European Journal of Obstetrics & Gynecology and Reproductive Biology 149 (2010) 195–198

Contents lists available at ScienceDirect

European Journal of Obstetrics & Gynecology and Reproductive Biology

journal homepage: www.elsevier.com/locate/ejogrb

A new approach to postoperative peritoneal adhesions: Prevention of peritoneal trauma by aloe vera gel

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ARTICLE INFO

Article history:
Received 6 May 2009
Received in revised form 24 September 2009
Accepted 23 November 2009

Keywords:
Adhesion
Aloe vera
Peritoneal
Prevention
Postoperative
New
Approach

ABSTRACT

Objective: Covering peritoneal surfaces with aloe vera gel may prevent peritoneal trauma and hence postoperative peritoneal adhesions.
Study design: Forty Wistar albino out-bred female rats (mean weight, 180 ± 25 g; mean age, 6 months) were divided into four groups. In Group 1, 0.1 mL aloe vera gel was injected into the peritoneal cavities. In Group 2, peritoneal adhesions were induced. In Group 3, adhesions were induced and the modeled area was covered by 0.1 mL aloe vera gel. In Group 4, the area was covered with aloe vera gel prior to adhesion induction. The rats were sacrificed on postoperative day 10 and the adhesions were scored both microscopically and macroscopically.
Results: The mean macroscopic adhesion score in the four groups was 0, 5.8 ± 0.42, 5.2 ± 0.79, and 1.1 ± 1.2 respectively, with the difference between Group 4 and Groups 2 ($p < 0.001$) and 3 ($p < 0.05$) statistically significant. The mean histopathological fibrosis values were significantly higher in Group 3 than in Group 4 (2.6 ± 0.51 vs 1.2 ± 0.91, $p = 0.002$).
Conclusion: Aloe vera gel can effectively decrease adhesion formation if applied before, but not after, after peritoneal trauma. This effect is likely due not to its chemical properties but to its viscosity, providing a covering to prevent peritoneal trauma.

1. Introduction

Postoperative peritoneal adhesions (PPAs) are the most frequent causes of intestinal obstruction, infertility, and abdominal and pelvic pains [1]. Various materials and/or techniques have been investigated but no effective solution has been identified to date.

Aloe vera (synonym: *Aloe barbedensis* Miller) is a plant with yellow flowers and triangular leaves, similar to cactus. It belongs to the Liliaceal family, which comprises 360 species [2]. Plant leaves contain abundant amounts of mucilaginous fluid of high viscosity, called aloe vera gel. Peripheral bundle sheath cells of leaves contain a less viscous liquid, called aloe vera juice or aloe vera sap [2].

The aloe vera plant contains 75 potentially active substances, including vitamins, enzymes, minerals, sugars, lignin, saponins, salicylic acids, and amino acids [3]. Owing to its rich content, it is used in the treatment of many clinical diseases and has been found to be effective in many pathological conditions [4–6]. Among these is its positive wound-healing effect. Aloe vera gel accelerates many internal (e.g. peptic ulcer) and external (e.g. dermal/subdermal) wound-healing processes [6,7]. We have therefore assessed whether aloe vera can prevent PPAs, by applying aloe vera gel to the peritoneal cavity before and after the induction of traumatic adhesions.

2. Materials and methods

This research was approved by the local ethics committee for experimental animals and was performed at the Experimental Animal Production and Research Laboratory of Cerrahpasa Medical School, Istanbul University, Turkey.

Forty Wistar out-bred female albino rats (mean weight, 180 ± 25 g; mean age, 4.5 months) were divided into four groups of 10 rats each, providing a sample power of 0.9 with a 95% confidence interval. Group 1 rats were injected with 0.1 mL aloe vera gel into the peritoneal cavity using 22 French syringes. In Group 2 rats, standard intraperitoneal adhesions were induced by laparotomy. In Group 3 rats, the area of adhesion was covered with 0.1 mL aloe vera gel. In Group 4 rats, 0.1 mL aloe vera gel was used to cover the area prior to induction of adhesion.

2.1. Anesthesia

Each rat was placed in a jar containing ether for 40–60 s, followed by intramuscular injection with 40 mg/kg body weight ketamine (Ketalar®, Parke Davis and Co. Inc.).

