

Splenic Implant Assessment in Trauma

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Rezumat

Evaluarea implantului splenic autolog în traumatisme

Traumatismele reprezintă o problemă de sănătate globală, fiind a IV-a cauză de deces după bolile cardio-vasculare, neoplasme și bolile cronice pulmonare și principala cauză de deces în rândul persoanelor tinere, sub 45 de ani (1). Frecvența traumatismelor abdominale este de 10-12% din totalul politraumatismelor, iar dintre organele intra-abdominale, splina și ficatul sunt cel mai adesea implicate în cazul pacienților politraumatizați (2). Primul scop al unui management operațional de succes îl reprezintă controlul hemoragiilor active, iar cel de-al doilea constă în conservarea cât mai mult posibilă a organelor distruse. În ultimele decenii, tratamentul leziunilor traumatiche ale splinei s-a diversificat, mergând de la tratament nonoperator până la tratament chirurgical, de asemenea complex și diversificat: de la tratament conservator la splenectomie. În prezent, din punct de vedere terapeutic, tendințele în trauma splenică sunt orientate spre metode de conservare fiindcă datele clinice și experimentale au arătat că "este mai bine cu toată splina decât cu o parte și mai bine cu o parte decât cu nimic" (Raymond Hinshaw) (3).

Cuvinte cheie: implant splenic autolog, trauma.

Abstract

Trauma is a global health issue, being the 4th death cause after cardio-vascular disease, malignancies and chronic pulmonary diseases and the main death cause among young people, under 45 years (1). The frequency of abdominal trauma is 10-12% of all polytrauma, and from all abdominal organs, the spleen and liver are the most often involved in polytraumatized patients case (2). The first purpose of a successful operational management is the control of active bleeding, and the second is preserving as much as possible of the destroyed organs. Over the last decades, the treatment of spleen traumas had been diversified, from nonsurgical treatment to surgical, also complex and diversified: from conservative treatment to splenectomy. Currently, from a therapeutic standpoint, the trends in spleen trauma are orientated towards conservative methods as the clinical and experimental data have shown that "it is better with the entire spleen than part of it, and better with a part of it than with none at all" (Raymond Hinshaw) (3).

Key words: autologous spleen implant, trauma

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The importance of the spleen and splenectomy consequences

Actually, the spleen is considered to be the most important peripheral immune organ, having more lymphatic tissue

than all lymph nodes from the organism. To successfully fulfil the immunological functions at least 25% from the weight of a normal size spleen must be preserved along with a proper arterial flow (4).

The spleen's functions are multiple, various, controversial and incompletely clarified. Galen considered the spleen as an "organ full of mysteries", and some of them still persist.

The spleen is the main area of early exposure of the immune system to bacteria from the blood flow. The organ has two well defined histological regions: red pulp (or erythroid region) and white pulp (lymphoid region), with distinct metabolic and immunological functions (Table 1).

Thereby it has an important role in generating immune responses to antigens from the systemic circulation, or brought here by various antigen presenting cells (blood dendritic cells). According to the nature of these antigens, either cellular immune responses, or humoral immune responses may be generated (8).

In the immune system, the spleen has another important role: opsonin generation – serum factors which favors phagocytosis of foreign particles by interacting with them. While the liver's mononuclear phagocyte system is involved in opsonized particle destruction, the spleen is the only organ that can clean the blood flow of non-opsonized or insufficiently opsonized antigens and the only specialized in antibody production in a short time from antibodies' contact (9).

Another particularity of the spleen is the ability of generating tuftsin, a natural tetrapeptid which stimulates the activity and migration of phagocyte cells. In tuftsin acquired deficiency, encountered not only in splenectomy, there is an increased susceptibility to infections (8).

Characteristically, the spleen is involved in immune responses triggered against thymus-independent type 2 antigens (antigens TI-2), most of them polysaccharides, which is the main antigen component of encapsulated bacteria's capsule, *Streptococcus Pneumoniae*, *Haemophilus influenza* and *Neisseria meningitidis* types (10). Also, at spleen level occurs IgM synthesis in a short time after exposure to blood antigens, being the largest IgM producer in organism.

The spleen's role in reducing the risk of infections has been known since the 1800s. In 1911, Luckhardt, after comparative studies on healthy and splenectomized animals concludes: "these results clearly show that healthy animals produce specific hemolysins, hemagglutinins and hemopsonins faster and in higher concentrations than splenectomized animals; one conclusion is obvious, namely that the spleen has an active role in developing those immune particles " (11).

The consequences of splenectomy can be divided in immunological, effects and non-immunological, as they are summarized in Table 2.

The most important consequence of the absence of the spleen or reduction of its functions is represented by severe infections, life threatening – OPSI (Overwhelming Post Splenectomy Infections), concept introduced in 1969 by Diamond (13).

Post-splenectomy infections occur with an incidence of 0.23-0.42% / year with the highest frequency in the first 2-3

Table 1. *The spleen's functions (5,6,7)*

White pulp	Red pulp
1. Antibody synthesis	1. Filter function
2. Initiation of humoral responses	2. Phagocytosis (especially type TI.2 antigens – polysaccharide antigens of encapsulated bacteria)
3. Lymphocyte reservoir	3. Platelet and immature erythrocytes reservoir
	4. Hematopoiesis (fetal life)
	5. Tuftsin production
	6. Positive effect on serum VII factor level
	7. Participate in the reuse of Iron
	8. Inhibiting Angiotensin
	9. Complement activation

years (30% of these episodes occur in the first year post-splenectomy and 50% in the first two years). Yet, serious infections may occur after 5 or even 20 years from splenectomy (14). The studies show that a percentage of 25% of asplenic patients will develop a serious infection sooner or later (15). The late risk for OPSI/serious infections at splenectomized adults is 2.2% including trauma and incidental splenectomy and 2.1-5.9% just for trauma. The real incidence of OPSI after posttraumatic splenectomy is not well known, but it is estimated at around 1.5% (16).

In a splenectomised patient's blood a decline of IgM concentration is observed, as well as of complement activity, on the alternate pathway, due to decrease of properdin and tuftsin levels, opsonins normally synthesized by spleen (17,18).

