ORIGINAL PAPERS

Adv Clin Exp Med 2004, **13**, 3, 411–417
ISSN 1230-025X

MAREK BOCHNIA[1], JERZY RAK[1], AGNIESZKA JABŁONKA[1], BEATA MACHOŃ[1], MICHAŁ JELEŃ[2]

Comparison of Two Sclerosants for Esophageal Varices – Histologic Effects of Sodium Morrhuate and Liquid Paraffin

Porównanie dwóch środków do sklerotyzacji żylaków przełyku – histologiczna ocena działania morhuatu sodu i płynnej parafiny

[1] Department of Otolaryngology, Wroclaw Medical University, Poland
[2] Department of Pathomorphology, Wroclaw Medical University, Poland

Abstract

Background. Sclerotherapy of esophageal varices is still the most popular endoscopic method of treatment, especially in children. Sclerosants, chemicals with strong tissue irritant properties, may be injected into the lumen of a dilated vessel or submucosally along the variceal column.
Objectives. The aim of this study was to compare the usefulness and efficiency of two sclerosants – sodium morrhuate and liquid paraffin with respect to the way of administration and the number of injections.
Material and Methods. Material of the research consisted of 40 mice belonging to Balb/c strain. Their evident reactivity to sclerotherapy has been proven already. The cohort was divided into 8 groups of 5 subjects. Each group received one of two sclerosants – sodium morrhuate or liquid paraffin, administered in one of two ways – into the caudal vein or around it, and maintained on one doze of a sclerosant or, after a period of 3 weeks, subjected to the another injection. Subsequently, vein sections stained with hematoxylin and eosin were performed to the microscopic estimation.
Results. Parietal thrombi and perivascular fibrotic changes were observed only after intravenous injections of sodium morrhuate and were more expressed after the second injection. Liquid paraffin, irrespective of the way of administration and its repetition, did not cause thrombus development.
Conclusions. Intravariceal injections of sodium morrhuate seem to be sufficient and safe in the final therapy of esophageal varices. Liquid paraffin seems to be useful but only as a remedy to stop acute bleeding (**Adv Clin Exp Med 2004, 13, 3, 411–417**).

Key words: esophageal varices, sclerotherapy, mice, sodium morrhuate, liquid paraffin.

Streszczenie

Wprowadzenie. Sklerotyzacja żylaków przełyku jest wciąż najczęściej stosowaną endoskopową metodą leczenia. Substancje chemiczne, wywołujące silny odczyn tkankowy, mogą być wstrzykiwane do światła rozszerzonego naczynia lub podśluzówkowo w jego sąsiedztwie.
Cel pracy. Celem pracy było porównanie przydatności i skuteczności dwóch środków – morhuatu sodu i płynnej parafiny, w zależności od drogi podania i liczby iniekcji.
Materiał i metody. Materiał badawczy stanowiło 40 myszy szczepu Balb/c, u których udowodniono wcześniej dużą reaktywość na środki sklerotyzujące. Kohortę podzielono na 8 grup po 5 sztuk. Zwierzęta z grup badanych otrzymały jeden z dwóch środków – morhuat sodu lub płynną parafinę, podane albo do światła żyły ogonowej, albo w jej pobliżu. Połowa zwierząt otrzymała jedną dawkę środka obliterującego, u drugiej części podano ją powtórnie po 3 tygodniach. Trzy tygodnie po pierwszej lub drugiej iniekcji, po barwieniu hematoksyliną i eozyną, zbadano mikroskopowo skrawki z fragmentem żyły.
Wyniki. Tylko po dożylnej injekcji morhuatu sodu obserwowano skrzepy przyścienne i włókniste zmiany okołonaczyniowe (bardziej nasilone po drugiej injekcji). Płynna parafina, niezależnie od drogi podania i liczby injekcji, nie powodowała powstania skrzepu.
Wnioski. Uzyskane wyniki potwierdzają, że morhuat sodu jest odpowiednim i bezpiecznym środkiem w leczeniu obliteracyjnym żylaków przełyku. Płynna parafina może znaleźć zastosowanie jedynie w doraźnym powtrzymywaniu krwawień (**Adv Clin Exp Med 2004, 13, 3, 411–417**).

Słowa kluczowe. żylaki przełyku, sklerotyzacja, myszy, morhuat sodu, płynna parafina.

