Intensive Care Med (2004) 30:162–166
DOI 10.1007/s00134-003-2050-2

BRIEF REPORT

Balasubramanian Venkatesh
Jenny Gough
David R. Ralston
Michael Muller
Stuart Pegg

Protein losing enteropathy in critically ill adult patients with burns: a preliminary report

Received: 17 February 2003
Accepted: 29 September 2003
Published online: 21 November 2003

B. Venkatesh (✉)
Intensive Care Unit,
Princess Alexandra Hospital,
University of Queensland,
Australia
e-mail: bala_venkatesh@health.qld.gov.au
Tel.: +61-7-32402111
Fax: +61-7-32407174

J. Gough
Department of Burns Surgery,
Royal Brisbane Hospital,
Brisbane, Australia

D. R. Ralston
Northern General Hospital,
Sheffield, S5 7AU, UK

M. Muller
South Auckland Burn Service,
Middlemore Hospital,
New Zealand

S. Pegg
Department of Burns Surgery,
Royal Brisbane Hospital,
Brisbane, Australia

Abstract *Objective:* Few data have been published regarding protein losing enteropathy in adult patients with burns. This study characterised the presence of protein-losing enteropathy in adults with burns and examined the relationship between the magnitude of burn size and the severity of protein loss. *Methods:* Twenty adult patients with burns (BSA 31±25%, range 2–80%) were studied. Fluid resuscitation was based on the Parkland's formula. Protein loss into the gastrointestinal tract was measured using faecal α_1-antitrypsin (FA-1-AT) concentrations. Serial measurements of serum protein and albumin concentrations were performed. *Results:* Fourteen patients demonstrated elevations in FA-1-AT levels. The mean peak FA-1-AT level was 3.6±4.2 mg/g dry weight of stool. Two patients demonstrated elevated FA-1-AT excretion 1.5 months and 3 months after the burns. There was a good correlation between burn size and FA-1-AT excretion (R^2=0.40). *Conclusions:* Protein losing enteropathy was demonstrable in patients with major burns. The magnitude of this phenomenon appears to be proportional to the burns size.

Keywords Burn · Critical illness · Gut · Mucosa · Protein losing enteropathy

Introduction

Abnormalities of gastrointestinal function in patients with major burns include increased mucosal permeability [1], mucosal oedema [2, 3], gastrointestinal haemorrhage, ischaemic necrosis [4], stress ulceration, and a malabsorption syndrome [5]. The cause of these abnormalities is multifactorial: post-burn vasoconstriction leading to splanchnic ischaemia as measured by gastric intramucosal pH [6, 7], neutrophil injury, endotoxin-induced mucosal alterations, and mucosal cell death by apoptosis [8].

Whilst considerable work has been carried out on the consequences of gut barrier failure in burns such as bacterial translocation [9], sepsis [10], multiple organ dysfunction syndrome [10] and death, a problem which has received little attention is barrier failure in the other direction, i.e. the issue of loss of protein across the mucosa into the gut lumen. This condition is termed protein-losing enteropathy (PLE). The rationale to consider this pathological process stems from a number of factors. Patients with major burns have protracted hypoalbuminaemia, and it is likely that gut losses are contributory. Pa-

tients with splanchnic ischaemia from other causes have been shown to develop protein loss across the gut [11, 12]. Mucosal oedema from non-burn pathologies has been found to be associated with protein losing enteropathy [13]. There is evidence of protein loss across other epithelial sites in burn patients including the burn wound [14] and the urinary tract [15]. Although a paediatric study examined this question [16], few published data exist regarding protein losing enteropathy in adult patients with burns.

The aims of this study were (a) to characterise the presence of protein losing enteropathy in adult patients with burns using faecal excretion of α_1-antitrypsin as a marker and (b) to examine the relationship between the magnitude of burn size and the severity of protein loss across the gut mucosa.

Materials and methods

Patients

The prospective observational study was approved by the Research Ethics Committee of Royal Brisbane Hospital, and informed consent was obtained from the patient or the next of kin depending on the conscious state of the patient. The study enrolled patients diagnosed with burns in the preceding 72 h. Patients were excluded if they had pre-existing gastrointestinal disease likely to cause protein losing enteropathy, or if stool samples could not be obtained. During the study period 31 patients with burns were admitted to the hospital, 11 of whom were excluded (2 refused consent, 3 spent less than 24 h in the hospital, and 6 presented more than 1 week after the burns occurred).