Another consequence that increases the risk of OPSI is reducing non-opsonised particle phagocytosis by the spleen's macrophages. The spleen mononuclear phagocyte system is different from the liver regarding the necessity of particle opsonisation.

The splenectomised patients have low concentrations of IgM, an altered process of changing IgM to IgG and a poor function of B lymphocytes in type T-independent responses, and present with long term lymphocytosis. In addition, a redistribution of lymphocyte T subpopulations from the peripheral blood has been observed, as a result of splenectomy. Thereby, the percentage of T CD4+ lymphocytes decreases and CD4/CD8 ratio also decreases (19)

The spleen has an important role in purging altered/imperfect erythrocytes of certain intracellular inclusions like

Table 2. *Effects of splenectomy (12)*

Immunological	Non immunological
1. ↓ phagocytosis activity of opsonized antigens	1. ↓ filtering function
2. ↓ Tuftsin	2. ↑ the number of reticulocytes
3. ↓ serum Ig M	3. ↑ the number of platelets
4. prolonged time in blood of lymphocytes	
5. ↓ complement functions	
6. ↑ auto – antibodies activity	
7. ↓ suppressor Limfocytes T	

Howell-Jolly bodies (formed from nuclear chromatin), Pappenheimer bodies (iron granules resulting from the degradation of hemoglobin), Heinz bodies (rests of pathological hemoglobin) in malaria parasites. After splenectomy, the body's ability to filter blood is significantly reduced. The presence of Howell-Jolly bodies in peripheral blood erythrocytes is an irrefutable argument sustaining the asplenia diagnosis (20).

A consequence of spleen absence is loss of function of reticulocyte maturation which will grow in peripheral blood. The reservoir function of the spleen is also abolished by splenectomy, which will determine hyperleucocytosis with granulocytosis and monocytosis.

In the case of splenectomised patients due to trauma, abnormal blood clotting has been described. Thereby, plasminogen activator inhibitor III (PAI-III), a glycoprotein which physiologically decreases the thrombin and factor X activation, preventing thrombosis, has low serum concentrations in splenectomised patients (21,22). Natural anticoagulants and endogenous fibrinolysis may also be reduced in those patients. Of natural anticoagulants, C and S proteins, which are normally designed to inhibit V and VIII coagulation factors and stimulate fibrinolysis, are also decreased (23). Due to reduction of these anticoagulants, there is a hypercoagulability condition. Another factor involved in blood clotting, plasminogen activator inhibitor has high levels, causing a decrease of plasmin production. The result of this decrease is reduction of fibrinolysis, emphasizing the hypercoagulability condition (24).

Postsplenectomy, thrombocytosis is another condition whose cause wasn't fully understood. On the one side, the splenic platelet reservoir is eliminated, and on the other, the platelets are not destroyed in the spleen anymore. Adams has reported reaching a maximum platelet number between days 10 and 14 postoperative. However, after a few months, it seems that the platelets' number returns to normal (25). In this period aggregating treatment is recommended for preventing thrombo-embolic complications.

Methods of preserving the spleen functions

Over the last two decades, spleen trauma treatment has undergone major transformations, a particular importance being attributed to conservative treatment (surgical and non-surgical), which has the goal to preserve immunological functions.

The easiest way to preserve the spleen is non-surgical treatment (NST). Only in NST failure, surgery has to be considered, which initially is focused on conservative methods (splenorrhaphy, partial splenectomy, hemisplenectomy, „splenic wrapping”, subtotal splenectomy, splenic artery ligation) (Table 3) (16).

NST in spleen trauma is a real possibility. Good knowledge of patient's selection algorithm for applying NST and the factors that allow the assessment of this therapy failure, makes non-surgical treatment the standard treatment in spleen trauma for hemodynamic stable patients.

Autologous spleen implant - heterotopic splenic autotransplants (HSAs)

The autotransplant concept is closely linked to the term splenosis, because both occur after spleen trauma, the autotransplant being a deliberate splenosis made by surgeons after spleen removal, while splenosis is an uncontrolled seeding of spleen tissue in various locations, after trauma.

Its purpose is maintaining the spleen's immunological role and preventing fulminant infection postsplenectomy.

Autologous spleen implant indications are: 1 - Gaucher disease; 2 - splenic trauma where total/partial preserving is not possible (4, 5 degree lesions); 3 - postschistosomal portal hypertension; 4 - chronic lymphocytic leukemia and 5 - myeloid metaplasia (caused by idiopathic myelofibrosis). Practically almost all cases where total splenectomy is needed (26-31).

Spleen implant contraindications are: 1 - abbreviated laparotomy techniques („damage control”); 2 - simultaneous small bowel - large bowel lesion (the devascularized spleen part would be a suitable place for developing septic complications); 3 - older age; 4 - surgeon preference (30,31).

According to Moore, simultaneous peritoneal contamination is not a spleen implant contraindication (30).

The implant site (location)

Most authors consider that the optimal site for implant is the great omentum (26-34). This option is supported by: 1 - rich vascularisation that allows increased growth factors, cytokines and inflammatory cells intake, great omentum vascularisation providing ideal conditions for nutrition by diffusion of the implant („per diffusionem”) in regeneration and neovascularisation stages (33); 2 - blood drainage to the liver through the port system (also the normal drainage of the orthotopic spleen). In fact, things are not very clear, not being able to say with certainty the an upper regeneration of omental implanted fragments is present compared to the ones with other sites (30). Other used locations were: subfascial properitoneal (Traub, 1987), mesocolon (31), muscle and subcutaneous. Spleen tissue transplanted in an extraperitoneal space does not satisfactorily remove *Streptococcus Pneumoniae* and, therefore, doesn't

Table 3. Therapeutic strategies in spleen rupture management (16)

Therapeutic method	Manner
NOM	clinical observation angiography
ST haemostatic agents	thrombin, collagens, etc.
arterial ligation	main trunk / segmental vessels
splenorrhaphy	manual sutures, omental folded gauze pad, absorbable nets
partial splenectomy	manual sutures / mechanical, laser coagulation
hemisplenectomy	
total splenectomy	autologous implant

NOM - non operative management; ST - surgical treatment

prevent infections (33). Muscular/subcutaneous implantation ensure viability for fragments but those, functionally speaking, are not enough (32).

