ANNALS OF SURGERY

Vol. 182 August 1975 No. 2

Effect of Nutrition, Diet and Suture Material on Long Term Wound Healing

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Although it is known that malnutrition hinders early wound healing, it has not been determined whether this occurs because of formation of a poor scar or a slow rate of normal healing; the ultimate fate of the malnourished wound is unknown. Malnutrition was produced in rats by short gut syndrome. Elemental diet was compared to rat chow and silk was compared with polyglycolic acid suture. Nutritional deficiency was seen in short gut rats for two weeks postoperatively. Thereafter adaptation allowed partial recovery, but relative deficiency persisted. Morbidity and mortality of short gut rats doubled that of controls and all wound complications were limited to this group, occurring within the first two weeks. Malnourished animals surviving for 60 days had wound strength equal to the control rats as determined by gut anastomosis bursting strength, skin wound breaking strength and wound hydroxyproline content. Neither diet nor suture material altered ultimate wound strength. Improved nutrition allowed more animals and wounds to survive, but ultimate healing in survivors was indistinguishable from that of normal controls. Thus early weakness probably results from slow healing rather than formation of poor scar. Nutrition plays an important role in early strength and survival, but not in ultimate wound healing.

RELATIVELY LITTLE is known about the effects of nutritional deficiency on long term wound healing. Koback et al.[4] have shown a three day increase in the lag phase in protein deficient animals, using bursting pressure of abdominal wounds in rats. This measurement does not allow determination of wound strength beyond 12 days. More recent work on the effect of protein depletion on healing[6,10] shows that even at 21 days there is a significant difference in wound strength of malnourished animals. These findings confirm the clinical observation of a higher rate of anastomotic leak and wound dehiscence in malnourished patients. Such complications occur early in the course of healing and it is rare for wounds to disrupt once healed, except in specific deficiency states such as scurvy. This suggests that wounds in surviving malnourished patients eventually gain sufficient strength for normal function, but does not allow relative comparison of strength with normal wounds.

The ultimate fate of the weaker wounds remains unknown and the cause of early weakness, observed both clinically and experimentally, has not been determined. Wounds in malnourished animals could be weaker because a structurally inferior scar is made or because normal wound healing progresses at a slower rate, presumably due to a shortage of building blocks. By examining wounds at a late stage of healing, the effects of retardation could be eliminated and a definitive answer re-

Submitted for publication February 12, 1975.

Partly supported by a grant from the Medical Research Council of Canada.

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garding relative wound strength could be reached, based solely on the relative structural quality of the scar. Levenson[5] has shown that, in the rat, 85% of the final wound strength has been achieved by 2 months. Comparing wounds at this time should enable elimination of the effects of slower healing and the comparison should very closely reflect the ultimate wound strength.

During an earlier nutritional study in the short gut syndrome[8] we created a model where nutritional depletion could be acutely produced and maintained for some 2 weeks. At the end of this time gut adaptation allowed slow recovery, thus preventing mortality from continued malnutrition; the gain in these rats, however, never approached that of controls and therefore they remained in a relative state of malnutrition when compared to the controls at all times. The purpose of the present study is to use this model to examine the effects of such sustained malnutrition on the long term effects of wound healing in order to determine whether structurally inferior scars or a slower rate of wound healing accounts for the early weakness in wounds of malnourished animals. Because we have had a clinical impression that elemental diet promotes better wound healing we have also examined the effect of this on the long term wound. Finally, because of interest in the new suture material polyglycolic acid (PGA), with evidence suggesting its superiority in the earlier stages of wound healing,[3] we decided to incorporate into our study an assessment of its contribution to the ultimate wound strength as compared to silk.

Method

Sixty mature Sprague-Dawley rats of comparable age and weight were subjected to laparotomy. An 85% enterectomy or 2 cm biopsy was randomly carried out, followed by an end-to-end everted anastomosis using a single layer of interrupted 5-0 silk sutures. The linea alba was closed with 2-0 silk and the skin closed with 4-0 silk for half its length and 4-0 PGA (Dexon), alternating the suture materials randomly between the top and bottom half of the wound. Both resected and biopsied animals were randomly allocated to be fed either standard rat chow or elemental diet (Flexical, Mead Johnson Co. of Canada, Ltd.) postoperatively. The nutritional specifications as well as details of the model have been outlined elsewhere.[8] Weights were recorded at weekly intervals and all animals sacrificed after 60 days. Hemoglobin and serum albumin concentrations were determined at the time of the original operation and at sacrifice.