Care of the patients

The Parkland's formula was used to guide the approximate total volume of fluids required during the resuscitation phase. The fluids were titrated to achieve a target urine output of 0.5–1 ml/kg per hour and a mean arterial pressure less than 60 mmHg. Maintenance fluid therapy was administered at 45 ml/kg per day as enteral feed supplemented by intravenous dextrose saline as required. All patients received a proprietary enteral formulation (Perative, Abbott Australasia, New South Wales, Australia) This is a high-protein feed designed for catabolic patients. Immune-enhancing feeds were not used. Prokinetic agents such as metoclopramide were used only if there was evidence of gastroparesis. No patient received steroids or any other immunosuppressive therapy. Routine blood tests included full blood counts, serum electrolytes, liver and renal functions and total protein and albumin concentrations.

Faecal α_1-antitrypsin (FA-1-AT) determination

Samples of faeces passed post-burn were sent for FA-1-AT level. Watery stools were excluded from the study because of the potential for diarrhoea per se to increase faecal clearance of A-1-AT, which may result in the overdiagnosis of excess gastrointestinal protein leakage [17]. For analysis of FA-1-AT an aliquot of the stool sample was diluted with 0.9% saline and centrifuged at 3000 rpm for 15 min. The supernatant was then used in the analysis for the determination of FA-1-AT using immunonephelometry. The analytical range of the assay was 0.1–36 g/l and the coefficient of variation ranged from 4.8% to 6.1%. The normal reference range for FA-1-AT is less than 1.5 mg/g dry weight of stools. As the presence of blood in the faeces may produce false-positive elevations in FA-1-AT, a simultaneous faecal occult blood assay was performed on all samples.

Data analysis

Summary statistics were employed to examine differences in demographic data, burn size, changes in serum biochemistry and FA-1-AT levels. Since only 14 patients had multiple measurements of FA-1-AT at various time periods, it was not possible to perform analysis of repeated measures or examine changes with time. Hence we chose to examine the peak FA-1-AT excretion, which was defined as the highest reported value of FA-1-AT on single or multiple measurements. The relationship between burn size and the FA-1-AT levels was determined using linear regression. The proportion of patients with faecal occult blood positivity in groups with raised and normal FA-1-AT were compared using Fisher's exact test. The proportion of patients fulfilling criteria for systemic inflammatory response syndrome or sepsis manifesting PLE was also examined using Fisher's exact test.

Results

The demographic details of the study population were as follows: 15 men, 5 women; mean age 34 years, range 18–58. They included four patients with inhalational burns. Eleven were admitted to the intensive care unit (ICU) and nine to the burns unit. The mean extent of burns was 31±25% (range 2–80%), and the mean Acute Physiology and Chronic Health Evaluation II score of the ICU patients was 16±6. Patients stayed a mean of 30±24 days in hospital. There were no deaths among the study population during the study.

The baseline total serum protein and albumin concentrations were 57±11 g/l (normal 62–83) and 31±7 g/l (normal 33–47), respectively. A statistically significant decrease in both total protein and albumin concentrations were observed reaching nadir values of 47±13 g/l ($p<0.01$) and 20±9 g/l ($p<0.001$), respectively.

The median day of stool collection for the initial FA-1-AT assay was day 9 (range 3–22). Fourteen patients had more than one stool sample analysed for FA-1-AT. Fourteen of the 20 patients (70%) demonstrated elevations in FA-1-AT levels (>1.5 mg/g of dry weight). The individual FA-1-AT data are summarised in Table 1. The mean peak FA-1-AT level was 3.6±4.2 mg/g of dry weight of stool (range 0.2–14.8). Faecal occult blood (FOB) was positive in 1 of the 14 patients with elevated FA-1-AT, whilst 2 of the 6 patients with normal FA-1-AT tested positive on FOB ($p=0.13$). The presence of faecal occult blood did not impact on the FA-1-AT concentration. Two patients, one with 70% burns and the other with 80% burns, were found to have elevated FA-1-AT excretion on 1.5 and 3 months after the burns, respectively.