Implanted spleen tissue quantity

Several authors (Patel - 1982, Holdsworth - 1991, Iinuma - 1992 and Resende - 2002) (26) have suggested that 25 % of normal spleen tissue, orthotopic and vascularised, is enough for maintaining the complete spleen function. This is equivalent, in humans, to a quantity of 35 grams. Weber considers a quantity of 50 grams from the original spleen necessary, in order to provide the complete function (33).

The evolution of the implant

The sequence of histological events of autotransplant regeneration has been described on mice using optic and electronic microscopy by Tavassoli and collaborators in 1973 (35) and, later, in 1989, by Johnson & Weiss (36).

The histological evolution of implanted spleen tissue has 3 stages: 1 - necrosis, 2 - regeneration, 3 - vascularisation development.

Histological studies on mice have shown that after a few hours from surgery, the fragments become necrotic (5), excepting one thin band of reticular cells just below the capsule. All lymphocytes from the transplanted tissue die, and only a few reticular cells and erythrocytes are kept.

The surrounding vessels increase their flow growing towards vessels from inside the tissue which subsequently will make an anastomosis (37). The next days, capillary and reticular fibres grow to form vascular spaces located in the subcapsular area.

Regeneration begins by forming large spaces from the periphery of the implant (35). After one week, the regeneration tissue is divided in an internal zone and an external one. In the internal zone are reticular cells, and in the external one spleen sinuses. The lymphocytes begin to immigrate forming the red pulp after two weeks from transplant. The next weeks the white pulp compartments begin to develop themselves approximately in the same order as in ontogenesis. First of all, PALS (periarteriolar lymphoid sheath) is formed, followed by follicular and marginal zones. Later, a series of regenerative strips is developing from still viable tissue which will grow, having, after 5 weeks from the procedure, the same structure as the spleen (37). Histological characteristics of spleen implant are dependent on local blood supply (38-39).

Neovascularization responsible for implanted spleen tissue irrigation occurs the second day after transplant (38). Before the advent of anastomosis between the new vessels and old ones, the implanted spleen tissue is fed by the nearby vascularisation. Alves and collaborators (39) have made an experimental study on mice in which the spleen was removed and after that spleen implants were placed in the abdominal cavity (in the gastro-splenic ligament, mesocolon and perirenal fat). By injecting fluorescent polystyren microspheres in mice's orbital plexus, the researchers found that the microspheres

were placed at the implant's periphery, as early as 3 days from procedure. The microspheres' concentration has grown in time, being found deep in the spleen parenchyma, the outside of white pulp, red pulp and transition zone; 10 weeks from implant, the marginal zone, typical structure of any intact spleen, is absent or rudimentary (39). Microspheres were not found earlier than 3 days, in vessels from nearby tissue and not in so-called "vascular spaces" described by Felle (39), which occur by dilatation of subcapsular vessels 2 days from implant.

In conclusion, the revascularisation of splenic grafts begins after 2 days from implant, grows centripetal from periphery to inner parenchyma, and anastomosis occurs after day 3 (39).

After 3-6 months from implant, the small splenic fragments have a complete histological structure including germinal centers, showing functional capacity of white pulp (7).

Holdsworth said that splenic implants return to normal structure and fulfil their functions after 3 months from surgery, while they follow the mentioned morphological stages (necrosis, regeneration and vascular development) (40).

Although the histological architecture of transplanted spleens is similar to the normal spleen, the compartment's dimensions are different. PALS and marginal zone are reduced by 50% compared to the physiological spleen. More than that, in the central region of transplants fibrotic tissue zones occur.

The factors involved in regeneration control are not exactly known. Orthotopic spleen seems to have a suppressive effect on autotransplant regeneration, but suppressive or stimulator factors are unknown. Pabst suggested that a stimulator factor could be increased intake of nutrients to implants (5)

Surgical technique

Splenic microfragmentation technique ("splenic omelet procedure" - splenic homogenate). According to Lucas' theory, excised spleen is sectioned in multiple fragments maximum size of 2 mm. The quantity used is 50 % of spleen mass and is placed in omental "pockets" made by rolling the great omentum and fixed with dissolving threads (41).

Moore technique uses 5 spleen fragments measuring 40 x 40 x 3 mm placed in pockets made in the great omentum and possibly marked with metal clips (to facilitate paraclinical tracking). Moore has proved low rate of complications in 43 patients compared with other methods of spleen saving. He did not reveal complications related to spleen implant (33).

Ionescu technique uses 20 x 25 x 5 mm decapsulated fragments (42).

Di Carlo I technique: after splenectomy, the organ was weighed and the undamaged part was cut transversely to prepare a segment of approximately 4 x 3 x 2 cm in size and of 35 g of weight to be transplanted. The greater omentum was pedunculated in its left lateral portion, and the previously prepared splenic tissue was implanted in a pouch created at the lower edge of the omentum. The omental peduncle containing the splenic tissue was fixed to the