A 10 cm segment of gut, with the anastomosis in the center, was removed, ligated and distended with air through an apparatus attached to a mercury manometer. The pressure of bursting was noted and was recorded as occurring either above or below 300 mm Hg. The abdomen was shaved and any remaining sutures were removed. The midline skin wound was excised with 3 cm of skin to either side of the scar. Wound breaking strength was measured on strips of uniform width according to the method of Crawford et al.[1] In 10 randomly selected animals separate strips were measured from the top and bottom half of the scar to determine the effect of PGA and silk on wound strength. In all other specimens a strip from the middle was used. After breaking, a 1 mm margin of skin containing the scar was submitted for hydroxyproline determination by acid hydrolysis.

Results

Nutritional

Nine of the resected animals and 5 of the biopsied animals died before the end of the experiment. Nonlethal complications occurred in 8 resected and 4 biopsied rats. When deaths and complications due to technical reasons were excluded, the incidence of both still remained twice as high in the resected animals. One ventral hernia and one superficial wound infection were seen at the time of sacrifice; both occurred in enterectomized rats. Three of the deaths were related to intra-abdominal abscesses communicating with anastomotic leaks; these were again seen in the enterectomized animals only, occurring at the 4th, 5th and 14th postoperative day.

Weight changes are shown in Fig. 1. The operation caused significant weight loss in both groups, greater in the resected animals. Whereas the biopsied animals started to regain their weight immediately, reaching the preoperative weight after the first week, the enterectomized animals did not start to gain any significant weight until the 3rd postoperative week and reached their preoperative weight in the 5th postoperative week. From the 5th week on, the curves are parallel with no indication that the short gut rats will reach the weight of their biopsied peers. At the conclusion of the experiment the biopsied animals weighed 14% more than the resected ones (418 gms versus 367 gms, $P<0.001$).

Preoperative albumin values were comparable for both groups. At the conclusion a small and statistically insignificant drop had occurred in both groups, the value being 2.1 in the biopsied animals and 1.9 in the resected one ($P>0.05$).

Preoperative hemoglobin was 17.3 gms per cent for all rats with no difference between groups. At sacrifice the value was 17.7 for the biopsied rats and 16.7 for the resected ones. Neither differences between the groups nor between preoperative values were statistically significant ($P>0.05$).

Dietary manipulation alone seemed to have no significant effect on these nutritional parameters. The weight, albumin and hemoglobin values for rats fed chow were

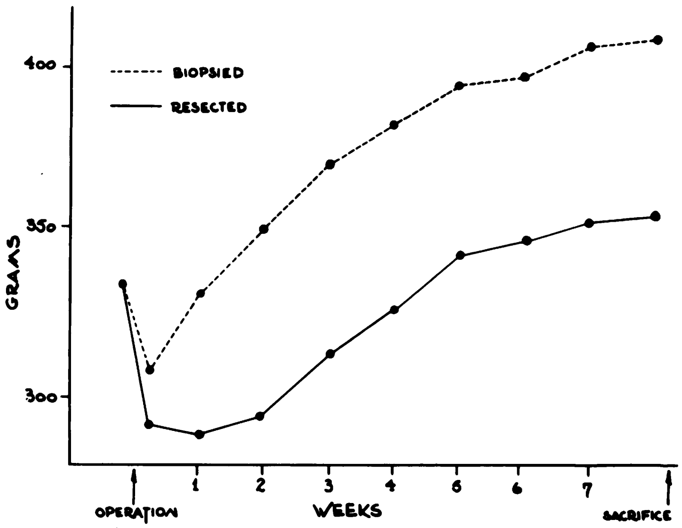

FIG. 1. Weight curves for the rats during the experiment. The preoperative weights of both groups of rats were indistinguishable. A weight loss occurred with the operation in both groups, twice as much weight being lost following enterectomy than after biopsy. Biopsied rats showed an immediate recovery with regain of lost weight by the end of the first postoperative week. A slow and steady gain was seen thereafter. Short gut rats continued to lose weight after the operation and were unable to show any significant weight gain until the third postoperative week. Preoperative weight was recovered in the fifth postoperative week and a slow gain was seen thereafter. This gain seemed to level off and did not indicate a tendency to catch up to that of the control group. The differences between the two groups were statistically significant (P<0.001) for all postoperative measurements. At the end of the experiment there was a 14% weight difference between the two groups; the biopsied rats had exceeded their preoperative weight by 20% and the short gut rats by 9%.