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doi:10.1016/j.ejogrb.2009.11.019

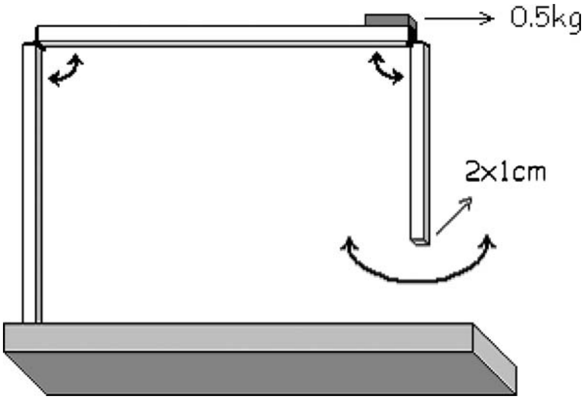

Fig. 1. Standard experimental peritoneal adhesion model constitution apparatus.

2.2. Adhesion model

All adhesions were induced using the apparatus described, which had been designed by the authors of this study to induce experimental peritoneal adhesion in small test animals (Fig. 1). The setup consists of a surgical table of size 20 cm × 10 cm, with three arms on this table, a vertical stable arm, a vertical moving arm and a horizontal moving arm. The vertical stable arm works as a shaft and fixes the other two arms onto the surgical table. A weight of 0.5 kg is positioned on the part of the horizontal moving arm most distant from the shaft. This weight corresponds to the pressure applied by a surgeon's fingertips to the surface of the intestine during manipulation throughout laparotomies.

The horizontal moving arm transmits the pressure effect of the weight to the lower part of the system by providing vertical movements of the system. The other moving arm, with a free pendulum movement in the vertical line, applies mechanical trauma (abrasion) to the peritoneal surface below. The size of the arm surface in contact with the peritoneal area was 2 cm × 1 cm, which corresponds approximately to the area of an adult finger in contact with a surface (2 cm^2). During construction of the model, a finger of a sterile and powder free latex glove was put on this surface, thus sterilizing the area and augmenting the human finger simulation (Fig. 2).

The front surface of the cecum was placed under the vertical moving arm and visceral peritoneal trauma was provided by pendulum movement of this arm ten times.

Fig. 2. Standard peritoneal adhesion creation apparatus on movement.

2.2.1. Group 1

Following short-term general anesthesia with ether, 0.1 mL sterile aloe vera gel (Natural Aloe Vera Gel® Arifoglu A.S.) was percutaneously injected into the peritoneal cavity using a standard plastic syringe with a diameter of 22 French through a point above the midline of the anterior abdomen wall and 3 cm below the xiphoid. After 6 h, the rats were allowed food and water.

2.2.2. Group 2

Rats under general anesthesia were placed on their backs on the operating table, and their extremities were fixed on the surface by wound plasters. The midline of abdomen was shaved, and the area was coated with povidone iodine solution (Betadine®, Kurtsan Co.). The peritoneal cavity was penetrated through a vertical midline incision of 3 cm. The cecum was pulled out of the abdomen, and the anterior wall was placed face upwards over the left index finger of the surgeon. While in this position, the adhesion setup was placed just below the vertical moving arm. The surgeon moved this arm back and forth, initiating free oscillation of the pendulum, which was repeated ten times. The cecum was replaced into the abdomen, and the incision was closed with 000 polypropylene sutures (Prolene®, Dogsan Co.) using a continuous suture technique. After 6 h, the rats were allowed food and water.

2.2.3. Group 3

The protocol was identical to that in Group 2 except that, following induction of the adhesions, 0.1 mL sterile aloe vera gel was applied to this area.

2.2.4. Group 4

The protocol was identical to that in Group 2 except that the surface of the cecum was covered with 0.1 mL sterile aloe vera gel before induction of adhesions.

All rats were sacrificed 10 days postoperatively by ether inhalation for about 10 min. The peritoneal cavity of each rat was penetrated via a "reverse U" incision. Without damaging the formed adhesions, the anterior abdomen wall flap was pulled caudally. Adhesions were graded according to size and severity (Table 1). Other intraperitoneal pathologies were also recorded. The 2-cm^2 area of cecum, where the model was formed, was resected for histopathological evaluation and conserved in glass bottles containing formol.

