The Effect of Intrarectal Paclitaxel Lavage on the Healing of Rectal Anastomosis: A Comparison with Povidone Iodine

REKTUM İÇİ PAKLİTAKSEL YIKAMASININ REKTUM ANASTOMOZU İYİLEŞMESİ ÜZERİNE ETKİSİ: POVİDON İYOT İLE KARŞILAŞTIRMA

Ömer GÜNAL, MD,[a] Salah GHANDOURI, MD,[b] Arif ASLANER, MD[a]

[a] Department of General Surgery, Abant İzzet Baysal University, Düzce Faculty of Medicine, DÜZCE
[b] Department of Physiology, Marmara University Faculty of Medicine, İSTANBUL

Abstract

Objective: Intraluminal colonic washout with anticancer agents is an alternative option to prevent local recurrences after colorectal cancer resection. However, intracolonic washout with any agent can interfere with anastomotic wound healing. The current study aimed to investigate the effects of intracolonic paclitaxel and povidone-iodine administration on intestinal anastomotic healing.

Material and Methods: Localized intracolonic washout with paclitaxel (3 mg/kg) and povidone-iodine (5%) was achieved before and after colorectal anastomosis in two (each consisting of 10) groups of rats. The control group (n= 10) received no medical therapy. Anastomotic bursting pressures, anastomotic hydroxyproline content, and peri-anastomotic myeloperoxidase activity were determined.

Results: Bursting pressure was significantly less in the povidone group, while it was slightly elevated in the paclitaxel group when compared to the control group. The hydroxyproline level was significantly increased in the povidone group, while it was found to be decreased in the paclitaxel group. Myeloperoxidase levels were also significantly decreased in both groups when compared to the control group.

Conclusion: Although paclitaxel decreases hydroxyproline production, it does not appear to affect anastomotic bursting pressure. Conversely, povidone-iodine decreases anastomotic bursting pressure despite increased hydroxproline synthesis. These data imply that paclitaxel and povidone-iodine carry out their effects on anastomotic healing either by altering the quality or quantity of collagen synthesis.

Key Words : Paclitaxel, povidone-iodine, colon

Özet

Amaç: Kolorektal kanser rezeksiyonları sonrası lokal nükslerin önlenmesi için antikanser ajanlarla kolonun intraluminar olarak yıkanması, alternatif bir seçenektir. Her ne sebeple olursa olsun, herhangi bir ajanla kolon içinin yıkanması, anastomoz iyileşmesini etkileyebilir. Bu çalışma intrakolonik paklitaksel ve povidon iyot uygulamasının intestinal anastomoz iyileşmesine etkilerinin araştırılması için planlandı.

Gereç ve Yöntemler: Her biri 10 sıçandan oluşan 2 gruba kolorektal anastomoz öncesi ve sonrasında paklitaksel (3 mg/kg) ve povidon iyot (%5) ile lokal kolon içi yıkama yapıldı. Kontrol grubu (n= 10) hiçbir medikal tedavi almadı. Anastomoz patlama basınçları, anastomoz hidroksiprolin içeriği ve perianastomotik miyeloperoksidaz (MPO) aktivitesi değerlendirildi.

Bulgular: Patlama basıncı, kontrol grubuna göre povidon iyot grubunda belirgin olarak az iken, paklitaksel grubunda hafif artmış bulundu. Hidroksiprolin seviyesi, povidon iyot grubunda belirgin olarak artmış iken, paklitaksel grubunda azalmış bulundu. MPO seviyeleri, kontrol grubuna göre her 2 grupta da belirgin olarak azalmış olduğu bulundu.

Sonuç: Paklitakselin hidroksiprolin üretimini azaltmasına rağmen, anastomoz patlama basıncını etkilemediği, buna karşın povidon iyotun hidroksiprolin sentezini arttırmasına rağmen anastomoz patlama basıncını azalttığı görüldü. Bu sonuçlarla, paklitaksel ve povidon iyotun etkilerini kollajen sentezinin miktar ve niceliğini değiştirerek sağladığı düşünüldü. Bu etkilerinin; her 2 grupta da MPO düzeylerinin düşük olması nedeni ile, inflamatuar yanıttan bağımsız olabileceği sonucuna varıldı.

