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Applications of Polidocanol in Varicose Vein Treatment Assisted by Exposure to Nd:YAG Laser Radiation

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1. Introduction

The understanding of the interaction between Polidocanol (POL) and the target veins tissues is important in utilizing it in varicose veins diseases treatment. Generally, the development of new drug delivery routes may represent methods to improve the efficacy and/or safety of the active pharmaceutical ingredients. With this respect, the treatment involving POL administration as foam has gained widespread use **(**Cavezzi and Tessari, 2009). Although the main approach in the treatment of small diameter veins, in venulectasias and reticular veins of less than 4 mm in diameter (class I/II and III) is sclerotherapy (Alos et al., 2006; Nijsten et. al., 2009; Parsi et al., 2007; Railan et al., 2006), lasers, especially the Nd:YAG laser, have shown interesting and non-negligible capabilities in treating these cases (Redondo and Cabrera, 2005; Santos et al., 2008; Trelles et al., 2005). Clinical experimental results prove that the exposure of the tissues impregnated with POL to laser radiation emitted at 1.06 μm improves the efficiency of the treatment (Moreno Moraga, n.d.).

The aim of this chapter is to present an extensive study concerning the optical properties of commercially available Polidocanol (Aethoxysklerol 2%, Kreussler & Co. GmbH, Germany) and the possible modifications induced at molecular level in this medicine as supplied by the manufacturer, by exposing it to Nd:YAG laser radiation.

Almost 60% of the adult population of Europe and the USA present varicose or spider veins in the lower limbs and this is the seventh most common chronic vascular disorder, nine times more frequent than arterial diseases (Tibbs, 1997). In recent years, a significant, growing number of patients are consulting in order to correct the symptoms originated in this vascular disorder, as well as the aesthetic implications that cause this ailment.

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The lower extremities contain both superficial and deep veins. The superficial venous system consists of the greater saphenous vein draining most of the leg, the lesser saphenous vein draining the posterior and lateral lower leg, and the lateral (subdermal) venous system draining the lateral leg above and below the knee. Clinically, this last system may be observed as visible veins. Dilatations of the most superficial veins are called telangiectasias. These are usually bright red and measure 0.03 mm to 0.3 mm in diameter. Slightly larger are postcapillary venulectasias, which measure 0.4 mm to 2 mm, and may be red or blue depending on their oxygenation. Telangiectasias and venulectasias are commonly referred to as spider veins. These connect to larger reticular veins measuring 2 mm to 4 mm that are typically blue because of the Tyndall effect and their deeper location within the dermis (Kunishige et al., 2007).

The treatment of varicose veins reduces the symptoms and the complication rate of venous insufficiency and increases the patient's health related quality of life. It can roughly be divided into four groups: compression therapy, surgical treatment, sclerotherapy and endovenous thermal ablation. To improve efficacy and treatment satisfaction and to reduce serious side effects, costs, and postoperative pain, new minimally invasive techniques, such as ultrasound-guided foam sclerotherapy (UGFS), endovenous laser therapy (EVLT), and radiofrequency ablation (RFA), have been introduced in the last decade (Nijsten et. al., 2009). Also, sclerotherapy (mainly foam sclerotherapy) combined with 1.06 μm Nd:YAG laser beam therapy showed very encouraging clinical results. This new approach is ideal for treating varicose veins originating in the lateral subdermal plexus of the lower limbs or reticular veins of other locations. Veins of up to 4 mm calibre located at a depth between 2 mm and 4 mm can be treated. Telangiectasias or spider-veins, located between 1.2 mm and 1.7 mm below the surface of the skin, either associated with or independent from reticular veins in any location are also ideal for combined treatment. Surgery or EVLT continue to be recommended for the great saphenous veins (GSV) of a calibre greater than 8 mm. It is possible to treat truncal veins with combined POL and laser, but this requires particular expertise. In these cases ultrasound monitoring is well advised, both when puncturing and injecting the microfoam. When POL enters the veins it is detected by the ultrasound, so vein spasms, which occur after the injection, can be well controlled as well as their duration (Moreno Moraga, n.d.; Trelles et al., 2011).

Sclerotherapy is the targeted elimination of varicose veins by injection of a sclerosing substance into the vein lumen. Sclerosing agents cause a chemical irritation of the venous intimate that produces an inflammation of the endothelial lining of the vessel. Subsequently, a secondary, wall-attached local thrombus is generated and, in long term, the vein will be transformed into a fibrous cord (Redondo and Cabrera, 2005). Each class and individual type of sclerosant within the same class produce this effect in different ways associated with diverse and highly variable patterns of efficacy, potency, and complications (Artemi, 2007). According to their potency, sclerosing agents can be classified as major (alcohol, iodine, sodium tetradecyl sulfate), intermediate (sodium salicylate, POL), or minor (chromated glycerin).

Modern sclerotherapy is performed using sclerosing detergents such as POL. With their introduction in the 1920s and 1930s, detergent sclerosants, also known as fatty acids and fatty alcohols, soon became and are still the most popular sclerosant types used worldwide for the treatment of varicose veins. Detergent sclerosants, with increasingly favorable risk-

to-benefit ratios, rapidly replaced older sclerosants thought to be ineffective if they could not produce tissue necrosis or significant thrombosis at vasodestructive concentrations. After intravenous injection, liquid detergent sclerosants become protein bound and inactivated when mixed with blood. When the foam sclerosant is injected it fills the vein completely; it also displaces blood from the vein and ensures that all endothelium is exposed to the sclerosant agent and destroyed.

Medical observations prove that foam sclerotherapy is preferable instead of the use of liquid sclerosing substances (Ouvry et al., 2008). Detergent-like sclerosing agents can be transformed into fine-bubbled foam by special techniques. Orbach was the first who described the use of froth in sclerotherapy (Orbach, 1950). In 1997 new methods of transforming the sclerosing liquid into foam were proposed (Cabrera et al., 1997a, 1997b; Henriet, 1997; Monfreux, 1997) and Cavezzi and Frullini in 1999 reported their 13 month experience of duplex-guided sclerotherapy with sclerosing foam prepared by Monfreux's method (Cavezzi and Frullini, 1999). Sadoun and Benigni (Sadoun and Benigni, 1998) and Garcia-Mingo (Garcia-Mingo, 1999) suggested new ways of manufacturing sclerosing foam. In December 1999 Tessari described a safe and easy method to generate a fairly stable and compact foam (made of a mixture of micro-bubbles of detergent drug and air) using two plastic syringes and a three-way tap (Tessari, 2000). Subsequently Frullini (Frullini, 2000) and Gachet (Gachet, 2001) suggested other ways to produce a sclerosing foam. The ability to be agitated and foamed increases the potency of detergents from 2 to 4 times by mechanically displacing blood and thus maximizing the surface area and the contact time with the endothelium. This process has the advantage of using small and presumably less allergenic or less tissue-toxic concentrations and volumes of sclerosant, although greater risks may occur if foam passes in the ocular or cerebral circulation (Duffy, 2010).

The precise procedure to prepare the microfoam is the subject of a confidential agreement with a third party who is the proprietary. It is, however, based on the procedure described in granted European and US patents EP 656 203 and US 5 676 962, respectively (Cabrera and Cabrera Jr., 1997b). The Polidocanol microfoam produced by Provensis has successfully undergone phase III clinical trials in Europe and is currently in the process of obtaining approval from the Food and Drug Administration (BTG International Ltd., 2011; Redondo and Cabrera, 2005).

Dermatologic application of laser technology was initiated by Leon Goldman. His work was further elucidated by some authors (Anderson and Parrish, 1983), who described the principle of selective photothermolysis. The application of this principle shows how laser energy is preferentially absorbed by the intended target tissue or chromophore, resulting in their controlled destruction while minimizing collateral thermal damage to surrounding normal tissue. To achieve selective photothermolysis, a wavelength that is preferentially absorbed by the intended target tissue (in our case the vein tissue) or chromophore is selected. The pulse duration of the irradiating beam is also important for selectivity. If the pulse duration is too long, heat absorbed by the veins will diffuse out in the surrounding tissues so that the veins will be not selectively heated to the necessary degree in order to be destroyed. If the pulse duration is too short, the light absorbing chemical species such as hemoglobin in blood will be heated causing vaporization. This effect can cause purpura. Theory dictates that the proper pulse width should match the thermal diffuse time of the targeted structure. For smaller vessels, for example, this thermal diffusion time can be of the

order of hundreds of microseconds to several milliseconds, while for larger leg veins it could be 5-100 ms.

In the last decade, the treatment technique has been improved so that predictable, reproducible results can be achieved. Several investigations (Mordon et al., 2003) have enabled us to better understand the role played by the transformation of oxyhemoglobin (OxiHb) into methemoglobin (MetHb) in order to obtain a more marked, more selective laser light-blood chromophore interaction.

Long-pulsed (as mentioned above) Nd:YAG lasers offer great advantages for treating varicose veins of the lower limbs. They can penetrate to a depth of 4 to 6 mm and may be used to treat more deeply seated vessels (Rogachefsky et al., 2002; Ross and Domankevitz, 2005). Also, they are less absorbed by melanin (which means that Nd:YAG lasers can be used to treat dark phototypes). On the other hand, in order to produce penetration, high fluence is required and hence treatment is painful and requires cooling and even topical anaesthesia (Anderson and Parrish, 1983).

The light absorption of whole blood is different depending on the osmolarity, hematocrit, hemodynamics and red cell morphology variables. The MetHb and protoporphyrin IX (PpIX) have an absorption coefficient at 1,064 nm four times greater than Hb. It is shown that one pulse of the Nd:YAG laser causes substantial transformation of Hb into MetHb, followed by a drastic decrease in blood circulation speed (Mordon et al., 2003). Then, a second Nd:YAG laser pulse, shortly after the first one, would be four times more effective. Following this theory, a system was manufactured with two different lasers built into the same console. The energy of the two lasers is sequentially pulsed. First, a pulse of a dye laser emitted at 585 nm produces an increase in the rate of intraerythrocytic MetHb, changing the coefficient of absorption of the Nd:YAG laser beam which is delivered after the first sequence. The long pulse emitted by the 1,064 nm Nd:YAG laser induces vessel damage at lower fluence. In reality, the pulsed dye laser has low penetration and so its efficiency is limited to the most superficial dermal plexus, since the vessels located at a greater depth than 1.5 mm are not reached by these laser pulses. Hence, significant rates of MetHb cannot be formed in the deepest veins (Trelles et al., 2010).

Clinical experimental results have proved that the exposure of tissues impregnated with POL to laser radiation emitted at 1.06 μm improves the efficacy of the treatment (Moreno Moraga, n.d.; Trelles et al., 2011). It combines the effect of sclerotherpy with that of the laser therapy and more than this, with the capabilities of drug induced modifications following the exposure to the laser radiation. Sometimes, the exposure of a medicine to laser radiation yields a product that is more biologically active than the un-exposed parent (Pascu et al., 2011; Wainwright, 2008).

In this respect, the data reported in this chapter represent a step to better understand the interaction of POL with the target tissues in the context of its exposure to pulsed laser beams emitted at 1,064 nm by the Nd:YAG laser.

2. Description of Polidocanol

Detergent sclerosants produce endothelial damage by multiple mechanisms associated with a decrease in endothelial cell surface tension, interference with cell surface lipids, disruption

of intercellular cement and extraction of cell surface proteins. Detergents are most effective in the form of micelles (molecular aggregates) drawn in Fig. 1.