Portal hypertension is not a distinct disease but a complex syndrome of haemodynamic disturbances. Dominant causes of its development are pathologies which lead to the increase in resistance of blood flow through the hepatic vessels (especially cirrhosis) and disturbances of portal vein trunk patency. When hypertension persists for a long period, collateral circulation develops in the area of communication between the portal blood and the drainage-basin of the superior or inferior caval vein. One of 16 the described anastomoses is the way through the left gastric vein, submucosal plexus of the esophagus and azygos vein [1]. Lying in the submucosal membrane, esophageal veins possess a small quantity of strengthening perivascular tissue, are deprived of valvules and exposed to large pressure fluctuations in the passage from the abdominal cavity to the thorax. The presence of long-lasting stasis in the portal system makes the loaded plexus become gradually wider and transforms it into irregular varices. Variceal haemorrhage is the most dangerous complication in patients with portal hypertension. It always makes the prognosis much poorer. One of seven incidents is fatal, more than 50% of untreated patients die in the course of a year from the first bleeding [2, 3]. However the mechanism of variceal formation is well known, the direct cause of the haemorrhage frequently remains difficult to determine. Varix rupture may occur subsequently after a local injury or a sudden increase of portal pressure. The lesion may be a result of cardiac inflammation and injury by swallowing food. A massive rise in the portal pressure may appear during vomiting, defecation, spurt, or rarely after heavy meal and blood transfusion.

The goal of sclerotherapy in esophageal varices would be to stop active bleeding and diminish the risk of rebleeding as a final mode of therapy [4]. Sclerosants (chemicals with strong tissue irritant properties) can be injected into the lumen of the dilated vessel or submucosally along the variceal column. Intravenous injection ought to lead to the development of fibrotic tissue, intimal thickening, and formation of intraluminal thrombosis to obliterate the varix [1]. Injections in the neighborhood of the varix should make the submucasal layer thicker and fibrotic and push the dilated vein deeper into the esophageal wall to diminish the danger of its damage [5]. Combination of the intra- and paravariceal injections may be used to achieve both of these results. The aim of our study was to compare usefulness and efficiency of two sclerosants, i.e. sodium morrhuate (s.m.) (one of fatty acid derivatives) and liquid paraffin (l.p.) with respect to the way of administration and the number of injections.

Material and Methods

The material of the research included forty (n = 40), 5-month-old mice, both sexes, weighing about 25–30 g. each, belonging to Balb/c strain. A half of them (n = 20) received 0.05 ml of Varicocid (1 ml containing 55 mg of sodium morrhuate) and another half (n = 20) – liquid paraffin of the same amount. Each agent was administered in some mice (n = 10 + 10) to the caudal veins and in the others (n = 10 + 10) perivascularly, around the caudal veins, as presented in the Table 1. The place of injections was next pressed for about 2 minutes each time. Three weeks later half of the animal were put to sleep and specimens from the places of injections were taken for histopathological assessment. The remained living half of the cohort underwent the same injection procedure, thus a second injection of the irritating agent was given in the same way as before. After the following another three weeks, the rest of the animals were also put to sleep and as well their tails were subjected to histopathological examination. The obtained material was preserved in 4% formalin, than embedded in paraffin. The sections were stained with hematoxylin and eosin (HE) before microscopic estimation. Finally, we obtained eight group of 5 subjects/specimens (A-H; Tab. 1), different as regards the type of the sclerosant, the way of administration and the number of injections. Guidelines of these experiments on the animals were accepted by the local Bioethics Committee.

