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Early Experience with a Novel Gelatine-based Sponge for Local Haemostasis in Thyroid Surgery

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Early Experience with a Novel Gelatine-based Sponge for Local Haemostasis in Thyroid Surgery

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Abstract. *Aim: The aim of the present investigation was to assess the feasibility, efficacy and safety of a novel gelatine-based sponge as a haemostat in thyroid surgery. Patients and Methods: A questionnaire was completed by surgeons after having used the sponge in thyroid surgery. The product in general, its effectiveness as a haemostat, its absorption capacity and handling issues were rated. Moreover unexpected complications or side-effects were documented. Results: Whenever thyroid resections were performed by the members of our study group (11 consultant surgeons in 8 hospitals specialized in thyroid surgery) the new haemostat was used during the period of surveillance. It was mainly rated as “excellent” or “good” by the study group members who used the product in 87 thyroid resections. Its effectiveness as a haemostat, its absorption capacity and handling issues were also rated as excellent. No poor results were reported. Complications occurred in only 2% of cases and were related to inappropriate application. Conclusion: The evaluated data demonstrated that the sponge has an excellent safety and haemostatic efficacy in surgical application. The product is user-friendly and demonstrated its effectiveness as a haemostat and its excellent absorption capacity.*

Uncontrolled major or minor bleeding after or during surgical procedures is still a problem not to be underestimated in daily surgical routine. Especially in thyroid surgery, a dry operating field before closing the wound is crucial because of dangerous complications in the patients’ neck area.

One possible option to prevent bleeding complications is using a haemostat (1). While in some countries surgeons are more used to using a haemostat in thyroid surgery, the

situation in Germany is different: Only recently have German surgeons become more open-minded regarding the use of a haemostat in thyroid surgery.

The haemostat market offers an enormous number of different products (2-4). They are differentiated by the materials used and facultative supplementation of coagulation-inhibiting substances. The materials used are made of gelatine, collagen (5) or cellulose derivatives. Some of them have an active component, such as fibrinogen or thrombin (6-8).

In the present study we report on experience using a novel gelatine-based sponge with improved features such as faster haemostasis and instant blood absorption.

Patients and Methods

For this investigation a new gelatine sponge was used (GELITA-SPON® RAPID³ produced by Gelita Medical GmbH, Eberbach, Germany). This novel product is made of pharmaceutical-grade chemical cross-linked gelatine characterized by a high pore density, reduced ligaments, and high nanoscale of roughness of the surface. The sponge is free from any supplementation or any active substances.

Adequate and fast haemostasis in pre-clinical studies was described and it has been shown as a feasible option in clinical cases (9).

For proof of efficacy and safety of this product, structured clinical data were required. Therefore a questionnaire was developed to be completed by surgeons directly after use of GELITA-SPON® RAPID³ in thyroid surgery. Between May and September 2011, the novel sponge was tested in thyroid surgery in 87 patients. The questionnaire included information about the treating physician (name, position in the hospital, address data), anonymous patient data (age, gender, diagnosis, coagulation-inhibiting medical treatment or other relevant drugs), and indications (subtotal thyroid resection one/both sides, thyroidectomy or other).

In addition to these general data, the questionnaire included a five-topic rating of the product (excellent, good, satisfactory, moderate and poor): the product in general, and its effectiveness as a haemostat, absorptive capacity (uptake of blood/fluid), handling in general (ready-to-use), ease of application and adaption to surrounding tissue.

Subsequently four questions followed regarding unexpected complications or side-effects, current use of a similar product, chance of prospective use of the novel sponge and any reasons preventing leaving the sponge in the patient after the procedure.

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Key Words: Gelatin-based sponge, haemostat, blood absorption, biodegradation, thyroid surgery.

Finally the benefits or disadvantages of the new product and optional other indication areas, as well as general comments, were recorded.

All 11 members of our study group were consultant surgeons with an experience of more than 10 years in general surgery. Moreover, each of them was specialized in thyroid surgery due to performance of more than 100 thyroid resections a year.

For these reasons we concluded that experienced thyroid surgeons who are familiar with the risk of bleeding after thyroid surgery were especially suited for the evaluation of the new haemostat in any kind of thyroid surgery (partial resection procedures, hemi-thyroidectomy and total thyroidectomy) for benign or malign diseases of the thyroid gland.