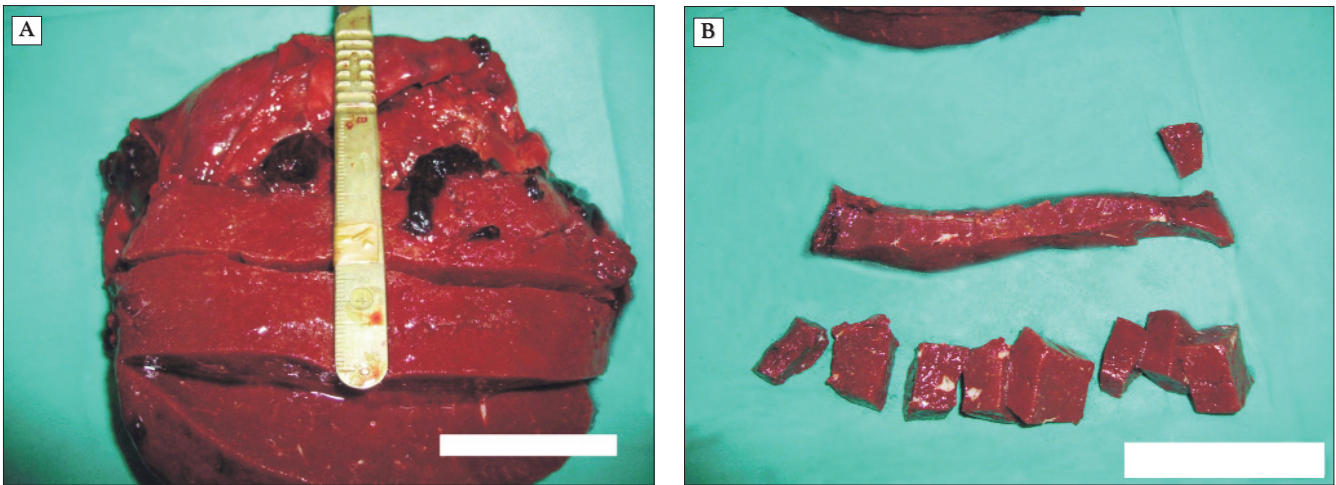


Figure 1. IIIrd degree spleen rupture (A) the incision of splenic tissue for preparing the implant fragments; (B) sectioned splenic fragments) – personal collection

parietal peritoneum of the posterior left upper quadrant of the abdomen where the native spleen was previously located (43).

Petroianu technique is actually used in Emergency Clinical Hospital. Average duration of the procedure is 5 minutes, practically being indicated in all cases of total splenectomy (excepting lesion control surgery). 20 splenic fragments are obtained, measuring 1-2 cm (1 x 1 x 2 cm), from which the capsule will be removed, that allowing the intimate contact between fragments and receiving zone and promoting angiogenesis. The weight of a fragment is 2-4 g (Fig. 1A and B).

Total weight of implanted fragments is more than 50 grams. The fragments are put in physiological serum (the serum is at room temperature). The great omentum is displayed, and on the front a wet dressing is applied. The fragments are sutured (through a simple surjet suture) on the outer layer of the great omentum strung on a catgut thread 3-0 (original technique) in "S" shape or in two parallel rows (splenic fragment - omentum - splenic fragment-omentum ...). Actually Vicryl 3-0 thread is used (Fig. 2).

The great omentum is folded over these fragments (with or without suture) so as not to come in direct contact with the anterior parietal peritoneum (Fig. 3).

Omental suture is not necessary because, in 24 hours when the patient can be mobilised, omento-parietal adhesions are formed (27-28).

Implant's complications

In 1988, Rhodes (41) describes for the first time a complication occurred after an autologous spleen implant by microfragmentation method. It is about an omental abscess in a patient to whom distal pancreatectomy and splenectomy for a pancreatic pseudocyst was performed, for whom a reintervention was performed on the 11th day postoperatively in order to evacuate it. The author suggested that coagulation necrosis is a fragile period with increased susceptibility to infections, and so, the

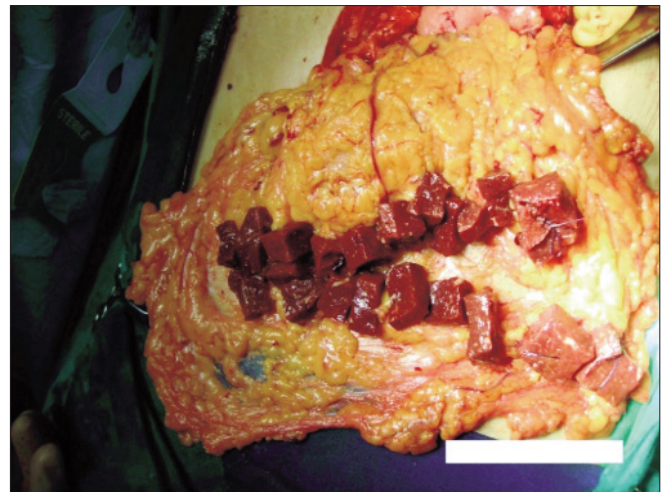


Figure 2. Splenic implants in the great omentum – arrangement in two parallel lines – personal collection

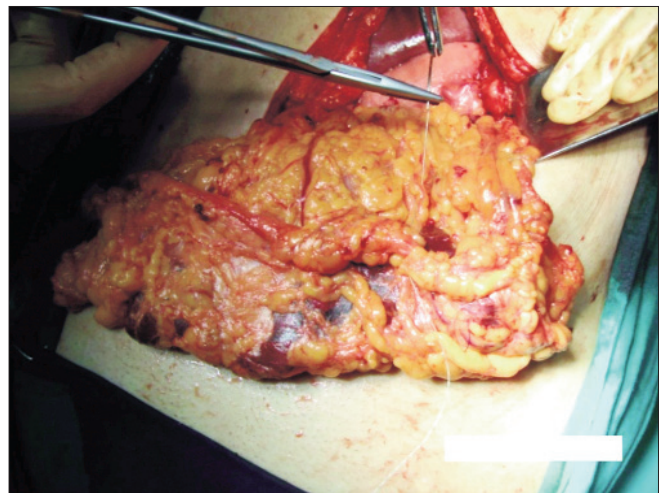


Figure 3. Rolling the great omentum over the implanted fragments – suture of great omentum – personal collection

method is contraindicated in peritoneal contamination.

The procedure's complications are rare. Weber and collaborators report a complications' rate of 2-3% (33). Petroianu didn't relate any complication (16, 27-28).

Was described:

- Intestinal occlusion. Have been published 2 cases, occlusion being secondary to adhesions around an implant abscess (37).
- Aseptic necrosis occurs more frequently in shock conditions, secondary to decreasing omentum's circulation (33).
- Implant's torsion, due to poor fixing of fragments in the great omentum (27).
- Implanted fragment's abscess; have been related 4 cases: 3 cases after omentum implant (one being secondary to spleen microfragmentation technique) and one case, after extra-peritoneal splenic implant (28).

