The potential effect of biological sealants on colorectal anastomosis healing in experimental research involving severe diabetes

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ABSTRACT

Colorectal anastomoses continue to pose a significant challenge in current surgical practice. Anastomotic leakage remains one of the most frequent and dramatic complications of colorectal surgery, even in centres of high specialisation. Diabetes is a well-established independent factor which results in higher anastomotic leakage rates. Fibrin sealants have been applied in experimental and clinical studies for the prevention of anastomotic dehiscence. However, little is known regarding their impact on diabetic patients. Several fibrin sealants have been proposed as adjunct to standard surgical techniques to prevent leakage from colonic anastomoses following the reversal of temporary colostomies, approved for general haemostasis. This review summarises current advances in colorectal anastomoses and provides evidence that may strengthen the need for tissue sealants in colorectal anastomoses of diabetic patients.

We searched Medline (1966–2016) and Scopus (2004–2016) for current evidence in the field. To date, there is no evidence to support the use of fibrin sealants as an adjunct in diabetic patients who undergo colorectal surgery. Experimental animal models with extreme diabetes could be of significant use in the present field and further research is needed prior to application of fibrin sealants in a clinical setting.

KEYWORDS

Colorectal – Anastomosis – Fibrin – Sealant – TISEEL

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Introduction

Surgery is currently the definitive treatment modality for colorectal cancer and other benign pathologies of the colon and rectum, including inflammatory bowel disease (IBD), inflammatory strictures, diverticulosis–diverticulitis, obstructive defecation, volvulus, fistulae and so on. Advancing knowledge in the fields of surgical anatomy, pathophysiology and mechanisms of lymph node metastasis in colorectal cancer have led to the implementation of novel surgical techniques, including total mesorectal excision (TME) and extra-levator abdominoperineal excision and routes (laparoscopic, robotic surgery).¹ These advances gradually modified current surgical techniques and the overall management of colorectal patients in an attempt to improve oncological and quality of life outcomes.

Despite the advances in surgical training and accumulated experience, anastomotic leakage remains one of the most frequent and dramatic complications, even in centres of high specialisation.^{2,3} Worldwide, the reported rates

range between 2.5% and 15% following rectal cancer surgery (10–28% for ultra-low anastomosis) and 3–7% following surgery for colon cancer.^{2,4}

The adoption of TME to decrease the local recurrence rate to acceptable levels and the use of circular staplers have enabled the performance of ultra-low colorectal anastomoses in rectal cancer surgery. The introduction of enhanced recovery after surgery (ERAS) in modern surgery offers significant postoperative advantages in colorectal wound healing.⁵ In this context, current management of colorectal patients suggests that oral mechanical bowel preparation should be avoided and that patients should receive a preoperative carbohydrate drink, as overnight fasting is not indicated. Long-acting sedation and pre-anaesthesia should also be avoided.⁵ The use of laxatives is encouraged to minimise postoperative ileus and postoperative nausea and vomiting should be treated as indicated. The use of nasogastric tube should be avoided. General anaesthesia plus use of epidural are indicated in cases of longer operative time and open surgery. The laparoscopic approach is recommended. Resection

site drainage (drain tubes placement) should be avoided. It is recommended to implement immediate postoperative diet. The urinary catheter should be removed on the first postoperative day and patients should be mobilised immediately.⁵

The impact of anastomotic dehiscence on patient morbidity and quality of life is devastating, even when it is detected within the limit of 60 minutes, as it usually leads to faecal peritonitis, uncontrollable sepsis and a permanent stoma.⁶ Anastomotic leakage results in higher morbidity and mortality with consequent longer postoperative hospitalisation and increased treatment costs. There is strong evidence that recurrence rates (both local and distal) are higher, thus increasing morbidity, mortality and costs, and seriously affecting the quality of life of patients.^{7,8}

Several risk factors seem to influence the odds of developing an anastomotic leakage.² A systematic review proposed that male sex, American Society of Anesthesiologists (ASA) grade above II, Charlson comorbidity score (renal disease, cardiac failure, malnutrition, diabetes) and history of radiotherapy are independent factors which predict anastomotic failure.³ Tumour-related factors also affect the quality of colorectal anastomoses. Distal site, size larger than 3cm, advanced stage, emergency surgery and metastatic disease appear to be the most important. Other risk factors which can be modified are smoking, obesity, poor nutrition, alcohol excess, immunosuppressants and therapy with bevacizumab. Several intraoperative risk factors are also associated with this serious complication, including severe blood loss, which necessitates blood transfusion and prolonged duration of surgery (greater than 4 hours).⁵