Whenever thyroid resections were performed by the members of our study group (11 consultant surgeons in 8 hospitals specialized in thyroid surgery) the new haemostat was used during the period of surveillance.

Every surgeon of our study group was supposed to use the haemostat in at least 5 operations. In each case a questionnaire was filled out by the operating surgeon immediately after surgery.

At the end of the surveillance we had collected data from 87 operations.

Results

The analysis of the 87 questionnaires prepared by the study group revealed a generally positive opinion regarding the product. It was rated mainly as excellent/good (73%) or satisfactory (19%), while poor results were not reported (Figure 1).

The handling in general (ready-to-use) of the specific sponge was judged as being excellent to satisfactory in 64% of enrolled patients. The applicability was reported as very good in 9%, good in 48%, and satisfying in 27% of the procedures, in contrast to 16% of moderate or poor application comfort (Figure 2).

In 71% of the surgical cases, the effectiveness of the novel sponge as a haemostat was excellent or good, while it was satisfying in 20% of the reports. Poor haemostasis was reported in only 1% of the cases (Figure 3).

The absorptive capacity for blood or liquids was assessed by the surgeons as very good in 15%, as good in 60%, and as satisfying in 23% of the procedures (Figure 4).

Adaptability to the tissue was judged as very good or good in 51% and as satisfactory in 33% of the occasions the novel sponge was applied.

The question regarding any unexpected complications was "No" in 98% of the cases. Complications occurred in only 2% and were related to inappropriate application (Figure 5).

None of the surgeons reported any reason for not leaving this gelatine sponge product in the patient after the procedure.

At 78% of the hospitals comparable haemostatic products were applied for thyroid surgery. A sponge with an active component was used in 45% and a cellulose-based haemostat in 33% of the participating hospitals, whereas 22% of the hospitals operated without the use of haemostatic materials.

Nearly two-thirds (65%) of the surgeons stated they would use GELITA-SPON® RAPID³ in the future.

Subsequently, a detailed subgroup analysis was carried out regarding the individual results of the different haemostat users by comparing thrombin sponge users, oxidized cellulose users and non-haemostat users in terms of general handling, blood absorption and haemostatic efficacy of this novel product.

While the thrombin sponge users rated general handling, blood absorption and haemostatic efficacy as very good or good from 30 to 41% of the cases, the oxidized cellulose users judged these capacities of the new sponge as very good or good in 51 to 77% of cases. The non-haemostat users were even more satisfied with this novel product. They rated general handling, blood absorption and haemostatic efficacy as very good or good in 84 to 95% of the cases.

These results of the three groups show differences in their decisions regarding using this new haemostat in the future.

While only 18% of thrombin sponge users would apply the novel sponge in the future (another 18% were still undecided), the oxidized cellulose users and non-haemostat users voted for its use in 72% and 79% of cases, respectively.

Discussion

The analysis of the results of these 87 questionnaires showed that the participating surgeons were satisfied with the performance of the new sponge. The same product had been tested for a different indication. A questionnaire-based field report of seven Ear Nose and Throat Centers in Germany with 62 patients undergoing nasal/sinus surgery was carried out (9).

The blood absorption was rated as excellent to satisfactory in 98% in thyroid surgery compared to 100% in nasal/sinus surgery. Both findings demonstrate outstanding performance of the novel product in absorptive capacity.

Haemostatic efficacy was 91% excellent to satisfactory in thyroid surgery, whilst 100% judged it to be so in nasal/sinus surgery. These results mean a high reliability of the new sponge as an effective haemostat in surgical procedures.

Complications occurred in only 2% of cases. One was caused by inappropriate application of the new product: dry use in a non-bleeding-area without any indication for the application of a haemostat. The other was related to repositioning of the product after the sponge had absorbed fluid.

The results of general handling were very good to satisfactory in 98% in nasal/sinus surgery compared to 64% in thyroid surgery. These different findings were reflected in the decisions to use the new haemostat in the future as well (nasal/sinus surgery 81%, thyroid surgery 65%).