Implant assessment

Viability and implant's functions are assessed after 3 months from surgery and consist in (31):

A. Implant's viability – assessment methods

1. Heat denatured erythrocytes scintigraphy marked with Tc99m (elective for implant's evaluation)

Quantitative assessment can be made by scintigraphy with Tc 99m Sulphur Colloid, but has limited value, the spleen receiving just 10-15 % from the injected radioactive substance's dose (37). Mere presence of spleen tissue scintigraphically proved doesn't involve a normal immune function. In early scintigraphy (6-7 weeks from surgery) the spleen

implants are viewed with difficulty having a lower radioactivity than the liver tissue (Fig. 4 A). In assessment scintigraphy (after 3-5 months) the intensity of tracer accumulation is significantly increased (Fig. 4 B).

2. Ultrasound and Doppler ultrasound with SonoVue contrast substance

Ultrasound reveals hypoechogenic masses, multinodular, lobulated, placed at low distance (1,5-2 cm) from the anterior abdominal wall, inferior from the pancreatic lodge, in the mesenterial space (Fig. 5).

With standard ultrasound current nodular structures cannot be emphasized and cannot be included in the differential diagnosis with tumours. In colour Doppler ultrasound with contrast substance primary spleen vessels are visible

Sono Vue is a contrast substance which contains sulphur hexafluoride microbubbles, being used for ultrasound imaging, for increasing blood's echogenicity, resulting in an improved Signal-Noise ratio.

SonoVue must be utilised only in patients where ultrasound images, without contrast improving, are not convincing.

SonoVue increases detection or exclusion of peripheral arterioles efficacy, by improving Signal-Noise ratio in Doppler examination.

In spleen implant assessment, SonoVue increases the quality of the Doppler image, regarding blood flow and signal duration clinically useful.

SonoVue improves the image of the spleen implants' vascularisation during a Doppler ultrasound, leading to a more specific characterisation of implanted tissue (44).

After administering 2 ml of Sono Vue contrast substance, many captive nodule structures can be distinguished in arterial

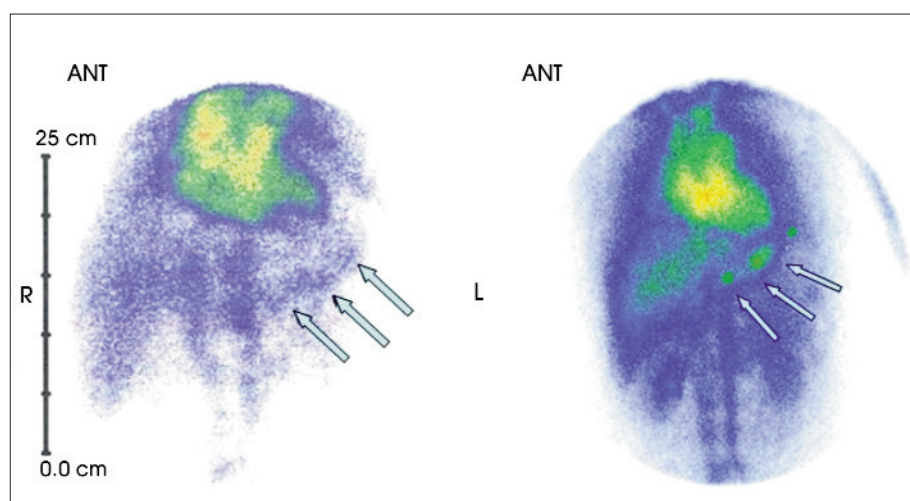


Figure 4. Scintigraphy with heat denatured erythrocytes and labelled with Tc99m –.
 (A) Early scans at 50 days after operation show the HSAs were faintly showed and the intensity of radioactivity in splenic autotransplants was lower than that in liver.
 (B) Follow-up scans at 149 days after operation showed that the intensity of radioactivity in splenic autotransplants was the same as the liver, but the slices of HSAs could not be distinguished - personal collection