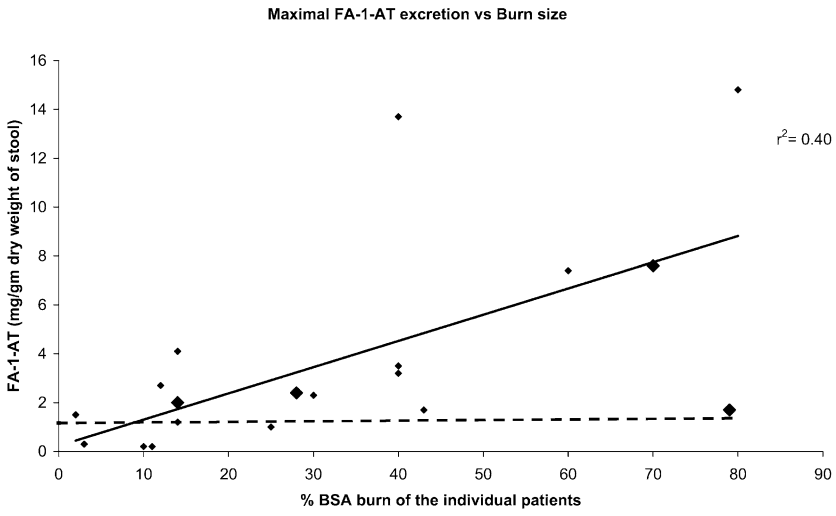

Fig. 1 Relationship between burn size and maximal FA-1-AT excretion. *Diamonds* individual patient values of FA-1-AT excretion plotted against their burn size; *continuous line* trend line; *BSA* burn surface area; *broken horizontal line* upper limit of excretion of FA-1-AT (as 2 patients had identical burn size and FA-1-AT excretion, there are only 19 visible data points because of 2 overlapping ones; however, there are 20 data sets); *larger diamonds* data points from four patients who also suffered from inhalational burns

Table 1 FA-1-AT excretion (mg/g dry weight of stool): individual data (*parentheses* day of stool collection)

Patient no.	% Burn	Sample 1	Sample 2	Sample 3	Sample 4
1	2	0.3 (5)	–	–	–
2	10	0.2 (9)	0.2 (10)	0.1 (13)	–
3	14	4.1 (7)	0.7 (13)	1.2 (16)	–
4[a]	14	0.7 (4)	1.2 (6)	2 (17)	–
5	40	3.5 (12)	1.2 (17)	1.5 (27)	–
6[a,b]	70	4 (11)	5.6 (11)	5.2 (13)	5.4 (14)
7	43	1.7 (6)	0.9 (20)	–	–
8	2	1.5 (4)	–	–	–
9	80	14.8 (16)	2[c]	2.6[c]	–
10[a]	28	2.4 (8)	0.2 (11)	–	–
11	25	1 (10)	0.3 (18)	–	–
12[a]	79	1.7 (22)	0.2 (30)	0.2 (63)	–
13	10	0.2 (9)	–	–	–
14	60	7.4 (19)	–	–	–
15	11	0.2 (7)	–	–	–
16	40	12.2 (16)	13.7 (17)	5.9 (23)	0.8 (40)
17	40	3.2 (12)	2.7 (24)	–	–
18	12	2.7 (4)	0.5 (8)	–	–
19	14	1.2 (2)	–	–	–
20	30	1.2 (14)	2.3 (17)	–	–

[a] Patients with inhalational burns
[b] Patient had 13 stool analyses, and FA-1-AT peaked at 7.6 mg/g dry weight of stool on day 23
[c] Stool collection >90 days

A good correlation was observed between the surface area of burn and FA-1-AT excretion (R^2=0.40, p<0.01; Fig. 1). No patients with a burn size less than 12% demonstrated elevated FA-1-AT excretion.