The primary outcome measure of this study was adhesion score, which is the sum of adhesion severity and adhesion size grading. The secondary outcome measure was fibrosis grading of the tissue samples extracted from the adhesion model area.

2.2.5. Statistical evaluation

Statistical analyses were performed using GraphPad Prisma V.3 software. Results were evaluated with a confidence interval of 95% and $p < 0.05$ level. In addition to descriptive statistical methods (mean, standard deviation, median), the Kruskal Wallis (KW) test for inter-group comparisons, the Dunn's multiple comparison test for comparison of subgroups, and the Chi-Square (χ^2) test and

Table 1
Definitions of size and severity grades of the peritoneal adhesions.

Grades	Adhesion size	Adhesion severity
0	No adhesion	No adhesion
1	Presence of adhesion in 25% of the model area	Spontaneously separating adhesion
2	Presence of adhesion in 50% of the model area	Separation of adhesion with traction
3	Whole model area covered with adhesion	Separation of adhesion with a sharp dissection

Table 2
Adhesion size grades of the groups (χ^2: 66.6, p: 0.0001).

Adhesion size grades	Group 1 (n)	Group 2 (n)	Group 3 (n)	Group 4 (n)
0	10	0	0	5
1	0	0	0	5
2	0	0	5	0
3	0	10	5	0

Table 3
Adhesion severity grades of the groups (χ^2: 48.8, p: 0.0001).

Adhesion severity grades	Group 1 (n)	Group 2 (n)	Group 3 (n)	Group 4 (n)
0	10	0	0	5
1	0	0	0	4
2	0	2	3	1
3	0	8	7	0

Fisher's exact test for comparison of nonparametric variables were used for evaluation of the data.

3. Results

The total adhesion scores of all subjects in all four groups are shown in Tables 2 and 3, and the mean and median scores of the groups are shown in Table 4. Group 1 was used specifically to assess whether aloe vera itself forms adhesions or has an effect on intraperitoneal pathology and no such effect was observed, so this group was excluded from further analyses.

Adhesion size and severity were significantly lower in Group 4 than in Group 2 ($p < 0.05$ and $p < 0.01$, respectively) and significantly lower in Group 4 than in Group 3 ($p < 0.05$ and $p < 0.01$, respectively). Groups 2 and 3 did not differ in total adhesion scores ($p > 0.05$). Total adhesion sizes and scores were significantly lower in Group 4 than in the combination of Groups 2 and 3 ($p < 0.001$ and $p < 0.05$, respectively).

The mean histopathological fibrosis score was significantly lower in Group 4 than in Group 3 (1.2 ± 0.91 vs 2.6 ± 0.51, $p = 0.002$).

4. Discussion

PPAs are caused by any kind of damage (including mechanical, ischemic, chemical, infective or inflammatory) to the peritoneum, which consists of a single layer of mesothelial cells. During PPA, there is fibrin-rich exudation into the damaged region. The fibrins form bands between surfaces that are in contact with this region. Fibrin bands and a hyaluronic acid-rich matrix, which fills the cavity between these bands, provide a suitable environment for collagen synthesis. Real adhesions occur after synthesis of collagen [1,5,8–10].

Several methods, materials, and agents have been assessed for their ability to prevent PPAs, including various surgical procedures, minimally invasive and laparoscopic techniques, pharmacological agents that target fibrin formation; and liquids, gels, and solids that can form a mechanical barrier between mesothelial surfaces. Although some of these methods or agents have been found beneficial, complete success was not achieved [1]. Recent studies have focused on the formation of a mechanical barrier between peritoneal surfaces. Among the substances tested as barriers are nonabsorbable solid materials (such as amniotic membrane), solid materials absorbed after liquefying at body temperature (such as hyaluronic acid), and liquid materials (such as methylene blue) [11–13]. Use of these materials is relatively easy and cheap, as well as suitable for peritoneal physiology, making them useful to prevent PPAs [14].

Various experimental PPA models have been developed, including those involving mechanical trauma (abrasion), local excision of the peritoneum, ischemic injury, placement of a foreign body (such as talcum powder) into the peritoneal cavity, thermal injury and bacterial contamination. We have utilized the mechanical trauma model, caused by a special apparatus developed by our group, owing to its ability to mimic the mechanical trauma that occurs after laparotomies. Any manipulation performed by hands or surgical instruments during laparotomy constitutes a mechanical trauma and this is the most frequent cause of PPAs [1,8].