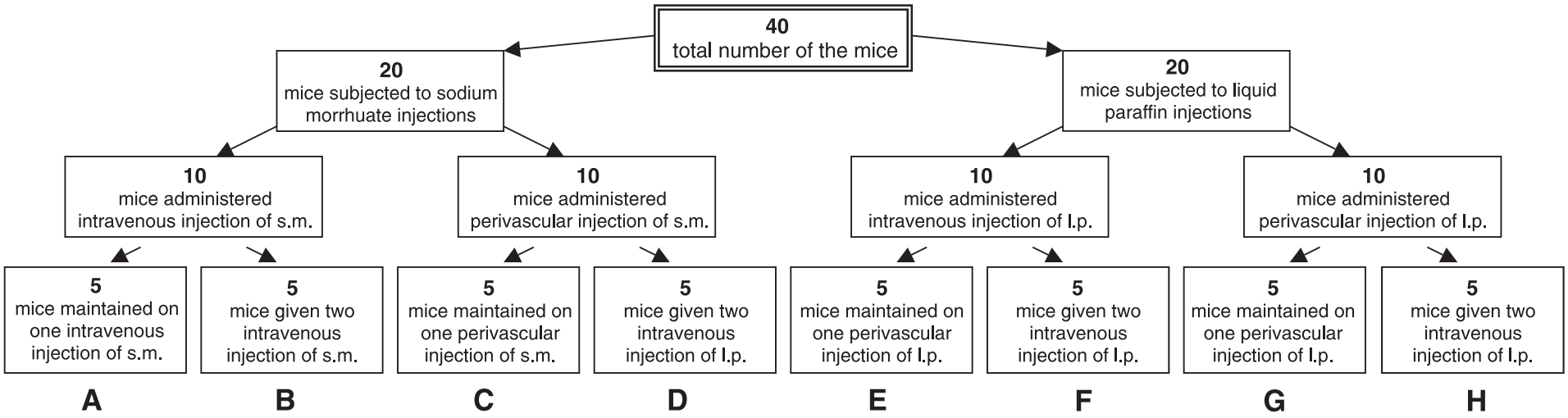
Results

1. In all mice from the group A, that is after one intravenous injection of sodium morrhuate, the macroscopic pictures were identical. Oval ulcerations of 3–4 mm in diameter, covered with brown, firmly adhered crusts were formed. The microscopic pictures in the places of injections presented some epithelial defects, covered by necrotic masses and deeper – granulocytic inflammatory infiltrations forming microabscesses. Lymphocytic infiltrations prevailed underneath. Numerous eosinophilic granulocytes and the giant cells of "foreign body" were found in some places. Fibroblasts proliferation and vascular hyperplasia, as in chronic inflammatory granulation, were also visible. Small calcifications occurred very seldom. In this subgroup there was evidence of endothelial necrosis and parietal thrombi without complete occlusion of the caudal veins lumen.

Progression of changes was observed in the group B, i.e. 3 weeks after second intravenous injection of s.m. The crusts and ulcerations were

Table 1. Mice distribution with respect to the type of the sclerosants, the way of administration and the number of injections

Tabela 1. Zestawienie badanych grup myszy w zależności od rodzaju środka obliterującego, sposobu jego podania i liczby ostrzykiwań



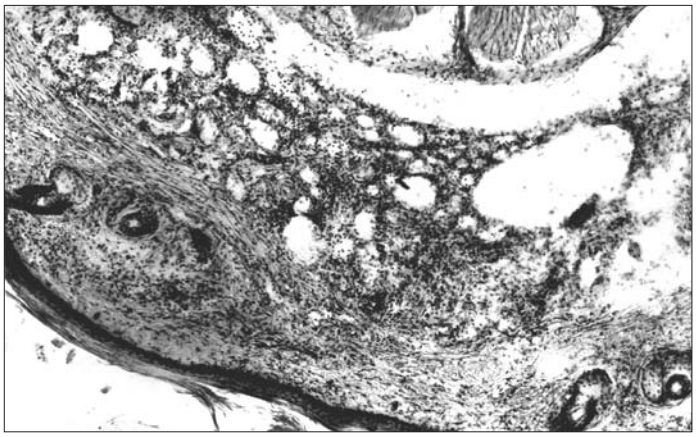


Fig. 1. Mouse's tail after 2 intravenous injections of sodium morrhuate. HE. Magnification about 200×

Ryc. 1. Ogon myszy po drugiej iniekcji s.m. do światła żyły ogonowej. Barwienie HE. Pow. 200×

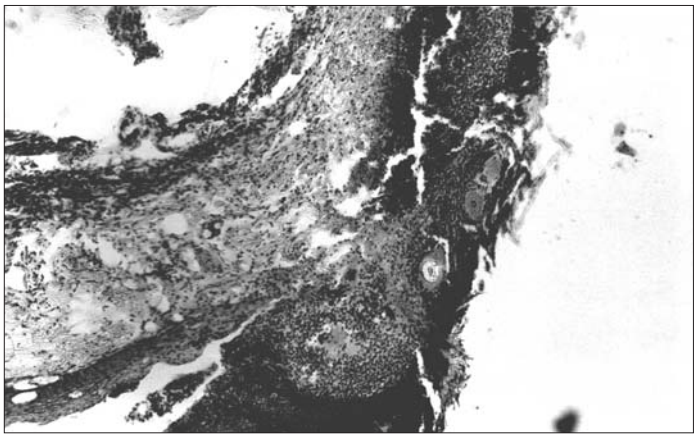


Fig. 2. Mouse's tail after 2 injections of sodium morrhuate around the caudal vein. HE. Magnification about 200×