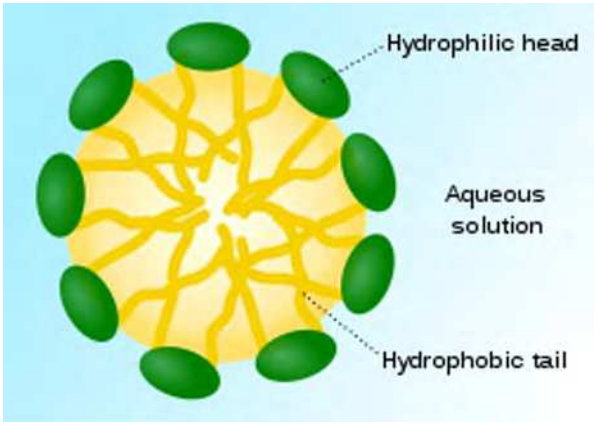


Fig. 1. Scheme of a micelle

At low concentrations and temperatures most detergent molecules are individually dissolved in solution, micelles are not formed and toxicity to endothelium is considered minimal. By increasing sclerosant concentrations at temperatures encountered in living tissue, a predictable threshold for endothelial damage occurs with a commensurate increase in sclerosing potency. The degree of damage can be controlled varying the detergent concentration and the time length of the sclerosant contact with the vessel wall. Unfortunately, due to the dilution with blood, it is not possible to determine the quantity of the sclerosing agent in intimate contact with the endothelial surface and the quantity which remains in circulation. Thus, the effects obtained are not always in direct proportion to their concentration; ''downstream'' effects with unintended damage to interconnecting vessels can always occur (Duffy, 2010).

2.1 Introduction of the compound

Polidocanol or Lauromacrogol 400, $C_{14}H_{30}O_2$ (CID 24750, molecular weight 230.3868 [g/mol]), is the pharmaceutical active ingredient of commercially available drugs listed in Table 1. It is a polyethylene glycol ether of Lauryl alcohol, where the average value of polymer is 9 (PubChem, 2011). This chemical compound is a viscous liquid at room temperature, having a melting point of 15–21°C. It is miscible in water, has a pH of 6.0–8.0 and has a density of 0.97 g/cm^3 at room temperature, closed to that of the water.

The 3D image of this molecule is shown in Fig. 2, where the carbon atoms are in dark grey colour, the hydrogen atoms are light grey and the oxygen atoms are red.

Polidocanol laurel macro gel 400 laureth-9 was first synthesized in 1936 and marketed as a topical and local anesthetic under the trade name SCH-600 (Chemische Fabrik Kreussler & Co. GmbH, Germany). This urethane local anesthetic differs from classic ester and amide anesthetic agents because it lacks an aromatic ring.

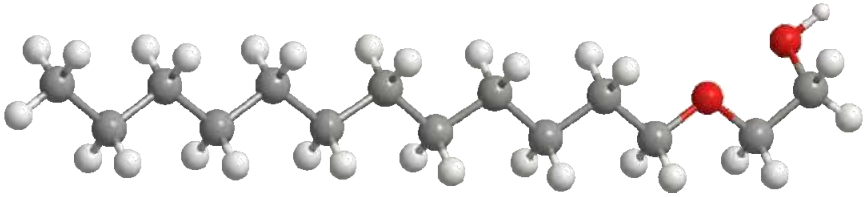


Fig. 2. 3D image of Polidocanol molecule, where the carbon atoms are figured in dark grey, the hydrogen atoms are light grey and the oxygen are in red color.

It is used as a topical anesthetic in ointments and lotions for skin irritation, burns, and insect bites and as an epidural anesthetic. The ability of POL to sclerose blood vessels without significant damage to surrounding tissues led to its use as a sclerosant in the 1960s. By 1967, it was registered in the Federal Republic of Germany as Aethoxysklerol, and it is now the only sclerosant approved for use in Germany (Duffy, 2010). Today, the leading sclerotherapy treatment in Europe is based on Aethoxysklerol. It is the only sclerosant approved by the Japanese Ministry of Health, Labor and Welfare (Eckmann, 2009). Recently - March 2010, the U.S. Food and Drug Administration approved Asclera (POL) injection (produced by Chemische Fabrik Kreussler & Co. GmbH, Germany, too) for the treatment of varicose veins in USA (U.S. Department of Health & Human Services, FDA., 2010).

In Table 1 are synthesized the main pharmaceutical companies, researchers, developers, manufacturers, distributors and suppliers to provide POL.

Companies	Product name
Chemische Fabrik Kreussler & Co., Germany	Aethoxysklerol, Asclera(USA)
Berlin Pharmaceutical Industry, Germany	Aethoxysklerol
Cem Farma Ilac, Turkey	Aethoxysklerol
Codali, Belgium	Aethoxysklerol
Felo, Denmark	Aethoxysklerol
Globopharm, Switzerland	Aethoxysklerol
IBI International, Czech Republic	Aethoxysklerol
Institute of Pharmaceutical Research and Technology, Grece	Aethoxysklerol
Inverdia, Sweden	Aethoxysklerol
Lomapharm Rudolf Lohmann, Germany	Aethoxysklerol
Nycomed, Switzerland	Aethoxysklerol
Repharma, Azerbaijan	Aethoxysklerol
Resinag, Switzerland	Sclerovein
Sigma-Tau, Italy	Aethoxysklerol
Tamro Distribution, Sweden	Aethoxysklerol

Table 1. The main pharmaceutical companies, researchers, developers, manufacturers, distributors and suppliers of POL

POL should be stored at room temperature (15–25^{0}C). No special precautions are necessary when disposing of this material. POL at concentrations of 0.5% to 1% is stable for 3 years

and should be stored between 15^{0}C and 30^{0}C (59–86^{0}F). There are no special precautions for the disposal of unused POL (product insert).

The common pharmaceutical presentation form of Aethoxysklerol is as ampoule of 2ml injection solution whose composition is given in Table 2.

Composition of injected solution in concentration of:	0.5%	2%	3%
Lauromacrogol 400 (active)	10mg	40mg	60mg
Ethanol (inactive)	0.10ml	0.10ml	0.10ml
Pure water (inactive)	2ml	2ml	2ml
Sodium hydrogen phosphate (buffer - inactive)			
Potassium dihydrogen phosphate (buffer - inactive)			

Table 2. Composition of Aethoxysklerol injected solution

2.2 Mechanism of the sclerosing action

Polidocanol exerts its sclerosant effects by causing concentration dependent differential cell injury (Eckmann and Kobayashi, 2005). Cellular calcium signaling and nitric oxide pathways become activated by the administration of the sclerosant, followed by cell death. The timing of endothelial cell death is predictable based on sclerosant concentration during exposure. This ultimately results in endothelial cell lysis but that can potentially also involve erythrocytes, platelets, and lead to platelet-derived microparticle formation (Parsi et al., 2008). Although hemolysis occurs experimentally in whole blood samples at Polidocanol concentrations greater than 0.45%, erythrocyte lysis, platelet lysis and platelet-derived microparticle formation have not been a significant concern reported in any clinical trials of sclerosant therapy.

2.3 Pharmacokinetics

POL: 12 hours after intravenous application, about 90% of the POL administered would have been eliminated from the blood. No accumulation is to be expected even after repeated doses at intervals which are normally used for sclerotherapy.

In a study made by Artemi and reported in 2007, the following values were determined after a single intravenous dose: protein binding 64%, terminal elimination half-life 4 hours, volume of distribution 24.5 l, total clearance 11.7 l/h, renal clearance 2.43 l/h and biliary clearance 3.14 l/h.

Ethanol: No data are available for the level of the ethanol absorption rate. The volume of distribution of ethanol is 0.68 l/kg for a man and 0.55 l/kg for a woman, and is reached very quickly. Ethanol enters the foetus and mother's milk. Ethyl alcohol is oxidised by alcohol dehydrogenase in the liver to form acetaldehyde, and acetaldehyde is in turn broken down by acetaldehyde dehydrogenase to form acetic acid. This metabolic breakdown method accounts for 90-96% in man. The rate of elimination is independent of concentration and is 0.1 g/kg/h for a man and 0.085 g/kg/h for a woman (hourly breakdown c. 0.15‰). Insignificant amounts are eliminated via the lungs (2-3%) and kidneys (1-2%).

2.4 Foaming process

There are many works providing good results about the use of POL foam sclerotherapy (Alos et al., 2006; Cabrera et al., 2000; Cavezzi and Frullini, 1999; Frullini, 2000; Gachet, 2001; Garcia-Mingo, 1999; Hsu and Weiss, 2003; Ouvry et al., 2008; Rabe et al., 2008; Redondo and Cabrera, 2005; Santos, 2008; Tessari, 2000). The ability to be agitated and foamed increases the potency of detergents from 2 to 4 times by mechanically displacing blood and thus maximizing surface area and time in contact with endothelium. This process has the advantage of using small and presumably less allergenic or less tissue-toxic concentrations and volumes of sclerosant, although greater risks may occur if foam passes in the ocular or cerebral circulation (Duffy, 2010). Since foam displaces the intravascular blood and is not diluted in it, as in the case of liquid sclerosant injection, the concentration of the sclerosing agent in the vessel is known and controlled. Foam instillation leads to a homogeneous distribution of the sclerosant within the vessel lumen, except in very large veins where gravitational effects maintain better contact between the upper venous wall and the foam, which is far less dense than blood. Foam can be produced to be sufficiently stable from breakdown so being possible to provide adequate therapeutic effects from relatively short times in which it is kept in contact with the lumenal surface of the vein, as reported (Rao and Goldman, 2005).

Generally, the sclerosing foam is defined as a mixture of gas and liquid sclerosing solution with tension-active properties. Besides being composed of different specific ingredients, foams can differ in their internal cohesion, which is related to the size of the air bubbles. Macrofoam contains bubbles larger than 500 μm, minifoam contains bubbles between (250 – 500) μm, and microfoam is composed of bubbles smaller than 250 μm (Frullini, 2011; Hsu and Weiss, 2003).

A given volume of liquid can be used to produce 4 or 5 times its volume in foam, depending on the utilized foaming method. This volume allows the use of a lower total dose of the sclerosant to achieve the desired effect. Sclerosing foam is characterized by variables such as: the type and concentration of the tension-active sclerosing agent, the gas type, the liquid-to-gas ratio, the preparation conditions (temperature, pH), the time between preparation and use, and the bubble size.

The foam homogeneity is an important prerequisite for its flow behavior (viscosity) and stability. The ideal foam would have uniform bubble diameter and inner gas pressure. The durability of foam is related to the bubble size, the tension-activity (surface tension properties) of the liquid solution and the conditions under which the foam is formed and kept. Foam should be sufficiently durable to avoid its separation into gas and liquid components during injection but sufficiently ephemeral to break down once injected. The gas must be physiologically tolerated at therapeutic doses. The mean bubble size of the foam should be considerably less than 100 μm (Redondo and Cabrera, 2005).

There are many methods and mechanisms that have been applied to mix and agitate liquid sclerosants with gas admissions to create foams for clinical use. With the exception of one self-contained formulation, POL microfoam for endovenous use (Varisolve®), foamed sclerosants are typically produced by cyclical mechanical agitation of the liquid agent in the presence of a gas to generate the froth used for intravascular injection. Such “home made” foams commonly employ ratios of gas to liquid ranging from 1:1 to 8:1, producing foams of

varying densities and rheological properties. In any event, the result is a froth containing 79% nitrogen and 21% oxygen and having a characteristic wide bubble size distribution (Cavezzi and Tessari, 2009).

