but the falling levels during reambulation could have resulted from the falling [PTH].

b. In the absence of an increase in serum [Ca], urinary calcium would be expected to decrease as a result of increasing PTH levels (49). A downward trend in calciuria was observed after the 7th week of bed rest, a period when [PTH] was still increasing. However, the levels of the urinary calcium were higher throughout bed rest than during the ambulatory periods, implicating mechanisms other than PTH.*

c. Urine phosphorus would be expected to increase (25); this was in fact observed, although the rise was not progressive as in the case of the [PTH].

d. Urinary excretion of hydroxyproline (40) and pyrophosphate (3) have been considered to be measures of PTH activity. Increases were

---

* Urinary calcium excretion is directly related to the filtered calcium load (12), which in turn depends on the glomerular filtration rate (GFR) and ultrafilterable calcium concentration ([Ca_{uf}]). GFR, as assessed by creatinine clearance, did not change during bed rest. [Ca_{uf}] was not measured directly but the lack of increase in [Ca] does not support a major increase. An alternate mechanism might be the known calciuretic effect of increased excretion of other cations such as sodium and magnesium (12). However the increase in the urinary content of these electrolytes was so small as to make this an unlikely explanation.

seen in the current study, although the pattern of the change did not exactly match that for [PTH].

e. The major skeletal effect of PTH is increased bone resorption (22). Disuse osteopenia, as observed in the current study, has been attributed to increased bone resorption in experiments employing microradiographic (7) and kinetic (24,46) and tetracycline labelling technics (33). Experimental parathyroidectomy in animals prevents the development of demineralization in immobilized limbs (7). Taken all together, the data suggest that a combination of local (mechanical) and systemic (PTH) mechanisms are operative in the etiology of disuse osteoporosis.

Other Metabolic Parameters

All three subjects tended toward a negative nitrogen balance during the first two months of bed rest, as has been observed in previous studies (11). However, on the average there was only a slight decrease during the bed rest period as a whole (+0.6 gm/day) when compared with the baseline period (+1.0 gm/day). The absence of a more negative balance suggests that lean body mass was maintained during bed rest, but it should be noted that failure to measure gaseous nitrogen loss causes a major systematic artifact in balance data in a positive direction (9). It is probable that in fact a negative nitrogen balance was present during the baseline and bed rest periods. In support of this speculation, loss of weight was observed during both periods, possibly due to caloric deficiency. Moreover, a decrease in limb girth occurred in the lower

extremities during bed rest, as has been previously observed (11). The more positive nitrogen balance during reambulation suggests an active anabolic process involving lean body tissue.

Both sodium and potassium balances tended to be more negative during bed rest than during the baseline period. This presumably reflects loss of both extra- and intra-cellular fluid and may correlate with the observed weight losses. Markedly positive balances of these electrolytes, an increase in weight, and pedal edema were observed during reambulation, suggesting accumulation of fluid. These observations extend similar findings of previous investigations (11,27), not only by increasing the duration of bed rest observation but also by providing concise quantitation of fecal and dermal losses.

The increased urine volumes during bed rest reflect the fact that oral fluids were encouraged to guard against possible urinary calculus formation. Under these conditions, there was no evidence of crystal or stone formation. The increased excretion of pyrophosphate during bed rest may have exerted a protective influence, since this substance is known to be an inhibitor of crystallization (34). Insensible fluid loss was decreased during bed rest, presumably reflecting reduced excretion of water by sweat and respiration which attended the lower level of physical activity.

A reduction in both plasma volume and red blood cell volume was observed during bed rest. The data show that these changes, which have been observed in previous short-term bed rest studies (27), persist for at least 30 weeks. The mechanism for the reduction in plasma volume is uncertain; the change is important, since it probably increases the individual's susceptibility to orthostatic hypotension (27). The reduced red cell volume cannot be attributed to venipuncture alone, since the value increased during reambulation when the frequency of blood withdrawal was not changed. Recent data implicate a reduction in bone marrow erythrocyte production during bed rest (32).

Magnesium balance data show a moderate negative mean balance during bed rest (-13 to -17 mg/day). Loss of magnesium during bed rest reflects loss of lean body mass primarily; dissolution of bone would account for a small component of this loss, since the bone mineral Mg:Ca ratio is about 1:100 (10). The more positive balance which was always observed during reambulation suggests the predominance of an active anabolic process.

Clinical evaluation of the patients during this study revealed no significant morbidity during bed rest other than a lack of incentive and a tendency toward mental as well as physical inactivity. During reambulation, all three subjects experienced severe pain and tenderness in the soles of the feet which persisted for three to four weeks. The

origin of these symptoms could not be clearly defined, but soft tissue or periosteal inflammation was suspect. Orthostatic symptoms were not prominent, although care was taken to avoid sudden orthostatic challenges. Easy fatigability and mild depression were observed in all three subjects during reambulation. Complete subjective return to normal was not attained until four to six months after reambulation.

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L E G E N D S T O T H E F I G U R E S

Figure 1. The effect of prolonged bed rest on urinary calcium excretion. The mean of the weekly values for the three subjects is plotted, with the range of observations depicted by vertical lines. The weeks shown are all those in which complete urinary collections in all three subjects were obtained: the last 4 weeks of the baseline period, the first 30 weeks of bed rest, and the first 4 weeks of reambulation. The mean control value is the average of all determinations durint the last 4 baseline weeks.

Figure 2. The effect of prolonged bed rest on calcium balance in Subject R.R. Calcium output by way of sweat, urine, and stool are plotted cumulatively on the ordinate, and time is in weeks on the abscissa. Calcium intake was 908 mg/day and the net calcium balance is shown on the right-hand scale.

Figure 3. The effects of prolonged bed rest on calcium balance in Subject R.R., plotted in the same fashion as Figure 2 except that 3-5 week averages are shown to facilitate interpretation.

Figure 4. The effects of prolonged bed rest on calcium balance in Subject G.B., plotted in the same fashion as Figure 2. The patient left the hospital for 3 days during the second week of reambulation to attend to a personal emergency.

Figure 5. The effect of prolonged bed rest on calcium balance in Subject G.B., plotted in the same fashion as Figure 3.

Figure 6. The effect of prolonged bed rest on calcium balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 7. The effect of prolonged bed rest on calcium balance in Subject C.S., plotted in the same fashion as Figure 3.

Figure 8. The effect of prolonged bed rest on serum calcium and phosphorus concentrations, plotted in the same fashion as Figure 1.

Figure 9. The effect of prolonged bed rest on urinary phosphorus excretion, plotted in the same fashion as Figure 1.

Figure 10. The effect of prolonged bed rest on phosphorus balance in Subject R.R., plotted in the same fashion as Figure 2.

Figure 11. The effect of prolonged bed rest on phosphorus balance in Subject R.R., plotted in the same fashion as Figure 3.

Figure 12. The effect of prolonged bed rest on phosphorus balance in Subject G.B., plotted in the same fashion as Figure 2.

Figure 13. The effect of prolonged bed rest on phosphorus balance in Subject G.B., plotted in the same fashion as Figure 3.

Figure 14. The effect of prolonged bed rest on phosphorus balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 15. The effect of prolonged bed rest on phosphorus balance in Subject C.S., plotted in the same fashion as Figure 3.

Figure 16. The effect of prolonged bed rest on serum parathyroid hormone concentration, plotted in the same fashion as Figure 1.

Figure 17. The effect of prolonged bed rest on urinary hydroxyproline excretion, plotted in the same fashion as Figure 1.

Figure 18. The effect of prolonged bed rest on urinary pyrophosphate excretion, plotted in the same fashion as Figure 1.

Figure 19. The effect of prolonged bed rest on urinary nitrogen excretion, plotted in the same fashion as Figure 1.

Figure 20. The effect of prolonged bed rest on nitrogen balance in Subject R.R., plotted in the same fashion as Figure 2.

Figure 21. The effect of prolonged bed rest on nitrogen balance in Subject G.B., plotted in the same fashion as Figure 2.

Figure 22. The effect of prolonged bed rest on nitrogen balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 23. The effect of prolonged bed rest on mean urinary magnesium excretion, plotted in the same fashion as Figure 1.

Figure 24. The effect of prolonged bed rest on magnesium balance in Subject R.R., plotted in the same fashion as Figure 2.

Figure 25. The effect of prolonged bed rest on magnesium balance in Subject R.R., plotted in the same fashion as Figure 3.

Figure 26. The effect of prolonged bed rest on magnesium balance in Subject G.B., plotted in the same fashion as Figure 2.

Figure 27. The effect of prolonged bed rest on magnesium balance in Subject G.B., plotted in the same fashion as Figure 3.

LEGENDS (cont.)

Figure 28. The effect of prolonged bed rest on magnesium balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 29. The effect of prolonged bed rest on magnesium balance in Subject C.S., plotted in the same fashion as Figure 3.

Figure 30. The effect of prolonged bed rest on mean serum magnesium concentration, plotted in the same fashion as Figure 1.

Figure 31. The effect of prolonged bed rest on mean urinary sodium and potassium excretion, plotted in the same fashion as Figure 1.

Figure 32. The effect of prolonged bed rest on sodium balance in Subject R.R., plotted in the same fashion as Figure 2.

Figure 33. The effect of prolonged bed rest on sodium balance in Subject G.B., plotted in the same fashion as Figure 2.

Figure 34. The effect of prolonged bed rest on sodium balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 35. The effect of prolonged bed rest on serum sodium and potassium concentrations, plotted in the same fashion as Figure 1.

Figure 36. The effect of prolonged bed rest on potassium balance in Subject R.R., plotted in the same fashion as Figure 2.

Figure 37. The effect of prolonged bed rest on potassium balance in Subject G.B., plotted in the same fashion as Figure 2.

Figure 38. The effect of prolonged bed rest on potassium balance in Subject C.S., plotted in the same fashion as Figure 2.

Figure 39. The effect of prolonged bed rest on body weight. Individual values for the three subjects are shown, the bed rest period being represented by open symbols and ambulatory periods by solid symbols.

Figure 40. The effect of prolonged bed rest on fluid balance in Subject R.R. Fluid intake is plotted downwards on the ordinate and urine output is plotted upwards from the intake value. The difference is shown as "insensible loss" and does not include a correction for changes in body weight or fecal water.

Figure 41. The effect of prolonged bed rest on fluid balance in Subject G.B., plotted in the same fashion as Figure 40.

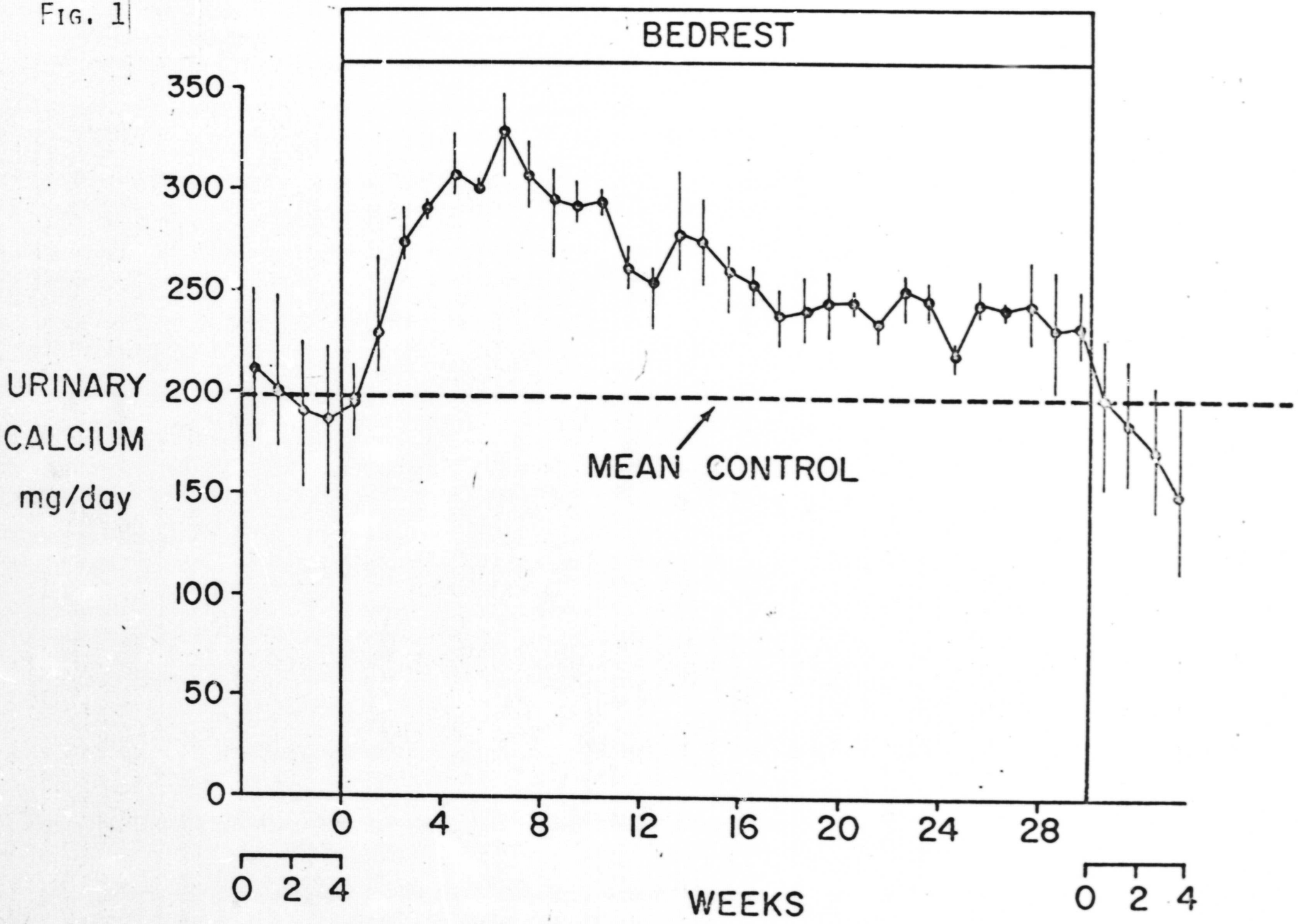
Figure 42. The effect of prolonged bed rest on fluid balance in Subject C.S., plotted in the same fashion as Figure 40.

Figure 43. The effect of prolonged bed rest on creatinine clearance, plotted in the same fashion as Figure 1.

Figure 44. The effect of prolonged bed rest on total blood volume, plasma volume, and red-blood cell volume. Mean values for the three subjects are shown.

Figure 45. The effect of prolonged bed rest on extra-cellular fluid volume (bromine space). Individual values for the three subjects are shown.

FIG. 1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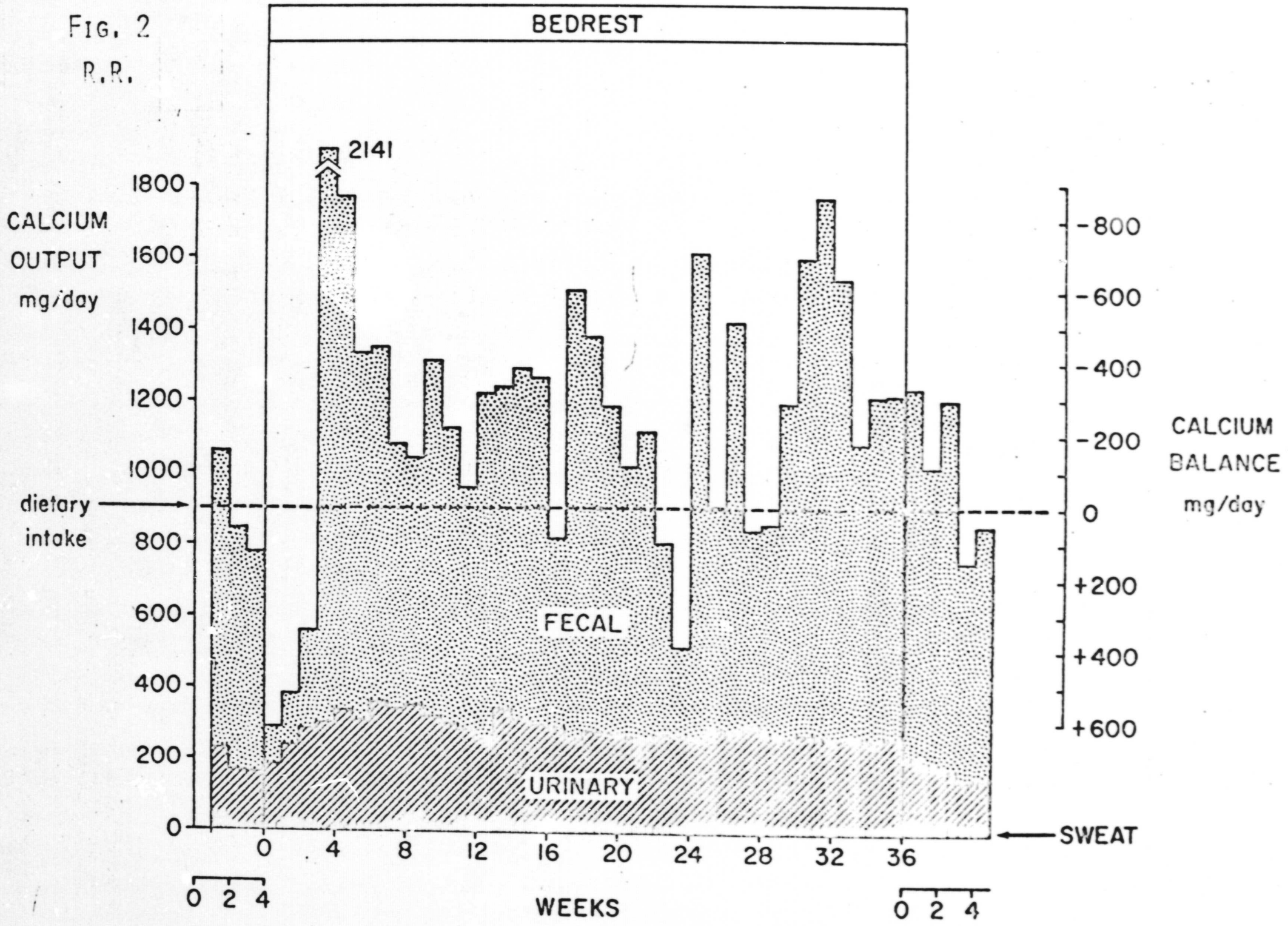
250
200
150
100
50
0
URINARY
CALCIUM
mg/day
MEAN CONTROL
0
4
8
12
16
20
24
28
0 2 4
WEEKS
0 2 4

FIG. 2

R.R.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1400
1200
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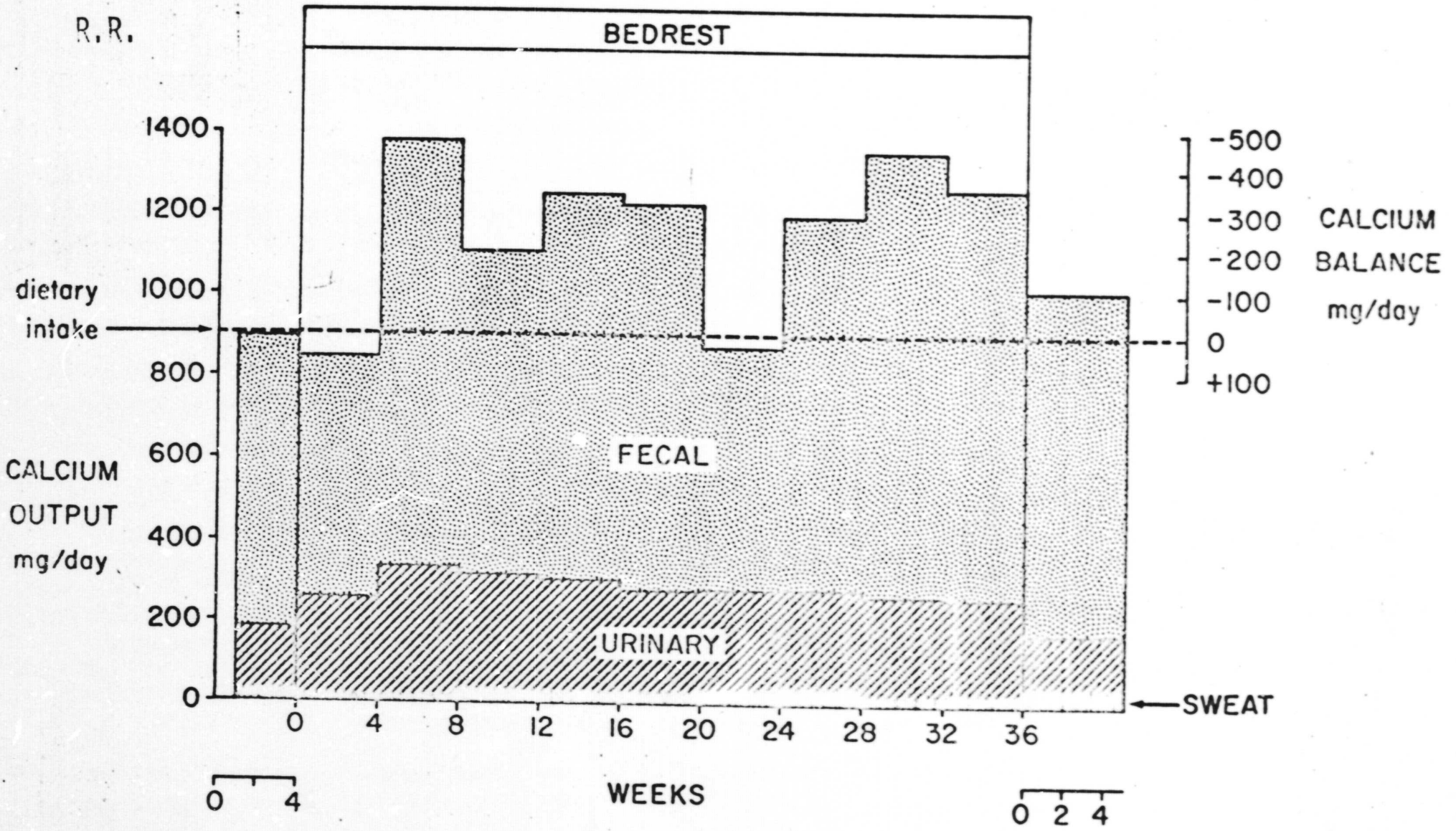
800
600
400
200
0
dietary intake
FECAL
URINARY
SWEAT
-800
-600
-400
-200
0
+200
+400
+600
CALCIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
0 2 4
WEEKS
0 2 4