Anahtar Kelimeler: Paklitaksel, povidon iyot, kolon

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Yazışma Adresi/*Correspondence*: Ömer GÜNAL, MD
Abant İzzet Baysal University,
Düzce Faculty of Medicine
Department of General Surgery,
Konuralp, DÜZCE
gunal@ibu.edu.tr

Currently, colorectal carcinoma is now the third leading cause of cancer related death in the USA[1] The surgery performed after an early diagnosis seems to be extremely vital. Prevention of anastomotic tumour recurrence has been the subject of investigation. Intraluminal

chemotherapy may be a good alternative for the prevention of local recurrences in anastomosis line after colonic cancer surgery. Shukla and his associates showed superiority of the intraluminal 5-FU administration in a clinical trial.[2] Intraluminal administration demonstrated that the appearance of 5-FU was prompt and that significantly higher levels continued in the colonic wall and liver. The way of administration of drugs has still been a matter of debate. Rectal or whole colonic washout with different agents to prevent the anastomotic recurrence is under investigation.[3,4] Whole colon washout with 5% povidone iodine (PI) was suggested to prevent the recurrent cancer.[3] Tumoricidal agents with different mechanism of action can be as effective as the PI when used as the colonic irrigation agent to prevent the anastomotic recurrence. Segmental intraluminal instillation of several tumoricidal agents including PI has been advocated to prevent anastomotic recurrence after colonic resection for colorectal cancer.[3-5] Paclitaxel (Pc) is one of the antineoplastic agents that carry on its effect by altering the tubulin assembly.[6] It has been shown that Pc decreases the in vitro colonic tumor cell line growth.[7] The effect of Pc on the colonic anastomosis healing has not yet been investigated. The application of the antineoplastic agents to the anastomosis line, especially in the early postoperative period may have harmfull effects on the anastomotic wound healing. The aim of this study was to investigate the effect of segmenter intracolonic Pc and PI administration on colonic anastomosis healing.

Material and Methods

30 Adult Wistar-Albino rats were divided into Pc treatment (group-1; n= 10), PI treatment (group-2; n= 10) and no treatment (group-3; n= 10) groups (Table 1). This study was approved by the Animal Studies Ethical Committee of the Düzce School of Medicine.

Preparation of Washout Solutions: Control group received no washing solution. In Pc group, 3 mg/kg Pc at temperature 35°C was diluted in 3 mL of serum physiologic. 5% PI solution was prepared by diluting in the 3 mL serum physiologic.

Anesthesia and Surgical Technique: Rats were anesthetized with intraperitoneal ketamine (100 mg/kg) and chlorpromazine (2 mg/kg). After shaving and cleaning the abdomen with PI, a midline 2.5 cm incision was made. Following a midline laparatomy, rectum was divided two cm above the anal verge. Cut ends of the rectum reanastomosed with a continuous 4/0 polypropylene suture material without creating a stricture. Rectal anastomosis site irrigation was performed with 3 mL Pc solution (3 mg/kg) through a No: 6 foley catheter that was indwelled through the anal canal. The bowel lumen was obliterated 1.5 cm above the probable anastomosis line with a No: 1 silk suture before starting the Pc irrigation. Distal part of the rectal lumen was obliterated by inflating the foley balloon. Similar washout process was accomplished with 1-3 mL prepared 5% PI solution. Exposure to Pc and PI has been continued for ten minutes, both before and after anastomosis. The

Table 1. Study groups.

Study groups	Specifications	Number of subjects (n)
Group-1	*Pc Group: Rectal anastomosis site irrigation was performed with 3 mL Pc solution (3 mg/kg) through a No: 6 foley catheter that was indwelled through the anal canal.	10
Group-2	*PI Group: Rectal anastomosis site irrigation was performed with 3 mL 5% PI solution through a No: 6 foley catheter that was indwelled through the anal canal.	10
Group-3	C Group: These subjects have not taken any rectal irrigation.	10

* Irrigation was accomplished before and after anastomosis.

abdomen was closed in one layer continuously with 2/0 silk suture material.