Orbach was the first who described the use of froth in sclerotherapy (Orbach, 1950). After 1995 new methods of transforming the sclerosing liquid into foam were described (Cabrera et al., 1997a; Cavezzi and Frullini, 1999; Garcia-Mingo, 1999; Henriet, 1997; Monfreux, 1997; Sadoun and Benigni, 1998). In December 1999 Tessari described a safe and easy method to generate a fairly stable and compact foam (made of micro-bubbles of detergent drug and air) using two plastic syringes and a three-way tap (Tessari, 2000). Nowadays this is the main method used in foam sclerotherapy. Subsequently, Frullini and Gachet suggested other ways to produce sclerosing foam (Frullini, 2000; Gachet, 2001).

In the Monfreux technique, negative pressure is generated by drawing back the plunger of a glass syringe, the tip of which is tightly closed. The resulting influx of air produces large-bubbled, fairly fluid foam. In the Tessari technique, the turbulent mixture of liquid and air in two syringes connected using a three-way stopcock produces the foam. It is fine-bubbled and fluid at low concentrations and rather viscous at high concentrations. The mixing ratio for sclerosant to air is 1:3 to 1:4. The double syringe system technique involves the turbulent mixing of POL with air in a sclerosant to air ratio of 1:4 in two syringes linked using a connector. The resulting product is a fine-bubbled, viscous foam (Rabe and Pannier, 2010).

Some authors described in 2008 a method to obtain standardized POL foam prepared by using the EASY-FOAM® kit (two 10 ml silicone-reduced syringes, connected by a two-way valve connector, with one syringe prefilled with 7.4 ml sterile air (Laboratoire Kreussler Pharma, Paris, France). After aspiration of 1.6 ml POL into the other syringe (1:5.6) standardized movements of the syringe plungers were achieved by using the Turbofoam® machine, with a controlled number of movements, speed, and power (Hamel-Desnos et al., 2005; Rabe et al., 2008).

Lately, BTG International (United Kingdom) has developed Varisolve® (polidocanol endovenous microfoam, PEM) as a first line treatment for incompetent great saphenous veins (GSV) and associated varicosities, above and below the knee and for use alongside endovenous thermal ablation. PEM has a controlled density, consistent bubble sizes, and proprietary gas mix that make it a simple and comprehensive treatment for symptomatic and aesthetic varicose veins.

PEM is a pharmaceutical form of micro-foam that emulates the foam originally produced and studied by Cabrera (Cabrera et al., 2000, 2003, 2004) in a standardized way. It is generated and dispensed using a pressurized canister mechanism depicted in Fig. 3.

An European Phase III clinical trial showed that 90% of patients treated with PEM had no reflux in the GSV at 3 months and fewer than 10% of patients had recurrence at 1 year. Patients can generally return to work or their usual activities the same day they are treated, and cosmetic results after PEM treatment are apparent at 6 weeks. PEM is progressing through three US Phase III trials to explore its safety and efficacy as a treatment for moderate to severe varicose veins. All studies are expected to be completed by the end of 2011 and potential approval in 2013 (BTG International Ltd., 2011).

The system contains the liquid agent and a gas mixture of oxygen and carbon dioxide with only trace (0.01–0.08%) nitrogen present. Passage of the gas and liquid under pressure through a microfoam producing system yields micro-structurally consistent 1% POL microfoam having reproducible rheological properties. The bubble size for Varisolve® foam is appreciably smaller than that resulting from manual foam production techniques, and the absence of nitrogen facilitates more rapid absorption of bubbles within the body. Both these considerations are important to the safety profile, given the possibility for gas embolism to occur with any type of foam sclerosant therapy (Eckmann, 2009).

Fig. 3. The Varisolve® PEM generation and dispensing canister.

3. Laser sources description

For laboratory measurements regarding the photophysical properties of POL, we used a Q-switched Nd:YAG laser, while for clinical applications long pulsed laser types with the same active medium were employed.

The difference between the two types of lasers is the fact that for long pulsed medical lasers the emission of the laser resonator is obtained from a simple laser cavity where elements characteristic to Q-switch mode are not present. In this way the laser pulse duration is given by the long life time of the upper laser level (of the order of milliseconds) and lamps emission modulation.

Our Q-switched laser is a Continuum Surelite II. The head of the laser consists in a 115 mm length rod of Nd^{3+} doped YAG pumped by a linear flash lamp. The high pumping efficiency is achieved through a closed coupled configuration surrounded by a high brilliance magnesium oxide diffuser. The flashlamps have Xe as discharge gas with a pressure of (1-3) atm.

The active Q-switch is performed by the combination of a polarizer, Pockels cell and $\lambda/4$ waveplate. The Q-switch allows the accumulation of high energy in the laser resonator till it opens and allows the cavity to oscillate. By this, fast and high peak power laser pulses are

produced. The typical laser pulses at 1,064 nm emission wavelength for our Surelite system have 5 ns pulse duration, 10 Hz repetition rate, and maximum peak energy of 685 mJ.

The fundamental laser beam emitted at 1,064 nm can be doubled, tripled, and quadrupled by using high harmonic generation BBO crystals. The characteristics of the beams obtained are: 532 nm, maximum energy per pulse 340 mJ; 355 nm, maximum energy per pulse 180 mJ; 266 nm, maximum energy per pulse 120 mJ.

The Nd:YAG lasers used for medical applications in leg veins treatment are long pulsed lasers, not having a Q-switch operation. Two different commercially available, long-pulsed 1,064 nm Nd:YAG laser systems were engaged for the clinical results presented here, namely:

- The Laserscope Lyra "i" (Laserscope, San Jose, CA, USA). To protect the epidermis from any adverse effects, Lyra-i system combines the long available wavelength with a unique flat beam profile to maximize patient safety and efficacy. The patients are protected by continuous parallel contact cooling that protects the epidermis. The cooling is carried out with a continuous flow of air (Zimmer Cryo 5® system, Zimmer Elektromedizin, Neu Ulm, Germany). The depth of penetration for 1,064 nm is due to its low scattering and epidermal transparency allowing the effective delivery of energy to deeply seated targets, such as blue and purple vessels while safely avoiding epidermal pigment. Computer adjusted fluence, delivered in adjustable pulse widths coupled with 1 mm to 5 mm continuously adjustable spot size is ideal for treating vessels from 0.5 mm to 4 mm.

The adjustable spot size and laser fluence allow the appropriate treatment procedure for several classes of veins. A spot size of 2 mm and fluence of 200 J/cm^2 are selected for spider (class I) and reticular (class II) veins, while 5 mm and 60 J/cm^2 are the laser system parameters for truncal veins (class III) treatment. The pulses delivered are 25 to 35 ms at a frequency of 5 Hz, for class I and II varicose veins, and 70 ms and 2 Hz, for class III varicose veins.

- A CoolTouch Varia® (CoolTouch Inc., Roseville, CA, USA) laser which has an adjustable pulsed cryogen cooling. The system is foreseen with an attached gas cooling canister that enables cooling the epidermis before and after laser pulses, at the same time interval between pulses. The thermal feedback is performed by precisely adjustment of the laser power to safely reach target temperature. Fluences used are similar, except for class I and II veins for which less than 150 J/cm^2 are programmed due to the fact that the minimum spot size is 3 mm and not 2 mm as in the Laserscope Lyra "i" system.

3.1 Laser tissue interaction

Basic requirements for a laser or a light source to treat leg veins are a wavelength that is proportionately better absorbed by the target (hemoglobin) than surrounding chromophores and penetration to the full depth of the target blood vessel. Sufficient energy must be delivered to damage the vessel without damaging the overlying skin, and this must be delivered over an exposure time long enough to slowly coagulate the vessel and its lining without damaging surrounding tissue.

Usually, optical radiation in the near-infrared wavelength range (from about 700 - 2,000 nm) is used, though when appropriate chromophores are available, visible wavelengths (e.g. green) can also be used. Photons launched into tissue are submitted to at least four optical processes: refraction, scattering, absorption, or output from the tissue.

In the context of this chapter, the light scattering in the tissues plays an important role since it regulates in a sense the penetration depth of the laser radiation in the tissue. There are different types of light scattering which may be produced in a tissue by its components, such as the Rayleigh scattering produced by very small particles, the Gans and Debye scattering produced by spherical and cylindrical macromolecules, globular proteins etc. (Grossweiner, 2005). Actually, the presence in the tissue of macromolecules, cells, water/liquid volumes, structures that have constituent layers (such as veins and arteries, cornea) enhance the light scattering within the tissue.

When photons are absorbed, the energy from the photon is converted into inter- and intra-molecular energy and results in generation of heat within the tissue. At the same time the good absorption in tissue limits the size of the lesion created by the laser irradiation. So, a compromise between good penetration and good absorption has to be found. After the initial absorption the temperature generated spreads through the tissue and enlarges the lesion somewhat, depending on the perfusion of the tissue. Large vessels will transport the heat away from the site and the effectively achieved temperature is reduced. For vascular structures the thermal relaxation time is of the order of milliseconds, so that lasers with pulse durations less than 1 ms are likely to produce little thermal injury.

Following Altshuler's expanded theory of selective photothermolysis, the chromophore is the Hb of the circulating blood. This captures the laser energy and turns it into heat. The chromophore must heat up sufficiently for thermal damage to occur in the vessel and especially on the vascular wall (Altshuler et al., 2001). The chromophore must heat intensely enough for heat to propagate to the wall of the vessel, but not to the extent that this becomes carbonized, which would involve the end of heat transfer in the vein structure. These data are highly important when deciding upon laser pulse dosimetry.

When the parameters (fluence and pulse width) are well chosen, intraparietal bleeding and rupture of the elastic fibers of the vein wall are observed immediately. Then, eosinophil appears in the muscle fibers of the treated vein. With the appropriate histology staining, there is a notable increase in the presence of beta-2-type tissue growth factor (TGF-beta 2). This intermediary acts on the fibroblasts increasing the secretion of proteins in response to thermal damage (heat shock protein 70), which are responsible for accelerating collagen and elastin fiber synthesis in the rough endoplasmic reticulum of the fibroblast (Black and Barton, 2004; Sadick et al., 2001). The first sequence is to induce vessel fibrosis and its re-absorption.

Optical absorption in the near-infrared spectral range is generally due to combination of overtone bands of fundamental molecular stretches. For wavelengths near 1,000 nm, water is a primary absorber of optical energy.

The choice of wavelength and pulse duration is related to the type and the size of the target vessel. Deeper vessels require a longer wavelength to allow penetration to their depth. Pulse duration must be matched to vessel size; the larger the vessel diameter, the longer the pulse

duration required to effectively damage the vessel thermally. To be most effective, thermal injury must encompass the full thickness and circumference of the vein wall endothelium, rather than just the most superficial aspect of the vein wall. The relative importance of the hemoglobin absorption peaks in green (541 nm) and red to infrared (800-1,000 nm) shifts as the depth and the size of the blood vessel changes. Absorption by hemoglobin in the long-visible to near-infrared range appears to become more important for vessels more than 0.5 mm in diameter and at least 0.5 mm below the skin surface.

This approach of using pulsed lasers to limit thermal effects can also be used to ablate tissue. The limiting pulse length would be determined by the thermal relaxation time of the material. This is the time for heat to diffuse out of the irradiated volume and it is determined by the thermal diffusivity of the tissue and the dimensions of the volume. When laser energy is deposited in pulses shorter than the thermal relaxation time, heat accumulates and high temperatures are achieved. The ablative event can then occur before the heat diffuses out of irradiated volume. This confinement of heat can reduce the thermal damage incurred by adjacent tissue.