FIG. 3

R.R.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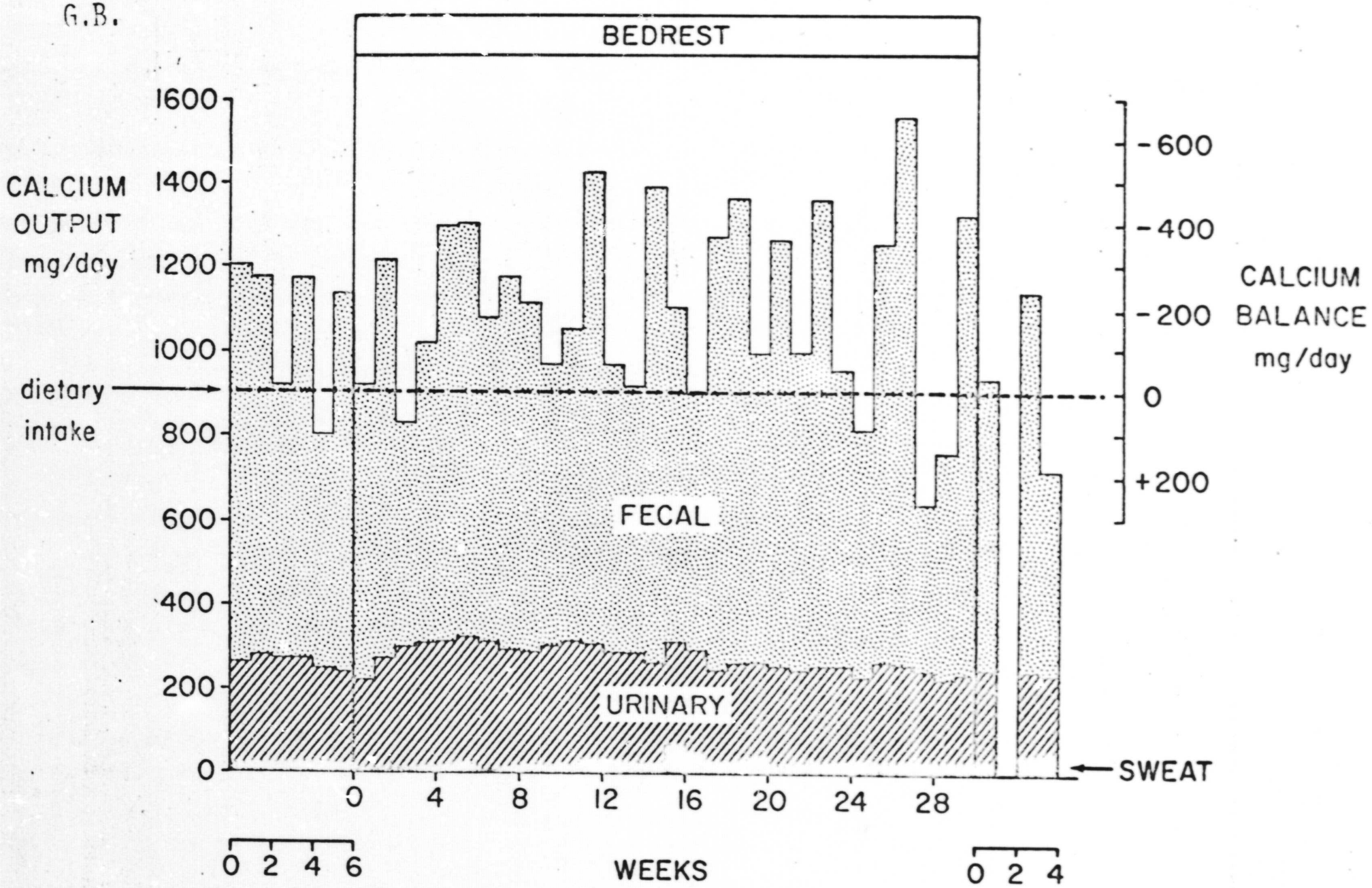
1400
1200
1000
800
600
400
200
0
dietary intake
CALCIUM OUTPUT mg/day
FECAL
URINARY
SWEAT
-500
-400
-300
-200
-100
0
+100
CALCIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
WEEKS
0 4
0 2 4

FIG. 4

G.B.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CALCIUM OUTPUT mg/day
1600
1400
1200
1000
800
600
400
200
0
dietary intake
FECAL
URINARY
SWEAT
-600
-400
-200
0
+200
CALCIUM BALANCE mg/day
0 4 8 12 16 20 24 28
0 2 4 6
WEEKS
0 2 4

FIG. 5

G.B.

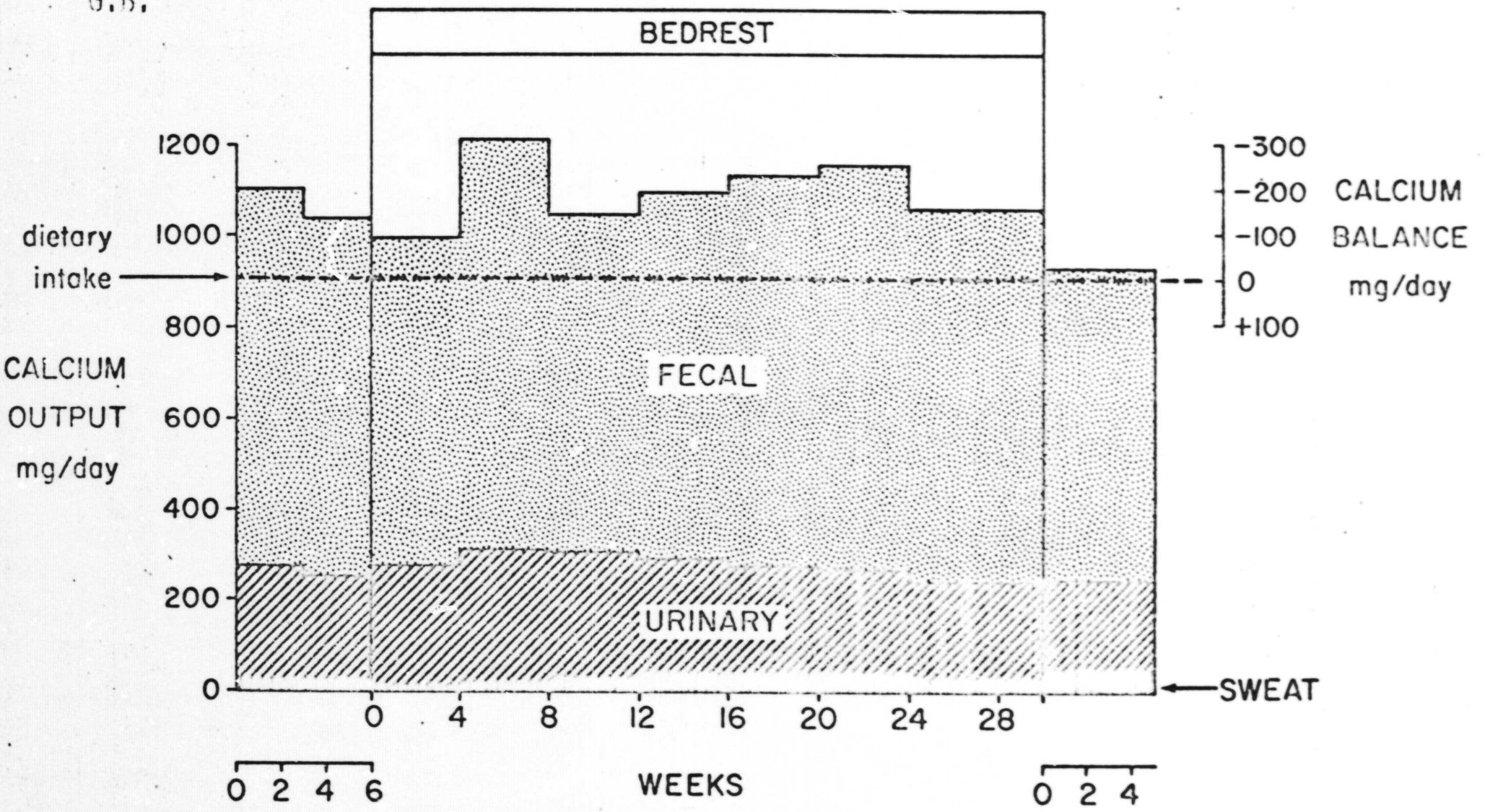
BEDREST
1200
1000
800
600
400
200
0
dietary intake
CALCIUM OUTPUT mg/day
FECAL
URINARY
SWEAT
-300
-200
-100
0
+100
CALCIUM BALANCE mg/day
0 4 8 12 16 20 24 28
WEEKS
0 2 4 6
0 2 4

FIG. 6

C.S.

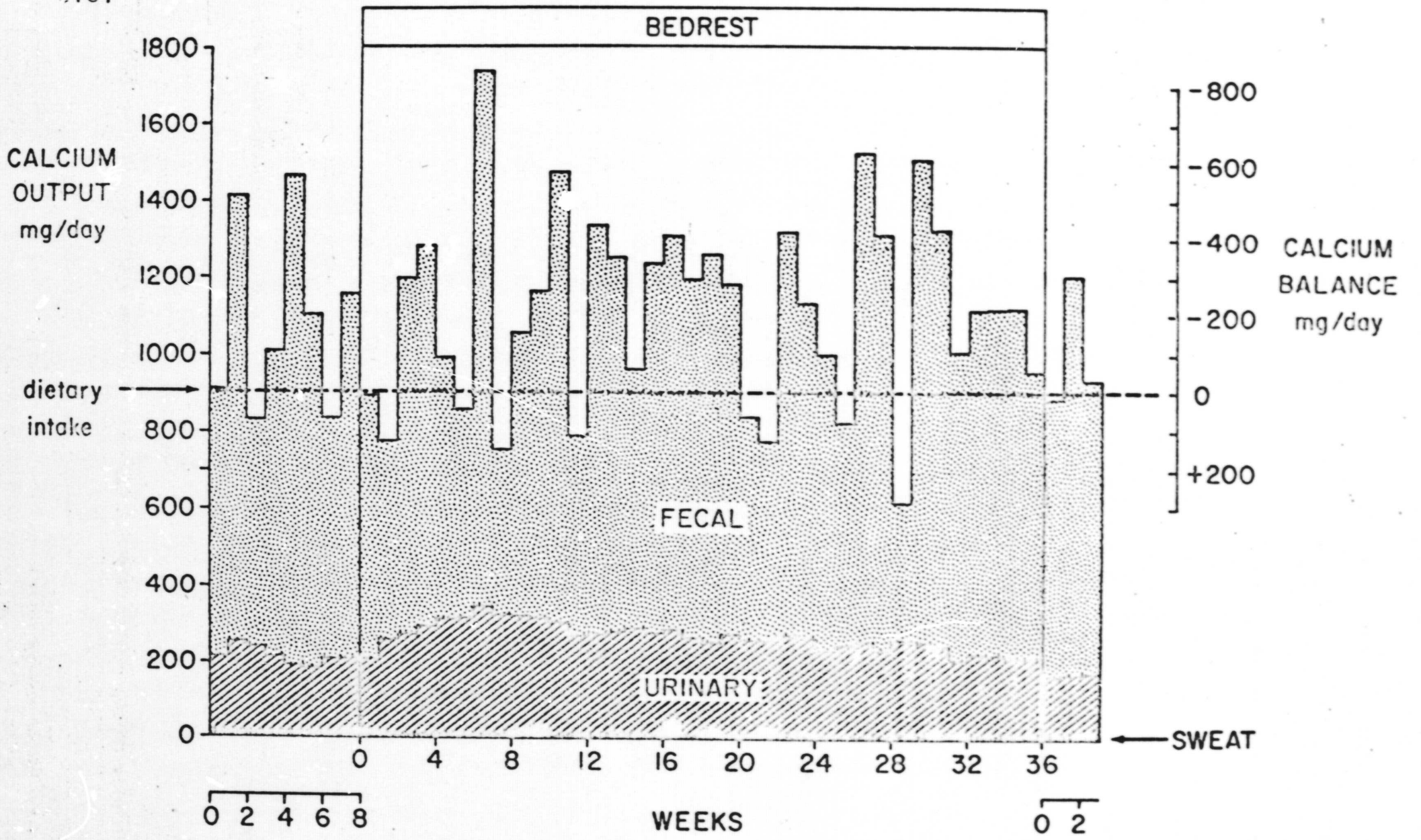
BEDREST
CALCIUM OUTPUT mg/day
1800
1600
1400
1200
1000
dietary intake
800
600
400
200
0
FECAL
URINARY
SWEAT
-800
-600
-400
-200
0
+200
CALCIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
0 2 4 6 8
WEEKS
0 2

FIG. 7

C.S.

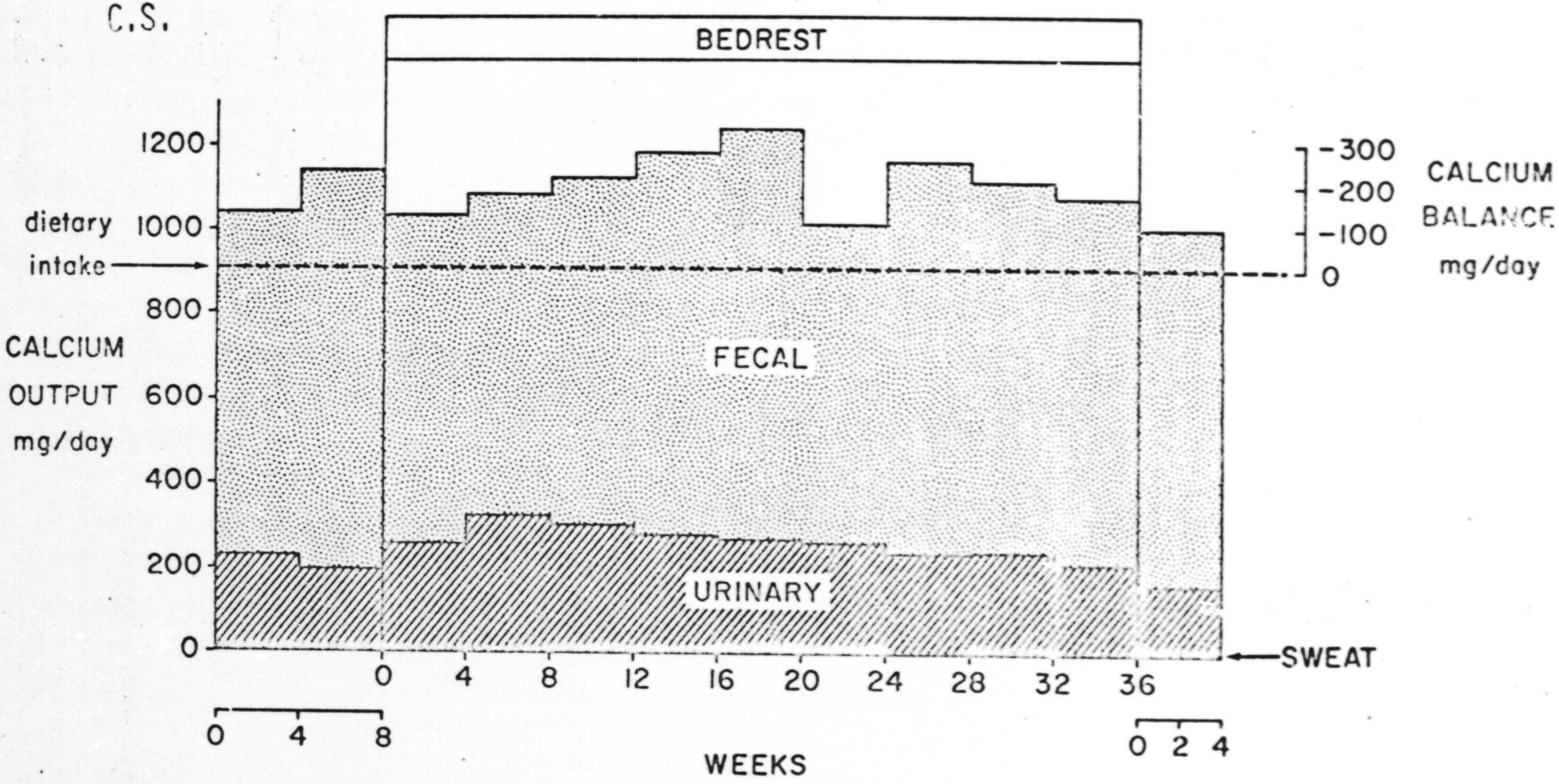
BEDREST
1200
dietary
1000
intake
800
CALCIUM
OUTPUT
600
mg/day
400
200
0
FECAL
URINARY
SWEAT
-300
-200
-100
0
CALCIUM
BALANCE
mg/day
0
4
8
12
16
20
24
28
32
36
0
4
8
0
2
4
WEEKS

FIG. 8

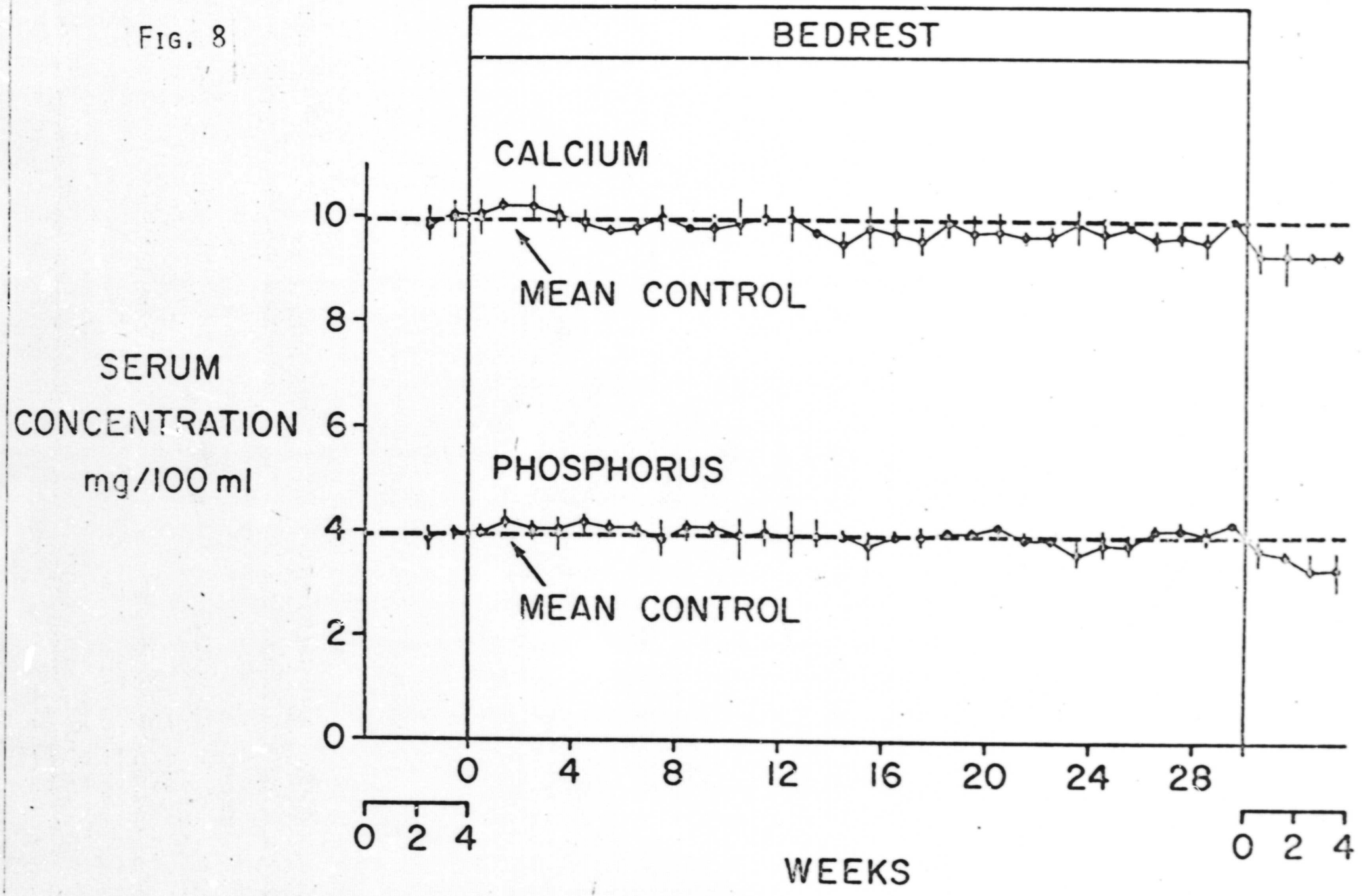
BEDREST
CALCIUM
MEAN CONTROL
PHOSPHORUS
MEAN CONTROL
SERUM CONCENTRATION mg/100 ml
10
8
6
4
2
0
0
4
8
12
16
20
24
28
0 2 4
0 2 4
WEEKS

FIG. 9

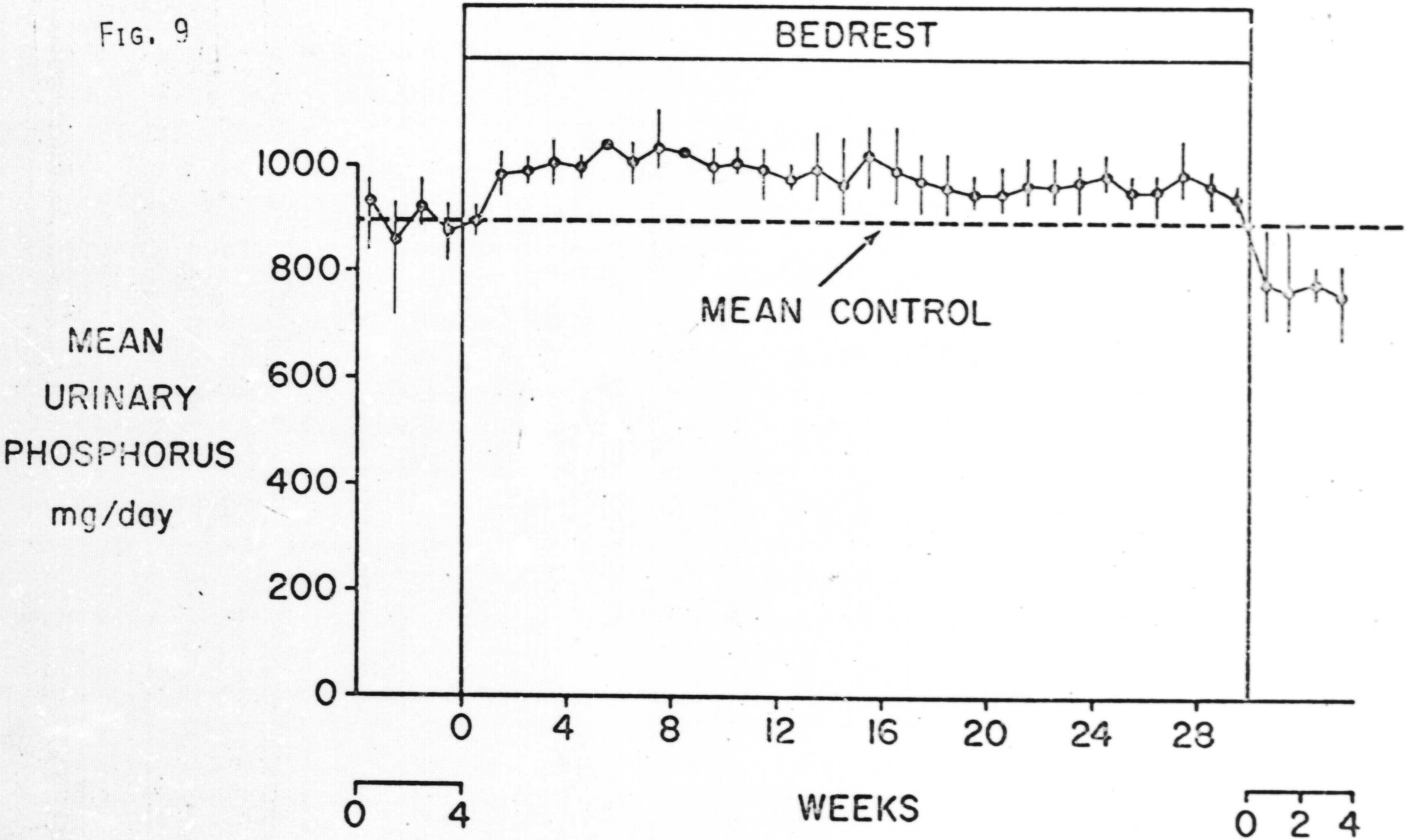
BEDREST
MEAN CONTROL
MEAN URINARY PHOSPHORUS mg/day
1000
800
600
400
200
0
0
4
8
12
16
20
24
28
0 4
WEEKS
0 2 4

FIG. 10

THE EFFECT OF PROLONGED BEDREST ON PHOSPHORUS BALANCE (R.R.)