All rats were sacrificed at postoperative 10th day. Two centimeter colonic segment which centered the anastomosis line was excised for the anastomosis bursting pressure (ABP) measurement, anastomotic hydroxyproline (HP) level and perianastomotic colonic tissue myeloperoxidase (MPO) level determinations.

ABP: At postoperative 10th day, anastomosed colonic segment was extirpated from the one centimeter distal and proximal of the anastomosis line. A catheter was indwelled from one end and ligated. A pressure transducer was placed at the other end. A steady pressure measurement or a sudden fall in pressure interpreted as the leakage of anastomosis. Maximum pressure value assumed as "ABP".

Tissue HP level: HP levels were assumed as the indicator of the perianastomotic collagen content. Tissue HP levels were determined as described in the literature.[8]

Tissue MPO level: It was calculated by the reduction of the o-dianidizine with hydrogen peroxide.[9]

Statistical analysis

Results were presented as mean ± s.e.m. (standard error of mean). Student's t test and Mann-Whitney-U Test was used to compare the results. $p< 0.050$ was considered to be statistically significant.

Results

No considerable weight loss was observed in the study groups. ABP was found to be significantly increased (232 ± 11.3 mmHg) (p= 0.018) in group-1 while decreased in group-2 (156.4 ± 9.7 mmHg) (p= 0.004) when compared to group-3 (197.4 ± 10.3 mmHg). Perianastomotic colonic tissue MPO levels were significantly less in group-1 and 2 (80.9 ± 7.5 U/100 mg tissue; p= 0.015; U': 33 and 64.4 ± 10.9 U/100 mg tissue; p= 0.002; U': 36) than the no irrigation group (143.6 ± 21.9 U/100 mg tissue). HP level in group-1 (292.4 ± 11.26 mg/g wet weight) was significantly less than the group-3 (399.5 ± 23.15 mg/g wet weight) (p= 0.002; U'89). However, HP level of group-2 (518.7 ± 21 mg/g wet weight) was found to be increased when compared to group-3 (p= 0.0002; U'95) (Table 2).

Discussion

Whole colonic irrigation for selective bacterial decontamination before the colonic surgery is one of the subjects of the modern gastrointestinal surgery. To date, preoperative or peroperative colonic irrigation has mainly been applied for the colonic decontamination. Recently, Basha proposed the rectal wash out with 5% PI after rectal tumor excision for the prevention of tumor recurrence at anastomosis site.[4] Due to their absorption from the intestinal mucosa, application of the anticancer agents through the intestinal lumen has been found to be noxious.[10] It may be harmful to apply the antineoplastic agent as a whole gut irrigation material due to their systemic effects or potential local effects such as inhibition of wound healing. We used the chemotherapeutic agent in a closed loop segment of the colorectal region which confined the colonic anastomosis line.

Anastomotic healing is affected by the degree of the inflammatory response, the rate of mucosal reepithelization and consequently the rate of collagen maturation.[11] Collagen fibers in the submucosal layer maintain the strength of an anastomosis.[12] Perianastomotic MPO levels as an indicator of the polymorphonuclear infiltration were found to be significantly decreased in both treatment groups when compared to no-treatment group. This reduction in MPO level was deeper in PI group than the Pc group.

Table 2. Parameters and results of the study.

Parameters	Group-1 (Pc)	Group-2 (PI)	Group-3 (C)
ABP (mmHg)	232 ± 11.3	156.4 ± 9.7	197.4 ± 10.3
MPO (U/100 mg tissue)	80.9 ± 7.5	64.4 ± 10.9	143.6 ± 21.9
HP (mg/g wet weight)	292.4 ± 11.26	518.7 ± 21	399.5 ± 23.15

ABP: Anastomosis bursting pressure measurements,
MPO: Myeloperoxidase levels,
HP: Hydroyproline levels.