4. Experimental devices

First of all, we measured the optical properties of Aethoxysklerol 2% (Chemische Fabrik Kreussler & Co., Germany) solution. The absorption spectra measurements in spectrophotometer cells of 10 mm optical length are performed in UV-VIS-NIR spectral ranges using a Perkin Elmer Lambda 950 spectrophotometer (PerkinElmer, Massachusetts, USA) that has UV/VIS resolution ≤ 0.05 nm, NIR resolution ≤ 0.20 nm, with an error limit for absorbance of ± 0.004 %. This high-performance spectrophotometer features a double beam, double monocromator and ratio-recording optical system. As for the IR spectral range we used a FTIR Spectrophotometer Nicolet Magna 550 (Nicolet Instruments Inc., Madison, USA), working at (4000 – 400) cm^{-1} spectral domain and resolution 0.125 cm^{-1}.

Secondly, we exposed medicine solution samples to laser radiation in order to detect the possible molecular modifications induced in drug by the laser beam. The irradiation experimental set-up is shown in Fig. 4. We exposed Aethoxysklerol solution samples at laser beam emitted by a pulsed Nd:YAG laser (Continuum, Santa Clara, USA,, Excel Technology, Surelite II Frequency 10 Hz, FTW 5 ns) at 1.06 μm, the laser radiation having the following characteristics: repetition rate 10 pps, beam energy on the sample 50 mJ.

The exposure time was made on samples in bulk, between 2 min and 30 min; the sample was introduced in spectrophotometer cells of 10 mm (C – Fig. 4) and the corresponding irradiation dose varied from 60 J/cm^2 to respectively 900 J/cm^2; 8-10 % of the laser radiation was directed through a beam splitter (BS) unto a powermeter (P).

The effect of the laser light may be enhanced if the POL is used as foam since the light scattering in the tissue becomes more important and the effective absorption of the laser beam becomes larger. So, one might have in this case a larger number of modified POL molecules and/or a larger volume of tissue which is exposed to laser radiation.

Using the Tessari method - two disposable 5 ml syringes (Luer Slip) connected to a three-way stopcock, we produced foam from a mixture of Aethoxysklerol solution and atmospheric air; the mixture ratio was 1:4 (Fig. 5). This batch was passed between the two syringes about 40 times and the resulting hand-made foam was stable for 5-6 min.

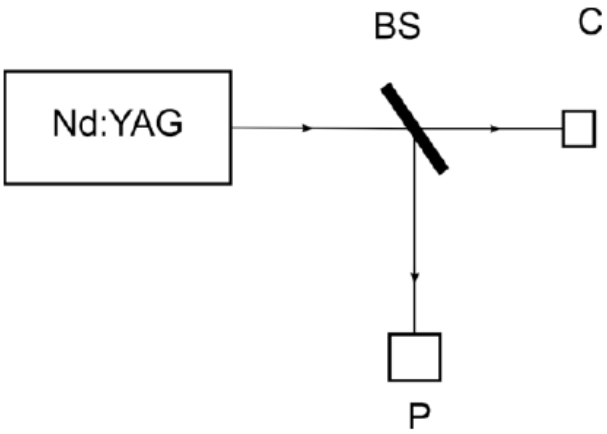
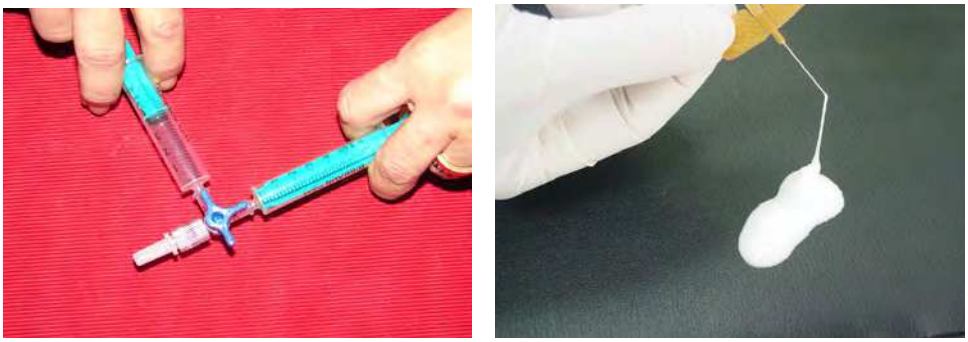


Fig. 4. Experimental set-up for laser irradiation of POL samples (P- powermeter, C- sample cell, BS- beam splitter)



(a) (b)

Fig. 5. The Tessari foaming method (a) and foam consistency (b)

Foam and solution samples of POL were investigated by Raman spectroscopy as shown in the experimental set-up in Fig. 6. The laser radiation used to excite the Raman emission is the second harmonic of the same pulsed Nd:YAG laser emitting pulsed radiation at 10 Hz repetition rate, pulse duration 5 ns, and 250 mJ energy at 532 nm. The complete description of the laser source was made in Section 5. The detection and analysis of Raman signal are made by a spectrograph (SpectraPro 2750, Acton Research) and ICCD camera (PI-MAX 1024RB, Princeton Instruments).

The SpectraPro 2750 is a 750-mm, f/9.7-aperture, triple-grating monochromator/ spectrograph that features a versatile multiport optical system, 0.0025 nm drive-step size, built-in computer compatibility, and a wide scanning mechanical range up to 1,400 nm). As a monochromator, it offers built-in stepping-motor scanning and 0.023 nm resolution, plus easy integration into automated spectral-data acquisition systems. As a spectrograph, the SpectraPro 750 provides 1.1 nm/mm dispersion, a large 14 mm high by 27 mm wide focal plane, and interchangeable triple grating turrets. The grating types are as follows:

holographic UV, 2,400 lines/mm, for (185-375) nm; holographic VIS, 2,400 lines/mm, for (300-800) nm, blaze 1 μm and 600 lines/mm, for (650-1,500) nm.

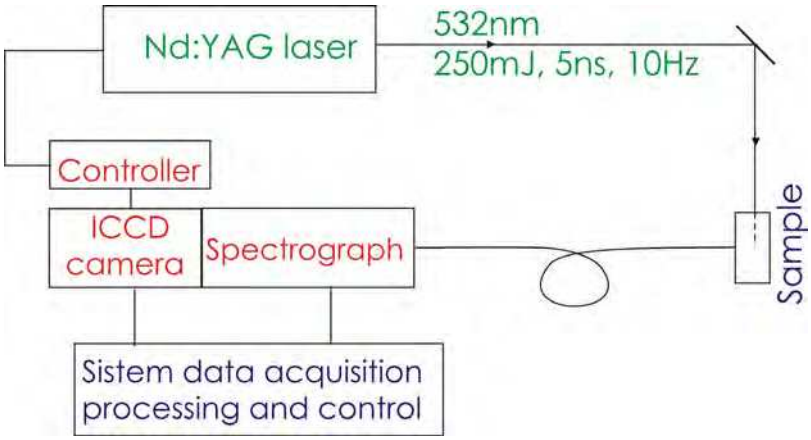


Fig. 6. The Raman spectroscopy system

The PI-MAX 1024RB from Princeton Instruments is a high performance intensified camera system featuring a spectroscopy format CCD. The imaging array is 1,024 x 256 and the spectral range (185-900) nm. It is fiber optically coupled to Gen II intensifiers with wide spectral coverage, quantum efficiency between (10-30)% in spectral range (200- 550) nm and 54 to 64 lp/mm resolution. Sub-nanosecond gating capability and an integrated programmable timing generator (PTG) make these ICCD cameras suitable for time-resolved spectroscopy applications.

5. Target patients groups and specific materials and method

From the clinical point of view and to illustrate this chapter we present the evaluation of leg telangiectasias and reticular veins (classes I to III) which were treated with the Nd:YAG laser following POL micro-foam injections. From our large casuistry we have selected a representative sample that underwent a prolonged control and follow-up.

Materials and method

This review corresponds to 200 patients, ages from 21 to 76 years (mean 37.2 y.o.a), treated with POL and Nd:YAG laser (Table 3). All patients were followed up and assessed periodically up to two years after treatment. Permission was granted to check medical files from the Administrative Council of the Instituto Médico Láser, Vilafortuny. The patients were females presenting leg varicosities that were treated with the same protocol. No patient has been exposed to sunlight or artificial tanning with UV-B during the 2 months prior to treatment; neither had they used any oral contraceptives for at least 12 weeks before the treatment. Exclusion criteria included: patients less than 18 years of age, pregnant women, lactation, scars or skin infections in the treatment area, the use of anticoagulants, a history of photosensitivity, keloids and/or hypertrophic scars and repeated herpes infections. None of the patients has been previously treated with laser for varicose veins.

60 patients suffered red venulectasias of less than 0.5 mm in diameter (class I); 72 of blue venulectasias of 0.5 to 1.5 mm (class II), and 68 patients had reticular veins 2 to 4 mm in diameter (class III).

	CLASS I VEINS Total no. pat. 60	CLASS II VEINS Total no. pat. 72	CLASS III VEINS Total no. pat. 68
AGE			
20-35 years	15	6	4
36-50 years	27	23	27
51-over	18	43	37
VEIN COLOR			
Red	41	39	12
Blue	6	5	42
Red/blue	13	18	14

Table 3. Patients demographics

All patients signed an informed consent for treatment and prior to treatment underwent a Duplex Ultrasound test (Soniline 050, Siemens, Issaqua, Japan) to screen for presence of reflux in the deep system, saphenous axes and in the perforant veins.

The micro-foam was obtained using two 10 ml Omnifix syringes with a Luer-Lock connection, a 3-way stopcock to connect the syringes and a 15G load needle with an air micro-filter. Two ml of POL (Aetoxysclerol® tamponné/lauromacrogol 400 (Kreussler Pharma, Germany) were used at 0.3% and 8 ml of air. Pumping and passing from one syringe to the other 15 times, produces stable micro-foam following the Tessari technique (Cavezzi et al., 2002; Frullini, 2000; Hamel-Desnos et al., 2003; Tessari, 2001; Tessari et al. 2001; Wollmann, 2004). The resulting foam is rich in nitrogen, with an irregular bubble size and a highly internal cohesion (Redondo and Cabrera , 2005).

The 200 patients received two treatments. The second treatment was given six weeks after the first. Parameters and methodology were the same for both treatments. The laser used was a 1,064 nm long pulse Nd-YAG laser, *Laserscope Lyra* "i"™ (Laserscope, San Jose CA, USA). The laser had a variable spot diameter, from 1 mm to 5 mm. The laser energy per pulse was delivered according to the treatment program at the same time that constant cooling of skin surface occurred because of the glass chamber that is adapted to the tip of the hand piece nozzle. The coolant is constantly circulated to cool the skin. In addition, a continuous flow of cold air that is capable of giving up to 1,000 l/m was used.

The device offers various programs to select the air flow. Program #5 was used which gave 600 l/m. The nozzle was pointed, at all times towards the place where the laser beam was directed. The temperature of the cold air was of 4ºC. This was measured by pointing the thermometer under the air flow coming out of the hose nozzle. The approximate time of the air flow focusing on the treatment area was of one second; according to the external temperature detected with an IR thermometer (Laser Infrared Thermometer Center 350®, Center Technology Corp., Taiwan) the skin temperature was reduced to 22ºC from the basal temperature of 33ºC. The cooling device (Cryo 5, Zimmer Elektromedizin, Neu Ulm, Germany) followed the vein path treated with the laser pulses.