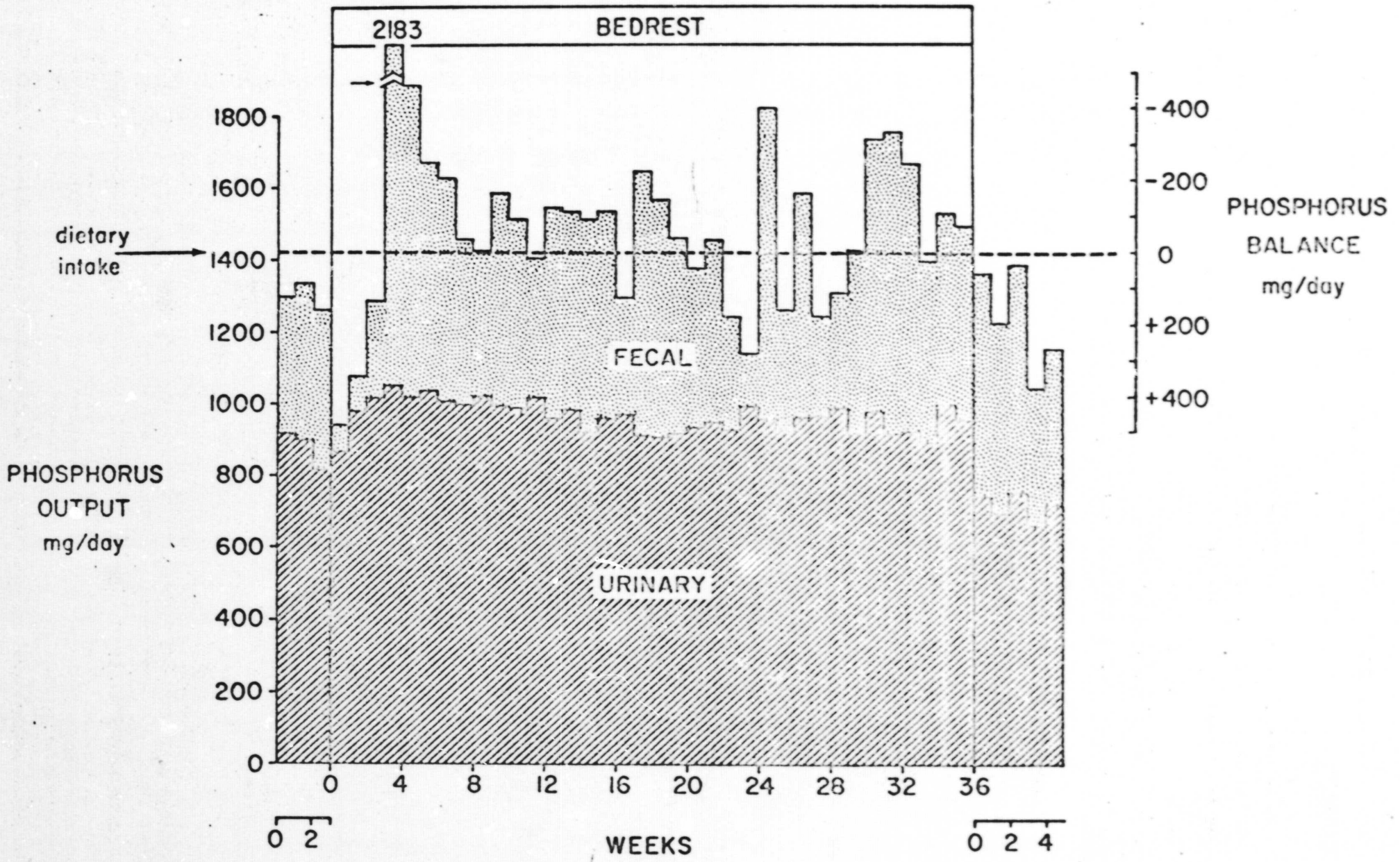

FIG. 11

THE EFFECT OF PROLONGED BEDREST ON
PHOSPHORUS BALANCE (R.R.)

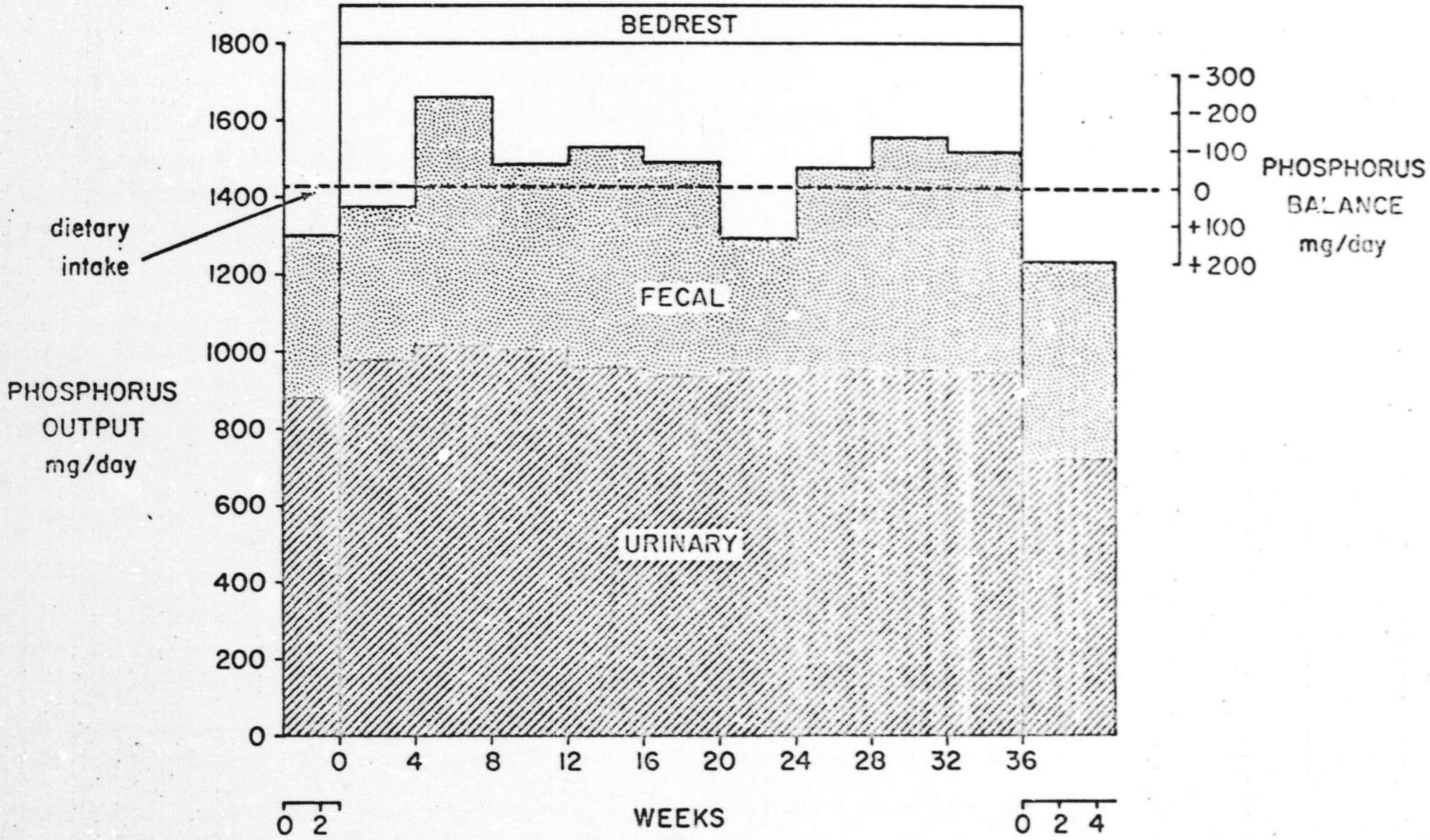

FIG. 12

THE EFFECT OF PROLONGED BEDREST ON PHOSPHORUS BALANCE (G.B.)

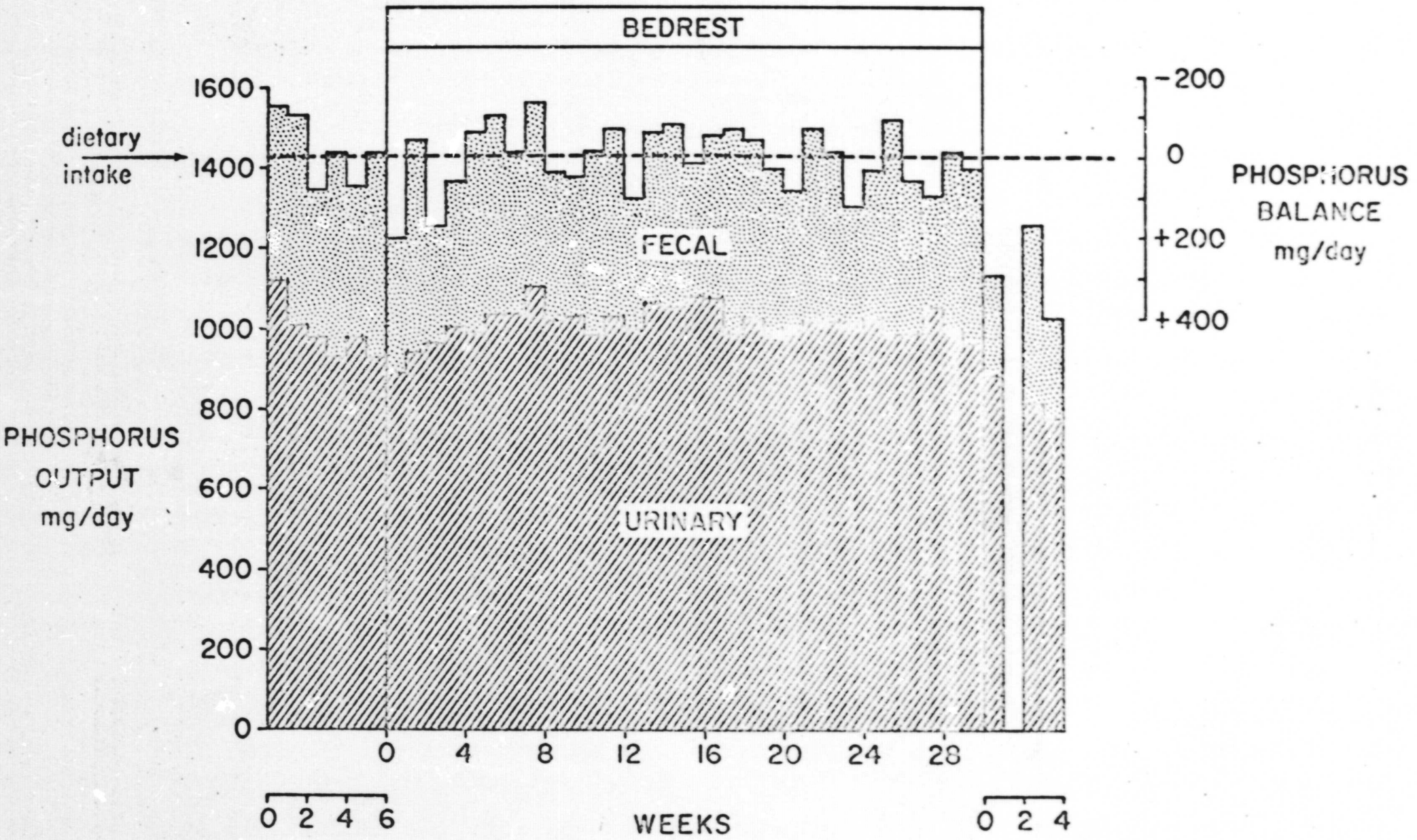

FIG. 13

THE EFFECT OF PROLONGED BEDREST ON PHOSPHORUS BALANCE (G.B.)

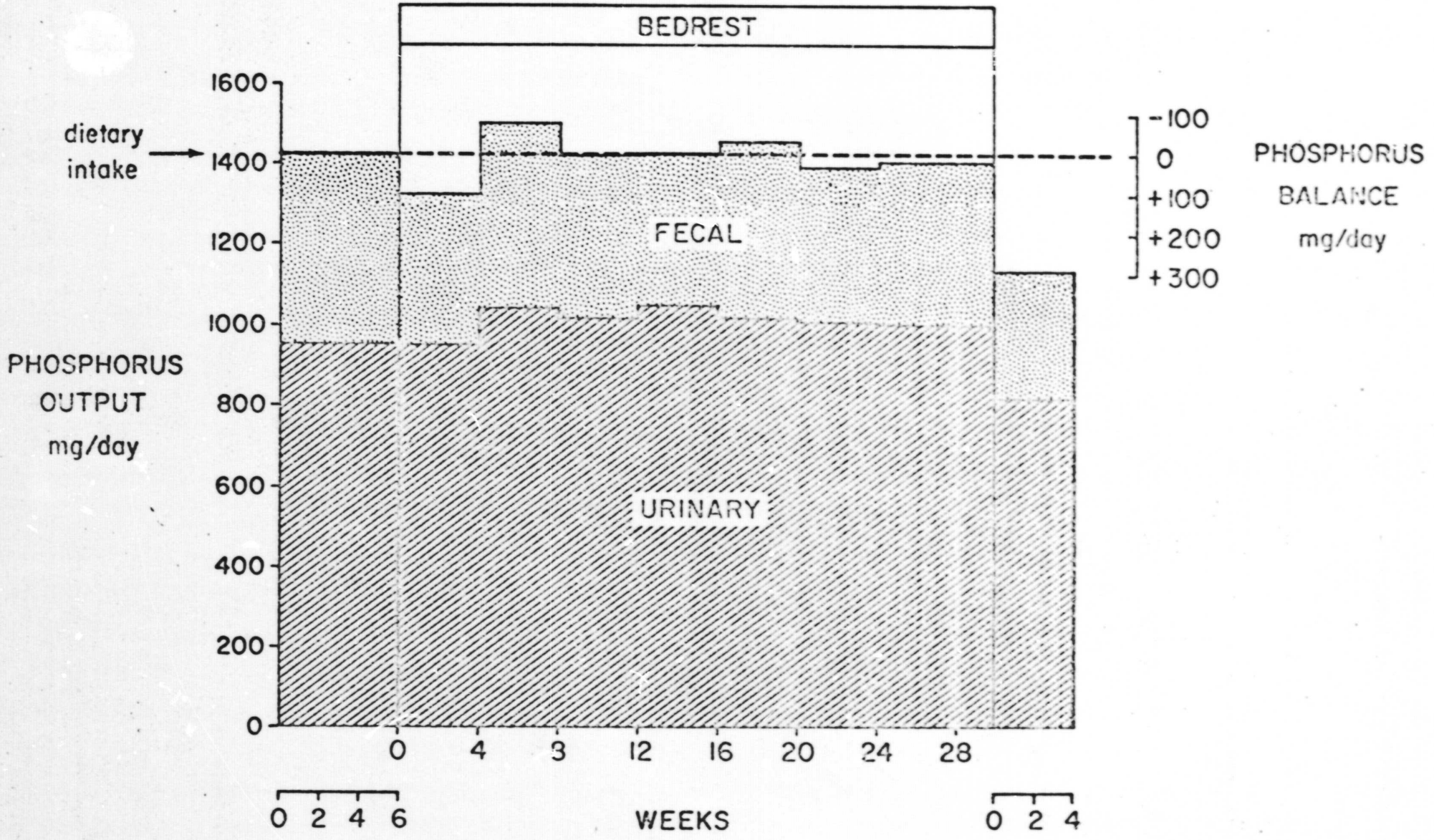

FIG. 14

THE EFFECT OF PROLONGED BEDREST ON PHOSPHORUS BALANCE (C.S.)

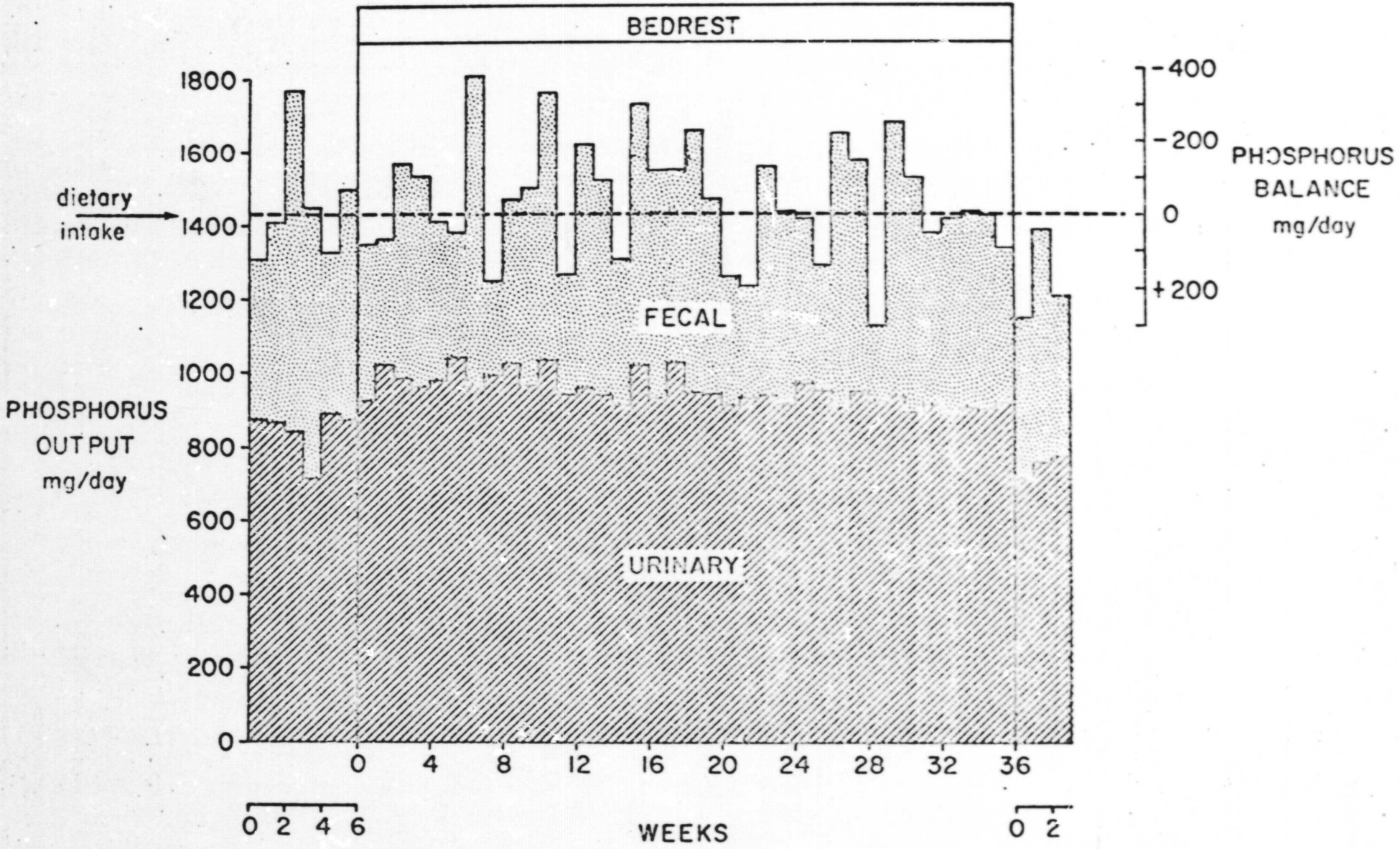

FIG. 15

THE EFFECT OF PROLONGED BEDREST ON
PHOSPHORUS BALANCE (C.S.)