383 gms, 2.1 gms per cent and 17.5 gms per cent respectively, and these same values for rats fed elemental diet were 390 gms, 1.8 gms per cent and 17.1 gms per cent, respectively. When the effect of diet was analyzed separately, the following was noted: 1) Biopsied rats gain more weight with elemental diet; 2) Resected rats gain less weight with elemental diet; 3) The albumin level is significantly higher at sacrifice in biopsied rats fed elemental diet; 4) The hemoglobin value is significantly lower at sacrifice in resected rats fed elemental diet.

Wound Healing

Bursting pressure of the healed anastomosis exceeded 300 mm Hg in 79% of the biopsied rats and 67% of those with short gut syndrome. This determination was 63% and 84% for rats fed chow and elemental diet, respectively. When the effect of diet upon each operative manipulation was analyzed, this parameter was 64% for biopsied rats fed chow, 94% for those fed elemental diet, 60% for resected rats fed chow and 73% for resected rats fed elemental diet. None of these differences in bursting strength were statistically significant. Bursting was never noted to occur at the anastomosis, but always immediately proximal or distal to it.

TABLE 1. *Relation of Nutritional State to Ultimate Wound Healing.*

	Anastomotic Bursting Strength*	Wound Breaking Strength	Wound Hydroxyproline Content**
Biopsied	79	1674	1.5
Resected	67	1500	1.6
P	>0.05	>0.05	>0.05

*Per cent bursting above 300 mm Hg.
**mg hydroxyproline per 100 mg tissue

The breaking strength of the healed abdominal skin wound was 1674 gms for the biopsied animals and 1500 gms for the resected animals. Rats fed chow had a wound breaking strength of 1588 gms and those fed elemental diet had a breaking strength of 1690 gms. The average skin wound breaking strength was 1615 gms for biopsied rats fed chow, 1715 gms for biopsied rats fed elemental diet, 1553 for resected rats fed chow and 1655 gms for resected rats fed elemental diet. The breaking strength of wounds repaired with silk was 1441 gms and those repaired with PGA was 1575 gms. There was no statistically significant difference between these breaking strengths in any of the groups above. The breaking point was very definite and always occurred at the wound.

The average hydroxyproline content of the abdominal skin wound, expressed as mg per 100 mg tissue, was 1.5 for the biopsied rats and 1.6 for those with the short gut syndrome. This value was 1.6 for both rats fed chow and those fed elemental diet. The average wound hydroxyproline content was 1.6 for biopsied rats fed chow, 1.5 for biopsied rats fed diet, 1.6 for resected rats fed chow and 1.7 for resected rats fed diet. The hydroxyproline content for wounds closed with silk was 1.7 and for those closed with Dexon it was 1.8. There was no statistically significant difference between any of the hydroxyproline contents in the above groups.

Tables 1, 2 and 3 summarize some of the wound healing results.

Discussion

The nutritional data from this experiment closely parallel and confirm our earlier data investigating the use of elemental diet in this model.[8] Severe nutritional depletion persisted for 2 weeks in rats after 85% enterectomy. Whereas control rats started to gain weight on the first postoperative day, having regained all their lost weight

TABLE 2. *Relation of Diet to Ultimate Wound Healing.*

	Anastomotic Bursting Strength*	Wound Breaking Strength	Wound Hydroxyproline Content**
Rat Chow	63	1588	1.6
Elemental Diet	84	1690	1.6
P	>0.05	>0.05	>0.05

*Per cent bursting above 300 mg Hg.
**mg hydroxyproline per 100 mg tissue

TABLE 3. *Relation of Suture Material to Ultimate Wound Healing.*

	Wound Breaking Strength	Wound Hydroxyproline Content*
Silk	1441	1.7
PGA	1575	1.8
P	>0.05	>0.05

*mg hydroxyproline per 100 mg tissue

by the end of the first postoperative week, the resected ones lost more weight and did not begin to gain significantly until the third week postoperatively. Both clinical and experimental data have shown that healing wounds are weakest during this time, as confirmed in our experiment where all anastomotic breakdowns occurred during this time. All wound complications, anastomotic, fascial (ventral hernia) and skin (infection), occurred in the resected group. After the third week sufficient adaptation had occurred that these rats began to gain weight, although slower than their biopsied controls, regaining their preoperative weight by the 5th week after operation. Then a small rate of weight gain continued to occur, never at a rate to enable them to catch up with their biopsied peers.