These apparent differences between both field reports regarding satisfaction with the ready-to-use handling and prospective use were examined in an additional appraisal. In a detailed analysis of the thyroid surgery survey, the current

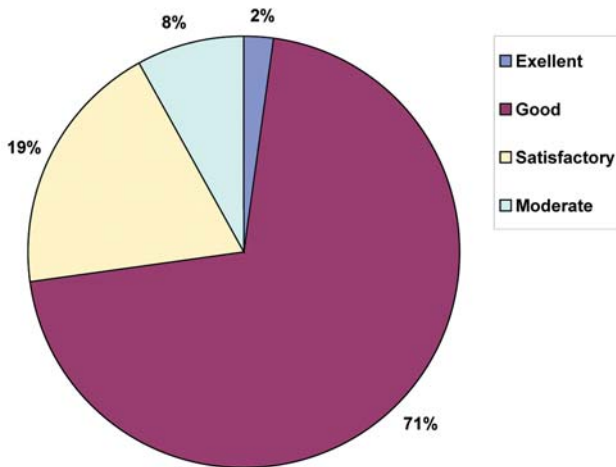


Figure 1. How do you rate the product in general?

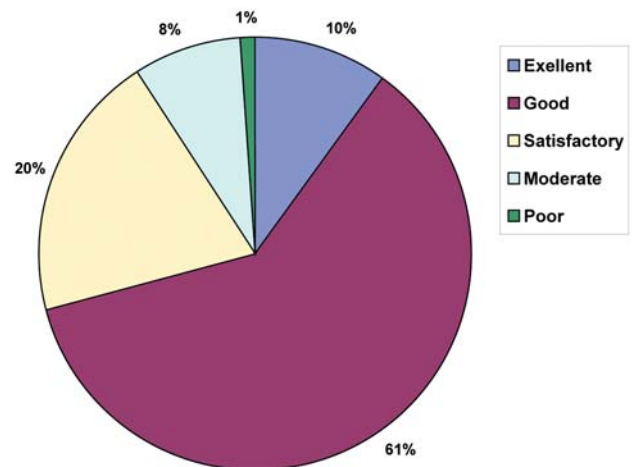


Figure 3. How do you rate the haemostatic efficacy?

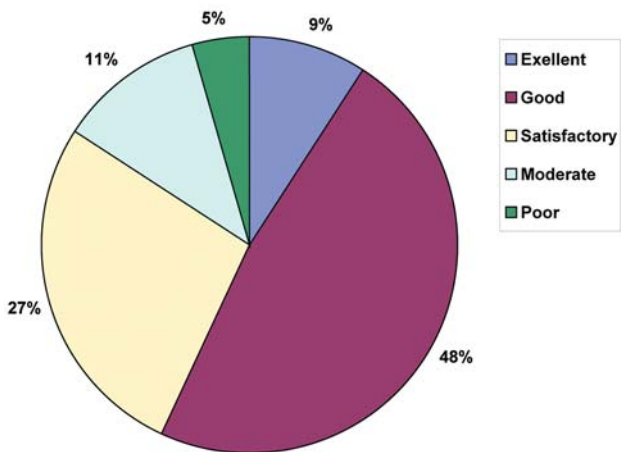


Figure 2. How do you rate the applicability?

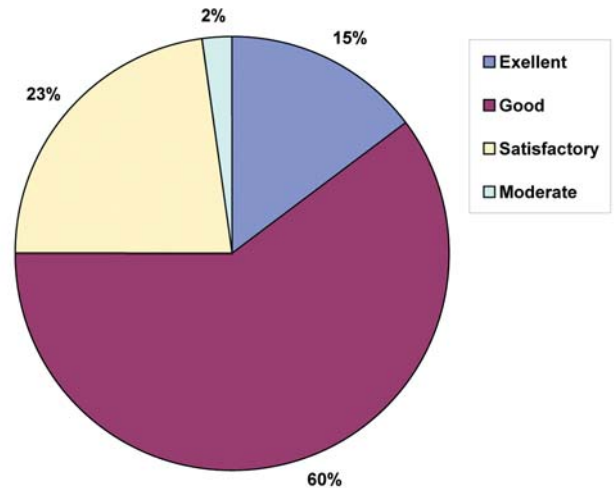


Figure 4. How do you rate the absorptive capacity (uptake of blood/fluid)?

use of haemostats for this indication in Germany was compared with the results in this recent questionnaire-based field report. While there are an estimated 10% thrombin sponge users in Germany for this indication, the proportion of such users was significantly higher in this recent survey (45%). Even the proportion of the non-haemostat users in thyroid surgery was different (22% compared to an estimated 65% in Germany). Oxidized cellulose users are estimated to be 25% in Germany; in the present report, there were 33% oxidized cellulose users.