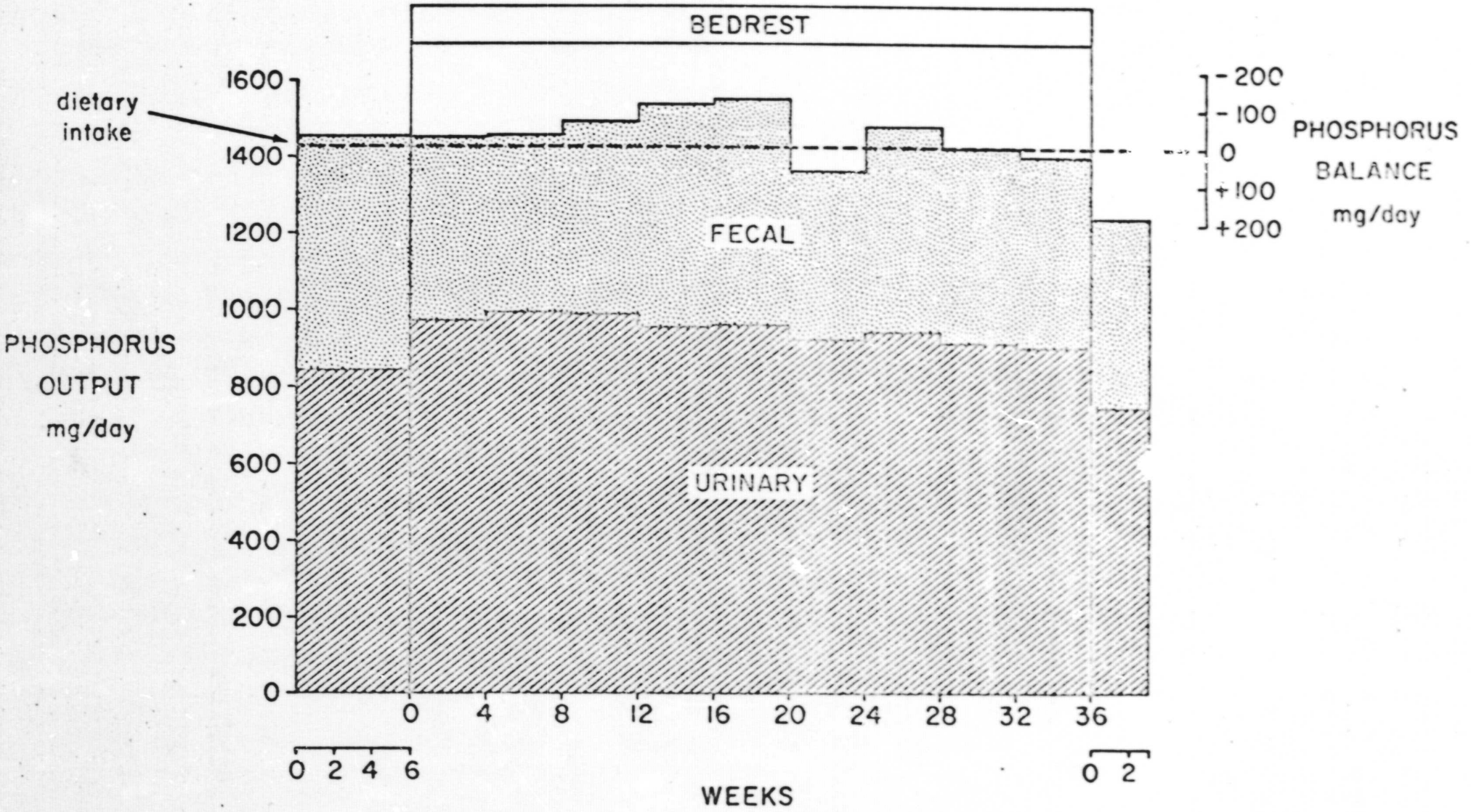

FIG. 16

THE EFFECT OF PROLONGED BEDREST ON
MEAN SERUM
PARATHYROID HORMONE CONCENTRATION (PTH)
(3 SUBJECTS)

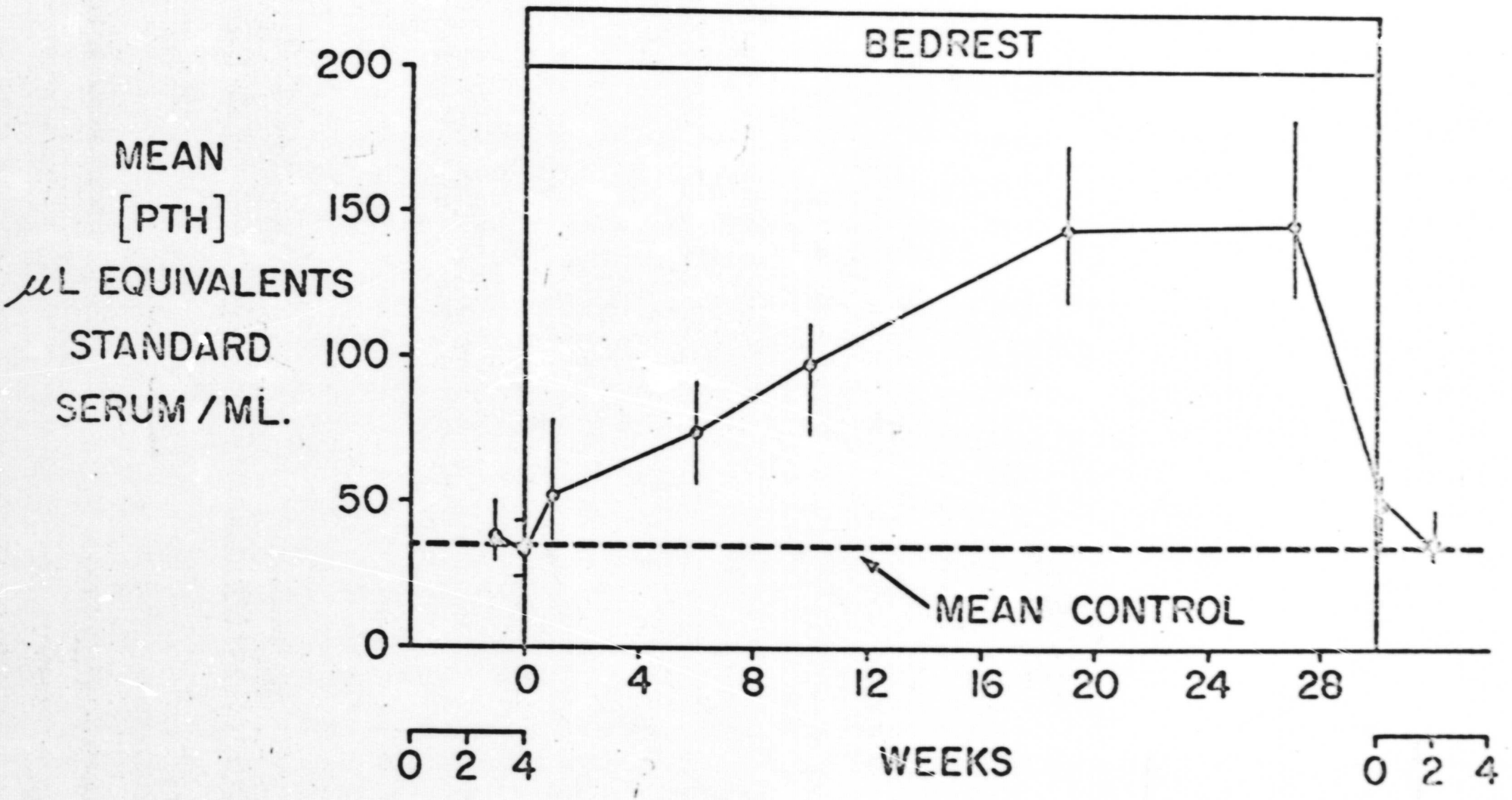

FIG. 17

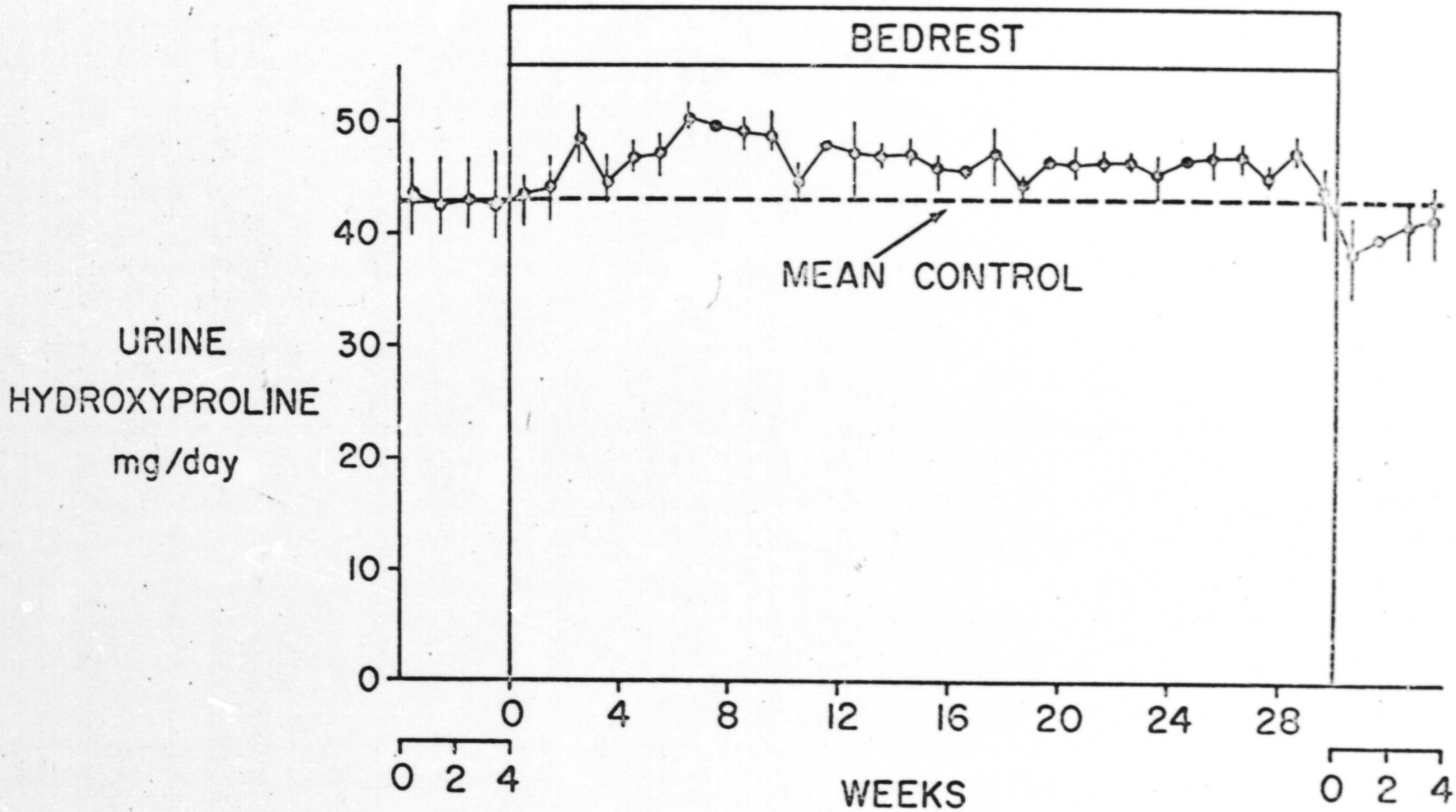
BEDREST
MEAN CONTROL
URINE
HYDROXYPROLINE
mg/day
50
40
30
20
10
0
0
4
8
12
16
20
24
28
0 2 4
WEEKS
0 2 4

FIG. 18

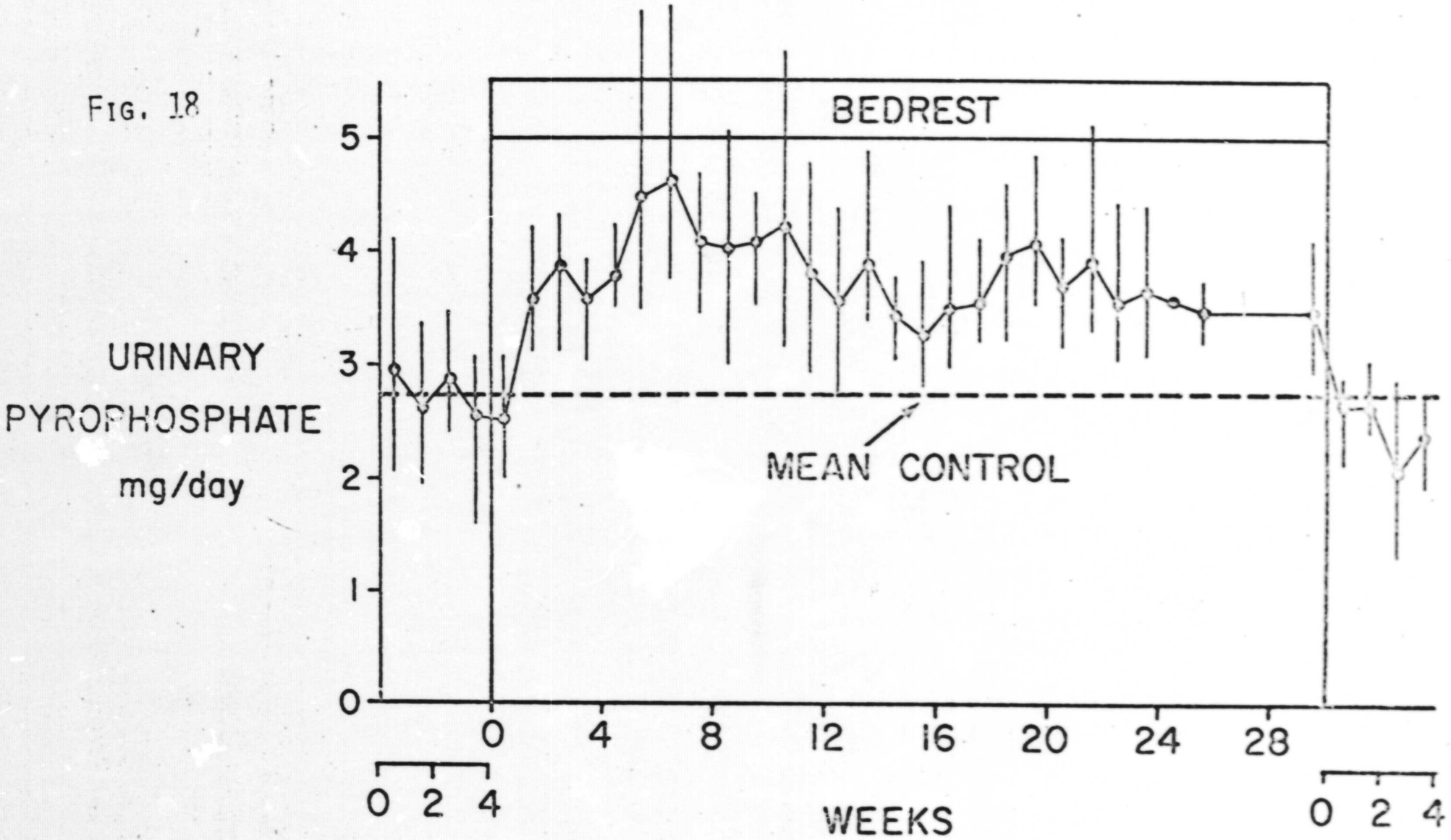
BEDREST
URINARY
PYROPHOSPHATE
mg/day
MEAN CONTROL
5
4
3
2
1
0
0
4
8
12
16
20
24
28
0 2 4
WEEKS
0 2 4

FIG. 19

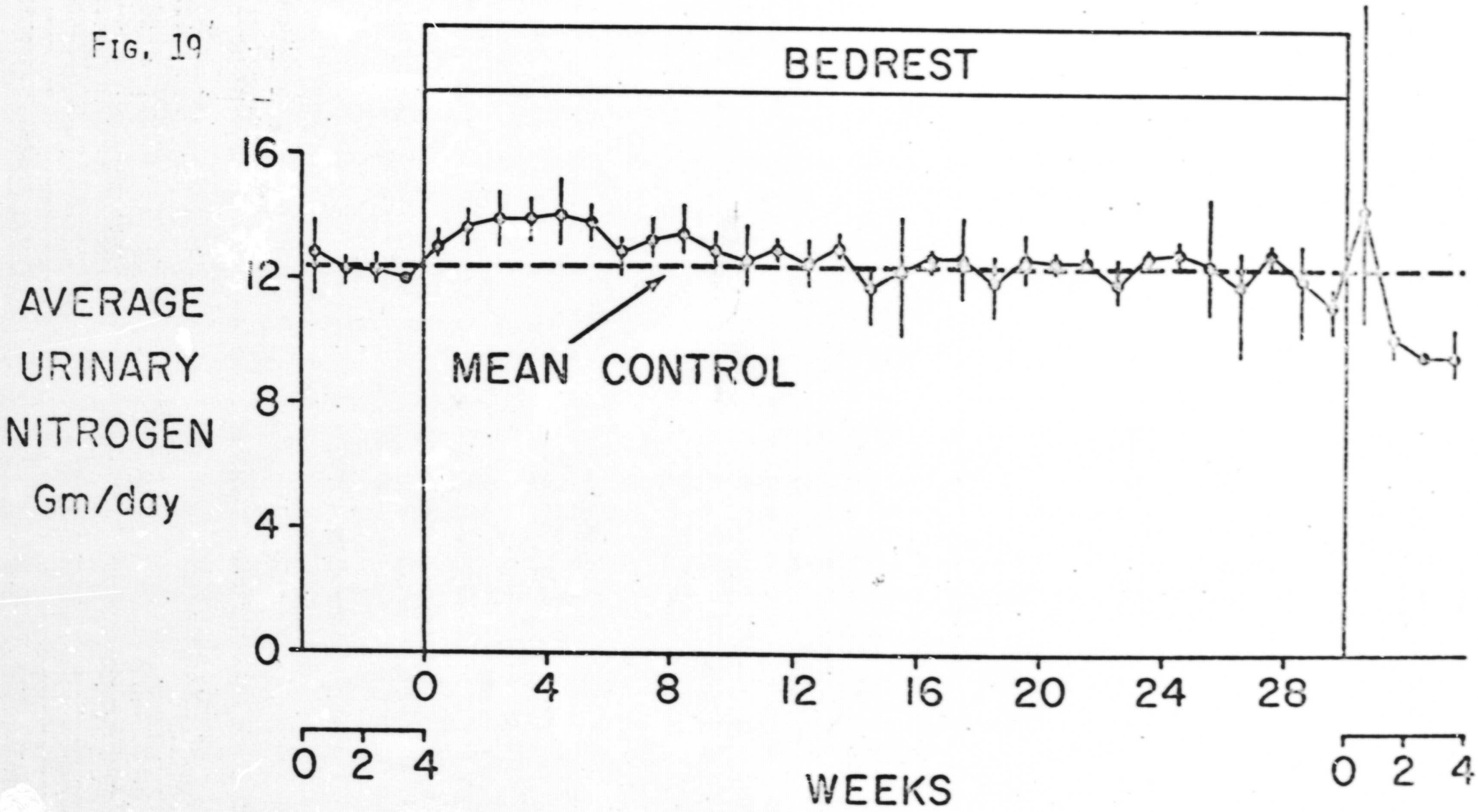
BEDREST
AVERAGE URINARY NITROGEN Gm/day
16
12
8
4
0
MEAN CONTROL
0
4
8
12
16
20
24
28
0 2 4
WEEKS
0 2 4

FIG. 20

R.R.

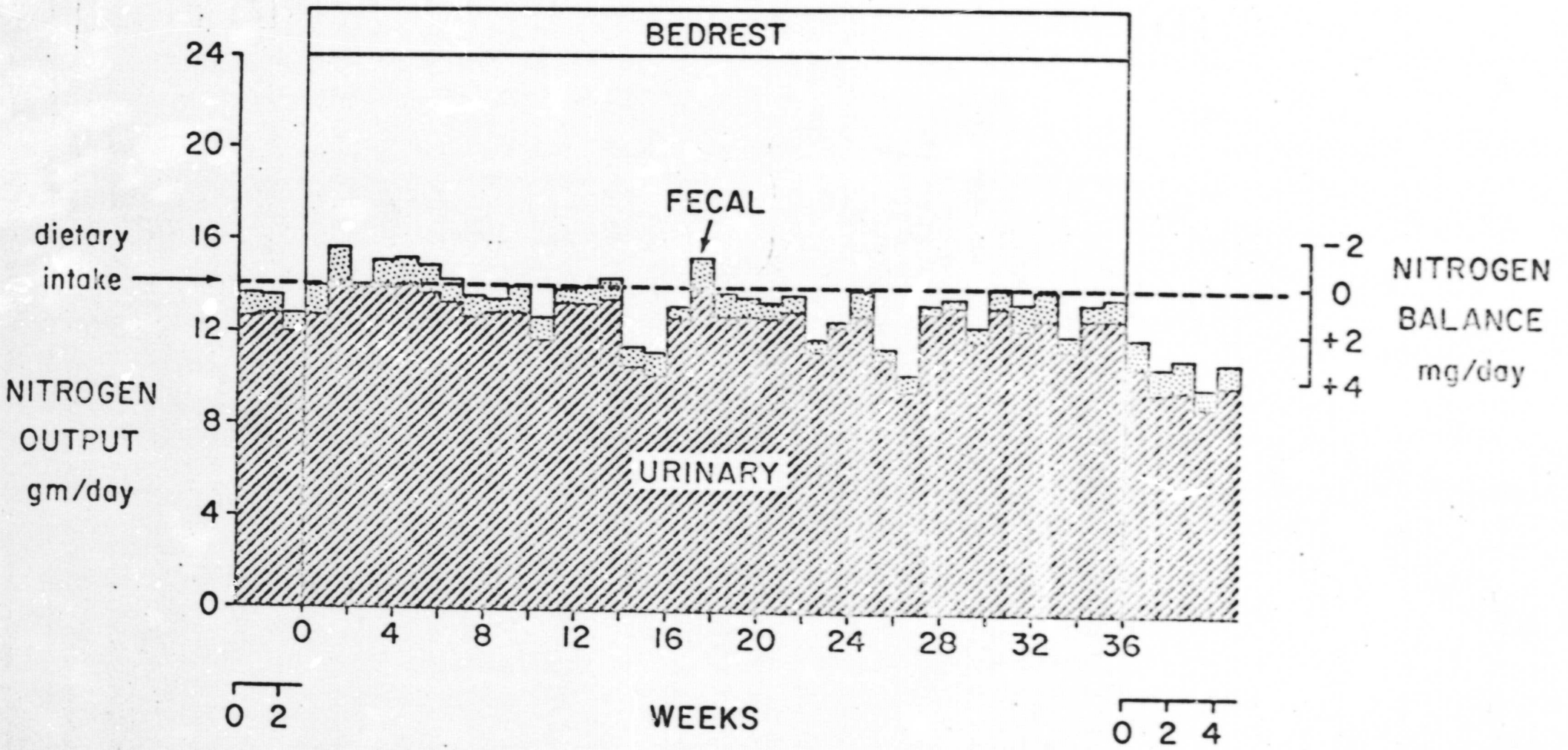
BEDREST
FECAL
dietary intake
URINARY
NITROGEN OUTPUT gm/day
24
20
16
12
8
4
0
0 4 8 12 16 20 24 28 32 36
WEEKS
0 2
0 2 4
-2
0
+2
+4
NITROGEN BALANCE mg/day

FIG. 21

G.B.