There are two primary methods that can be used to prevent PPAs: preventing peritoneal trauma and preventing the traumatized peritoneal surface from adhering to any other surface. Manipulation at the first stage is simpler because it only requires formation of a physical barrier to prevent trauma, and more effective because inflammation and wound-healing processes occurring after trauma formation are quite complex and include unknown stages. Without clarification of the underlying physiopathology of these processes, their chance of success will be relatively low and probably incidental. Furthermore, whatever material/method is used after the occurrence of trauma, it should have no toxic effect on peritoneal mesothelial cells, as well as accelerating wound healing and/or preventing adhesion of mesothelial surfaces until completion of wound healing. Optimization of all these factors is difficult.

Aloe vera has been used clinically to prevent the formation of peptic ulcers and to accelerate their healing [15,16] owing to its cytoprotective [17], anti-inflammatory [19] and healing-accelerating effects [18,19]. Aloe vera also accelerates the healing of other types of wounds owing to its anti-inflammatory effects [20]. Polysaccharides in the structure of aloe vera have an important role in these anti-inflammatory effects. For example, acemannan, a beta-1,4-linked acetylated mannan interspersed with O-acetyl groups, is the most common polysaccharide fraction in the structure of the plant [21]. Aloe vera also has a radioprotective effect, mediated by the inhibition of pro-apoptotic protein expression, inhibition of over-expression of anti-apoptotic proteins, and inhibition of cell-cycle perturbation and regulation of p53 [22]. Aloe vera gel was routinely used for protection from the dermatologic side effects of radiation by 50% of the institutes affiliated with the Radiation Therapy Oncology Group [23]. In addition, aloe vera has been reported to have immunoregulatory activity [24] and to inhibit tumor growth in mice [25].

We tested aloe vera gel in our adhesion model because it has positive effects on wound healing and because it is a viscous liquid. Our first aim was to assess whether aloe vera has toxic effects in intact (not exposed to trauma) peritoneal cavity, by injecting 0.1 mL aloe vera into the peritoneal cavity of ten rats. In this group, we did not apply aloe vera via laparotomy to these rats because the trauma of the laparotomy incision may have given rise to adhesions, thus giving false-positive results. During repeat laparotomies in these rats, we did not observe any adhesion or any findings consistent with toxic reaction in the peritoneal cavity.

Table 4
Adhesion scores of the groups (KW: 33.71, p: 0001).

Groups	Mean ± standard deviation	Median
Group 1	0.0 ± 0.0	0
Group 2	5.8 ± 0.42	6
Group 3	5.2 ± 0.79	5
Group 4	1.1 ± 1.20	1

When we applied aloe vera over the region in which adhesion was induced, we found that PPAs were slightly reduced compared with the control group but that this difference was not statistically significant, indicating that aloe vera did not have positive wound-healing effects after trauma to peritoneal mesothelial cells. However, when the cecal surface was covered with aloe vera gel before adhesion induction, there were significantly fewer PPAs compared with control rats and rats treated with aloe vera following trauma induction.

These findings could not be explained by the chemical composition of aloe vera gel, because similar results would have been observed when aloe vera was applied before and after trauma induction. It is therefore likely that our results can be explained by the ability of aloe vera to protect the area to be exposed to trauma by forming a surface layer. This finding is supported by our histopathologic evaluation of fibrosis.

Application of aloe vera in the abdominal cavity is new, so its effects on gastrointestinal anastomoses, peritoneal vascular and lymphatic systems, abdominal incision healing process, etc., have not been investigated before. These subjects need investigation in the future.

In conclusion, our findings indicate that an aloe vera covering can decrease PPAs if applied before trauma formation. Moreover, covering peritoneal surfaces with viscous liquids such as aloe vera, which do not have toxic effects on vital tissues, before peritoneal trauma can decrease PPA formation. This is particularly true for peritoneal mesothelial cells (for example, during laparotomy).