For class I and II veins, a 2 mm laser beam spot size was used, whereas for class III veins a 5 mm beam was used. Fluences were 200 J/cm^2 for class I and II and 60 J/cm^2 for class III. The laser pulse width was 30 ms for class I and II, and 70 ms for class III with a pulse repetition rate of 5 Hz for class I and II and 2 Hz for class III veins. According to the utilized protocol, all patients received treatment of the whole limb in the same session. For this, areas of 30x30 cm were selected continuing until the whole leg was treated. Disinfection of the areas for treatment was carried out using hydrogen peroxide. No anesthesia (i.e. any type of anesthesia) was used. Then, the POL micro-foam injection was injected using a 2 ml Omnifix syringe with a 30G needle. The sclerosant concentration was 0.8% in all cases and the maximum amount injected was 30 ml per session which proved to be enough in all cases. The average amount of sclerosant injected in each of the two scheduled treatments was around 20 ml.

Injection caused vessels to blanch and then a delay time of (1-3) min was necessary for the vessels to recover a soft pink color. Following the injection, laser was applied on the whole length of the vessel in the 30 x 30 cm area until it was totally covered with laser pulses. Because the total varicosity that the leg presented were treated in one session (with a repetition session after six weeks), the average number of laser pulses per session varied from 600 to 900.

After 6 weeks, the treatment was repeated with the same parameters. The patients returned for control at 18 weeks after the first treatment and the final assessment was at 2 years, taking as a reference the last (second) treatment carried out. At the 18-week control, complications were evaluated and further follow-up of their evolution was carried out at the 2 year assessment.

At this final assessment, all patients answered questionnaires to score in percentages their subjective impression regarding results. The percentages scored were: >90%, Very Good Results; 70 to 89%, Good Results; 50 to 59%, Fair Results and < 50%, Poor Results. Patients' satisfaction (PS) was determined by adding up the Very Good and Good results.

To objectively determine vessel clearance, photographs were taken, at a distance of 18 cm, using a Canon EOS 400D photo camera, equipped with a macro-lens (Tokina ATX Pro 100 f 2.8 Macro, Sea&Sea Flash Macro DRF 14, Tokyo, Japan). Photographs were taken before first treatment and at 2 years after the second treatment and they were used as objective references for evaluation by two independent Doctors, familiar with leg vein treatment with laser.

6. Physical and clinical results and discussions

It is important to provide a thorough understanding of the basic physical principles that underlie the use of coherent light in therapeutic applications. In this way, with respect to the new method to treat the varicose veins by combining POL microfoam and pulsed 1,064 nm Nd:YAG laser beam we must consider three factors: the drug sclerosis abilities, the action of the laser beam on the surrounding tissues and the possible increasing of the drug's potency under the laser light exposure. Of course, in practice it is important to adjust each of the parameters involved in the exposure to laser radiation in order to obtain the best results.

6.1 Optical results and discussions

Lasers used for the percutaneous treatment of varicose veins of the lower limbs must fulfil the principles of selective photothermolysis: a wavelength that is preferably more absorbed by Hb than by other similar chromophores; sufficient penetration as to achieve the depth at which the vessel for treatment is found; the appropriate energy to damage the vessel without damaging the skin; and release of energy slowly so that the heat reaches the vein wall without damaging the adjacent tissues (Suthamjariya et al., 2004). In order to achieve the above, the following variables must be taken into account: laser beam spot size, fluence, pulse width, pulse repetition rate and the technique used to apply the treatment.

Our laser irradiation and Raman spectroscopic measurements are made with experimental devices that involved time width pulses, energies, and irradiation doses that are different from those used in medical practice. However, they are made in order to better understand the physical processes specific to the interaction of the laser light with the tissues of medical interest in the treatment of the varicose veins.

The light propagation in turbid biological media is jointly governed by the absorption and scattering properties of the medium. On the other hand, the optical properties (absorption and scattering of light) of the marketed sclerosing substance may be subjects for studies in order to elucidate the mechanisms involved in this particular case in which combined sclerotherapy and light therapy of varicose veins are involved.

The absorption spectra of the medicine, shown in Fig. 7 and Fig. 8, indicate no significant absorption in the UV-VIS spectral range, and very weak peaks in NIR at 900 nm, 1.06 μm, 1.19 μm, 1.69 μm and 1.72 μm; they are the result of the superposed absorption of all the compounds included in the commercially available Aethoxysklerol 2%.

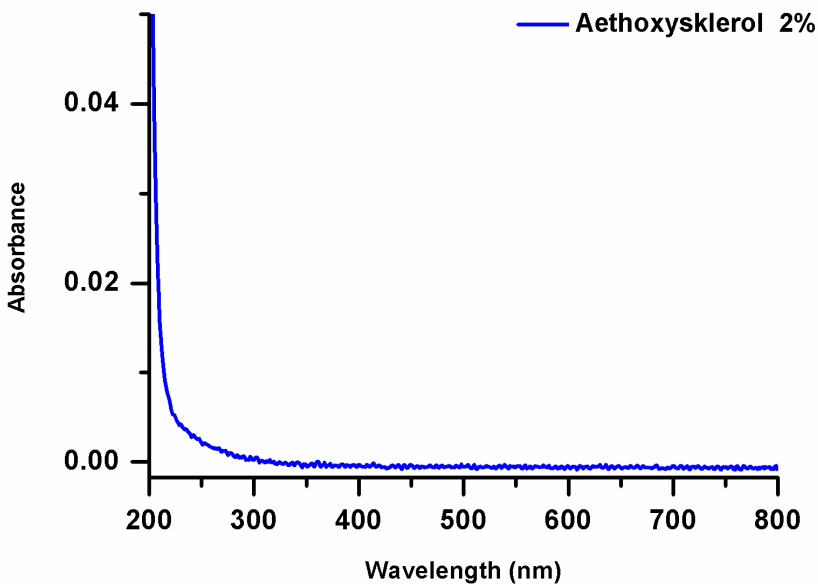


Fig. 7. UV-VIS absorption spectrum of Aethoxysklerol 2%

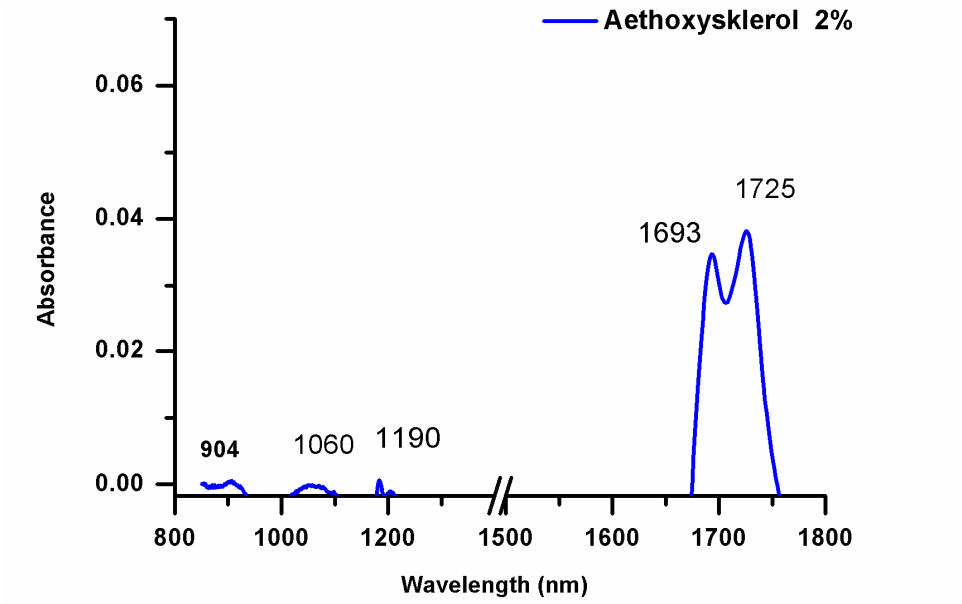


Fig. 8. NIR absorption spectrum of Aethoxysklerol 2%

Also, the FT-IR spectrum of POL shown in Fig. 9 is influenced by the absorption properties of water contained in the commercially presentation of the Aethoxysklerol solution.

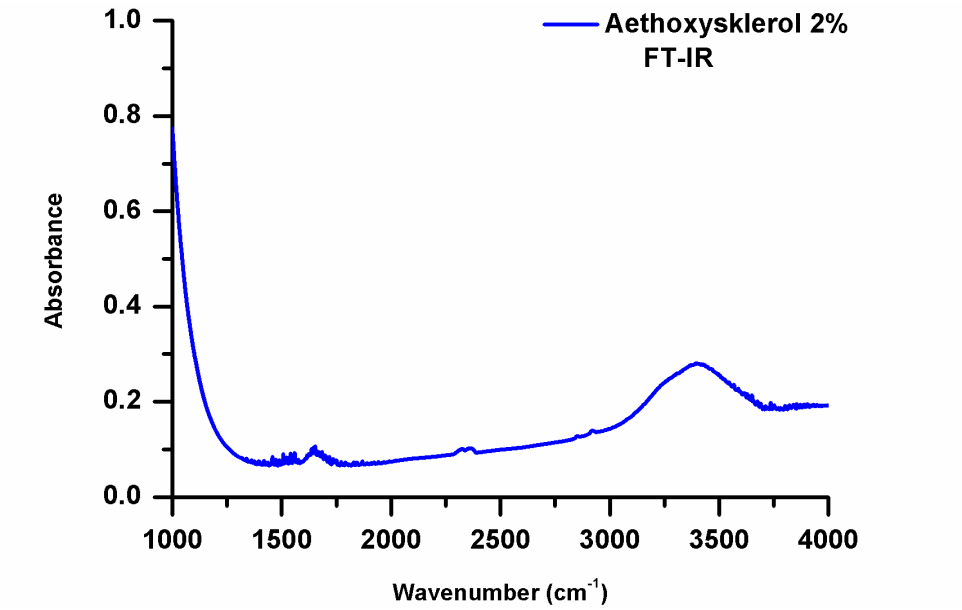
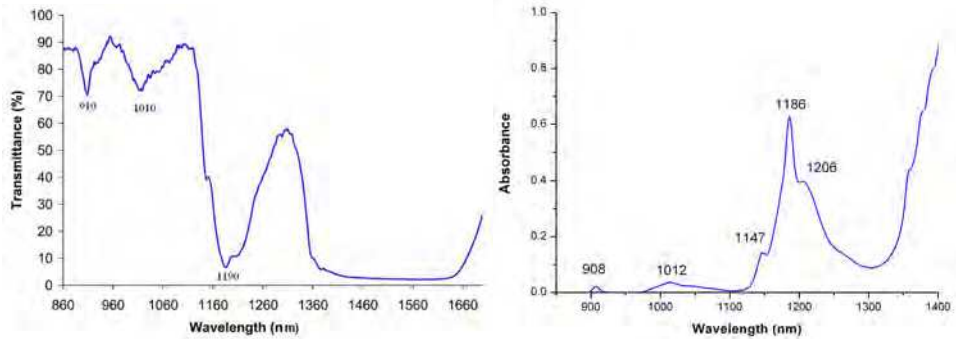


Fig. 9. The FT-IR spectrum of Aethoxysklerol 2%

The ethyl alcohol, for instance, is one of the inactive substances included in the commercially available Aethoxysklerol and has relatively significant absorption peaks at around 900 nm, 1 μm and 1.2 μm as it can be observed in Fig. 10. The absorbance measured in laboratory is in good concordance with literature reports (see Fig.10.a for recent literature reports).



a) b)

Fig. 10. The spectra of Ethanol in NIR: a) the transmission spectrum (Barun and Ivanov, 2010); b) the absorption spectrum (Smarandache et al., 2010)

Also, water presents broad absorption bands in NIR and middle IR, as it is shown in Fig. 11.