Diabetes is a well-established independent factor associated with higher anastomotic leakage rates. McDermott et al found that, even if controlled, the rate of leakage remained higher compared with otherwise healthy patients.⁵ The same meta-analysis provided strong evidence to suggest that diabetes is significantly and independently associated with an increased risk of mortality from anastomotic leakage in colorectal surgery.⁵

A number of important studies in human and animal models have been published to determine the molecular basis for impaired healing in patients with diabetes, looking at the healing processes in wounds, soft tissues, bone and tendons.⁹ The impact of diabetes in tissue healing has been described in several studies.^{10–12} Inflammation seems to play a major role in the pathophysiology of the cardiovascular, neurological and ischaemic features of diabetes. Recently, this fact has received more attention and a number of studies have been published on the resolution of inflammation pathways. Data analyses have demonstrated that diabetes promotes an enhanced, macrophage-mediated inflammation, promoting a proinflammatory, profibrotic and dysfunctional phenotype. Other diabetes-related factors, including hyperglycaemia, also act as positive regulators of inflammation. The resolution of the inflammation is the basis of the healing process and several factors are involved. It has been recognised that the peroxisome proliferator-activated receptors (PPARs) induce inflammation resolution in different ways, in particular through their activity on

macrophages, which promotes their transition from proinflammatory to healing-associated phenotypes. In fact the downregulation of PPAR gamma in diabetic wounds is responsible for the defect in this transition and the consequent healing impairment. Mirza et al conducted an important study which concluded that nucleotide-binding oligomerisation domain-like receptor 3–interleukin-1 β is one of the key pathways in this problem.¹² The importance of the leucocytes in healing is well recognised and we know that neutrophils are the main subtype involved; the mechanisms of their action have been thoroughly studied. In particular, forming extracellular traps of chromatin with cytotoxic proteins can also damage healthy tissues other than microbes. One study showed that in individuals with diabetes (human and mice), neutrophils were primed to form extracellular traps (NETosis).^{10,11} A study of diabetic mice by Gallagher and colleagues has demonstrated that bone marrow derived circulating endothelial progenitor cells are mobilised into the circulation by hyperoxia.¹⁵ Ischaemia induces neoangiogenesis as a physiological response and these endothelial progenitor cells are the cornerstone of this process. In patients with diabetes, peripheral angiopathy resulting in decreased peripheral blood flow and ischaemic conditions are common complications of the process of healing.¹⁴ In addition, colorectal resections involving devascularisation by means of the high ligation of the main vessels aggravates the ischaemia, since the surgical resection forces the blood supply to rely exclusively on the marginal artery for a certain length at the proximal (colonic) end of the colorectal anastomosis.¹⁵

More than 100 physiological factors which contribute to wound healing impairment in individuals with diabetes have now been identified. The most important among them are reduced or abnormal growth factor production, adequate angiogenic response, normal macrophage function, collagen concentration and degradation, the epidermis barrier function, the quantity of granulation tissue, keratinocyte and fibroblast migration and proliferation, the number of nerves in the area of interest, bone healing and the balance between the accumulation of extracellular matrix components and their remodelling by matrix metalloproteinases. Importantly, the pathogenic markers associated with impaired wound healing have been also identified via molecular analysis on biopsies retrieved from the epidermis of patients. The two most important markers among them are the overexpression of *c-myc* and the nuclear localisation of beta-catenin. Furthermore, when epidermal growth factor receptor reduction and abnormal localisation and activation of the glucocorticoid pathway are associated, they result in inhibition of keratinocyte migration.¹⁶

Current evidence

Animal models of colorectal anastomosis

Since 1991, when Van Der Ham used the first rat model of colorectal anastomosis to test the effect of fibrin glue, hundreds of similar models have examined the efficacy of other potential factors, including hyperbaric oxygen, prokinetics, nonsteroidal anti-inflammatory agents, amino acids,

cianoacrylate.^{17–20} Different techniques and routes to perform the anastomosis and to identify the left colon have been also proposed. The clinical evidence for rupture of the anastomosis was studied initially, then a large number of different techniques to test for microscopic leakage and the anastomotic strength were implemented (biopsies for pathology, collagen concentration and degradation measuring the hydroxyproline, neutrophilic granulocyte infiltration and bursting pressure).^{21–25}