Ryc. 2. Ogon myszy po drugiej iniekcji s.m. w pobliżu żyły ogonowej. Barwienie HE. Pow. 200×

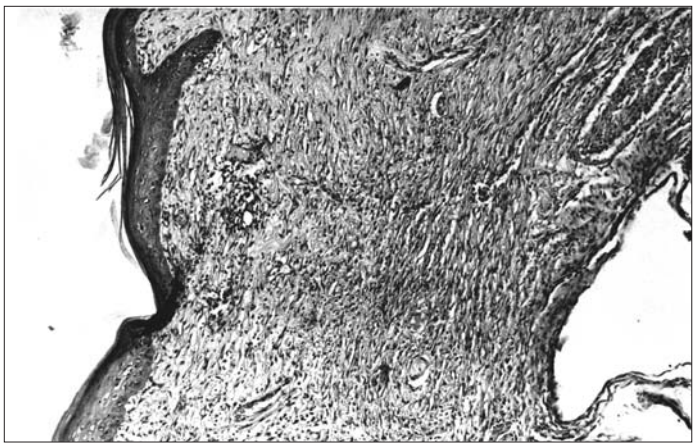


Fig. 3. Mouse's tail after 2 liquid paraffin intravenous injections. HE. Magnification about 200×

Ryc. 3. Ogon myszy po drugiej iniekcji płynnej parafiny do światła żyły ogonowej. Barwienie HE. Pow. 200×

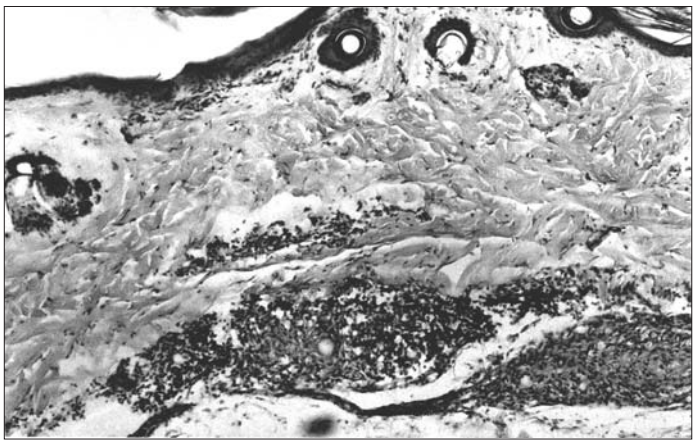


Fig. 4. Mouse's tail after 2 paraffin injections around the caudal vein. HE. Magnification about 200×

Ryc. 4. Ogon myszy po drugiej iniekcji parafiny w pobliżu żyły ogonowej. Barwienie HE. Pow. 200×

here 1/3–1/2 larger in the diameter. The necrotic--inflammatory infiltrations were also larger and in 3 cases (60%) the lumen of vessel was completely obliterated by thrombus. Thrombi were organised in stages of recanalization.

2. The macroscopic lesions observed after perivascular and intravenous injections of sodium morrhuate were similar to each other in all animals from groups A, B, C and D. Nevertheless, after 2 perivascular injections (group D), crusts were larger and they often occupied above one-half of caudal circumference. The microscopic changes situated around the vessels were also nearly equal, independent on the way of application of the sclerosant, and their progression after repeated dose of s.m. was observed. For a change, in all the mice from the group C and D, the walls of veins were thickened with distinct surrounding fibrosis, especially after two injections. Besides, endothelial necrosis and thrombus formations were not detected here.

3. All cases from the group E and F, after l.p. intravenous injections, showed no macroscopic changes. Microscopic pictures were nearly identical after first and second injection. The wall of the caudal vein was slightly thickened with small disseminated inflammatory lymphocytic infiltrations visible around. Progression of changes after 2 injections was only insignificantly expressed.

4. Also, injections of l.p. in the neighbourhood of caudal veins resulted in none macroscopic changes in all mice from the group G and H. Small inflammatory infiltrations composed of lymphocytes and eosinophilic granulocytes prevailed in the microscopic picture. Induction of fibrotic changes with fibroblast proliferation was also visible. Walls of the vessels were thickened without intimal injuries and without formation of parietal thrombi. The progression of mild fibrosis was noted after second perivascular injection (group H).