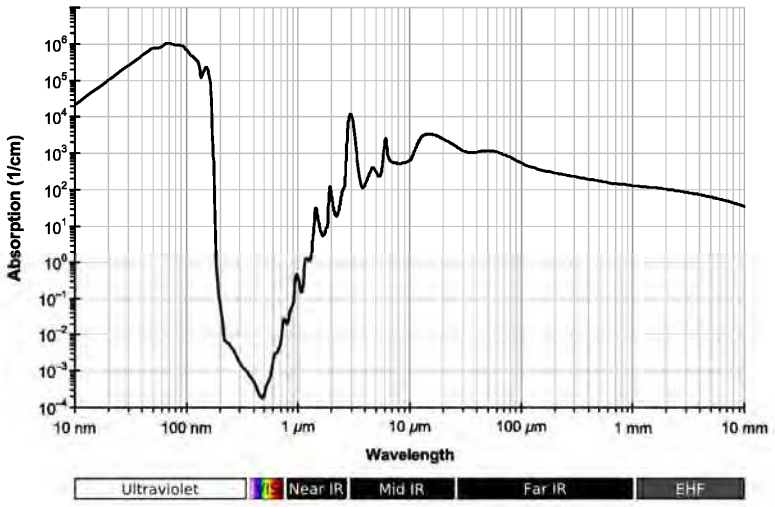


Fig. 11. The absorption spectrum of ultrapure water (Querry and al., 1998)

Some authors have assumed that the molecules of drugs can be photoactivated by assisting the hydrolysis process that results from excitation of vibration levels of the water molecule in the spectral range of 1-2 μm, following the absorption of infrared radiation by the water molecules (Fumarel et al., 2009).

We exposed the commercially available Aethoxysklerol 2% at laser beam emitted by a pulsed Nd:YAG laser at 1.06 μm, between 2 min and 25 min (exposure procedure as shown in Fig. 4). Following the irradiation, the absorption spectra of the medicine was measured (Fig. 12).

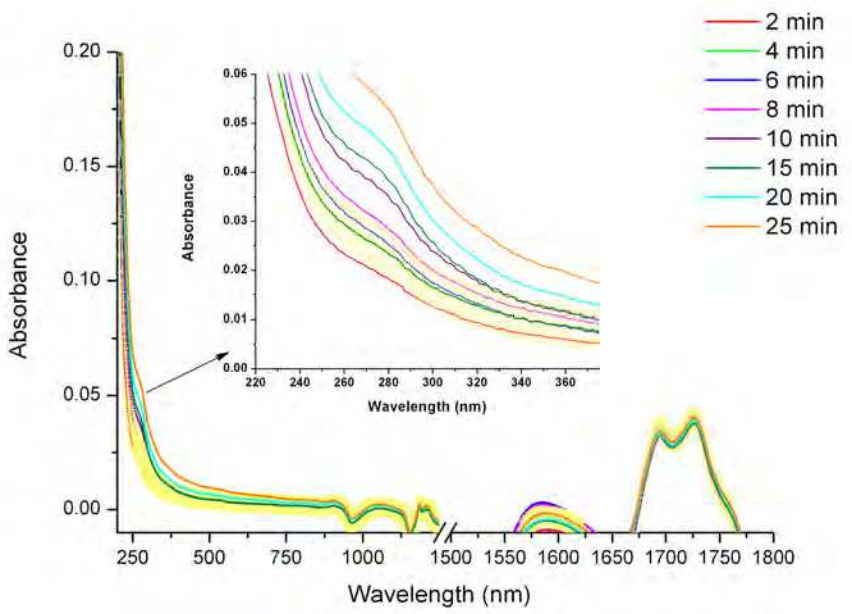


Fig. 12. The absorption spectra of Aethoxysklerol 2% exposed to 1.06 μm Nd:YAG laser radiation.

These spectra indicate that for wavelengths that exceed 500 nm they remain within the measuring error limits (±0.004 %). We assume that in the spectral range (1,000-1,100) nm the absorption is due rather to the main components of the tissue (melanin, water and hemoglobin/ oxihemoglobin); methemoglobin, especially, has the absorption three times greater than oxihemoglobin in this spectral range (Lee et al., 2006), (Fig.13).

Derivatives of blood hemoglobin, molecular oxygen dissolved in all components of the biological tissue, different ferments and other tissue substances that absorb light are considered as primary photoacceptors. Several photoinduced processes are known to take place during irradiation of an organism, such as photodissociation of oxyhemoglobin and the light-oxygen effect (Barun and Ivanov, 2010). Both mechanisms involve the absorption of light and the formation of oxygen in different forms which have a biological effect. The efficiency of these processes depends quantitatively on the way the light propagates and on the absorption coefficients of individual chromophores in the tissue of interest.

Modifications that as a trend are above the error limits are obtained in the spectral range (250-285) nm, as it is shown the detail in Fig. 12. In some cases (exposures at 4 min, 6 min, 8 min) the absorption curves may be considered within the error limits of the measuring system but they evaluate according to the trend.

A possible explanation of this behavior of the curves is that nonlinear absorption effects take place in Aethoxysklerol, such as the absorption of 4 photons at 1.06 μm, which would correspond to a transition at 266 nm. This modification may show that after absorbing four photons at 1.06 μm the Polidocanol molecules change their structure. The interaction mechanisms of laser radiation to the investigated solution are not completely elucidated and it is expected that further studies give a better understanding of them. More, it is expected that the modified Polidocanol is more efficient in destroying the varicose veins than the unirradiated solution (Smarandache et al., 2010).

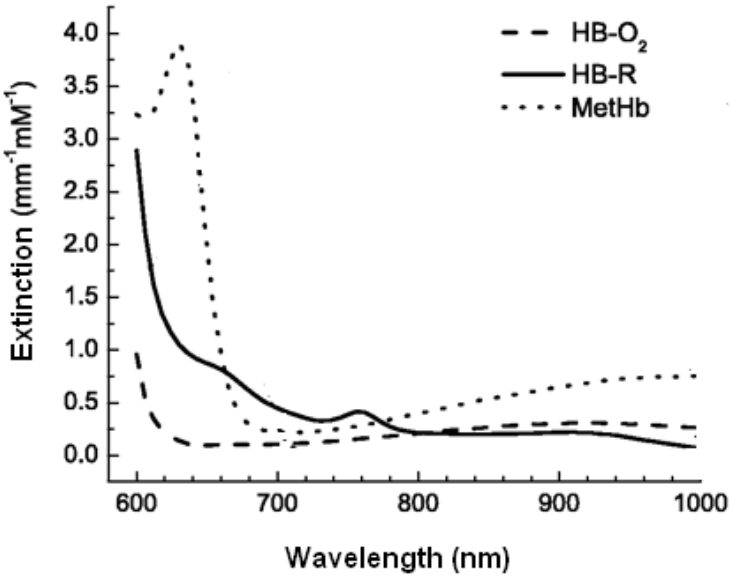


Fig. 13. The absorption spectra of hemoglobin (Hb-R), Oxihemoglobin (Hb-O_2) and Methemoglobin (MetHb) (Lee et al., 2006)

In an attempt to change the chromophore to obtain significant rates of MetHb, while seeking to increase photon absorption, J. Moreno Moraga & all (n.d.) started the combined application of microfoam sclerosants, proven to be efficient for vein sclerosis, with the action of pulsed 1.06 μm Nd:YAG laser beams.

The measurements performed at the National Institute for Laser, Plasma and Radiation Physics at Bucharest (Smarandache et al., 2011) have shown that the use of POL in the foam form increases the optical path of the laser radiation in the foam by light scattering, which leads to an increase of the total absorption, since this is proportional with the product between the absorption coefficient, the optical path length in the sample and the concentration of the absorbent. Laser energy absorption in the foam can be boosted by the multiplication of the impacts of the photons at the collisions with the gas bubbles. Moreover, under these circumstances, the number of changed POL molecules could also increase.

Following the line of increasing the efficacy of the laser beam if the POL is used as foam introduced in the tissue, the foam (produced by the Tessari method and the procedure

described in Section 2) and solution samples of POL were investigated by Raman spectroscopy. The obtained Raman signals were more intense for foam than for simple solution samples (Fig. 14).

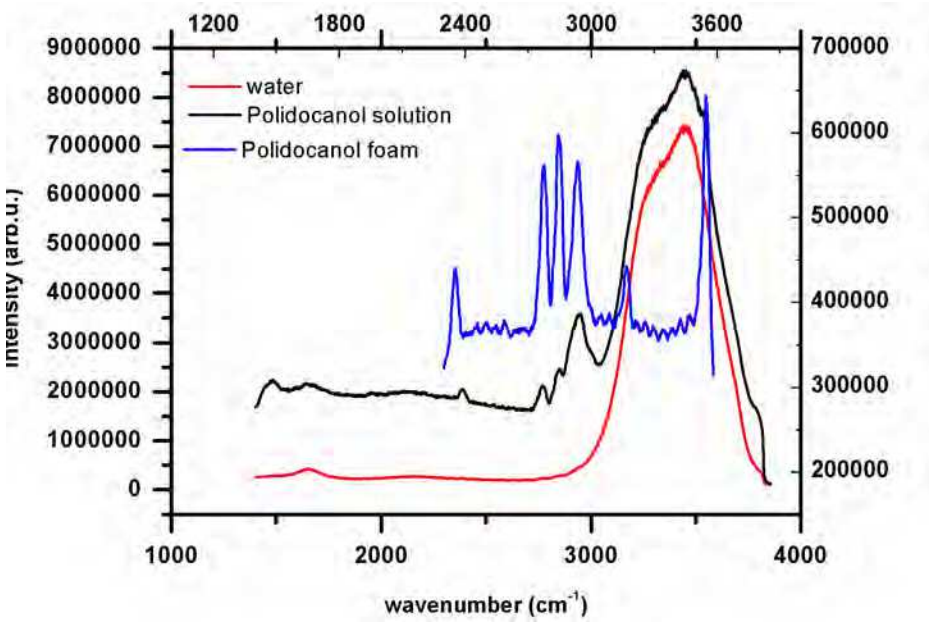


Fig. 14. The compared Raman spectra of the Polidocanol in solution and foam presentation

This laser light scattering enhancement is due to a longer optical path of the laser beam in the foam sample. The Raman vibrational lines corresponding to foam sample are more structured and stronger.

Also, the Raman signals were acquired at different time moments after the preparation of the foam samples. The results indicate that there are some parameters which must be taken into account, such as the bubble dimensions referred to the foam cohesion; these are important with respect to the moment of exposure of the varicose vein injected before with foam POL and exposed in the tissue to laser radiation. As it can be seen in Fig. 15 the best scattering signals were obtained at 2 minutes after foam preparation, in our experimental conditions. After few minutes the foam is destroyed and the intensity of the characteristic vibrational lines decreases drastically.

It is very important that physicians who use laser irradiation of the organism for therapeutic purposes and/or irradiated drugs have a physically well-founded quantitative instrument which would permit an analysis of the significance of the processes involved as a function of the spectral range of the irradiating light, its duration, and power. In addition, the development of a physical basis for the interaction of light with such a special object as biological tissue and the development of recommendations for the optimum interaction parameters are of fundamental scientific interest for medical applications.