Overall, the evaluation of the attending surgeons seems to be dependent on the current practice in using a haemostat. Whilst non-haemostat users (79%) and oxidized cellulose users (72%) seemed to be satisfied with the performance of the new

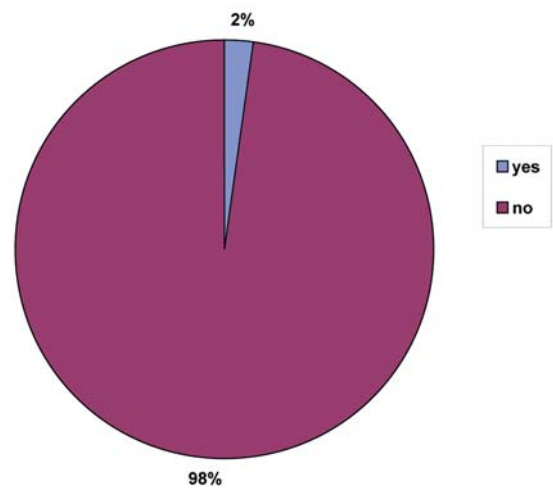


Figure 5. Were there any unexpected complications?

sponge and would use it in the future, the thrombin sponge users would not change their haemostat so easily. Apart from that, active sponges are found in a different price range.

Regarding the general handling, blood absorption and haemostatic efficacy, the non-haemostat users attested to the best results with the gelatine sponge, followed by the oxidized cellulose users. Again, the thrombin sponge users were not as satisfied with the performance of the product as the other groups, which could also be due to the familiar good performance of thrombin sponges. The disadvantage of the extensive documentation required when applying a thrombin sponge was not part of this survey.

In summary, it can be stated that more than nine out of ten (92%) users in thyroid surgery rated this product in general as satisfactory or better. This showed a high level of satisfaction with the performance of the GELITA-SPON® RAPID³.

Conclusion

The application of this sponge seems to be an improvement of haemostasis and safety in thyroid surgery. Moreover, the novel product could be an improvement in other fields of indication. However, a greater pool of clinical data is required.

Acknowledgements

In consequence of several round table discussions with experts we realized the need for an evaluation of haemostatic materials in thyroid surgery as there are still few data available.

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References

- 1 Erdogan D and Van Gulik TM: Evolution of fibrinogen-coated collagen patch for use as a topical hemostatic agent. *J Biomed Mater Res B Appl Biomater* 85: 272-278, 2008.
- 2 Haeton N: Advances and methods in liver surgery. *Hemostasis. Eur J Gastroenterol Hepatol* 17(Suppl1): 2-12, 2005.
- 3 Michel O: Generation Marshmallow - Neue Materialien zur Blutstillung. *HNO-Nachrichten* 2: 32-36, 2010.
- 4 Carbon RT: Evaluating the *in vitro* adhesive strength of biomaterials: Biosimulator for selective leak closure. *Biomaterials* 24: 1469-1475, 2002.
- 5 Walsh PN: Platelet coagulation-protein interactions. *Semin Thromb Hemost* 30: 461-471, 2004.
- 6 Holcomb JB, Pusateri AE, Harris RA, Charles NC, Gomez RR, Cole JP, Beal LD, Bayer V, MacPhee MJ and Hess JR: Effect of dry fibrin sealant dressings versus gauze packing on blood loss in grade V liver injuries in resuscitated swine. *J Trauma* 46: 49-57, 1999.
- 7 Kheirabadi BS, Acheson EM, Deguzman R, Sondeen JL, Ryan KL, Delgado A, Dick EJ and Holcomb JB: Hemostatic efficacy of two advanced dressings in an aortic hemorrhage model in Swine. *J Trauma* 59: 25-34, 2005.
- 8 Pusateri AE, Holcomb JB, Harris RA, MacPhee MJ, Charles NC, Beall LD and Hess JR: Effect of fibrin bandage fibrinogen concentration on blood loss after grade V liver injury in swine. *Mil Med* 166: 217-222, 2001.
- 9 Hajosch R, Suckfuell M, Oesser S, Ahlers M, Flechsenhar K and Schlosshauer B: A novel gelatin sponge for accelerated hemostasis. *J Biomed Mater Res Part B: Appl Biomaterials* 94B: 372-379, 2010.

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