days at least. In good agreement with the results obtained on experimental animals it is also apparent that there is not a complete adjustment immediately following a change in routine. The synchronization with a new time schedule also is apparent in the electrolyte excretion.

Summary. 1) Excretion of 17-hydroxycorticosteroids in the urine was measured at frequent intervals throughout 30 hours at 5 different times following a shift from CST to Japanese and Korean time. The excretion was synchronized with CST immediately after arrival in Japan, but became synchronized with the new time schedule by 9 days. 2) Sodium and potassium excretion patterns are similar to that of a previous study on Central Standard Time but synchronized with the new time schedule also in 9 days.

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Wound Healing: Glycoproteins of Wound Tissue. I. Studies of Hexosamine, Hexose, and Uronic Acids Content.* (24675)

M. R. SHETLAR, EUGENIA G. LACEFIELD, BETTY N. WHITE AND JOHN A. SCHILLING
Depts. of Surgery and Biochemistry, University of Oklahoma School of Medicine, Oklahoma City

The importance of connective tissue in wound healing is well recognized. However, little is known about the complicated sequence of events by which connective tissue is formed. Kao, Boucek, and Noble(1) recently reported studies of connective tissue formed following implantation of polyvinyl sponge. Robertson, *et al.*(2) found that following injection of carrageenin suspension into scorbutic guinea pigs, granulomatous repair tissue contained more hexosamine containing polysaccharide material than that formed under the same conditions by normal animals. Slack(3), using a similar suspension, found that increase in polysaccharide material in scorbutic guinea pig tissue was due to increase of hyaluronic-acid-like material, but that chondroitin sulfate decreased. Schilling *et al.*(4) have described a technic for study of wound fluid utilizing stainless steel cylinders which are implanted subcutaneously in animals. This procedure allows removal of numerous samples at various time intervals for studies of tissue and fluid components. The following describes serial studies of hexosamine, hexose, and uronic acid contents of wound tissue formed around such cylinders.

Methods. Stainless steel cylinders were implanted subcutaneously in dogs as previously described for guinea pigs(4). The cylinders used were of #46 mesh and were 1 cm in diameter by 5.5 cm long. Two lucite plastic rings 9.5 mm in diameter and approximately 3 mm thick were placed in each cylinder to keep it from collapsing during the healing process. At stated intervals the cylinders were removed, the fluid within cylinder was drawn off, and extraneous tissue was stripped from the outside of cylinder. The cylinder was opened by longitudinal scissor cut, and the tissue was lyophilyzed still attached to the cylinder. The dry tissue was stripped by hand from the stainless steel mesh and extracted in Soxhlet extractor for 24 hours with diethyl ether to remove lipid material. The extracted tissues were ground to pass through

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TABLE I. Uronic Acid, Hexosamine, and Hexose Contents* of Wound Repair Tissue.

Days after implantation	Avg wt of tissue, mg	Uronic acid	Hexosamine	Hexose
5	200	.57 ±.06	5.79 ±.12	20.9 ± .2
7	157	.72 ±.14	6.80 ±.10	19.4 ± .7
14	221	1.04 ±.14	7.07 ±.25	26.0 ±1.7
21	238	1.37 ±.21	6.19 ±.25	27.4 ±2.9
28	683	1.77 ±.15	7.60 ±.36	22.3 ± .3
28	316	2.35 ±.17	8.47 ±.16	30.3 ± .3
42	402	2.27 ±.09	7.32 ±.23	29.7 ±1.0
56	392	2.80 ±.33	6.73 ±.17	26.6 ± .7

* All results are based on analyses of 6 separate samples and calculated as mg/g of dry lipid-free material. Figures following ± sign are stand. errors.

a 60 mesh screen. Aliquots of ground material were used for chemical analyses. Uronic acid was determined by the carbazole method (5) following preliminary hydrolysis with Dowex 50 resin as previously described(6). A correction for the reaction of hexose with carbazole was made by determining hexose in each Dowex hydrolysate. Under conditions of carbazole reaction, 100 mg of hexose was equivalent to 7 mg of uronic acid. Hexosamine was determined by method modified from that of Boas(7). Hexose was determined by an anthrone method.

Results. Results of analytical studies are presented in Table I. It is readily apparent that the uronic acid (expressed as glucuronic acid) is relatively low at 7 days, increases to maximum level at about 28 days and remains relatively constant thereafter for at least 42 days. On the other hand, hexosamine and hexose levels are already comparatively high at 7 days and do not increase strikingly in later samples.