Saline is one of the most frequently administered intraoperative lavage fluids and acts by decreasing the number of bacteria in the colonic lumen through its mechanical bulk-reducing effect.[13,14] PI has been incorporated in lavage fluids to reduce the population of colonic bacteria through its bactericidal effects.[15] Both of these agents thus limit the degree of the inflammatory response and facilitate anastomotic healing. Basha has also shown that 5% PI has tumoricidal activity when used as whole colon washout.[10] Either mechanical or bactericidal effect of PI might be responsible for having the least MPO level among the three groups in the current study.

The continuing search for effective adjuvant therapy after resection of colorectal carcinoma has resulted in a growing interest in the regional chemotherapy.[16] The wound healing process is characterized by massive cell migration and proliferation. Cytostatic drugs are by nature anti-proliferative and may therefore be expected to interfere with wound healing.[17] Indeed 5-FU administered daily from the day of operation onwards until sacrifice after 7 days severely impairs the anastomotic healing in the rat intestine.[18] In our study, despite not statistically significant, Pc increased the ABP. However, it caused a significant reduction in both anastomotic HP concentration and tissue MPO levels. This implicated that Pc works on anastomosis wound healing by inhibiting the inflammatory cell proliferations MPO level reduction in Pc group may be due to this immune-suppressive effect of the Pc. This fact needs to be further studied. It has been shown that Pc, in vitro binds reversibly to the beta subunits of tubulin as well as promoting microtubule formation and preventing their depolimerization.[19] In addition to being an essential component of mitotic spindle, microtubules are involved in a wide variety of cellular activities, such as cell mobility and transport with the cell.[19,20] It has also been shown that cells treated with this drug are blocked in G2 + M phase of the cell cycle. This effect can explain the reduced MPO activity and HP level in the current study.

Although the Pc decreased the HP synthesis at anastomosis site, it did not decrease the ABP. Similar controversial effect has been observed with PI. Despite a significant increase in HP synthesis at anastomosis site with PI washout, PI caused a significant reduction in ABP. This discrepancy may be due to the different effects of both agents on collagen synthesis quality. Although the PI induced excess amount of HP production, this synthesized collagen could not afford to obtain anastomosis strength as sufficient as in control group. Similar conflicting data has been reported in the literature which was studied with different agents.[11] The incorporation of Nitric oxide synthase inhibitors in intraoperative colonic lavage fluids resulted in a lower ABP, but a similar HP concentration compared with the control group.[11] This effect has been attributed to the impairment of the hydroxylation of proline and lysine, which is essential for the stability and cross-linking of collagen fibers.[11,21] 5% PI might cause a similar effect on collagen cross-linking or stability. Unchanged and even increased ABP with Pc despite the reduced HP content can be explained by the same mechanism at the same time. Pc seems to have a regulatory effect on HP production that makes the collagen more stabilized. Hopkins and associates demonstrated decreased abdominal wound strength in rats which were treated with Pc intraperitoneally.[22] They also found a reduction in wound thickness at abdominal wall as a marker of the decreased wound healing. Our study was done on colonic anastomosis and we used different parameters for the evaluation of the anastomosis healing.

In conclusion; Pc and PI have vice-versa effects on the colonic anastomosis healing in the name of HP synthesis and ABP. Both agents seem to have changed the collagen quality and collagen quantity. Intracolonic Pc can safely be used after colorectal anastomoses to prevent tumor recurrence without afraid of anastomosis healing impairment. Although the PI seems to improve the wound healing due to increased HP production but that did not turned out to be so.