Experimental models of diabetes, colorectal anastomosis and application of biological sealants

To date, several experimental studies suggest that the implementation of biological sealants might be effective as an adjunct factor that could ensure the integrity of colorectal anastomoses.^{24–26} Vakalopoulos et al evaluated the efficacy of Histoacryl® (TissueSeal, Ann Arbor, MI), BioGlue® (Cryo-Life Inc, Kennesaw, GA), Dermabond® (Ethicon), Tissucol® (Baxter Healthcare Corporation, Newbury, Berks), Dura-seal® Xact (Integra LifeSciences Corporation, St Priest, France), gelatin-resorcinol-formaldehyde and Glubran 2® (GEM srl, Viareggio LU, Italy) on sutureless sealing of colonic defects.²⁶ They observed that Histoacryl Flex exhibited significantly higher collagen formation at day 10 compared with other sealants. In a previous systematic review from the same institution, researchers concluded that experimental and clinical studies in the field appear promising.²⁵ TISSEEL® (Baxter Healthcare Corporation, Newbury, Berks) has been also used as an adjunct to standard surgical techniques, such as suture and ligature, to prevent leakage from colonic anastomoses following the reversal of temporary colostomies approved for general haemostasis. It contains two components: the sealer protein solution, which contains human fibrinogen and aprotinin (a synthetic fibrinolysis inhibitor which helps prevent premature degradation of fibrin clot) and the thrombin solution, which contains human thrombin and calcium chloride. When mixed together, the two components combine and mimic the final stages of the body's natural clotting cascade to form a rubber-like mass that adheres to the wound surface and achieves haemostasis and sealing or glueing of tissues.

In a previous experimental study, van der Vijver et al argued the beneficial effect of fibrin sealants on patent anastomoses in low risk conditions.²⁷ To date, none of the aforementioned fibrin sealants has been studied among severely diabetic colorectal patients who require anastomosis. We believe that the application of this new material will be able to achieve an improved healing of the colorectal anastomosis and thus reduce anastomotic leakage rate in severely diabetic subjects.

The rationale for colorectal anastomosis in a diabetic animal model with the application of fibrin sealants

Performing colorectal anastomosis with a greater degree of safety is still a challenge. It would of great socioeconomic importance to identify methods, factors and/or instrumentation to reduce postoperative complications, especially for patients at higher risk of anastomotic complications.

To our knowledge the use of glues has been one of the first factors to be studied in animal models without additional risk factors. Although there has been increasing interest in testing the healing of colorectal anastomosis in animal models and, in one study, fibrin glue has been applied on the colorectal anastomosis in peritonitic and non-peritonitic rats.²¹ Previous systematic reviews failed to show a clear benefit of the application of glue on the colorectal anastomosis.²⁸ New sealants with a licence to be used in colorectal anastomosis have not so far been tested in animal models or, more importantly, in animal models of severe diabetes.

Conclusion and implications for future research

The dramatic results and the unaffordable high rate of anastomotic leak, especially for rectal operations, have directed colorectal institutions to adopt and recommend different techniques to limit the consequences of anastomotic leak, including diverting defunctioning loop ileostomies for single low anterior resections,^{29,50} trans-anal drainage tubes, trans-anal sponge and other systems of drainage.^{31,52} Even if it has been recognised, the fact that the diverting stomas limit the sequels of the anastomotic leak rather its occurrence, a Swedish trial concluded to a reduction of the rate of anastomotic leakage from 28% to 10.3% in patients randomised to a stoma.²⁹

Recent clinical and experimental studies confirm the presence of new molecular targets and biomarkers in the field of diabetic wound healing. To date, diabetes remains a major predisposing factor for colorectal anastomotic leakage. Even if controlled, the risks of developing this complication remain high and there are no effective modifications which could further decrease its incidence. Biological sealants could potentially benefit patients with diabetes, but there seems to be a complete absence of consensus in this field. Given the fact that their cost is reasonable, their implementation in current clinical practice could be justified if they would lead to reduced rates of the anastomotic ruptures. Experimental models give us the opportunity to conduct pilot experiments in extreme biological conditions which are otherwise incompatible with human life. In this context, we strongly believe that future investigation should be conducted in this field, to provide a basis for further clinical studies.

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