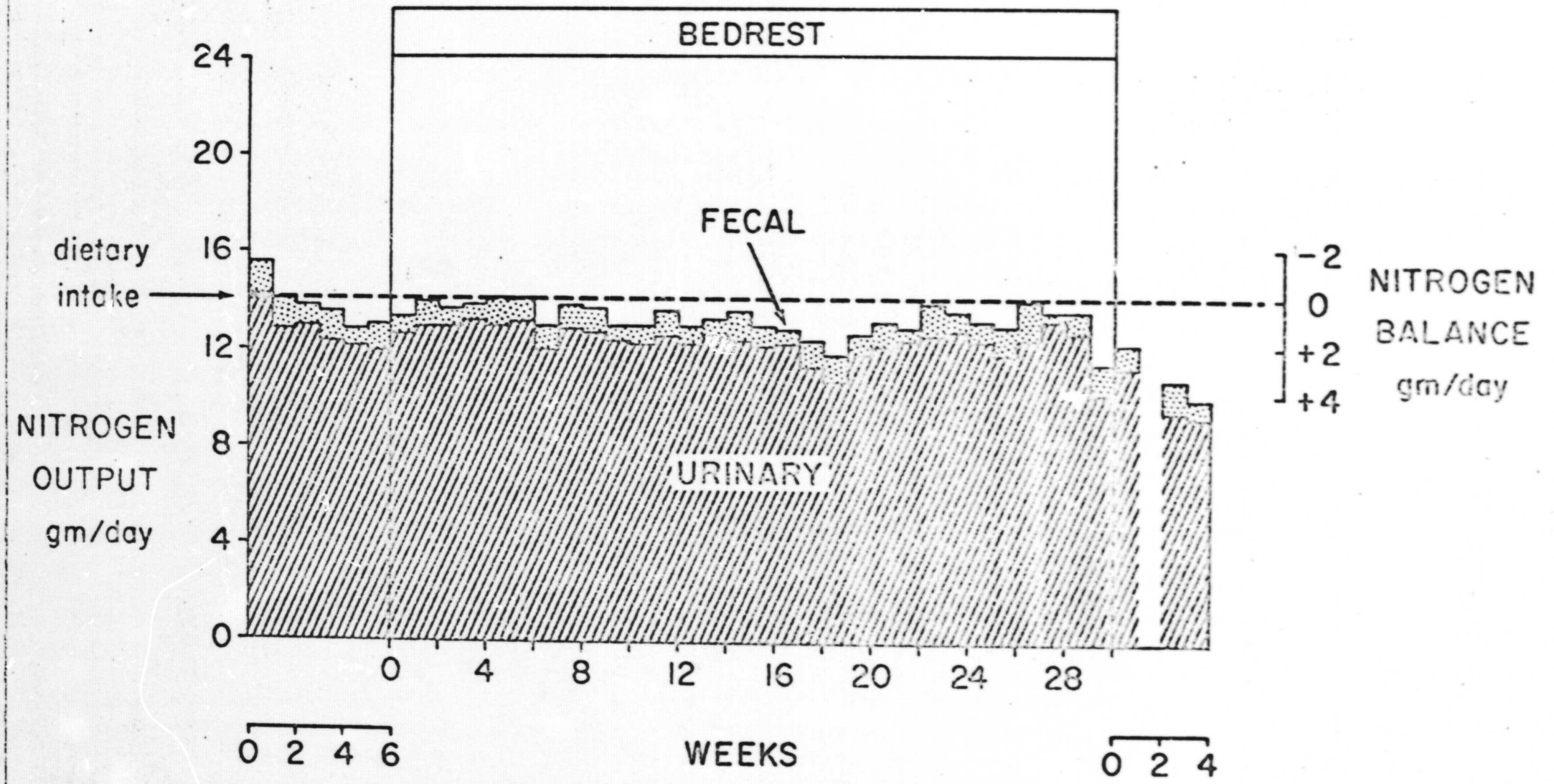
BEDREST
24
20
16
12
8
4
0
dietary intake
FECAL
URINARY
NITROGEN OUTPUT gm/day
-2
0
+2
+4
NITROGEN BALANCE gm/day
0 4 8 12 16 20 24 28
WEEKS
0 2 4 6
0 2 4

FIG. 22

C.S.

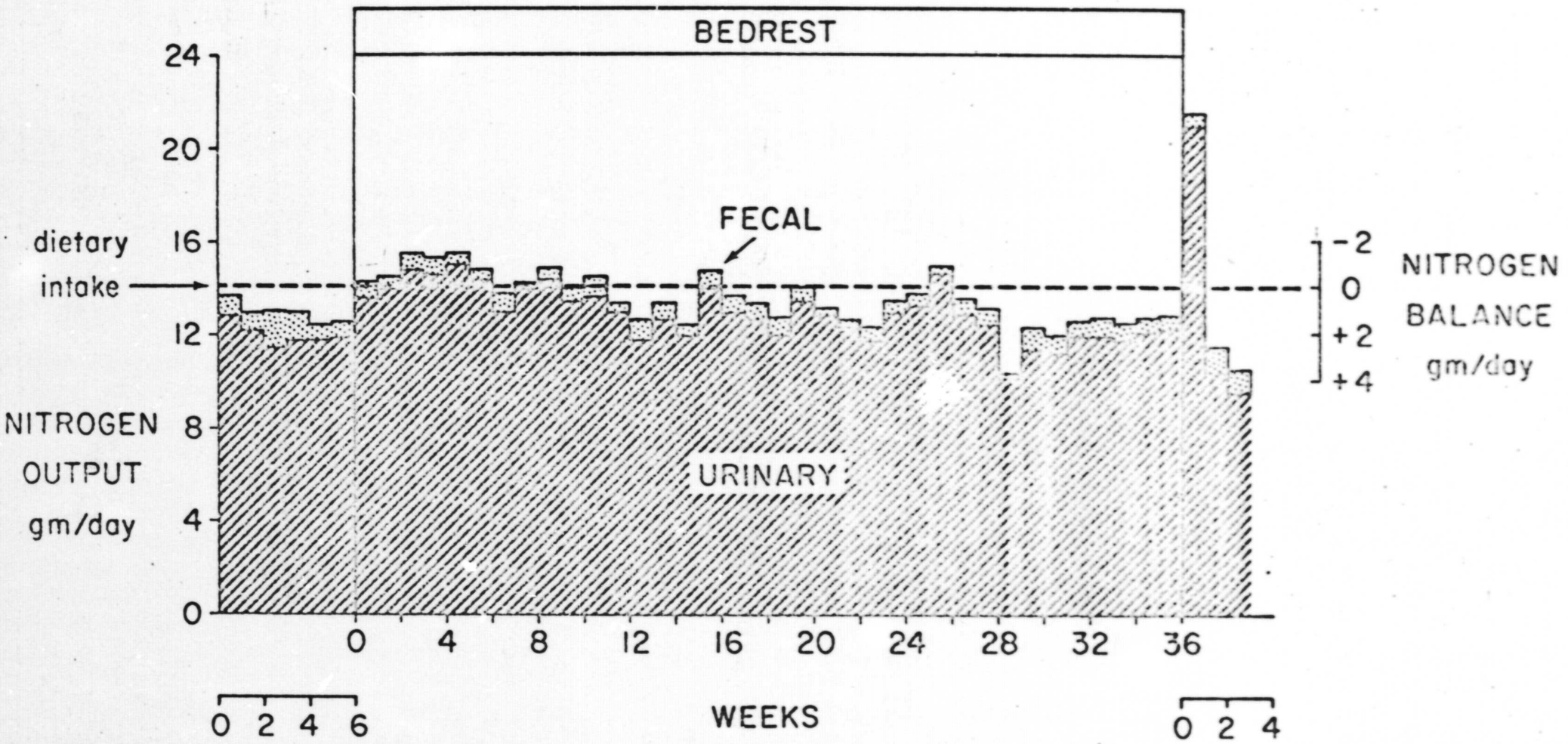
BEDREST
FECAL
URINARY
dietary intake
NITROGEN OUTPUT gm/day
24
20
16
12
8
4
0
0 4 8 12 16 20 24 28 32 36
WEEKS
0 2 4 6
0 2 4
-2
0
+2
+4
NITROGEN BALANCE gm/day

FIG. 23

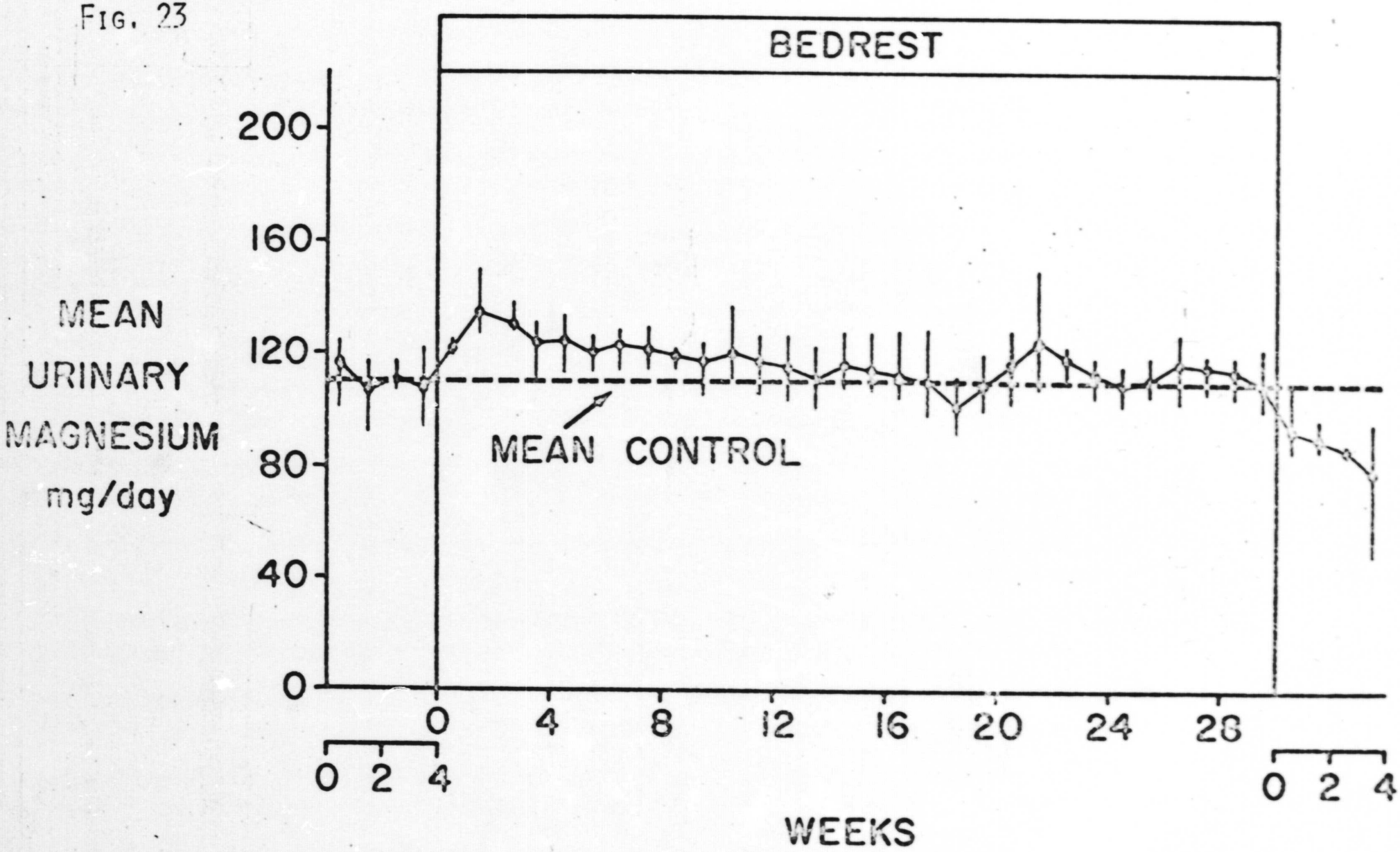
BEDREST
MEAN URINARY MAGNESIUM mg/day
200
160
120
80
40
0
MEAN CONTROL
0
4
8
12
16
20
24
28
0 2 4
0 2 4
WEEKS

FIG. 24

R.R.

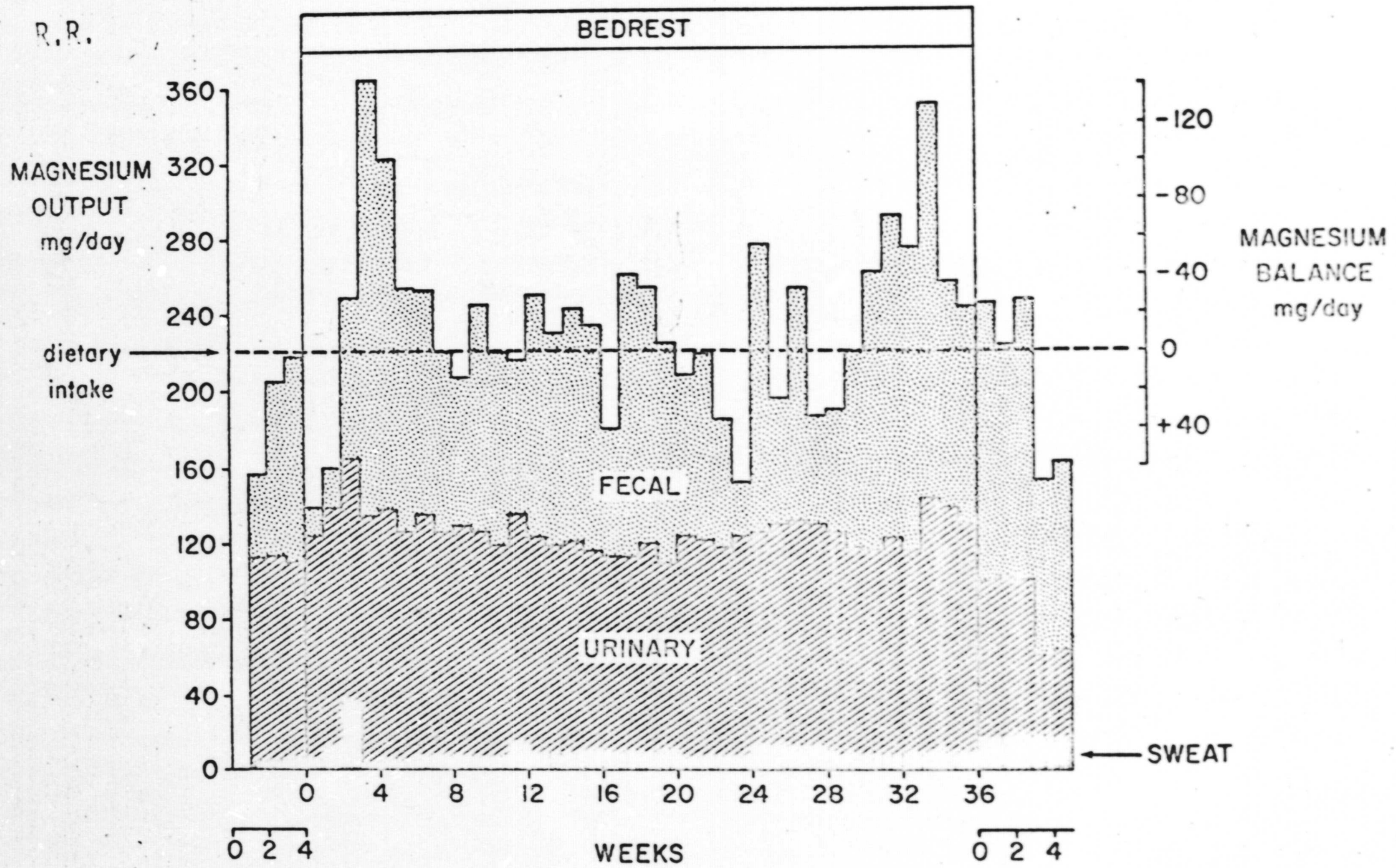
BEDREST
MAGNESIUM OUTPUT mg/day
360
320
280
240
200
160
120
80
40
0
dietary intake
FECAL
URINARY
SWEAT
-120
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
WEEKS
0 2 4
0 2 4

FIG. 25
R.R.

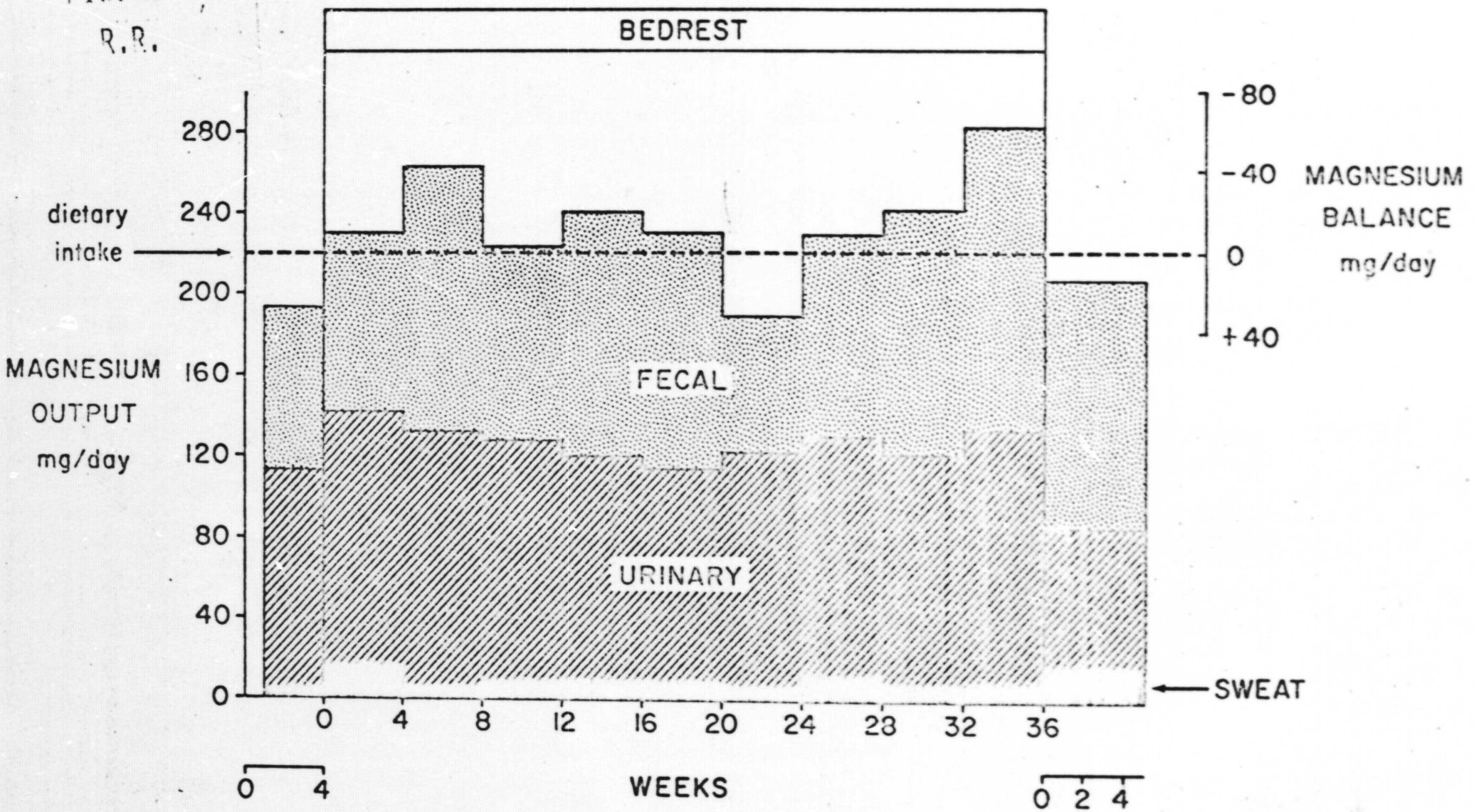
BEDREST
280
240
200
160
120
80
40
0
dietary intake
MAGNESIUM OUTPUT mg/day
FECAL
URINARY
SWEAT
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
WEEKS
0 4
0 2 4

FIG. 26

G.B.

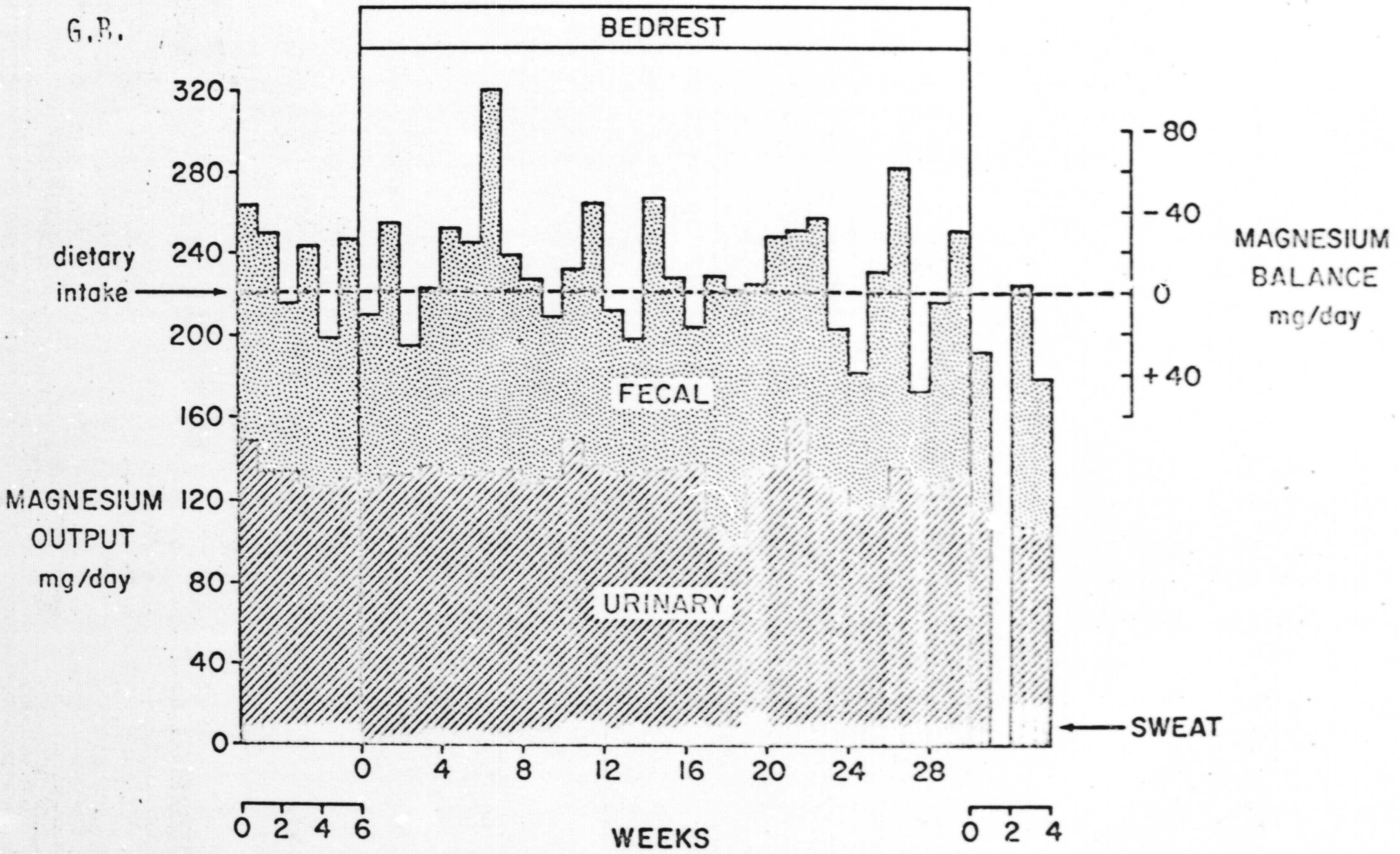
BEDREST
320
280
240
200
160
120
80
40
0
dietary intake
MAGNESIUM OUTPUT mg/day
FECAL
URINARY
SWEAT
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
0 4 8 12 16 20 24 28
WEEKS
0 2 4 6
0 2 4

FIG. 27

G.B.