Discussion

In the last few decades many surgical procedures were used in the treatment of portal hypertension. Each time it seemed that the recently proposed portocaval shunt can solve the problem of bleeding esophageal varices. Lately, many hopes have been put in the transjugular intrahepatic portosystemic shunt (TIPS), which was introduced to clinical practice in 1988 and became widely accepted [6]. Still, all the traditional methods of treatment do not always improve long–term survival in cirrhotic patients [7]. What is more, even such a non-invasive and haemostatically efficient TIPS appeared to be connected with high risk of encephalopathy [8].

Rubber bands application was introduced in 80th with the hope that it would be as efficacious as sclerotherapy and be associated with fewer complications [9]. Its methodology is continually developed. For example, numerous devices were constructed to allow more ligatures to be made during a single endoscopic insertion [10]. It was even suggested that sclerotherapy as technically easier should be performed if a surgeon concedes a lack of expertise in ligation or in desperation. Anyhow sclerotherapy was not eliminated by this mechanical method. While the two techniques are equally effective in controlling acute bleeding, there was observed significantly higher esophageal variceal recurrence after ligation [11].

Sclerotherapy of the esophageal varices was proposed by Crafoord and Frenckner in 1939 [12]. This procedure is still the most popular and safe endoscopic method in the esophageal varices management, especially in children [13] and in patients with fundic varices [14]. Dangers of the method are mostly connected with the usage of sclerosing agents which are chemicals of strong irritant properties. One of the most frequent complications are ulcers at the area of injection. Sarin observed their occurrence in about 90% of patients after the sclerotherapy and even after intravariceal injections [15]. He characterized them as benign and thought they arise generally as a result of sclerosant extravasation. A dangerous problem may also be dissemination of the chemicals being introduced to the circulation. Even such a distant complication as a cerebral abscess was described after the sclerotherapy of the esophageal varices [16]. Thus, these undesirable sequels have to be connected not only with the irritant properties of an agent, but also with the way of its administration. It's then questionable which kind of injection, or both of them, is better, i.e. more efficient and safer, with reference to a given sclerosant.

Of necessity, clinical studies of the variceal sclerotherapy are not blinded. Searching for an ideal sclerosant with optimum thrombogenic and minimum necrotic properties and selection of its way of administration has to be supported, then, by animals experiments. Previous investigations were performed on various animal species. Efficacy of aquesous and oily chemicals was examined on rats [17], rabbits [18], and dogs [19]. Animal tissue reactivity sometimes differs from that of humans and agents, suspected to be ineffective, have later been found to be useful in clinical trials [15]. Applying some studies to humans also remains questionable, because conclusions have been drawn by observing effects of few injections, whereas the sclerotherapy requires repeated courses of the procedure [20].

In our experiment, sodium morrhuate and liquid paraffin were used. They are the most popular irritating agents because of their necrotico-thrombotic and inflammatory properties [20]. They are commonly used where the sclerotherapy of esophageal varices is carried out, also in Poland [21]. The tested animals were mice of Balb/c strain. We chose mice because of their proved significant reactivity to sclerotherapy. Three-week intervals between the courses of the experimental sclerotherapy were set, based on clinical observations. A shorter period causes a rise in patients' complaints, a longer pause probably increases the risk of rebleeding [22]. The place of injection was pressed each time for about 2 minutes in order to keep the drugs in the veins and to prevent bleeding. On the basis of our investigations, we can state that simultaneous intra- and paravariceal injections are not imperative with the use of 5% solution of s.m. Because of its powerful necrotic-inflammatory properties, s.m. may be injected only intravariceally. After the intravenous injection, not only parietal thrombi but also perivascular fibrotic changes are observed. Subcutaneous administration of this chemical does not provoke the formation of thrombus. What is more the ulceration at the site of injection is much larger. Intravariceal injections of s.m. ought to be sufficient and safer in the final therapy of esophageal varices. Liquid paraffin, a sclerosant of weak inflammatory properties, irrespectively of the way of administration, does not cause the occurrence of thrombus. As opposite to s.m., and lacking an other sclerosant, it may be no more but a remedy to stop acute bleeding.

References

[1] Schier J: Sclerotherapy as a method of management of oesophageal varices (in Polish with English abstract). Pol Arch Med Wewn 1977, 57, 177–182.

[2] Graham DY, Smith JL: The course of patients after variceal hemorrhage. Gastroenterology 1981, 80, 800–809.

[3] Jurczyk G: Massive bleeding from the upper gastrointestinal tract (p.I) (in Polish) Gastroenterologia 2002, 6, 6–16.