Discussion

The principal finding in this study is the clear demonstration of protein loss into the gastrointestinal tract in adult patients with major burns. This is the first demonstration of this phenomenon in a prospective clinical study in critically ill adult patients. The other major finding is the linear relationship between burn size and magnitude of FA-1-AT excretion. There was evidence of protracted protein loss into the gastrointestinal tract in two patients with major burns.

Possible mechanisms of protein-losing enteropathy

Although a diverse group of gastrointestinal disorders can result in PLE [18], mucosal damage resulting from a number of mechanisms such as ischaemia, oedema, neutrophil injury, ulceration and apoptosis may be responsible. Data from our previous study showed a strong correlation between burn size and intramucosal pH [6]. The present study demonstrates a strong correlation between burn size and FA-1-AT excretion. However, the presence of an inhalational injury may increase the burn wound, which can influence the correlation between the two. Although intramucosal pH was not measured in this study, it is tempting to speculate that mucosal acidosis is associated with PLE. Other possibilities to be considered for the raised FA-1-AT excretion include diminished enteral nutrition and gastrointestinal bleeding. Mucosal atrophy

from lack of enteral nutrition can lead to altered epithelial permeability. In this study enteral nutrition was established within 24 h of admission to the hospital, and all patients tolerated enteric feeds. Blood in the gastrointestinal tract can confound FA-1-AT interpretation. However, in our study only one of the three patients who tested positive for FOB had raised FA-1-AT levels, and there was no significant difference in the proportion of patients who tested positive for FOB between those with raised and normal FA-1-AT.

Clinical implications

There are a number of clinical implications. Firstly, does the protein loss contribute to hypoalbuminaemia? Support for this concept is present in some of the published data which report an elevation in plasma α1-antitrypsin clearance of more than five times associated with albumin concentrations less than 30 g/l. In our study some patients demonstrated excretion of FA-1-AT that was more than eight times the normal level. This is likely to have contributed to the hypoalbuminaemia. However, it is important to recognise that burns patients have diverse causes of hypoalbuminaemia, including protein loss through the burn wound [14], urinary albumin losses [15] and decrease in hepatic albumin synthesis [19]. In the presence of an inhalational burn the lung capillary leak of protein may accentuate the hypoalbuminaemia [20]. Determining the precise contribution of faecal protein losses to hypoalbuminaemia amongst a plethora of causes is possible only with simultaneous measurements of plasma α1-antitrypsin clearance, losses through other sites and estimation of hepatic synthetic capacity. Another important issue arising from the results of this study is that the presence of ongoing protein loss should raise the possibility of ongoing mucosal insults, and there should be a heightened vigilance for covert splanchnic ischaemia or mucosal injury.

Critique of the study

Although this study is limited by the small sample size and the limited frequency of stool collection, our findings demonstrate the presence of PLE with statistical significance. The marked variability in the timing of stool sample availability for FA-1-AT assay in our study limits more detailed analysis of data. While some investigators recommend the use of plasma clearance of α1-antitrypsin as an index of PLE, FA-1-AT has been shown to be closely correlated with ^{51}Cr-labelled plasma protein clearance [21].

In conclusion, PLE is a real phenomenon in patients with burns occurring in up to 70% of patients. The magnitude of this phenomenon appears to be proportional to burn size. The documentation of PLE in critical illness adds one more piece to the puzzle of the complex role of the gut in the development of multiple organ dysfunction syndrome. Future large-scale studies examining this phenomenon in various ICU populations must fully delineate the incidence and severity of this process.

Acknowledgements These data were presented as a free paper in the Annual Scientific Meeting of the Australian New Zealand Burn Association in Hobart in 2001. The study was supported through a grant from the Royal Brisbane Hospital Research Foundation. We also acknowledge the nursing staff of the Burns and Intensive Care Units for their assistance with the study.