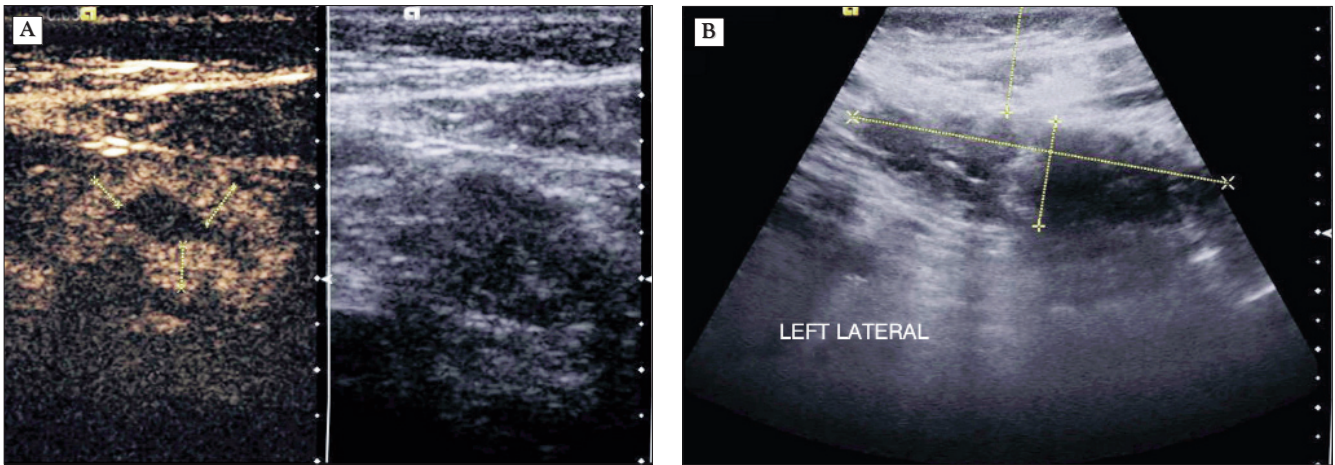


Figure 5. Soft tissue ultrasound – personal collection

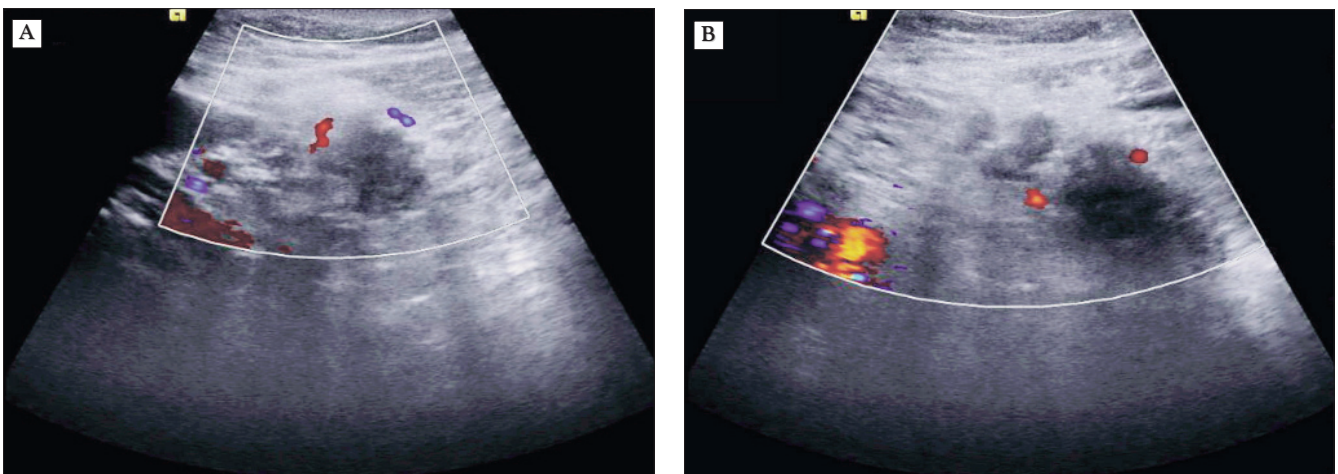


Figure 6. Doppler ultrasound with contrast agent SONO VUE – personal collection

and venous time. The average diameter of these nodules varies between 6.5 and 9.5 mm. A perinodular vascular net tends to be the arch well represented. The nodules look stitched, tending to form lymphocyte rings - possibly similar to ones present in infantile digestive structures (Fig. 6).

3. CT+SPECT (Single-Photon Emission Computed Tomography) – quantitative determination

Cothren (37) and collaborators have studied the aspect and evolution of splenic implants by CT scan. So, they have observed different images of splenic implant evolution in certain periods. Early CT scan shows multiple intra-abdominal fluid collections, similar to abscesses. CT guided percutaneous drainage of these collections was sterile, which denied possibility of existence of postoperative abscesses. The authors followed the CT characteristics of implanted tissue, the changing of its morphological aspect in time and its differentiation of intra-abdominal abscesses. The observations were:

- Intestinal obstruction. 2 cases have been published, obstruction being secondary to adhesions around day 9: fluid collections unaccompanied by inflammatory events;
- Day 14: decreasing of implant's dimensions with peripheral loading; without adjacent inflammatory events;
- Day 52: decreasing by 50% of implant's dimensions and peripheral loading, fluid collections without inflammatory events;
- Day 113: decreasing with another 25% of splenic implant (from 5 cm to 1.2 cm); the absence of inflammatory modifications; peripheral load persisted, but was considerably diminished.

CT aspect of the implant's transformation in abscess is suggested by gas bubbles in spleen fragments, local inflammatory modifications and emphasizing of peripheral loading.

Imagistic differential diagnosis between splenic implant and infected/with abscess implant is very important for avoiding a possibly unnecessary laparotomy.

Recently Weber stated the possibility of measuring the implant's dimensions using SPECT (Single-Photon Emission Computed Tomography) to make a quantitative determination (33) (Fig. 7,8).

B. Filter function

Is highlighted by disappearance of Howell-Jolly bodies (peripheral smear, Giemsa colouration) and modified platelets number.

C. Phagocytic and immunological function:

- a) determining the tuftsine's level (by ELISA), IgM level, NBT test (evaluates granulocyte's phagocytic function);

- b) determining CD3, CD4, CD8, CD4/CD8 in peripheral blood (has low levels in the first week postoperatively, becoming normal after 3 months from surgery);
- c) B lymphocytes have high levels after implant (33,34);
- d) determining the anti-pneumococcal antibodies titre (ELISA).

Tuftsine (endocarboxypeptidase) is produced only by the spleen and has a significant correlation with residual splenosis secondary to posttraumatic splenectomy (34).

After 6 months from surgery the assessment will include the next determinations: Howell-Jolly bodies, poikilocytes, thrombocytosis (27,37).

Immunological answer assessment (the implant's functionality) is represented by increase of: 1 - bacterial clearance; 2 - anti pneumococcal antibodies; 3 - Ig M; 4 - tuftsine; 5 - oxidative capacity of phagocytes (phagocyte oxidative burst).

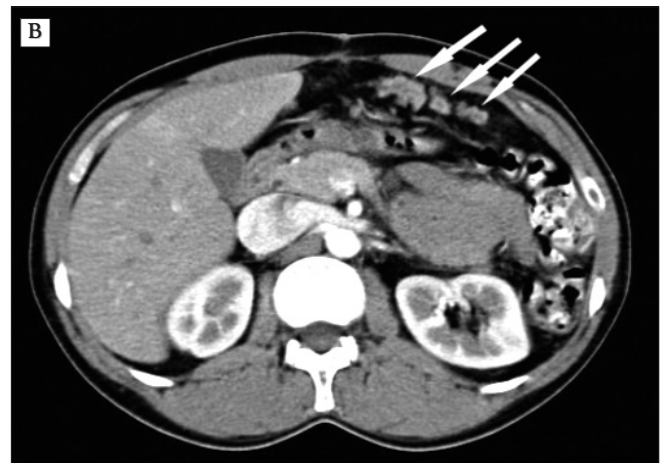
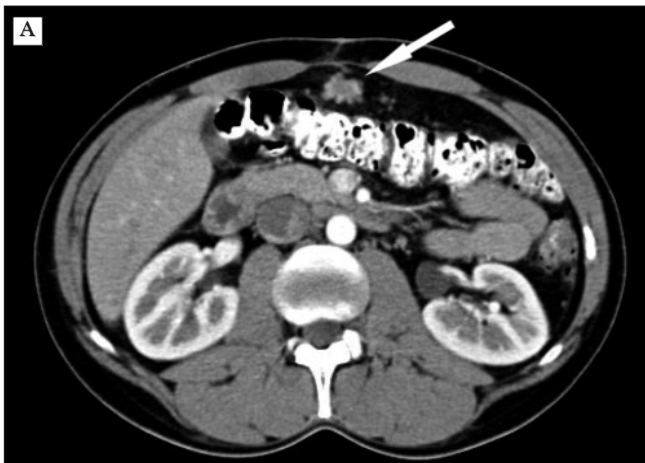