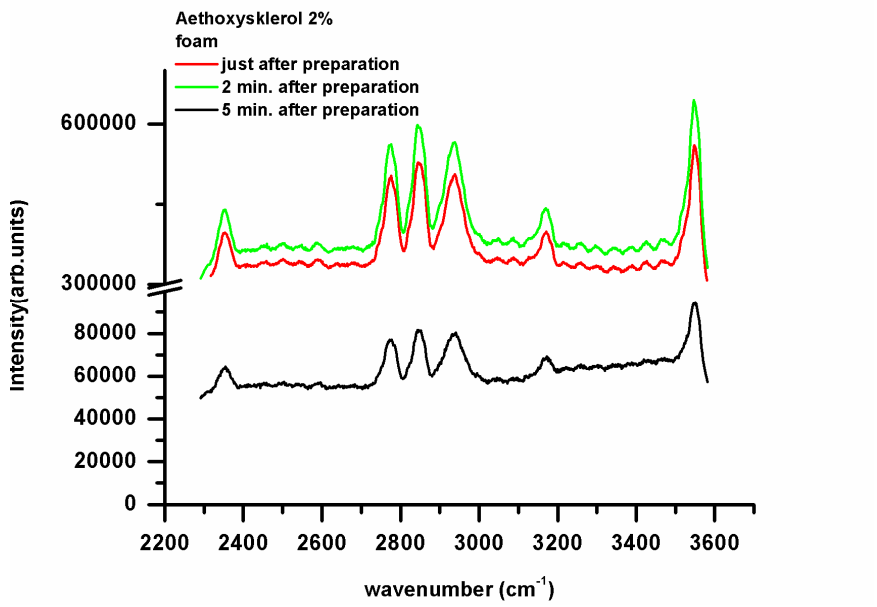


Fig. 15. The influence of foam separation (in two phases: liquid and remaining foam) on the Raman spectra of a POL foam sample

6.2 Clinical results and discussions

Immediately after treatment, darkening of the intravascular pigmentation with perivenous erythema was noticed. These alterations had completely disappeared when patients came back after 6 weeks for the second treatment. In spite of not having used any topical anesthesia, tolerance to treatment was good. Out of the 200 treated patients, 6 patients did not show up for the 2 year assessment, i.e. 3 class I patients, and 3 class III.

Answers to questionnaires regarding results corresponding to the 2 year assessment are presented in Table 4. The results correspond to scores presented by patients (subjectively) and those given by Doctors (objectively). Total satisfaction index (SI) was deduced by adding Very Good and Good scores given by patients. So, SI was 170 out of 194 patients, corresponding to 87,6% that showed up for the 2 years after assessment taking into account that 200 patients received treatment initially.

Nº pat.	Vein type according to vessel diameter	PATIENT EVALUATION			DOCTOR EVALUATION		
		Very Good	Good	Fair	Very Good	Good	Fair
57	Class I	1	46	10	4	48	5
60	Class II	17	41	2	22	37	1
65	Class III	39	26	0	43	22	0

Table 4. Assessment of patients at 2 years

At 18 weeks, hyper-pigmentation and matting were noticed in 8 patients, all of them presenting class I veins. However these complications have disappeared at the 2 year assessment. The greatest success was achieved in class II and III veins. The results obtained in the evaluation at week 18 remained stable even at the 2 year assessment (Fig. 16 – Fig. 18).

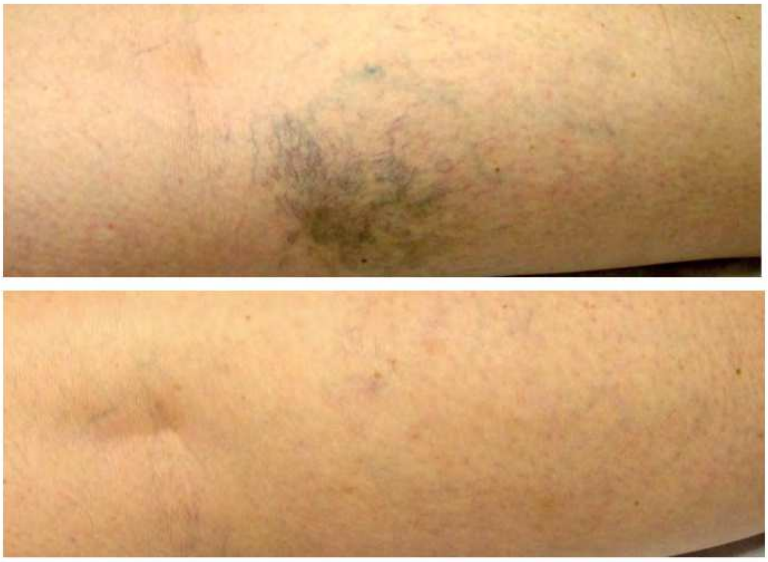


Fig. 16. Vein Class I. Aspect before and at 2 year assessment

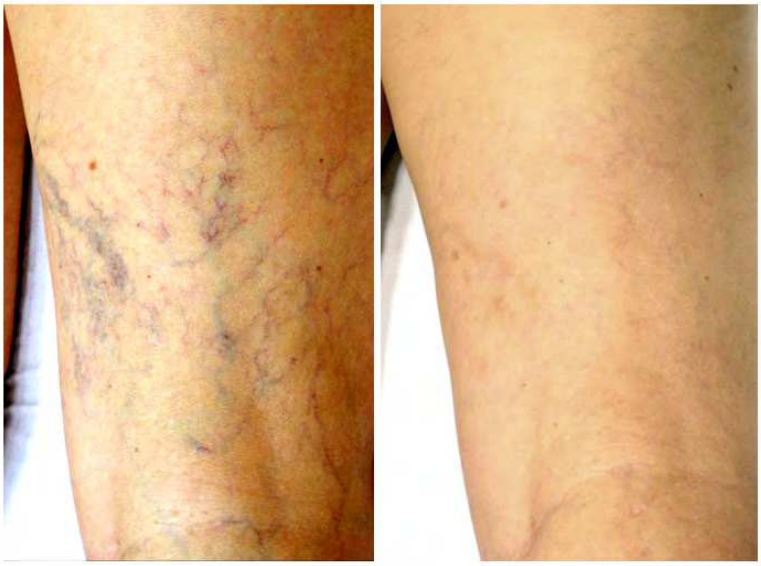


Fig. 17. Vein class II. Before and results 2 years after POL+ laser treatment

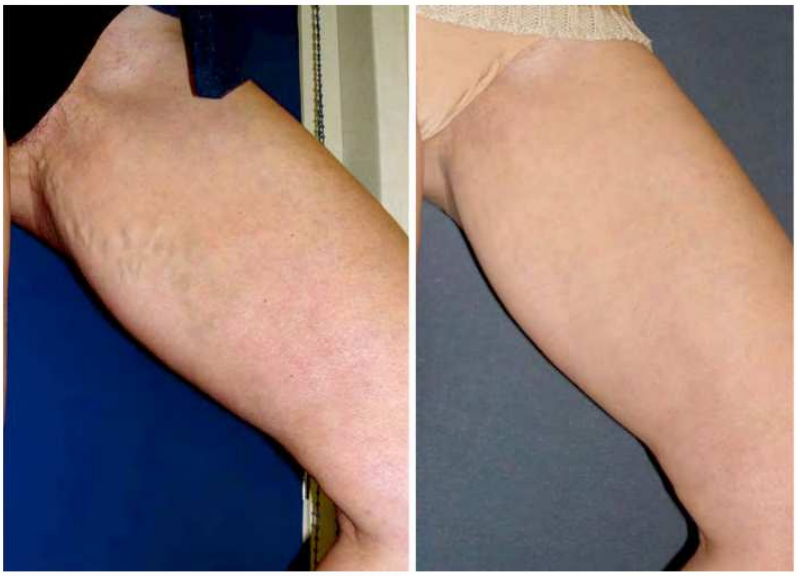


Fig. 18. Vein class III. Patient before and 2 years after treatment with described technique

The reported clinical results show that the use of POL foam combined with the exposure of the patient's tissue which contains it at pulsed laser beams of 1.06 μm wavelength allow obtaining better effects in the treatment of the varicose veins.

7. Conclusions

Clinical results prove that foam sclerotherapy combined with photothermolysis based on laser therapy increases the efficiency of the varicose veins treatment (Trelles et al., 2011).

The foamed form of the detergent sclerosing drugs has resulted in improvements in the efficacy of sclerotherapy. Foam sclerotherapy reduces the dose and concentration of injected drug and assures a better intimate contact of the active substance with the target tissues.

Concerning the foaming procedure, it is recommended to achieve a compromise between the bubbles dimension and the foam stability in time, so that the value of the surface tension is big enough to produce sclerosis of the target tissue and, nevertheless, the foam does not selfdestroys too quickly and dilutes in the blood stream before acting on the vein.

On the other hand, pulsed Nd:YAG lasers emitting at 1,064 nm offer important advantages for treating varicose veins of the lower limbs. They penetrate more and are less absorbed by melanin, which means that they can treat dark phototypes. A laser pulse at 1,064 nm converts Hb into the more spherically shaped MetHb, which has a 4 times higher absorption coefficient. After initial irradiation, further delivering of energy is more effective at heating blood and the surrounding vessel (Kunishige et al., 2007). In order to produce penetration, high fluences are required and hence, treatment is painful and requires cooling and even topical anaesthesia, which are considered as disadvantages of this treatment method.

Comparing the spectroscopic measurements data with the clinical experimental results, we might conclude that the improvement of the action of Polidocanol commercially available as Aethoxysklerol 2% on the varicose vein by exposure of the impregnated tissues with 1.06 μm laser beam is possible due to the following mechanisms:

- for wavelengths around 260 nm, the laser radiation may be absorbed by the Polidocanol proper. The mechanisms of interaction between the veins tissues and the medicine under the influence of laser radiation are not elucidated yet, but it is possible that nonlinear absorption effects take places in the tissue such as absorption of 4 photons at 1.06 μm (which would correspond to a transition at 266 nm), which may be responsible for further effects on the tissue;
- at λ=1.06 μm the absorption may be produced by the Ethyl alcohol and the main chromophores such as hemoglobin, especially methemoglobin and melanin; this may contribute to the sclerosis of the veins in the exposed area but it remains to clarify the possible mechanisms which lead to this effect. The modifications of the molecular structure of Polidocanol may be produced by the Nd:YAG laser radiation once this drug is introduced in the tissue.

The effect of the laser light may be enhanced if the Polidocanol is introduced as foam since than, the light scattering in the tissue becomes more important and the absorption of the laser beam becomes larger. So, we might have in this case a larger number of modified Polidocanol molecules. The Raman spectroscopy measurements prove that the Raman signals were more intense for foam than for simple solution samples. This laser light scattering enhancement is due to a longer optical path of the laser beam in the foam sample. The vibrational lines corresponding to foam sample are more structured and stronger.

As for the proper/most recommended moment to expose the varicose vein injected before with foam POL to laser radiation, the results of Raman spectroscopy study, performed at different moments of foam lifetime, indicate that there are some parameters which must be taken into account, such as bubble dimensions referred to the foam cohesion.

As for the direct clinical application of the Polidocanol, assisted by Nd:YAG laser radiation, the combined treatment of leg veins with Polidocanol (micro)foam and 1,064 nm Nd:YAG laser pulses shows promising possibilities for coagulation of varicles of the lower limbs with efficacy, without the damage of the skin surface. This kind of treatment, using low fluency, permits carrying out treatment sessions on larger areas with patient comfort. Efficacy may be substantially due to Polidocanol injection given to the patient before his/her exposure to laser pulses. The Polidocanol micro foam sclerosant would change the optical absorption of the Nd:YAG emitted at 1,064 nm, increasing the efficacy of the treatment and the vein closure. The obtained promising results are backed by evidence-based clinical outcome, and efficacy has been assessed up to 3 years (Trelles et al., 2011).