References

1. Ryan CM, Yarmush ML, Burke JF, Tompkins RG (1992) Increased gut permeability early after burns correlates with the extent of burn injury. Crit Care Med 20:1508–1512
2. Sir O, Fazal N, Choudhry MA, Goris RJ, Gamelli RL, Sayeed MM (2000) Role of neutrophils in burn-induced microvascular injury in the intestine. Shock 14:113–117
3. Sir O, Fazal N, Choudhry MA, Gamelli RL, Sayeed MM (2000) Neutrophil depletion prevents intestinal mucosal permeability alterations in burn-injured rats. Am J Physiol Regul Integr Comp Physiol 278:R1224–R1231
4. Wilson MD, Dziewulski P (2001) Severe gastrointestinal haemorrhage and ischaemic necrosis of the small bowel in a child with 70% full-thickness burns: a case report. Burns 27:763–766
5. Jenkins ME, Gottschlich MM, Kopcha R, Khoury J, Warden GD (1998) A prospective analysis of serum vitamin K in severely burned pediatric patients. J Burn Care Rehabil 19:75–81
6. Venkatesh B, Meacher R, Muller MJ, Morgan TJ, Fraser J (2001) Monitoring tissue oxygenation during resuscitation of major burns. J Trauma 50:485–494
7. Tokyay R, Zeigler ST, Traber DL, Stothert JC Jr, Loick HM, Heggers JP et al (1993) Postburn gastrointestinal vasoconstriction increases bacterial and endotoxin translocation. J Appl Physiol 74:1521–1527
8. Spies M, Chappell VL, Dasu MR, Herndon DN, Thompson JC, Wolf SE (2002) Role of TNF-alpha in gut mucosal changes after severe burn. Am J Physiol Gastrointest Liver Physiol 283:G703–G708
9. Herndon DN, Zeigler ST (1993) Bacterial translocation after thermal injury. Crit Care Med 21:S50–S54
10. Pastores SM, Katz DP, Kvetan V (1996) Splanchnic ischemia and gut mucosal injury in sepsis and the multiple organ dysfunction syndrome. Am J Gastroenterol 91:1697–1710

11. Effenterre G van, Haut J, Brezin A, Le Mer Y, Rambaud JC, Galian A et al (1989) Retinal and choroidal ischemic syndrome, digestive tract and renal small vessel hyalinosis, intracerebral calcifications and phenotypic abnormalities: a new family syndrome. Graefes Arch Clin Exp Ophthalmol 227:315–322
12. Matsushita I, Hanai H, Sato Y, Arai H, Iida T, Hosoda Y et al (2000) Protein-losing enteropathy caused by mesenteric venous thrombosis with protein C deficiency. J Clin Gastroenterol 30:94–97
13. Waldmann T (1985) Protein losing gastroenteropathies. In: Haubrich W, Kalser M, Roth J, Schaffner F (ed) Gastroenterology. Saunders, Philadelphia, p 1814
14. Waxman K, Rebello T, Pinderski L, O'Neal K, Khan N, Tourangeau S et al (1987) Protein loss across burn wounds. J Trauma 27:136–140
15. Yu H, Cooper EH, Settle JA, Meadows T (1983) Urinary protein profiles after burn injury. Burns Incl Therm Inj 9:339–349
16. Matoth I, Granot E, Gorenstein A, Abu-Dalu K, Goitein K (1991) Gastrointestinal protein loss in children recovering from burns. J Pediatr Surg 26:1175–1178
17. Strygler B, Nicar MJ, Santangelo WC, Porter JL, Fordtran JS (1990) Alpha 1-antitrypsin excretion in stool in normal subjects and in patients with gastrointestinal disorders. Gastroenterology 99:1380–1387
18. Brasitus T, Bissonnette B (1998) Protein-Losing Gastroenteropathy. In: Feldman M, Sleisenger M, Scharschmidt B (ed) Sleisenger and Fordtran's gastrointestinal and liver disease, 6th edn. Saunders, Philadelphia, pp 369–375
19. Dickson P, Bannister D, Schreiber G (1987) Minor burns leads to major changes in synthesis rates of plasma proteins in the liver. J Trauma 27:283–286
20. Turnage R, Nwariaku F, Murphy J, Schulman C, Wright K, Yin H (2002) Mechanisms of pulmonary microvascular dysfunction during severe burn injury. World J Surg 26:848–853
21. Florent C, L'Hirondel C, Desmazures C, Aymes C, Bernier JJ (1981) Intestinal clearance of alpha 1-antitrypsin. A sensitive method for the detection of protein-losing enteropathy. Gastroenterology 81:777–780