References

[1] Senthilkumar MP, Dreyer JS. Peritoneal adhesions: pathogenesis, assessment and effects. Trop Gastroenterol 2006;27:11–8.
[2] Vogler BK, Ernst E. Aloe vera: a systematic review of its clinical effectiveness. Br J Gen Pract 1999;49(447):823–8. Oct; Review.
[3] Atherton P. Aloe vera revisited. Br J Phytotherapy 1998;4:176–83.
[4] Newall CA, Anderson LA, Phillipson JD. Herbal medicines. A guide for healthcare professionals. London: The Pharmaceutical Press; 1996.
[5] Townsend J. Aloe vera. The UK reference guide to complementary medicine. London: Chartwell House Publishing; 1998.
[6] Davis RH, Leitner MG, Russo JM, Byrne ME. Wound healing. Oral and topical activity of aloe vera. J Am Podiatr Med Assoc 1989;79:559–62.
[7] Davis RH, Leitner MG, Russo JM. Aloe vera. A natural approach for treating wounds edema and pain in diabetes. J Am Podiatr Med Assoc 1988;78: 80–8.
[8] Davey AK, Maher PJ. Surgical adhesions: a timely update, a great challenge for the future. J Minim Invasive Gynecol 2007;14:15–22.
[9] Ellis H. The causes and prevention of intestinal adhesions. Br J Surg 1982;69:241–3.
[10] Holmdahl L, Risberg B, Beck DE, et al. Adhesions: pathogenesis and prevention-panel discussion and summary. Eur J Surg Suppl 1997;577:56–62.
[11] Tayyar M, Turan R, Ayata D. The use of amniotic membrane plus heparin to prevent postoperative adhesions in the rabbit. Tokai J Exp Clin Med 1993;18:57–60.
[12] Ito T, Yeo Y, Highley CB, Bellas E, Benitez CA, Kohane DS. The prevention of peritoneal adhesions by in situ cross-linking hydrogels of hyaluronic acid and cellulose derivates. Biomaterials 2007;28:975–83.
[13] Dinc S, Ozaslan C, Kuru B, et al. Methylene blue prevents surgery-induced peritoneal adhesions but impairs the early phase of anastomotic wound healing. Can J Surg 2006;49:321–8.
[14] Matthews BD. Absorbable and nonabsorbable barriers on prosthetic biomaterials for adhesion prevention after intraperitoneal placement of mesh. Int Surg 2005;90:30–4.
[15] Blitz JJ, Smith JW, Gerald JR. Aloe vera gel in peptic ulcer therapy: preliminary report. J Am Osteopathic Assoc 1963;62:731–5.
[16] Kandil A, Gobran W. Protection of gastric mucosa by aloe vera. Bull Islam Med 1982;2:503–11.
[17] Mahattanadul S. Antigastric ulcer properties of aloe vera. Songklanakarin J Sci techno 1996;18:49–57.
[18] Robert A, Neamis JE, Lancaster C, Hamchar AJ. Cytoprotection by prostaglandins in rats. Gastroenterology 1979;77:433–43.
[19] Teradaire R, Shinzato M, Beppu H, Fujita K. Antigastric ulcer effects in rats of aloe arborescens Miller var. natalensis berger extract. Phytotherapy Res 1993;7:34–6.
[20] Strickland FM, Daevill A, Albersheim P, Eberhald S, Pauly M, Pelley RP. Inhibition of UV-induced immune suppression and interleukin-10 production by plant oligosaccharides and polysaccharides. Photochem Photobiol 1999;69:141–7.
[21] Pugh N, Ross SA, El Sohly MA, Pasco DS. Characterization of aloeride. A new high molecular weight polysaccharide from aloe vera with potent immunostimulatory activity. J Agric Food Chem 2001;49:1030–4.
[22] Wang Z, Zhou J, Huang ZS, et al. Aloe polysaccharides mediated radioprotective effect through the inhibition of apoptosis. J Radiat Res 2004;45: 447–54.
[23] Fisher J, Scott C, Stevans R, et al. Randomised phase III study comparing best supportive care to biafine as a prophylactic agent for radiation-induced skin toxicity for women undergoing breast irradiation: radiation therapy oncology group (RTOG) 97-13. Int J Radiat Oncol Biol Phys 2000;48:1307–10.
[24] Strasser A, O'Connor L, Dixit VM. Apoptosis signals. Annu Rev Biochem 2000;69:217–45.
[25] Wu W, Yang RJ, Kong XT. Effects of estradiol on radiation-induced apoptosis in immunocytes of mouse. Chin J Radiol Med Prot 2000;20:98–101.