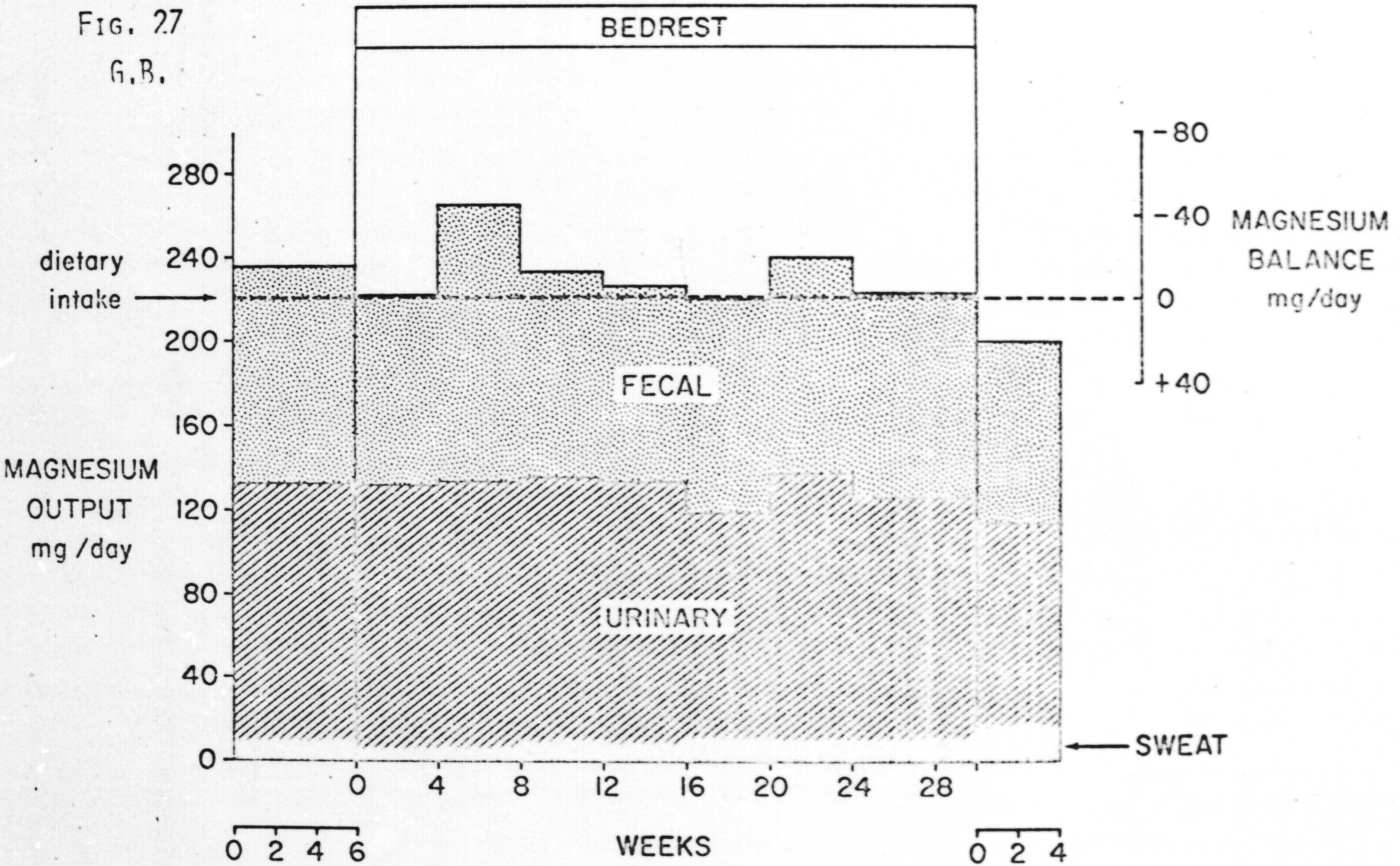
BEDREST
280
240
200
160
120
80
40
0
dietary intake
MAGNESIUM OUTPUT mg/day
FECAL
URINARY
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
SWEAT
0 4 8 12 16 20 24 28
WEEKS
0 2 4 6
0 2 4

FIG. 28

C.S.

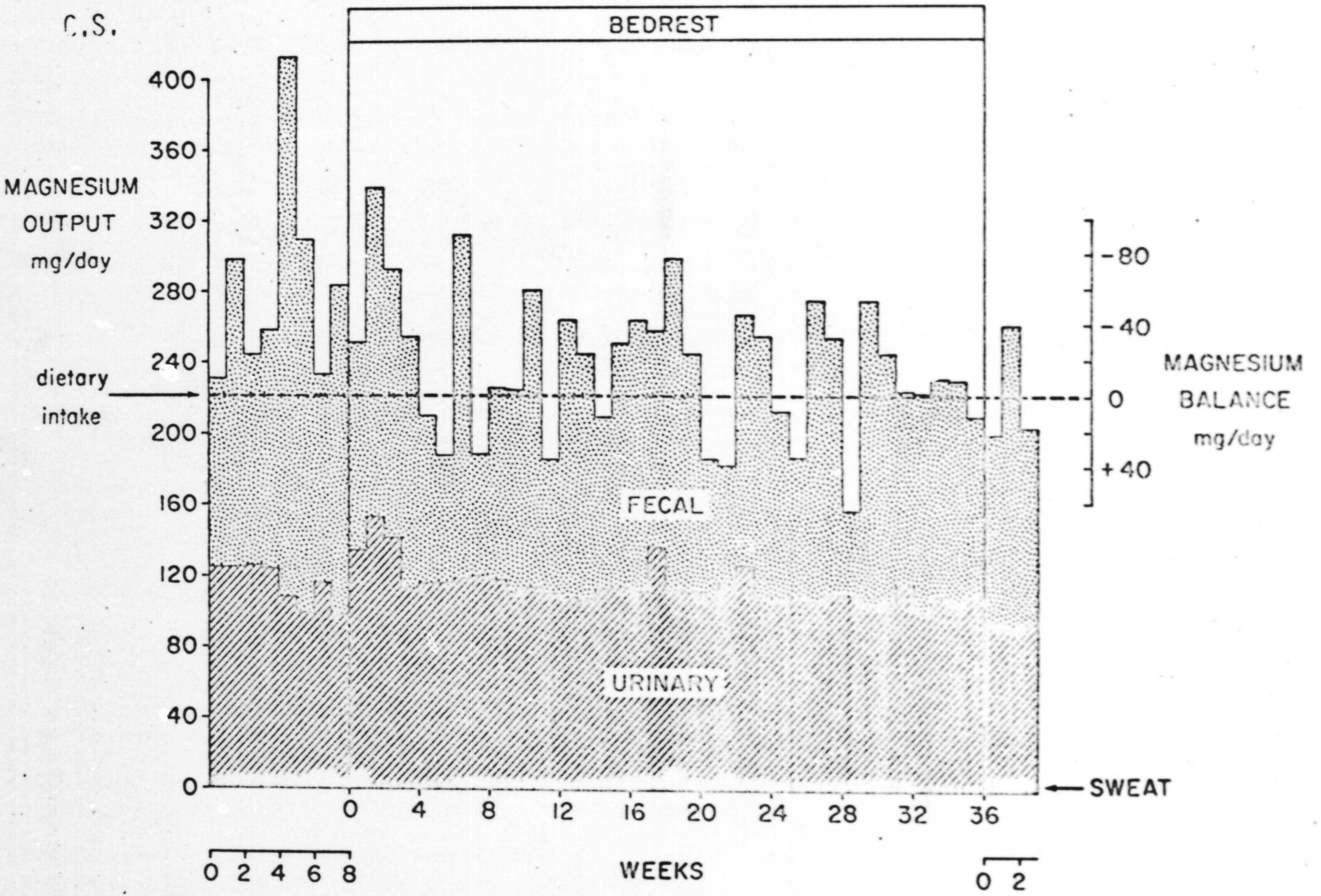
BEDREST
MAGNESIUM OUTPUT mg/day
400
360
320
280
240
200
160
120
80
40
0
dietary intake
FECAL
URINARY
SWEAT
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
0 4 8 12 16 20 24 28 32 36
WEEKS
0 2 4 6 8
0 2

FIG. 29

C.S.

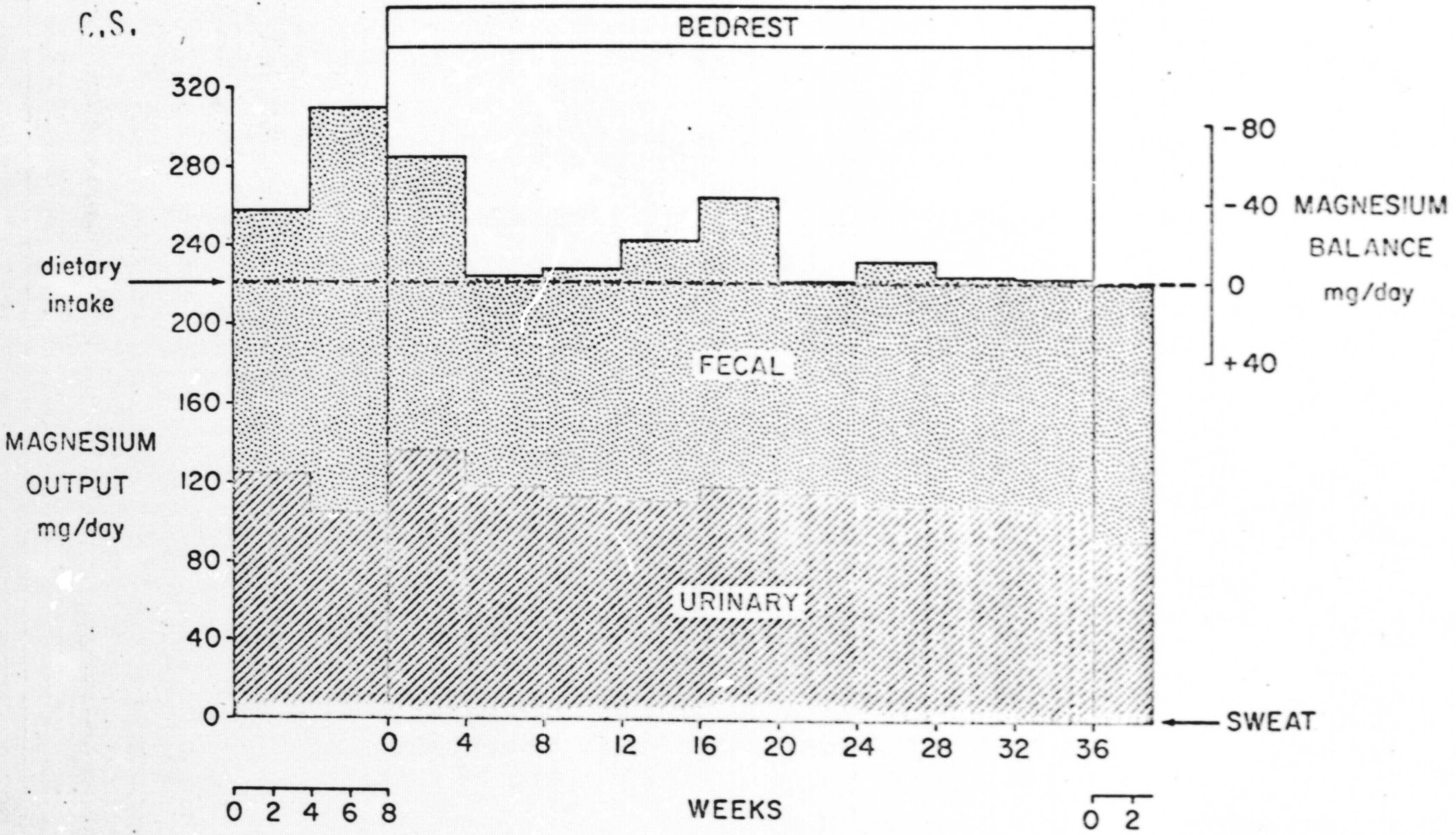
BEDREST
320
280
240
dietary intake
200
160
120
80
40
0
MAGNESIUM OUTPUT mg/day
FECAL
URINARY
-80
-40
0
+40
MAGNESIUM BALANCE mg/day
SWEAT
0 4 8 12 16 20 24 28 32 36
0 2 4 6 8
WEEKS
0 2

FIG. 30

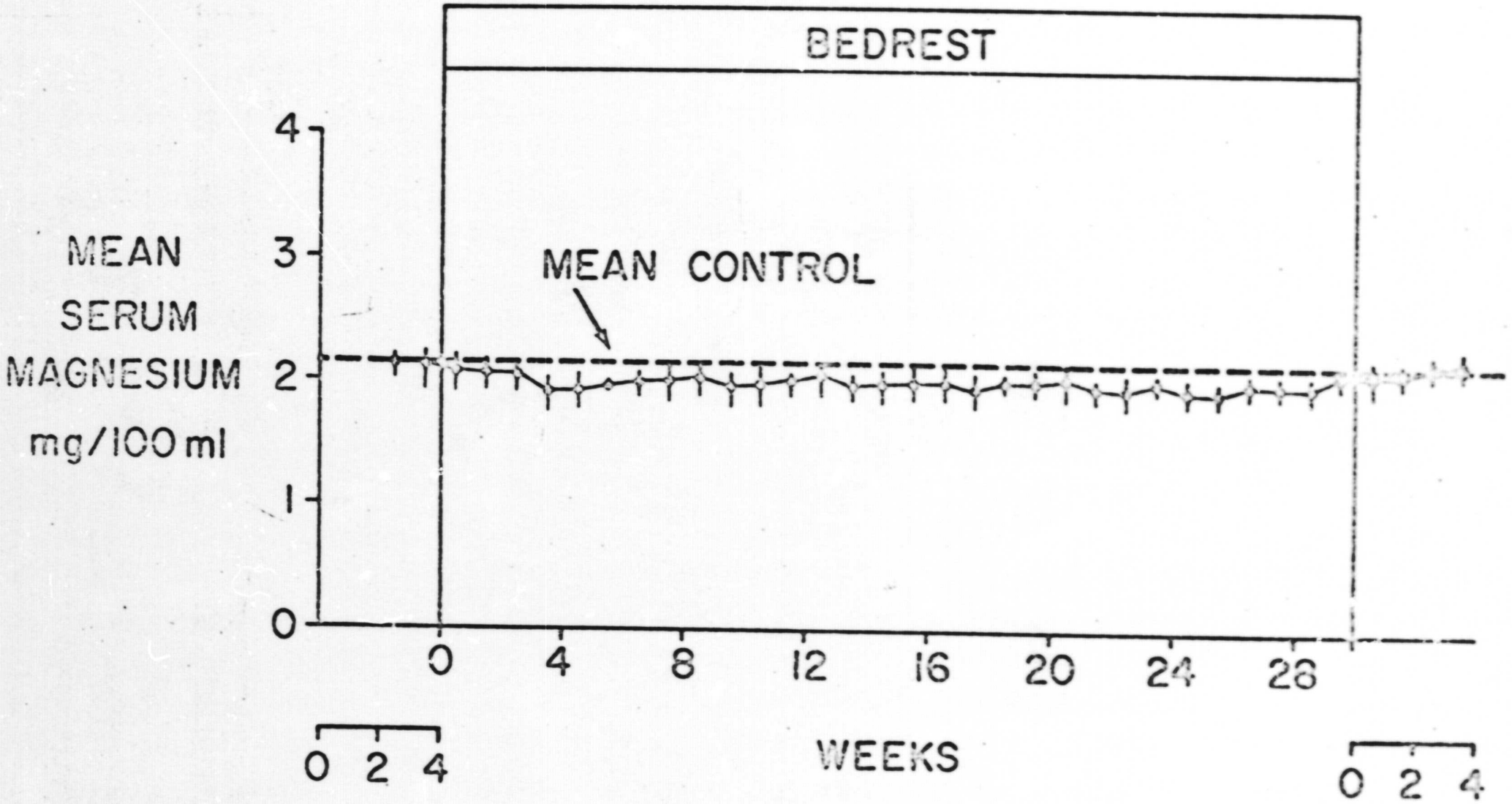
BEDREST
MEAN CONTROL
MEAN SERUM MAGNESIUM mg/100 ml
4
3
2
1
0
0
4
8
12
16
20
24
28
WEEKS
0 2 4
0 2 4

FIG. 31

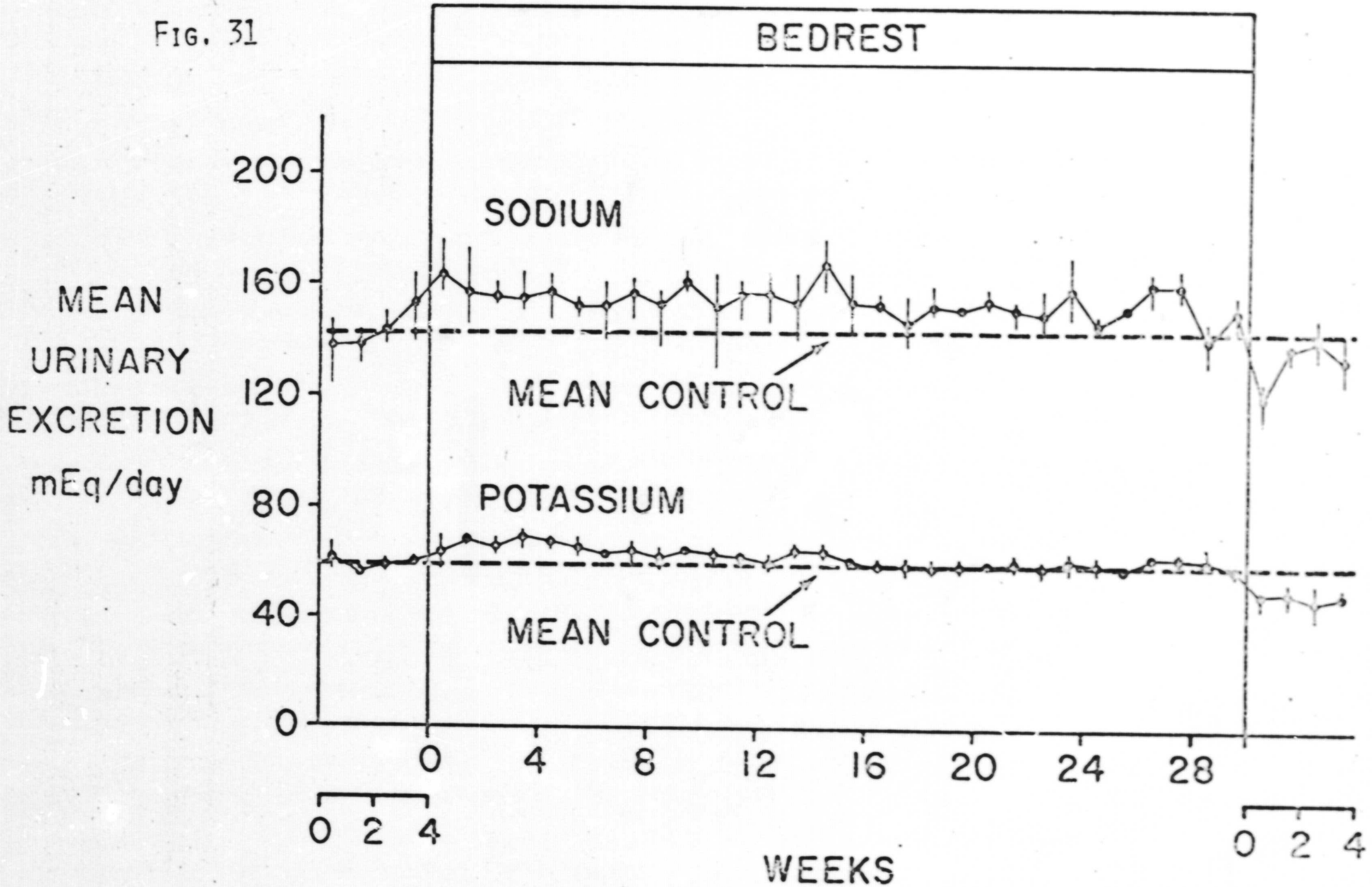
BEDREST
200
160
120
80
40
0
SODIUM
MEAN CONTROL
POTASSIUM
MEAN CONTROL
MEAN URINARY EXCRETION mEq/day
0 4 8 12 16 20 24 28
0 2 4
0 2 4
WEEKS

FIG. 32

R.R.

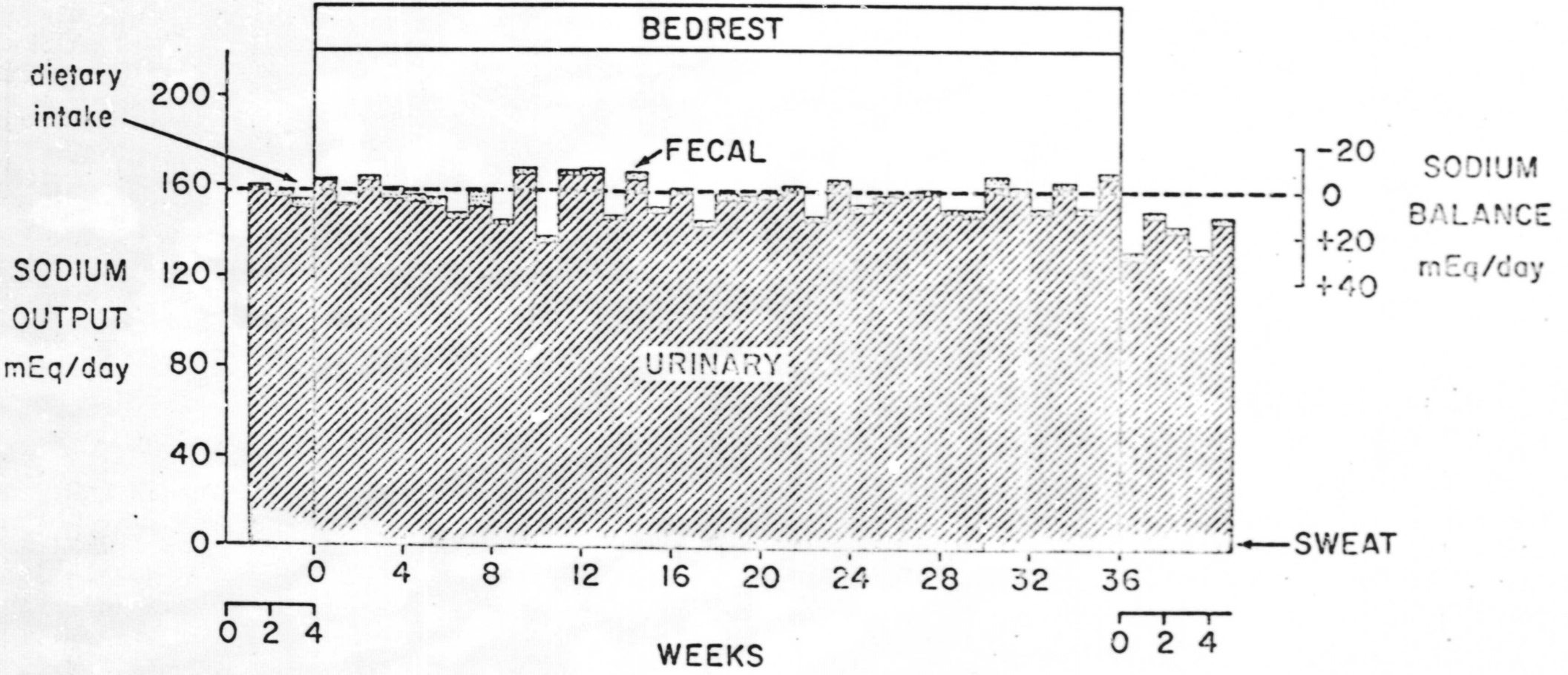
BEDREST
dietary intake
200
160
120
80
40
0
SODIUM OUTPUT mEq/day
FECAL
URINARY
SWEAT
-20
0
+20
+40
SODIUM BALANCE mEq/day
0 4 8 12 16 20 24 28 32 36
0 2 4
0 2 4
WEEKS

FIG. 33

G.B.