[4] Paquet KJ, Lazar A: Current therapeutic study in bleeding esophageal varices in babies and children and long--terms results of endoscopic paravariceal sclerotherapy over twenty years. Eur J Pediatr Surg 1994, 4, 165–172.

[5] Helpap B, Bollweg L: Morphological changes in the terminal esophagus with varices, following sclerosis of the wall. Endoscopy 1981, 13, 329–333.

[6] Richter GM, Noeldge G, Palmaz JC, Roessle M: The transjugular intrahepatic portosystemic stent-shunt (TIPSS): results of a pilot study. Cardiovasc Intervent Radiol 1990, 13, 200–207.

[7] Cabrera J, Maynar M, Grandos R, Gorriz E, Reyes R, Pulido-Duque JM, Rodriguez SanRoman JL, Guerra C, Kravetz D: Transjugular intrahepatic portosystemic shunt versus sclerotherapy in the elective treatment of variceal haemorrhage. Gastroenetrology 1996, 110, 832–839.

[8] Conn HO, Lebrec D, Terblanche J: The treatment of oesophageal varices: a debate and a discussion. J Int Med 1997, 241, 103–108.

[9] Swain CP, Mills TN, Northfield TC: Experimental sudies of new mechanical methods of endoscopic haemostatis: Stitching, banding, clamping, and ulcer removal (abstr.). Gut 1995, 26, A1151.

[10] Saed ZA: Endoscopic esophagogastric variceal ligation with a six-shot multiple ligation device. Am J Gastroenterol 1995, 90, 1570.

[11] Sarin SK, Govil A, Jain AK, Guptan RC, Issar SK, Murthy NS: Prospective randomized trial of endoscopic sclerotherapy versus variceal band ligation for esophageal varices: influence on gastropathy, gastric varices and variceal recurrence. J Hepatol 1997, 26, 826–833.

[12] Crafoord C, Frenckner P: New surgical treatment of varicous veins of the oesophagus. Acta Otolaryngol (Stockh) 1939, 27, 422–429.

[13] Nijhawan S, Patni T, Sharma U, Rai RR, Miglani N: Endoscopic variceal ligation in children. J Pediatr Surg 1995, 30, 1455–1456.

[14] Takeuchi M, Nakai Y, Syu A, Okamoto E, Fujimoto J: Endoscopic ligation of gastric varices. Lancet 1996, 348, 1038.

[15] Sarin SK: Endoscopic sclerotherapy for esophago-gastric varices: a critical reappraisal. Aust NZ J Med 1989, 19, 162–172.

[16] Cohen FL, Koerner RS, Taub SJ: Solitary brain abscess following endoscopic injection sclerosis of esophageal varices. Gastrointest Endosc 1985, 31, 331–333.

[17] Blenkinsopp WK: Comparison of tetradecyl sulfate of sodium with other sclerosants in rats. Br J Pathol 1968, 49, 197–201.

[18] Martin DE, Goldman MP: A comparison of sclerosing agents: clinical and histologic effects of intravenous sodium tetradecyl sulfate and chromated glycerin in the dorsal rabbit ear vein. J Dermatol Surg Oncol 1990, 16, 18–22.

[19] Szczepanik A, Schier J, Milewski B: Investigatrions on the effect of varicocid, a preparation used for varices obliteration on the enzymatic function of the liver in experimental ascites in dogs (in Polish with English abstract). Pol Tyg Lek 1979, 34, 1351–1353.

[20] Sarin SK, Kumar A: Sclerosants for variceal sclerotherapy: a critical appraisal. Am J Gastroenterol 1990, 85, 641–649.

[21] Bochnia M, Ziemski Z, Rak J, Dziewiszek W: The role of sclerotherapy in the treatment of oesophageal varices in children (in Polish with English abstract). Otolaryngol Pol 1992, 46, 156–160.
[22] Clark AW, Macdougall BR, Westaby D, Silk DB, Strunin L, Dawson JL, Williams R: Prospective controlled trial of injection sclerotherapy in patients with cirrhosis and recent variceal hemorrhage. Lancet 1980, 2, 552–554.

Address for correspondence:

Marek Bochnia
Department of Otolaryngology
Wroclaw Medical University
Chałubinskiego 2
50-368 Wroclaw
Poland

Received: 14.07.2003
Revised: 20.08.2003
Accepted: 17.09.2003

Praca wpłynęła do Redakcji: 14.07.2003 r.
Po recenzji: 20.08.2003 r.
Zaakceptowano do druku: 17.09.2003 r.