The definitely higher levels of hexosamine as compared to uronic acid (as determined by the carbazole reaction) levels are of interest. It appears that the polysaccharides which do not contain uronic acid are present in much greater amounts in wound healing tissue than are uronic acid containing mucopolysaccharides. This is particularly true of the early tissue. As verification of these data, preliminary studies of constituent monosaccharides of hydrolysates of wound tissue were performed. Wound tissue was hydrolyzed using Dowex 50 W × 8 as catalyst at 97-100° C. Under these conditions, 75-95% of the glucuronic acid of chondroitin sulfate was still present. These hydrolysates were concentrated under vacuum and subjected to paper electrophoresis in borate buffer. Definite, but weak, bands characteristic of uronic acid were found as compared to relatively strong bands of glucose, mannose, and galactose (Fig. 1). The uronic acid band was decidedly stronger in later sample. All or most of the glucose in the hydrolysates may be assumed present in the tissue as glycogen; however, galactose and mannose must be constituents of polysaccharides or glycoproteins which have not yet been identified.

Summary. A serial study of uronic acid, hexosamine, and hexose content of wound repair tissue was made. Uronic acid content was quite low in the 7-day sample, but increased until about 28 days. Hexosamine in considerable excess of uronic acid was found indicating presence of polysaccharides or glycoproteins which did not contain uronic acid.

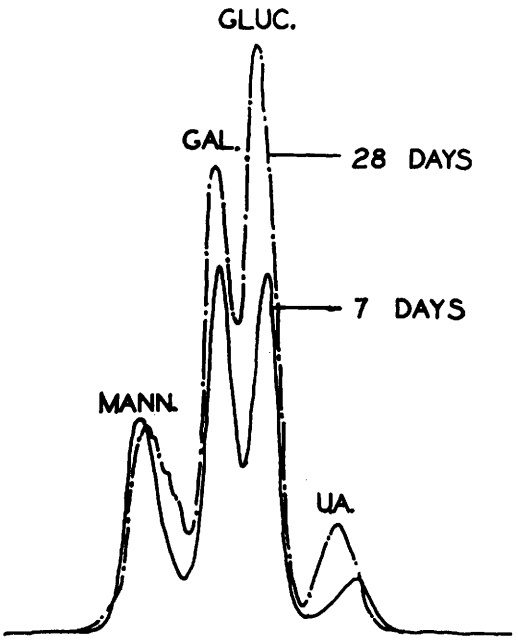

FIG. 1. Densitometer tracings of paper electrophoretic patterns of sugars in hydrolysates from wound repair tissue. Borate buffer .05 M, pH 9.2. Solid line, tissue at 7 days; broken line, tissue at 28 days. Peaks typical of mannose are indicated by "Mann."; galactose by "Gal."; glucose by "Gluc."; and uronic acid by "U.A.". Note increase in uronic acid in 28 day sample.

Sugars with electrophoretic mobility in borate buffer of galactose, mannose, and glucose were identified in hydrolysates of wound repair tissue.

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Acute Effect of Psychologic Stimuli upon Plasma Non-Esterified Fatty Acid Level.* (24676)

MORTON D. BOGDONOFF, E. HARVEY ESTES, JR. AND DAVID TROUT
(Introduced by E. A. Stead)
Dept. of Medicine, Duke University Medical Center, and Vet. Admin. Hospital, Durham, N. C.

Several recent studies have pointed up the important role of non-esterified fatty acids (NEFA) as major transport form of plasma lipid(1,2,3). This segment of plasma lipid undergoes rapid turnover (half-life of approximately 2 minutes)(3), and is promptly altered by administration of such substances as glucose(1,2,6), glucagon(5), insulin(4,6), epinephrine(1,2), and heparin(2). Lability of this lipid fraction provided an opportunity to study the relationship of fat metabolism to acute alterations in affect state within the structure of a short-term, controlled experiment. Other serum lipid fractions varied in response to changes in life-situational patterns, but these studies were usually conducted over long periods of time(7,8,9). The results herein reported clearly indicate that acute alterations in emotional arousal had a distinct effect upon fat homeostasis.

Methods and procedures. Four volunteer subjects (medical students) were each asked to report in early morning in fasting state. They were informed that only a single needle was to be inserted and that samples were to be drawn during sleeping and waking periods during the morning. #18 Cournand needle was placed in antecubital vein under procaine anaesthesia, and the first sample drawn about 10 minutes later. The room was darkened; the subject was asked to relax completely and to sleep if possible. Samples were drawn at approximately 30 minute intervals for one to 2 hours. Then the lights were turned on and a non-directed, light conversation was carried on between various persons. After 15 to 30 minutes, blood was again withdrawn, and all experimenters except one, excused themselves for various reasons. The remaining experimenter engaged the subject in casual conversation, which was quickly focused upon areas of personal conflict, anxiety, anger, and hostility, uncovered during the interview. All subjects became more or less heatedly involved in conversation, all becoming restless, showing changes in facial expression and some changes in skin color. Samples were obtained during this period by another member of the experimental team, who entered only for the period required for obtaining the sample. The conversation was terminated by reentry of other members of the team, who continued with a superficial non-directed conversation for another 15 to 30 minutes, after which the final sample was drawn and the needle withdrawn. Samples were withdrawn into heparinized syringes, which were placed in ice and immediately taken to the analytical laboratory where centrifugation was quickly carried out and the plasma measured into the extraction mixture. NEFA determinations were carried out by the

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