Repair mechanisms in abdominal tissue

Chapter 10

Summary

Tissue healing is a biological response of traumatized tissues to restore the structural origination and normal function after surgical injury. Failure of the restore process in abdominal fascial incision can result in incisional hernia (IH) formation while over-restored fibrous tissues can form intraperitoneal adhesions. IH and intraperitoneal adhesions are two of the most common complications following laparotomies. This thesis aims to investigate the normal healing process of abdominal fascia overtime and the efficacy of antiadhesive barriers that could prevent the formation of intraperitonal adhesions.

Animal experiment plays an indispensable role in translational research, although alternative models have been developed. High quality of paper reporting maximizing details of animal experiments can increase the reproducibility of the study and decrease the number of animal use. **Chapter 2** presents the effect of ARRIVE-2010 and ARRIVE-2020 ((Animal Research: Reporting of In Vivo Experiments) guidelines on improving the reporting quality of articles using IH animal models repaired with meshes. Compared to articles published in 5 years before the publication of ARRIVE-2010 guidelines (pre-ARRIVE), articles published in the latest 5 years until the update of ARRIVE-2020 guidelines (post-ARRIVE) were found with significantly higher mean numbers of fully reported (sub)items listed in ARRIVE. Compared with pre-ARRIVE articles for per (sub)item, post-ARRIVE articles were significantly different in a few (sub)items, mostly reported with improvements while few reported worse post-ARRIVE. A large number of (sub)items remained unreported or inadequately reported post-ARRIVE. This was also found in articles published in the latest year using ARRIVE-2020 as the scoring criteria. Therefore, ARRIVE-2010 guidelines may have a limited effect on improving the reporting quality of animal experiments. There is much room left for further improvement of reporting for animal experiments. Furthermore, the reporting quality of articles between pre-ARRIVE and post-ARRIVE were also compared for specific species (rats, rabbits, and pigs, respectively), suggesting unbalanced effects of ARRIVE guidelines on the improvement of reporting in animal studies.

Animal gender should be taken into account in the design of animal experiments. **Chapter 3** demonstrates the difference between male and female rats regarding the intraperitoneal adhesion formation as well as normal abdominal wall healing rate. No gender-related difference of adhesion score and abdominal wall wound score were identified between male and female rats using an ischemic-button adhesion model. However, we found male rats seem to gain weight faster than females during the one-week follow-up postsurgery.

Failure of fascial healing in abdominal incision can form IH, while over-restored fibrous tissues can lead to intraperitoneal adhesions. Despite the importance of normal

fascial healing, few studies have been focusing on fascial healing process. **Chapter 4** displays the temporal changes of cellular population and gene expression levels in normal fascial healing. Animals received laparotomy closed with a single layer continuous suturing technique for fascia were allocated evenly to four different time points. Granulocytes macrophages were found with significant alteration during the normal healing process. Gene expression levels of markers for the two subtype macrophages (classically activated macrophages and alternatively activated macrophages) were significantly altered at three postoperative days. These results indicate that the inflammatory cells predominate from granulocytes to macrophages in early postoperative days. Furthermore, macrophage polarisation , a switched predomination from classically activated macrophages to alternatively activated macrophages, occurs in early fascial healing process.

Besides cells in the normal fascial healing, the temporal changes of lipid molecules in the healing process are important as potential treatment targets promoting fascial healing. **Chapter 5** focuses on the investigation of changes of lipid spatial distribution related to inflammatory cells. Animals healed normally were sacrificed at seven different time points. Lipids were analyzed using matrix‐assisted laser desorption/ionization combined with time‐of‐flight mass spectrometers (MALDI-TOF). The relationship between the intensity of any *m/z* species and the healing time was analysed with linear regression using R. A significant difference was found in the analysis in a total of 35 mass to charge (m/z) values for negative ion mode and a total of 18 m/z values for positive ion mode. Five different lipid species, belonging to phosphatidylcholines, phosphatidylethanolamines, and gangliosides, were found with correlation to inflammatory response or fibroblast growing postsurgery. These lipid species are potential biomarkers for predicting incisional hernia and could be the therapeutic targets preventing incisional hernia.

Chapter 6 compares the antiadhesive efficacy of five novel antiadhesive barriers using an ischaemic button animal model. These physical barriers were placed intraperitoneally isolating internal organs and peritoneum from ischaemic buttons. One of these antiadhesive polymer barriers (A2 barrier) was found with significantly lower numbers of buttons involved with adhesions in comparison to others. However, no significant difference of adhesion score was found between these antiadhesive barriers, using Nair's score (quantity score) or Zühlke score (quality score)**.** Therefore, the antiadhesive efficacy of A2 barrier needs further investigation. **Chapter 7** evaluates the antiadhesive efficacy of a novel hyaluronic acid gel using the aforementioned ischaemic button model. Compared to the hyaluronic acid/carboxymethyl cellulose gel, the novel gel significantly reduced intraperitoneal adhesions only when measured with the number of involved organs. Results from pathophysiological analysis of the

adhesion tissues together with ischaemic buttons indicate a prolonged activation of alternatively activated macrophages (M2-like phenotypes).

Antiadhesive barriers coated on meshes are commonly used to prevent tissue-to-mesh adhesion formation. A large number of previous animal experiments were used to test the antiadhesive efficacy of mesh coatings, lacking consistent results. **Chapter 8** summarizes the antiadhesive performance of mesh coatings. A systematic review and meta-analysis was performed aiming to make comparisons between meshes with coatings and meshes without coatings. In our integrated results, polypropylene meshes coated with hyaluronic acid/carboxymethyl cellulose showed a significant reduction of intraperitoneal adhesion formation compared to polypropylene meshes. Heterogeneity was low for this integration. There was no significant reduction of adhesion formation for polyester meshes coated with collagen compared to polypropylene meshes. However, heterogeneity for this comparison was high while the source of the heterogeneity was complex. These findings indicate that hyaluronic acid/carboxymethyl cellulose as a mesh coating shows antiadhesive property in animal models.