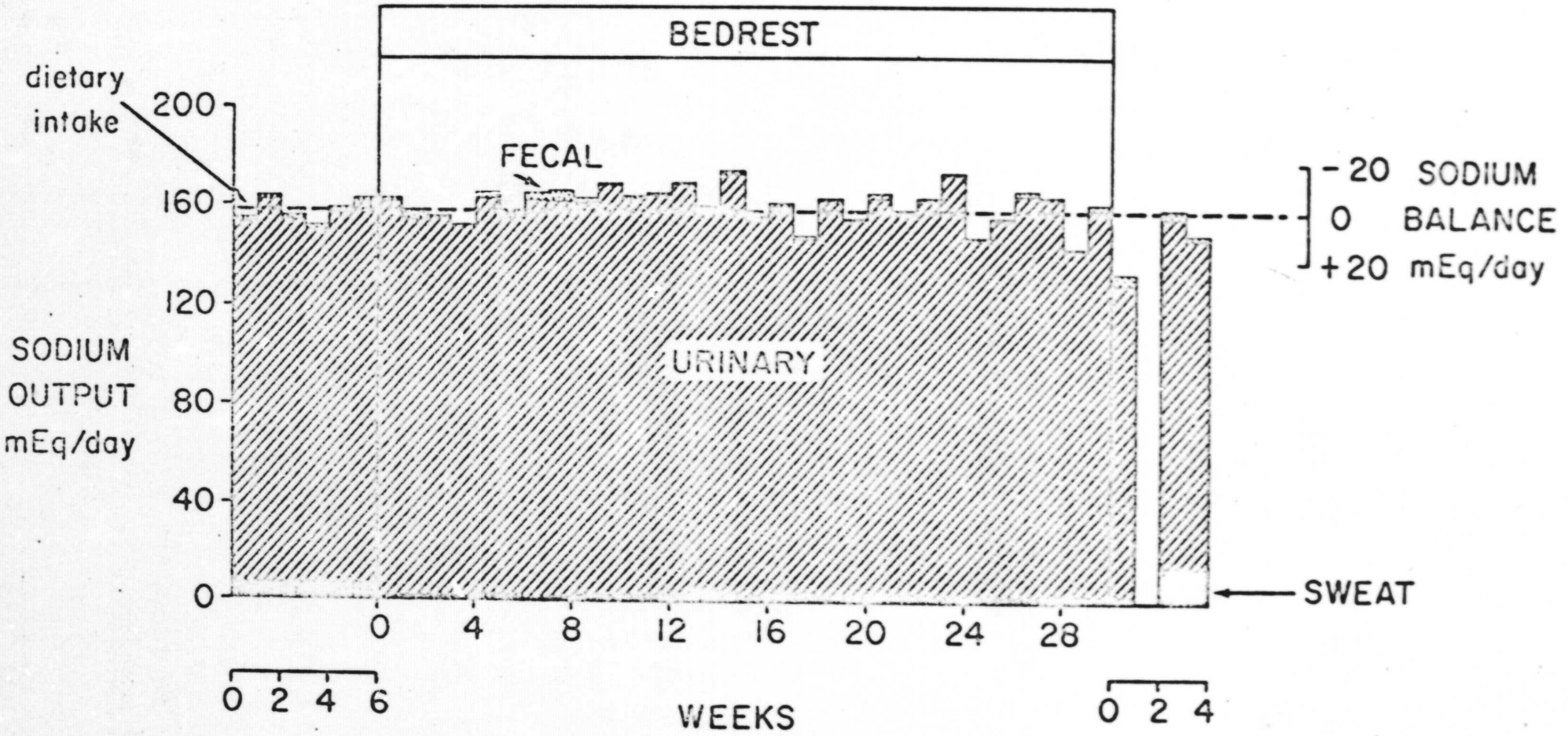
BEDREST
dietary intake
200
160
120
80
40
0
FECAL
URINARY
SODIUM OUTPUT mEq/day
-20 SODIUM
0 BALANCE
+20 mEq/day
SWEAT
0 4 8 12 16 20 24 28
0 2 4 6
0 2 4
WEEKS

FIG. 34

THE EFFECT OF PROLONGED BEDREST ON SODIUM BALANCE (C.S.)

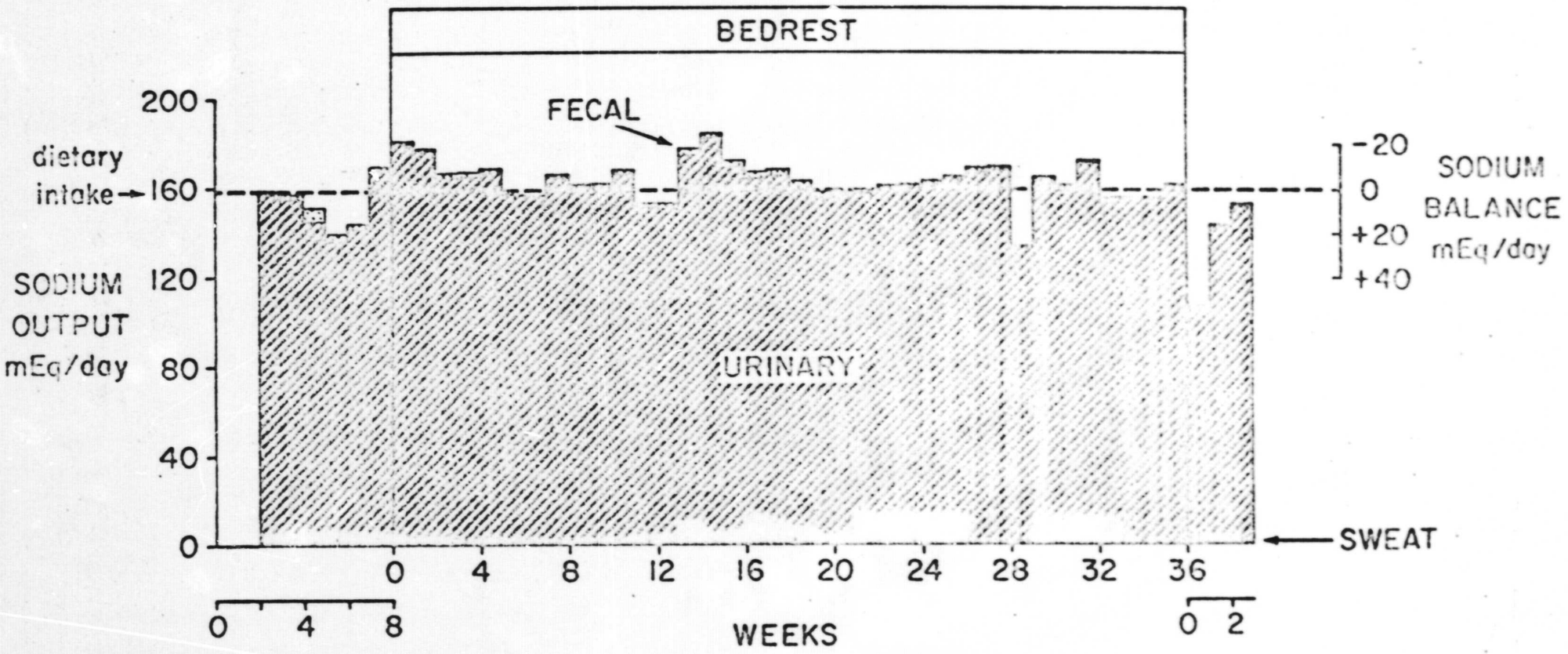

FIG. 35

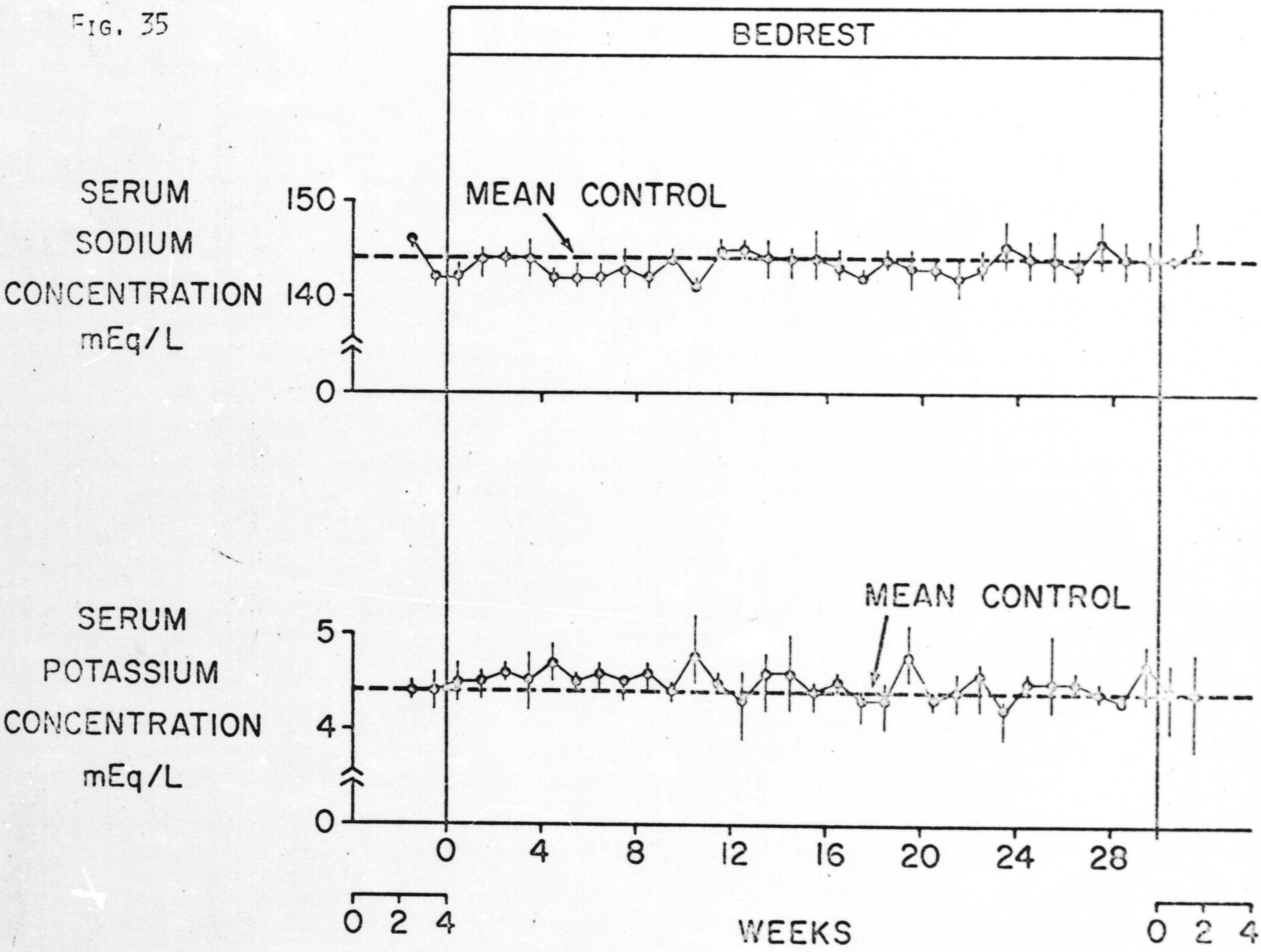
BEDREST
SERUM SODIUM CONCENTRATION mEq/L
150
140
0
MEAN CONTROL
SERUM POTASSIUM CONCENTRATION mEq/L
5
4
0
MEAN CONTROL
0 4 8 12 16 20 24 28
0 2 4
WEEKS
0 2 4

FIG. 36

R.R.

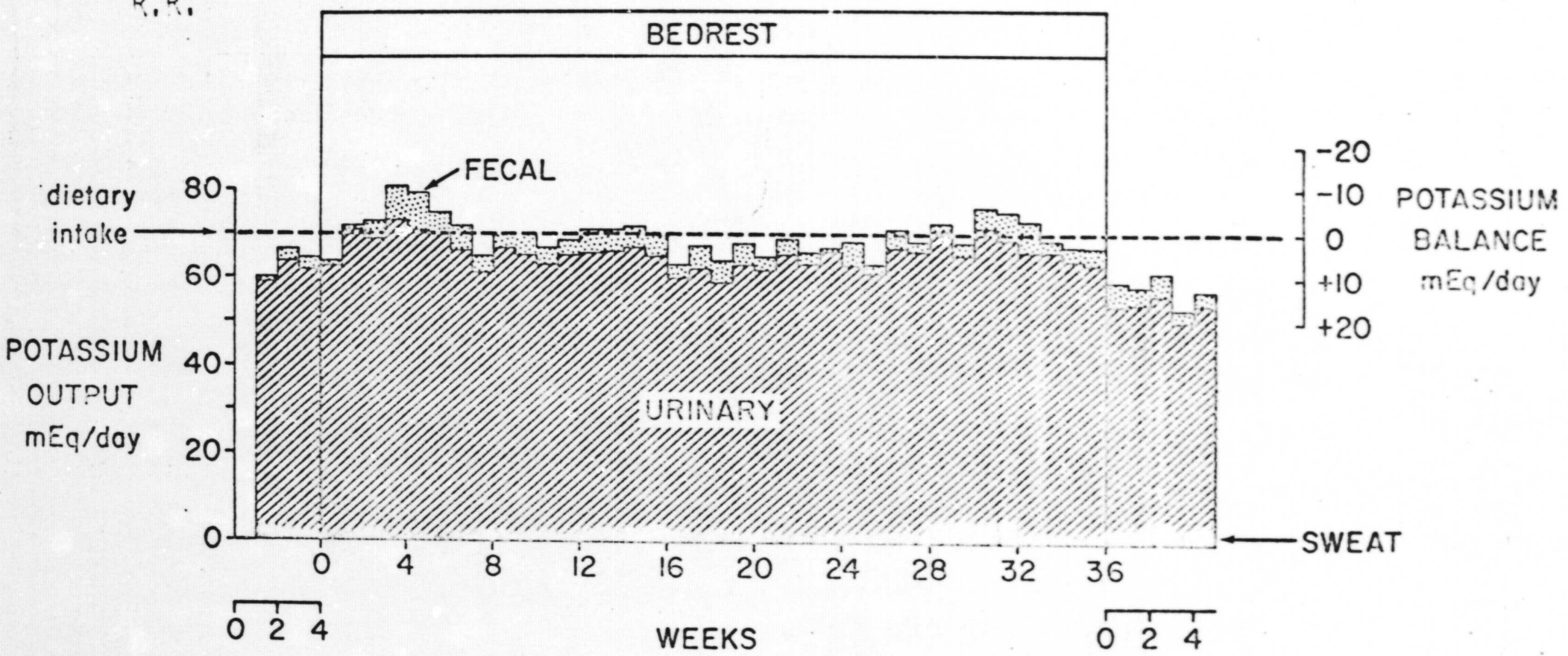
BEDREST
FECAL
dietary intake
80
60
40
20
0
POTASSIUM OUTPUT mEq/day
URINARY
SWEAT
-20
-10
0
+10
+20
POTASSIUM BALANCE mEq/day
0 4 8 12 16 20 24 28 32 36
0 2 4
WEEKS
0 2 4

FIG. 37

G.R.

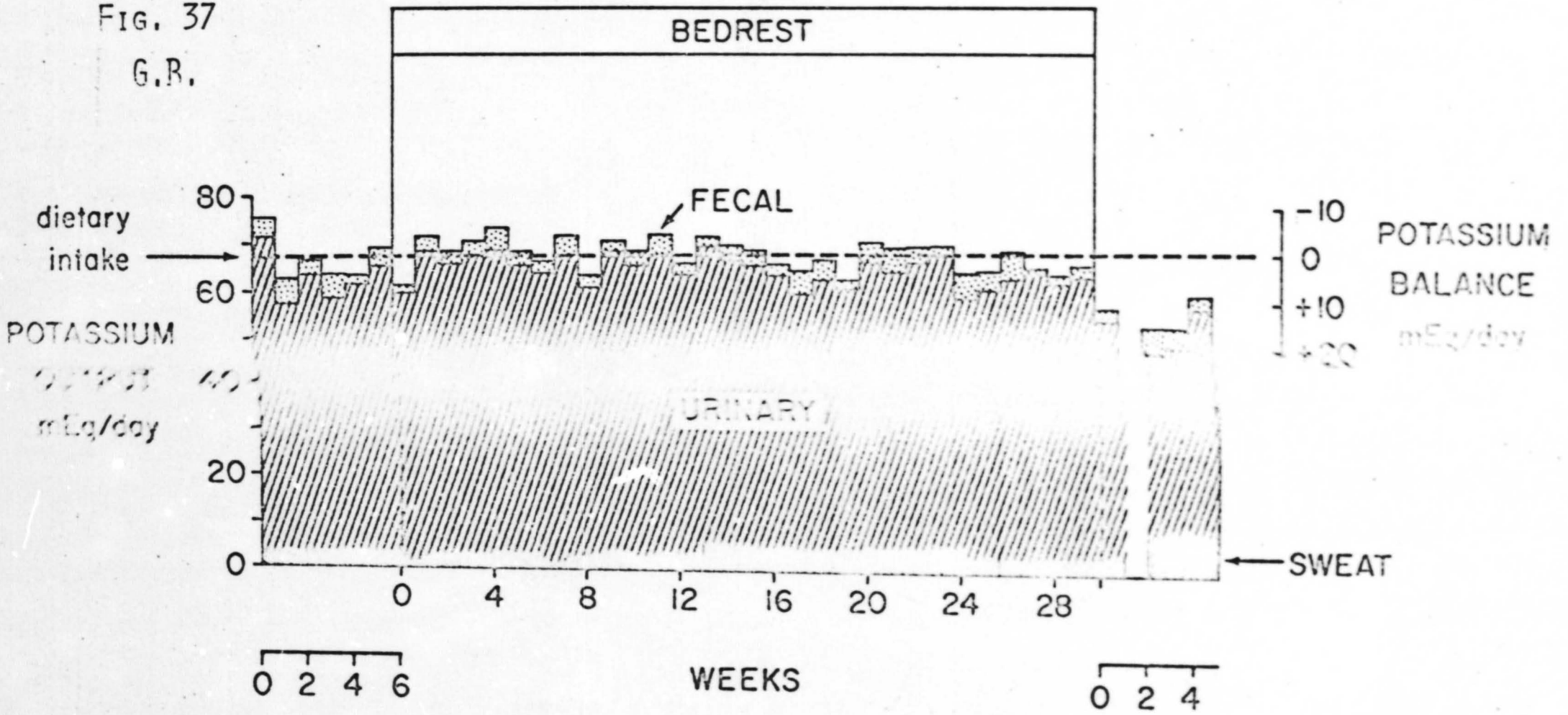
BEDREST
FECAL
dietary intake
POTASSIUM
mEq/day
80
60
20
0
URINARY
SWEAT
-10
0
+10
POTASSIUM BALANCE
mEq/day
0 4 8 12 16 20 24 28
WEEKS
0 2 4 6
0 2 4

FIG. 38

THE EFFECT OF PROLONGED BEDREST ON POTASSIUM BALANCE (C.S.)

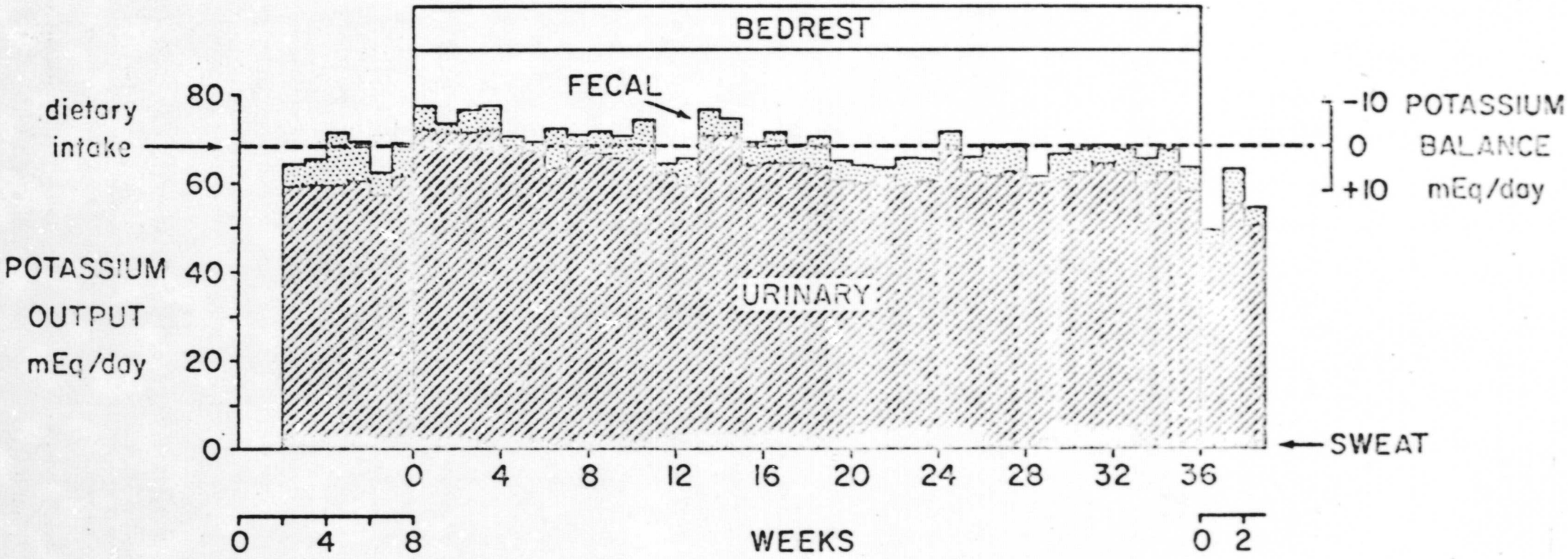

FIG. 39

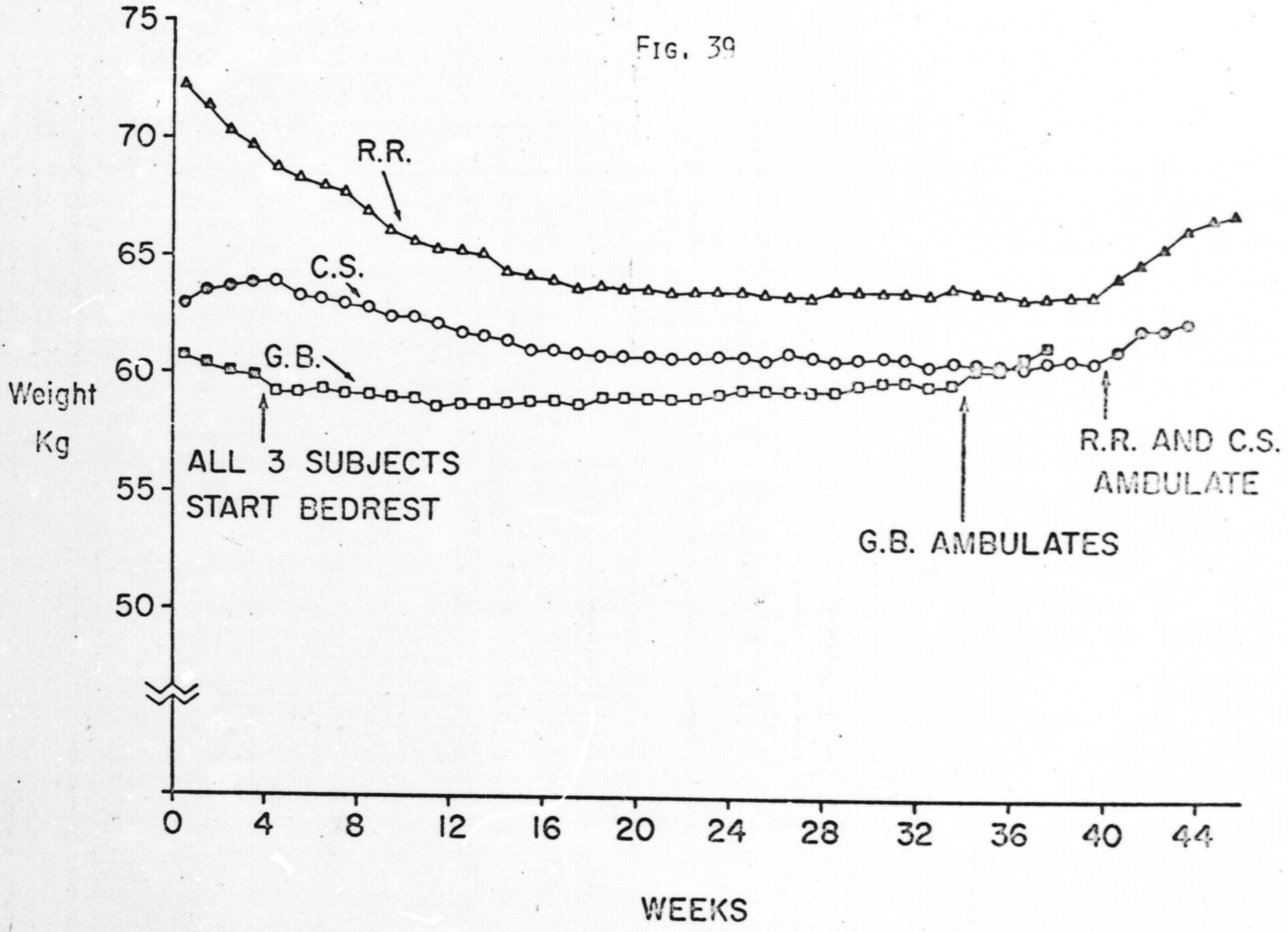
75
70
65
60
55
50
Weight
Kg
R.R.
C.S.
G.B.
ALL 3 SUBJECTS
START BEDREST
G.B. AMBULATES
R.R. AND C.S.
AMBULATE
0
4
8
12
16
20
24
28
32
36
40
44
WEEKS

FIG. 40

R.R.