REFERENCES

1. Landis SH, Murray T, Bolden S, Wingo PA. Cancer statistics. CA Cancer J Clin 1998;48:6-29.
2. Shukla HS, Hughes LE, Davis PW, Whitehead RH, Leach KG. Distribution of 5-FU to body tissues compared after intraluminal, intravenous and intramural administration in gastrointestinal cancer. Am J Surg 1977;133:46-50.
3. Jenner DC, de Boer WB, Clarke G, Levitt MD. Rectal washout eliminates exfoliated malignant cells. Dis Colon Rectum 1998;41:1432-4.
4. Basha G, Penninckx F, Mebis J, Geboes K, Yap P. Prevention of anastomotic tumour take by on-table colon washout with povidone-iodine, an experimental study in rats. Eur Surg Res 1999;31:202-9.
5. Rosin RD, Exarchakos G, Gilmore OJ, Ellis H. Topical noxythiolin in colonic healing. Br J Surg 1978;65:603-6.
6. Horwitz SB. Taxol (paclitaxel): Mechanisms of action. Ann Oncol 1994;5(Suppl 6):S3-6.
7. Sheppard BC, Rutten MJ, Meichsner CL, et al. Effects of paclitaxel on the growth of normal, polyposis, and cancerous human colonic epithelial cells. Cancer 1999;85:1454-64.
8. Reddy GK, Enwemeka CS. A simplified method for the analysis of hydroxyproline in biological tissues. Clin Biochem 1996;29:225-9.
9. Gulluoglu BM, Kurtel H, Gulluoglu MG, et al. Role of endothelins in trinitrobenzene sulfonic acid-induced colitis in rats. Digestion 1999;60:484-92.
10. Basha G, Penninckx F, Mebis J, Filez L, Geboes K, Yap P. Local and systemic effects of intraoperative whole-colon washout with 5 per cent povidone-iodine. Br J Surg 1999;86:219-26.
11. Erbil Y, Çalış A, Berber E, Mercan S. The effect of intraoperative colonic lavage with NG-nitro-L-arginine methyl ester (L-NAME) on anastomotic healing in the presence of left-sided colonic obstruction in the rat. Surg Today 2000;30:421-5.
12. Single-stage treatment for malignant left-sided colonic obstruction: A prospective randomized clinical trial comparing subtotal colectomy with segmental resection following intraoperative irrigation. The SCOTIA Study Group. Subtotal Colectomy versus On-table Irrigation and Anastomosis. Br J Surg 1995;82:1622-7.
13. Dudley HA, Radcliffe AG, McGeehan D. Intraoperative irrigation of the colon to permit primary anastomosis. Br J Surg 1980;67:80-1.
14. Foster ME, Johnson CD, Billings PJ, Davies PW, Leaper DJ. Intraoperative antegrade lavage and anastomotic healing in acute colonic obstruction. Dis Colon Rectum 1986; 29:255-9.
15. Pollock AV, Playforth MJ, Evans M. Peroperative lavage of the obstructed left colon to allow safe primary anastomosis. Dis Colon Rectum 1987;30:171-3.
16. Pahlman L. Open trials in colorectal cancer. Eur J Surg Oncol 1995;21:347-51.
17. de Waard JW, Wobbes T, de Man BM, van der Linden CJ, Hendriks T. The effects of 5-fluorouracil and interferon-α on early healing of experimental intestinal anastomoses. Br J Cancer 1996;74:711-6.
18. de Waard JW, Wobbes T, de Man BM, van der Linden CJ, Hendriks T. Postoperative levamisole may compromise early healing of experimental intestinal anastomoses. Br J Cancer 1995;72:456-60.
19. Rowinsky EK, Cazenave LA, Donehower RC. Taxol: A novel investigational antimicrotubule agent. J Natl Cancer Inst 1990;82:1247-59.
20. Valenti AM, Niero A, Monti G, Marangolo F, Marangolo M. Paclitaxel and N-methylformamide: In vitro interactions in human colon cancer cell line. Anticancer Res 1997;17:2491-7.
21. Carrico TJ, Mehrhof AI, Cohen IK. Biology of wound healing. Surg Clin North Am 1984;64:721-33.
22. Hopkins MP, von Gruenigen VE, Holda S, Weber B. The effect of intermittent release intraperitoneal chemotherapy on wound healing. Am J Obstet Gynecol 1997;176:819-23.