Figure 7. CT scan – personal collection

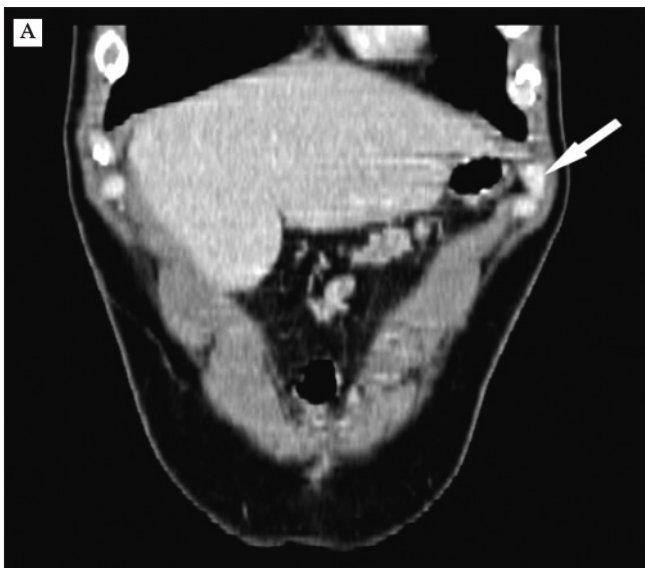


Figure 8. CT reconstruction – personal collection

Conclusions

Autologous spleen implant is required when total splenectomy is necessary, being the most efficient preservation method of spleen functions. Placing it intra-omentally is superior to splenectomy but, obviously, inferior to „in situ” spleen preservation. It is a simple procedure that associates a minimal morbidity, being considered a safe technique that can be made even when hollow organ lesions are present.

Immunoprophylaxis is mandatory in splenectomised patients or in those who have less than 50% intact spleen and has firm indications after heterotopic splenic autotransplant. It consists in administration of vaccines against *Streptococcus pneumoniae*, *Haemophilus influenza* and *Neisseria meningitidis*. The optimal moment is the 14th day postoperatively (12,45-46).

Experimental studies have described: low sepsis in animals with spleen implants vs. asplenic control groups (12). The animals with splenic implant had more frequently infections than control groups (with intact spleen) or the ones who had partial or subtotal splenectomy (46,47). The absence of abnormal particles peripheral smear, positive scintigraphic image, high level of Ig, especially IgM (usually produced by the spleen), suggest the implant's functionality (12,46-48). After omental splenic implant, the clearance for *Streptococcus Pneumoniae* is maintained at 50 %, while after splenectomy no bacteria is eliminated (27,28,33,46,48).