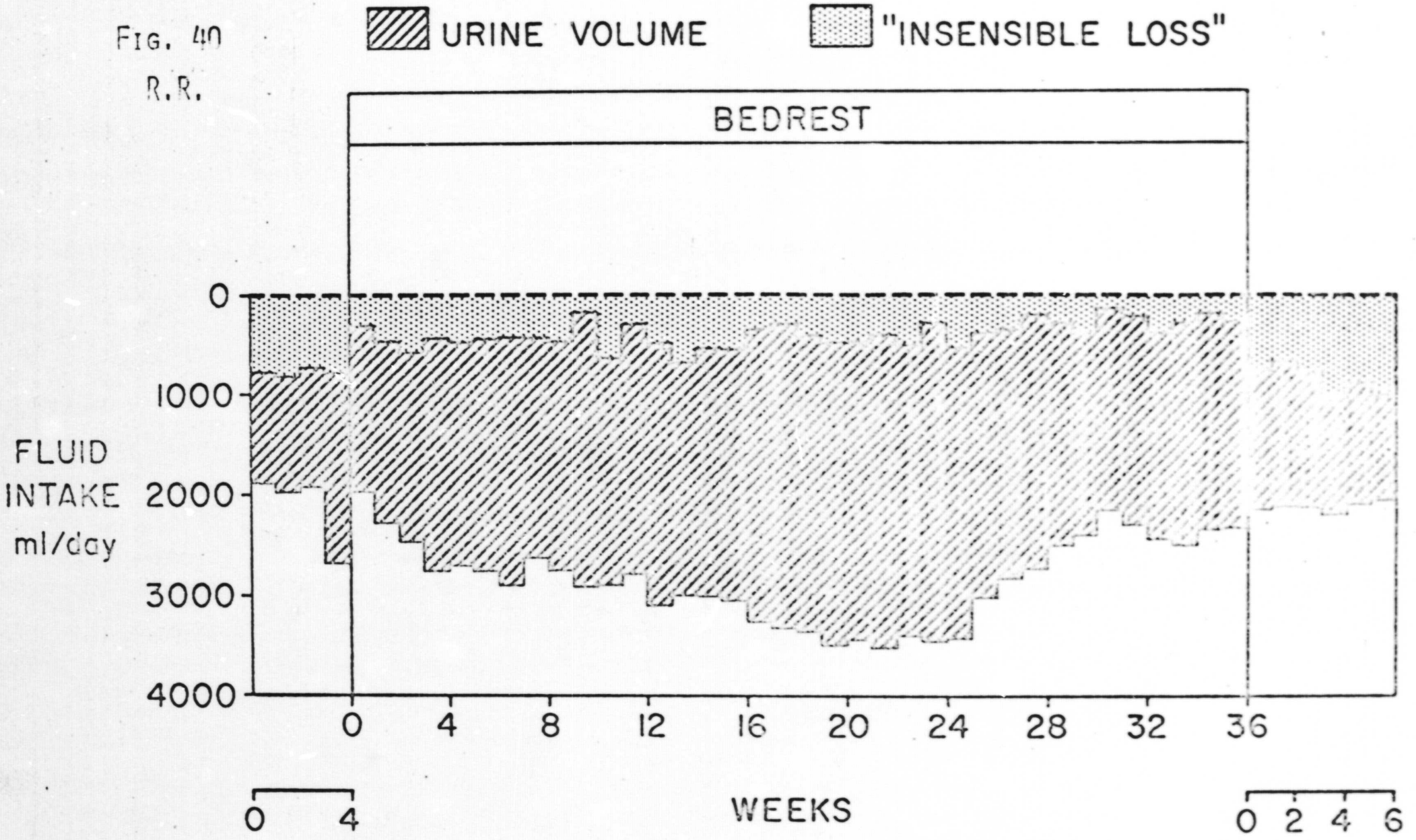
URINE VOLUME
"INSENSIBLE LOSS"
BEDREST
0
1000
2000
3000
4000
FLUID INTAKE ml/day
0 4 8 12 16 20 24 28 32 36
WEEKS
0 4
0 2 4 6

FIG. 41

G.B.

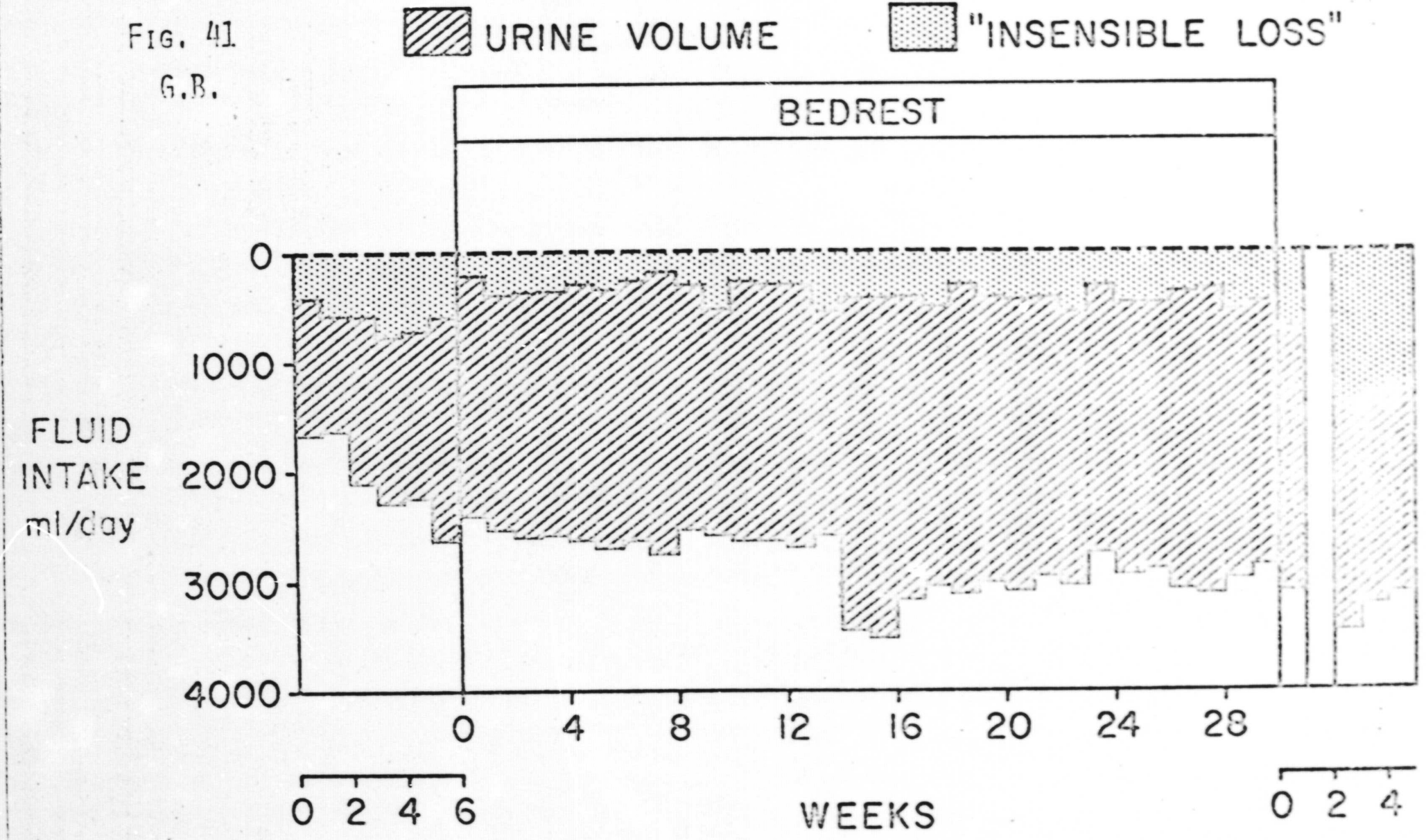
URINE VOLUME
"INSENSIBLE LOSS"
BEDREST
FLUID INTAKE ml/day
0
1000
2000
3000
4000
0 4 8 12 16 20 24 28
0 2 4 6
WEEKS
0 2 4

FIG. 42

C.S.

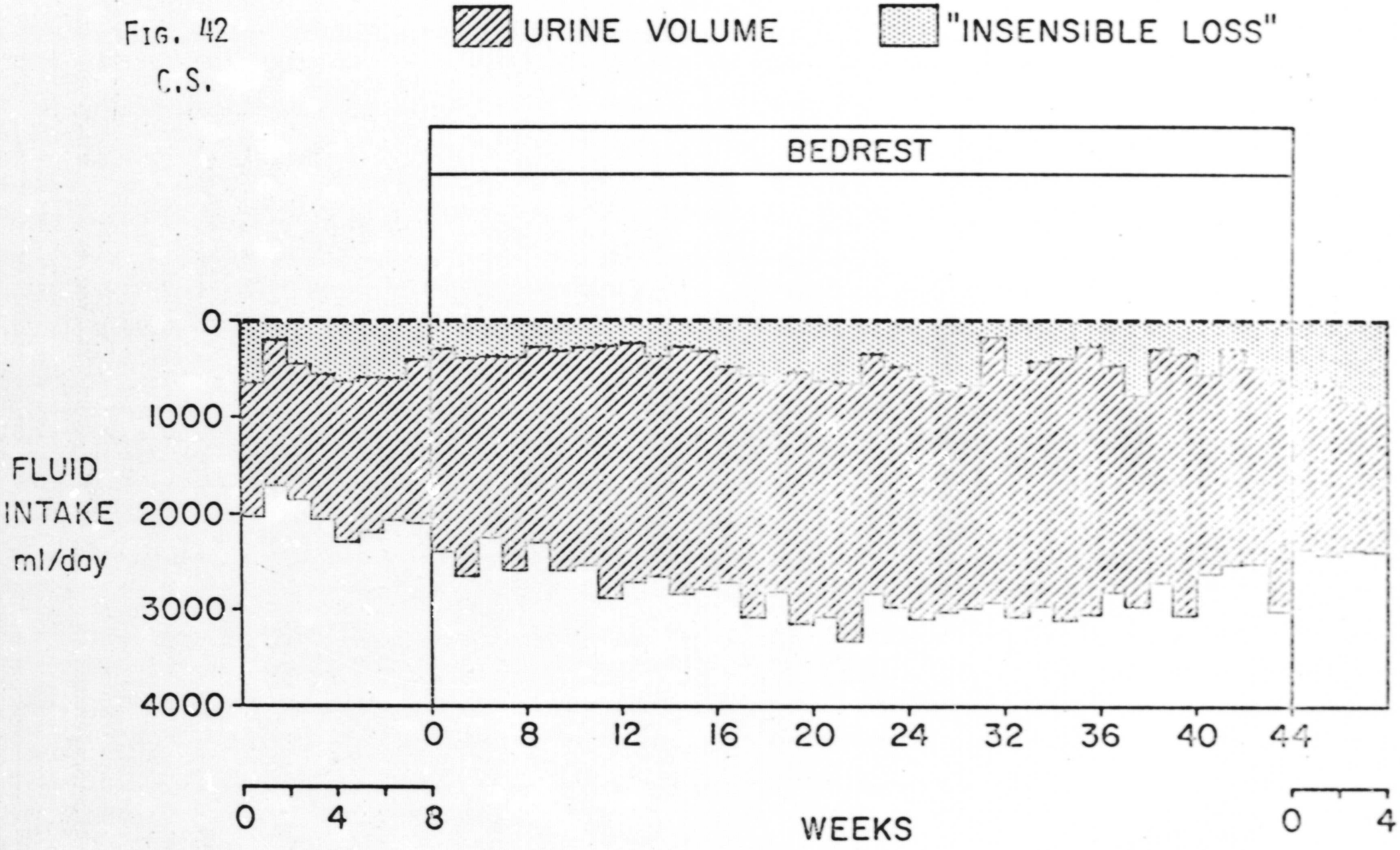
URINE VOLUME
"INSENSIBLE LOSS"
BEDREST
0
1000
2000
3000
4000
FLUID INTAKE ml/day
0
8
12
16
20
24
32
36
40
44
0
4
8
WEEKS
0
4

FIG. 43

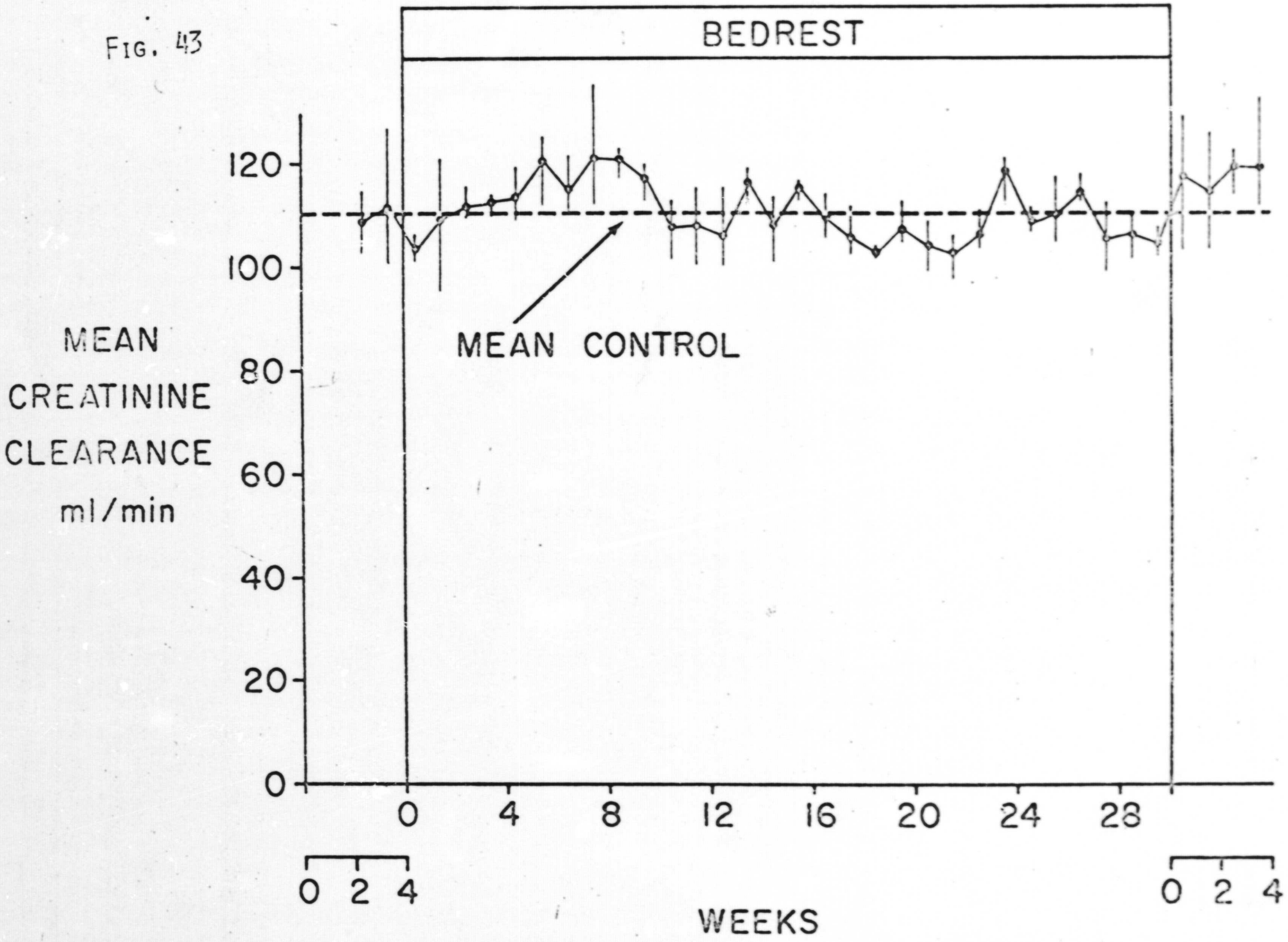
BEDREST
MEAN CONTROL
MEAN CREATININE CLEARANCE ml/min
120
100
80
60
40
20
0
0
4
8
12
16
20
24
28
0 2 4
0 2 4
WEEKS

FIG. 44

THE EFFECT OF PROLONGED BEDREST ON
MEAN RED BLOOD CELL VOLUME, PLASMA VOLUME, AND TOTAL BLOOD VOLUME
(3 SUBJECTS)

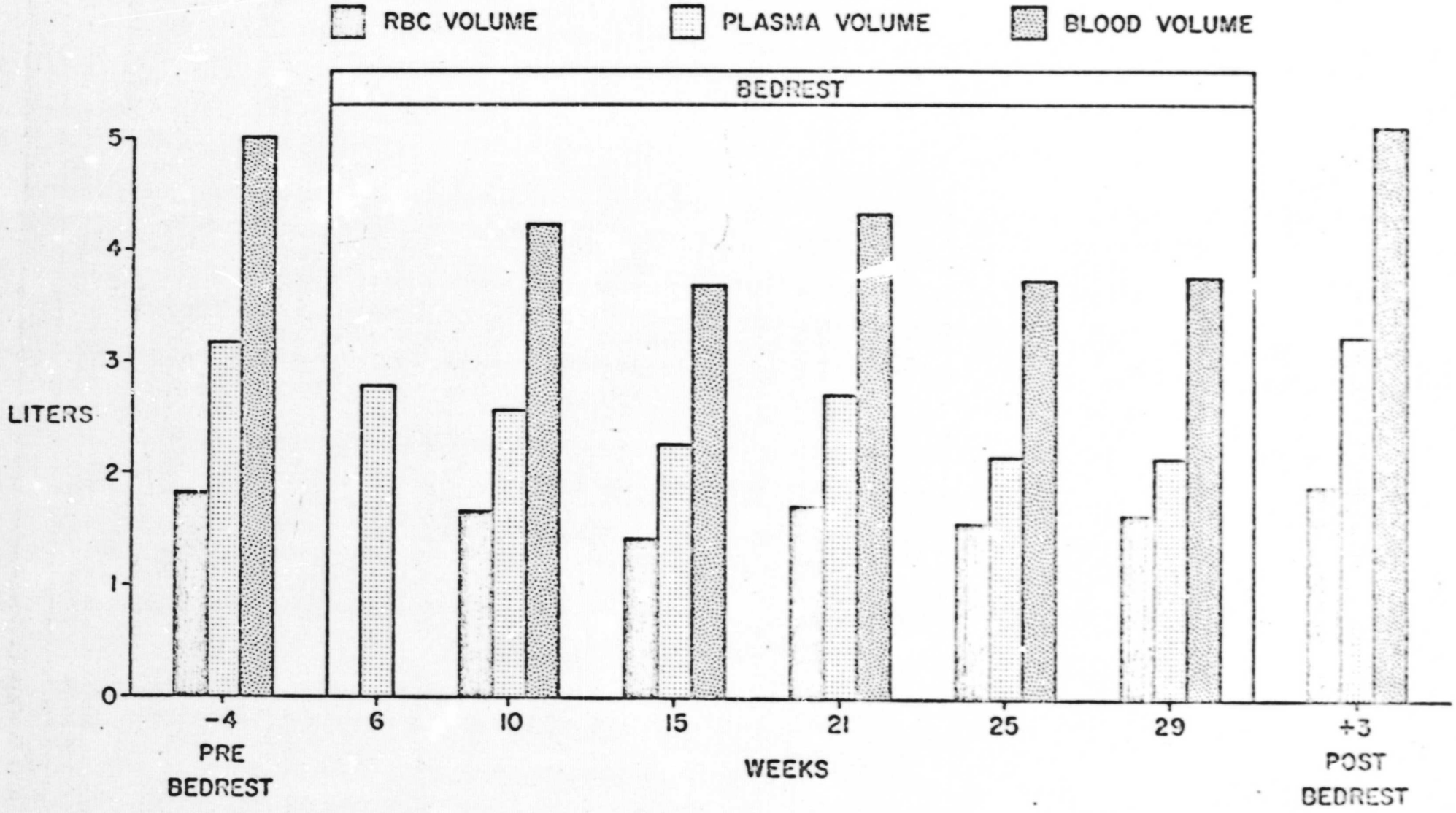

FIG. 45

THE EFFECT OF PROLONGED BEDREST ON EXTRACELLULAR FLUID VOLUME