Thus, although it would be incorrect to say that the malnutrition of the first two weeks had persisted, a relative malnutrition still existed because these rats were not able to catch up to their controls. Average hemoglobin and albumin levels were also lower in the short gut rats and although these differences were not statistically significant, the fact that all nutritional parameters showed a uniform direction of change makes it reasonable to suppose that the observed changes reflected real differences. The increased morbidity and mortality, twice that seen in the control rats, again indicates a nutritional deficiency in the resected group. When the effects of nutritional state and diet are simultaneously analyzed, it seems pertinent that the only significant drop in hemoglobin concentration occurs in one of the resected groups and the only significant rise in albumin concentration occurs in one of the biopsied groups. Although only concentrations of albumin and hemoglobin were measured, experimental data using a nutritionally depleted rat model,[2] as well as our own clinical experience[7,9] have shown that in states of malnutrition hemoglobin and albumin deficiency is even more marked if total plasma content rather than concentration is measured. Concentration differences which seem insignificant can sometimes reflect marked and significant differences in total content.[7]

The parameters of wound healing, anastomotic bursting strength, skin wound breaking strength and skin wound hydroxyproline content, show no difference in the scars at 60 days between animals subjected to small bowel biopsy and those given prolonged malnutrition from the short gut syndrome. The distribution of wound breakdown during the early stages of healing shows a definite weakness in the nutritionally depleted group. In surviving animals, this weakness has disappeared at 60 days, a time when 85% of the ultimate wound strength in the rat is achieved. This suggests that the weakness seen early in nutritionally depleted animals depends on a slower rate of wound healing rather than the formation of a structurally inferior scar. It seems that malnutrition leaves an animal with a shortage of building blocks for wound healing and that this shortage in turn limits the rate at which healing can take place. However, provided the animal can survive his disease and enough time is allowed, he will eventually go on to form a wound of normal strength. These data further suggest that wound healing takes precedence over other aspects of recovery in the malnourished animal, in as far as a normal wound had formed when other nutritional parameters were still less than normal.

A clinical impression that elemental diet promotes better wound healing than normal food has not been borne out by this experiment. It may be that a stronger wound could have been detected in elemental diet fed animals during the earlier stages of healing, but ultimate wound strength is the same regardless of diet used. As the clinical impression was gained from experience with malnourished patients, many of whom were unable to tolerate normal food, the comparison may well have been invalid. In such a situation elemental diet fed people will be provided with nutrition whereas other "similar" patients will be unable to avail themselves of any nutrition. In such a case, any difference in wound healing does not really reflect a difference of diet but rather a difference of nutrition or its lack.

Choice of suture material, whether absorbable or nonabsorbable, does not seem to effect the eventual outcome of wound healing. This suggests that choice of suture material should be predicated on considerations other than the requirement of ultimate wound strength. Thus, in rapidly healing exposed areas, such as the face, the cosmetic effect of the suture material should be of overriding importance. In contaminated wounds the likelihood of suture material harboring micro-organisms should be considered more than eventual strength. The wound complications do, however, suggest that where strength and holding power is required, the suture material should be so chosen as to allow maximal strength during the first two or three weeks of healing.

Although this study has shown that nutritional state does not influence the ultimate strength of the wound, it has also served to underline the importance of nutrition to wound healing. All the wound complications occurred in the malnourished group of animals. Similarly, the incidence of mortality and morbidity was double in the malnourished group. Comparisons were only carried out among surviving animals with normal wounds. Although the strength in survivors was comparable at 60 days, many more wounds and animals survived the 60 days to enable themselves to be compared in the nourished group than in the malnourished group.

If these data can be extrapolated to the clinical situation, they underline once more the importance of nutrition to healing, health and recovery. The data will also encourage those surgeons treating seriously stressed and chronically malnourished patients, for they suggest to us that if we can carry our patient through his illness, even if to only partial recovery, then in the end he will heal as well as his normal counterpart.

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