References

1. http://www.who.int/gho/publications/world_health_statistics/EN_WHS2011_Full.pdf
2. Caloghera C. Abdominal Trauma - Generalities. In: Abdominal Trauma, Caloghera C, ed. Timisoara: Ed. Facla; 1983. p. 1:11-42.
3. Demetriades D, Hadjizacharia P, Constantinou C, Brown C, Inaba K, Rhee P, et al. Selective nonoperative management of penetrating abdominal solid injuries. *Ann Surg.* 2006;244(4): 620-8.
4. Selman Uranues, Yusuf A. Kilic. Injuries to the Spleen, *European Journal of Trauma and Emergency Surgery.* 2008; 34(2):355-61.
5. Timens W, Leemans R. Splenic autotransplantation and the immune system: adequate testing required for evaluation of effect. *Ann Surg.* 1992;215(3):256-60.
6. Sills RH. Splenic function: physiology and splenic hypofunction. *Crit Rev Oncol Hematol.* 1987;7(1):1-36.
7. Timens W. The human spleen and the immune system: not just another lymphoid organ. *Res Immunol.* 1991;142(4):316-20.
8. Băra C. Essential Immunology. București: Editura All; 2002.
9. Lockwood CM. Immunological functions of the spleen. *Clin Haematol.* 1983;12(2):449-65.
10. Harms G, Hardonk MJ, Timens W. In vitro complement-dependent binding and in vitro kinetics of pneumococcal polysaccharide TI-2 antigens in the rat spleen marginal zone and follicle. *Infect Immun.* 1996;64(10):4220-5.
11. McClusky DA 3rd, Skandalakis LJ, Colborn GL, Skandalakis JE. Tribute to a triad: history of splenic anatomy, physiology, and surgery-part 2. *World J Surg.* 1999;23(5):514-26.
12. Beuran M, Grigorescu M. Medical and surgical actualities in digestive pathology vol. II. Cluj-Napoca: Ed. Medical University "Iuliu Hatieganu"; 2010.
13. Diamond LK. Splenectomy in childhood and the hazard of overwhelming infection. *Pediatrics.* 1969;43(5):886-9.
14. Davidson RN, Wall RA. Prevention and management of infections in patients without a spleen. *Clin Microbiol Infect.* 2001;7(12):657-60.
15. Braunwald E, Fauci AS, Kasper DL, Hauser SL, Longo DL, Jameson JL, Loscalzo J. *Harrison's Principles of Internal Medicine* 17th. ed. McGraw-Hill; 2008.
16. Venter MD. Traumatic lesions of spleen – PhD Thesis. Bucharest: Carol Davila University of Medicine and Pharmacy; 2008.
17. Bohnsack JF, Brown EJ. The role of the spleen in resistance to infection. *Annu Rev Med.* 1986;37:49-59.
18. Najjar VA, Nishioka K. "Tuftsin": a natural phagocytosis stimulating peptide. *Nature.* 1970;228(5272):672-3.
19. Karakantza M, Theodorou GL, Mouzaki A, Theodori E, Vagianos C, Maniatis A. In vitro study of the long-term effects of post-traumatic splenectomy on cellular immunity. *Scand J Immunol.* 2004;59(2):209-19.
20. Sumaraju V, Leon G, Smith SM. Infectious complications in asplenic hosts. *Infect Dis Clin North Am.* 2001;15(2):551-65, x.
21. Bagge L, Haglund O, Wallin R, Borg T, Modig J. Differences in coagulation and fibrinolysis after traumatic and septic shock in man. *Scand J Clin Lab Invest.* 1989;49(1):63-72.
22. Tilsney V. Antithrombin III in surgery. *Folia Haematol Int Mag Klin Morphol Blutforsch.* 1988;115(3):284-91.
23. Gillespie DL, Carrington LR, Griffin JH, Alving BM. Resistance to activated protein C: a common inherited cause of venous thrombosis. *Ann Vasc Surg.* 1996;10(2):174-7.
24. Kessler CM. Anticoagulation and thrombolytic therapy: practical considerations. *Chest.* 1989;95(5 Suppl):245S-256S.
25. Adams E. Postoperative thrombocytosis. *Arch Intern Med.* 1944;73:329-35.
26. Resende V, Petroianu A, Junior WC. Autotransplantation for treatment of severe splenic lesions. *Emerg Radiol.* 2002;9(4): 208-12. Epub 2002 Aug 9.
27. Petroianu A, Petroianu LP. Splenic autotransplantation for treatment of portal hypertension. *Can J Surg.* 2005;48(5):382-6.
28. Petroianu A1, Vidigal FM, Costa VC, De Oliveira SC. Splenic autotransplantation in Gaucher's disease. *Dig Surg.* 2000;17(2):181-3.
29. Martin LW. Autologous splenic transplantation. Editorial. *Ann Surg.* 1994;219(3):223-4.
30. Pisters PW, Pachter HL. Autologous splenic transplantation for splenic trauma. *Ann Surg.* 1994;219(3):225-35.
31. Haowei Wang, Zengli Liu, Jinchang Wu, Haorong Wu. Value of Spleen Imaging with 99mTc Labeled Heat-Damaged Erythrocytes in Evaluating The Function of Splenic Autotransplants *Ann Nucl Med Sci* 2001;14:91-95.
32. Petroianu A. Assessment of phagocytic function in remnants of subtotal spleen implantation. *Rev Bras Hematol Hemoter.* 2003;25:25-31.
33. Weber T, Hanisch E, Baum RP, Seufert RM. Late results of heterotopic autotransplantation of splenic tissue into the greater omentum. *World J Surg.* 1998;22(8):883-9.
34. Brandt CT, Maciel DT, Caneca OA, Castro CM, Araújo LB. Autotransplant of spleen tissue in children with schistosomiasis: evaluation of splenic function after splenosis. *Mem Inst Oswaldo Cruz.* 2001;96 Suppl:117-22.
35. Tavassoli M, Ratzam RJ, Crosby WH. Studies on regeneration of heterotopic splenic autotransplants. *Blood.* 1973;41(5):701-9.
36. Johnson J, Weiss L Electron microscopy study of subcutaneous

- and intraperitoneal splenules in the mouse. *American Journal of Anatomy*. 1989;158:89-100.
37. Cothren CC, Moore EE, Biffl WL, Ray CE Jr, Ciesla DJ, Johnson JL. Radiographic characteristics of postinjury splenic autotransplantation: avoiding a diagnostic dilemma. *J Trauma*. 2004;57(3):537-41.
 38. Sasaki K, Kiuchi Y, Sato Y, Yamamori S. Morphological analysis of neovascularization at early stages of rat splenic autografts in comparison with tumor angiogenesis. *Cell Tissue Res*. 1991; 265(3):503-10.
 39. Alves HJ, Viana G, Magalhães MM, Arantes RM, Coelho PM, Cunha-Melo JR. Kinetics of neovascularisation of splenic autotransplants in mice. *J Anat*. 1999;195 (Pt 3): 387-92.
 40. Holdsworth RJ. Regeneration of the spleen and splenic autotransplantation. *Br J Surg*. 1991;78(3):270-8.
 41. Lucas CE. Splenic trauma. Choice of management. *Ann Surg*. 1991;213(2):98-112.
 42. Ionescu NG, Szabo J. Implant and splenic autotransplant. *Spleen*. Cluj-Napoca: Ed. Dacia; 1993.
 43. Di Carlo I, Pulvirenti E, Toro A. A new technique for spleen autotransplantation. *Surg Innov*. 2012;19(2):156-61.
 44. Shatz DV, Schinsky MF, Pais LB, Romero-Steiner S, Kirton OC, Carlone GM. Immune responses of splenectomized trauma patients to the 23-valent pneumococcal polysaccharide vaccine at 1 versus 7 versus 14 days after splenectomy. *J Trauma*. 1998; 44(5):760-5; discussion 765-6.
 45. Fremont RD, Rice TW. Splenosis: a review. *South Med J*. 2007;100(6):589-93.
 46. Petroianu A, Resende V, Da Silva RG. Late follow-up of patients submitted to subtotal splenectomy. *Int J Surg*. 2006;4(3):172-8. Epub 2006 Feb 20.
 47. Herneth AM, Pokieser P, Philipp MO, Bader TR, Helbich TH, Mallek R, et al. Role of Doppler sonography in the evaluation of accessory spleens after splenectomy. *J Ultrasound Med*. 2001; 20(12):1347-51.
 48. Mazurek A, Szaluś N, Stembrowicz-Nowakowska Z, Hałka J, Chmieliński A. Detection of splenic tissue by ^{99m}Tc-labelled Sn-colloid SPECT/CT scintigraphy. *Nucl Med Rev Cent East Eur*. 2011;14(2):116-7.