Further studies are needed to elucidate the role of Polidocanol microfoam interaction with the Nd:YAG laser beam and both a possible molecular excitation and light scattering phenomenon that will enhance the observed results.

8. References

Alos, J.; Carreno, P.; Lopez, J.A.; Estadella, B.; Serra-Prat, M. & Marinello, J. (2006). Efficacy and safety of sclerotherapy using polidocanol foam: a controlled clinical trial. *Eur J Vasc Endovasc Surg*, Vol.31, No.1, (January 2006), pp. 101–107, ISSN 1078-5884

Altshuler, G.B.; Anderson, R.R.; Manstein, D.; Zenzie, H.H. & Smirnov, M.Z. Extended Theory of Selective Photothermolysis. *Lasers Surg Med,* Vol.29, No.5, (December 2001), pp. 416-432, ISSN 1096-9101

Anderson, R.R. & Parrish, J.A. (1983). Selective photothermolysis: precise microsurgery by selective absorption of pulsed radiation. *Science,* Vol.220, No.4596, (April 1983), pp. 524- 527, ISSN 0036-8075 (print), 1095-9203 (online)

Artemi P. (2007). Pharmacology of Phlebology, *ACP Australasian meeting*, Sydney, Australia, September 18-21, 2007

Barun, V.V. & Ivanov, A.P. (2010). Depth distributions of light action spectra for skin chromophores. *Journal of Applied Spectroscopy,* Vol.77, No.7, (March 2010), pp. 73-79, ISSN 0021-9037 (Print), 1573-8647 (Online)

Black, F.B. & Barton, J.K. Chemical and structural changes in blood undergoing laser photocoagulation. *Photochem Photobiol,* Vol.80, No1, (July 2004), pp. 89-97, ISSN 1751-1097

BTG International Ltd. (2011). Our Pipeline, In: *Development,* September 2011, Available from: http://www.btgplc.com/development/our-pipeline

Cabrera, G.J., Cabrera G.O.J. & Garcia-Olmedo M.A. (1997a). Elargissement des limites de la schlerotherapie: nouveaux produits sclerosants. *Phlebologie*, Vol.50, No.2, (1997), pp. 181–187, ISSN 0031-8280

Cabrera, G.J. & Cabrera, G.O.J. Jr. (1997b). BTG International Limited inventors; assignee Injectable microfoam containing a sclerosing agent. *US patent 5676962*. (October 1997) available at www.patents.com/us-5676962.html

Cabrera, J.; Cabrera, J. Jr. & Garcia-Olmedo, M.A. (2000). Treatment of varicose long saphenous veins with sclerosant in microfoam form: long-term outcomes. *Phlebology*, Vol.15, No.1, (2000), pp.19–23, ISSN 0268-3555

Cabrera, J.; Cabrera, J. Jr; Garcia-Olmedo, M.A. & Redondo, P. (2003). Treatment of venous malformations with sclerosant in microfoam form. *Arch Dermatol,* Vol.139, No.11 (November 2003), pp. 1409–1416, ISSN 1538-3652 (on-line)

Cabrera, J.; Redondo, P., Beccerra, A.; Garrido, C.; Cabrera, J.Jr.; Garcia-Olmedo, M.A.; Sierra, A.; Lloret, P. & Martinez-Gonzalez, M.A. (2004).. Ultrasound-guided injection of polidocanol microfoam in the management of venous leg ulcers. *Arch Dermatol* , Vol.140, (June 2004), pp. 667–673, ISSN 1538-3652 (on-line)

Cavezzi, A. & Frullini, A. (1999). The role of sclerosing foam in ultrasound guided sclerotherapy of the saphenous veins and of recurrent varicose veins: our personal experience. *Australian & New Zealand journal of phlebology,* Vol. 13, (1999), pp. 49–50, ISSN 1441-7766

Cavezzi, A.; Frullini, A.; Ricci, S. & Tessari, L. (2002). Treatment of varicose veins by foam sclerotherapy: two clinical series. *Phlebology,* Vol.17, (2002), pp. 13-18, ISSN 0268-3555

Cavezzi, A. & Tessari, L. (2009). Foam sclerotherapy techniques: different gases and methods of preparation, catheter versus direct injection. *Phlebology*, Vol.24, No.6, (December 2009), pp. 247-251, ISSN 0268-3555

Duffy, D. M. (2010). Sclerosants: A Comparative Review, *Dermatol. Surg.,* Vol. 36, Suppl.2, (June 2010), pp. 1010–1025, ISSN 1524-4725

Eckmann, D.M.; Kobayashi, S. & Li, M. (2005). Microvascular embolization following polidocanol microfoam sclerosant administration. *Dermatol Surg,* Vol.31, No.6, (June 2005), pp. 636–643, ISSN 1524-4725

Eckmann, D.M. (2009). Polidocanol for Endovenous Microfoam Sclerosant Therapy. *Expert Opin Investig Drugs*, Vol.18, No.12, (December 2009), pp. 1919–1927, ISSN 1354-3784

Frullini, A. (2000). New technique in producing sclerosing foam in a disposable syringe. *Dermatologic Surgery,* Vol.26, No.7, (July 2000), pp. 705–706, ISSN 1524-4725 (on-line), 1076-0512 (print)

Frullini, A. (2011). An Investigation into the Influence of Various Gases and Concentrations of Sclerosants on Foam Stability. *Dermatologic Surgery, Vol.37, No.1, (January 2011), pp. 17-18,* ISSN 1524-4725 (on-line), 1076-0512 (print)

Fumarel, R.; Murgoi, G.; Albert, P.; Hurduc, A. & Pascu, M.L. (2009). Increase of Cisplatinum therapeutic index through optical irradiation, *AIP Conf. Proc. of LASER FLORENCE 2008,* Vol.1142, pp.1-7, ISSN 0094-243X, ISBN 978-0-7354-0679-7, Florence, Italy, October 31-November 1, 2008

Gachet, G. (2001). Une nouvelle methode simple et economique pour confectionner de la mousse pour la sclerose echoguidee. *Phlebologie*, Vol.54, No. 1, (2001), pp. 63–65, ISSN 0031-8280

Garcia-Mingo, J. (1999). Esclerosis venosa con espuma: Foam Medical System. *Revista Espanola de Medicina y Cirugia Cosmetica,* Vol.7, (1999); pp.29–31, available at www.cavezzi.it/garciaen.html

Grossweiner, L.I. (2005), *The science of Phototherapy: An Introduction*, Springer, ISBN-13: 978-1402028830

Hamel-Desnos, C.; Desnos, P.; Wollmann, J.C.; Ouvry, P.; Mako, S. & Allaert, F.A. (2003). Evaluation of the efficacy of polidocanol in form of foam compared to liquid form in sclerotherapy of the long saphenous vein. *Dermatol Surg,* Vol.29, No.12, (December 2003), pp. 1170-1175, ISSN 1076-0512 (print), 1524-4725 (electronic)

Hamel-Desnos, C.; Ouvry; Benigni, J.P.; Boitelle, G.; Schadeck, M.; Desnos P. & Allaert, F.A. (2007). Comparison of 1% and 3% Polidocanol Foam in Ultrasound Guided Sclerotherapy of the Great Saphenous Vein: A Randomised, Double-Blind Trial with 2 Year-Follow-up. "The 3/1 Study", *European Journal of Vascular and Endovascular Surgery,* Vol. 34, No.6, (December 2007), pp. 723-729, ISSN 1078-5885

Henriet, J.P. (1997). Un an de pratique quotidienne de la sclerotherapie (veines reticulaires et teleangiectasies) par mousse de polidocanol: faisebilite, resultats, complications. *Phlebologie,* Vol.50, No.2, (1997); pp. 355–360, ISSN 0031-8280

Hsu, T.S. & Weiss, R.A. (2003). Foam sclerotherapy: a new era. *Arch Dermatol,* Vol. 139, No.11, (November 2003), pp. 1494-1496, ISSN 1538-3652 (on-line)

Kunishige, J.; Goldberg, L. & Friedman, P. (2007), Laser therapy for leg veins. *Clinics in Dermatology,* Vol.25, No.5, (September-October 2007), pp. 454–461, ISSN 0738-081X

Lee, J.; El-Abaddi, N.; Duke, A.; Cerussi, A.E.; Brenner, M. & Tromberg, B.J. (2006). *J. Appl. Physiol.* Vol.100, No.2, (February 2006), pp. 615-622, ISSN 8750-7587

Monfreux, A. (1997). Traitement sclerosant des troncs saphenies et leurs collaterales de gros calibre par le methode MUS. *Phlebologie*, Vol.50, No.2, (1997), pp. 351–353, ISSN 0031-8280

Mordon, S.; Brisot, D. & Fournier, N. (2003). Using a "non uniform pulse sequence" can improve selective coagulation with a Nd:YAG laser (1.06 micron) thanks to Met-hemoglobin absorption: A clinical study on blue leg veins. *Lasers Surg Med* , Vol.32, No.2, (2003), pp. 160-170, ISSN 1096-9101 (electronic)

Moreno Moraga, J.; Isarria Marcos, M.J.; Royo de la Torre, J. & Gonzalez Urena, A. (n.d.). Photodynamic therapy in the treatment of varicose veins, Available from http://www.institutomedicolaser.com/archivos/areacientifica/varices_070306.pdf

Nijsten, T.; van den Bos, R.R.; Goldman, M.P.; Kockaert, M.A.; Proebstle, T.M.; Rabe, E.; Sadick, N.S.; Weiss, R. & Neumann, H.A.M. (2009). Minimally invasive techniques in the treatment of truncal varicose veins. *J Am Acad Dermatol,* Vol.60, No.1, (January 2009), pp. 110-119, ISSN 0190-9622

Orbach E.J. (1950). Contribution to the therapy of the varicose complex. *J Int Coll Surg*, Vol.13, No.6, (June 1950), pp. 765–771, ISSN 0020-8868

Ouvry, P.; Allaert, F.A.; Desnos, P. & Hamel-Desnos, C. (2008). Efficacy of Polidocanol Foam versus Liquid in Sclerotherapy of the Great Saphenous Vein: A Multicentre Randomised Controlled Trial with a 2-year Follow-up. *Eur J Vasc Endovasc Surg,* Vol. 36, No.3, (September 2008), pp.366-370, ISSN 1078-5884

Parsi, K.; Exner, T.; Connor, D.E.; Ma, D.D.F. & Joseph, J.E. (2007). In vitro Effects of Detergent Sclerosants on Coagulation, Platelets and Microparticles. *Eur J Vasc Endovasc Surg*., Vol.34, No.6, (December 2007), pp. 731-740, ISSN 1078-5884

Parsi, K.; Exner, T.; Connor, D.E.; Herbert, A.; Ma, D.D. & Joseph, J.E. (2008). The lytic effects of detergent sclerosants on erythrocytes, platelets, endothelial cells and microparticles are attenuated by albumin and other plasma components in vitro. *Eur J Vasc Endovasc Surg* , Vol. 36, No.2, (August 2008), pp. 216–223, ISSN 1078-5884

Pascu, M.L.; Nastasa, V.; Smarandache, A.; Militaru, A.; Martins, A.; Viveiros, M.; Boni, M.; Andrei, I.R.; Pascu, A.; Staicu, A; Molnar, J. ; Fanning, S. & Amaral, L. (2011). Direct Modification of Bioactive Phenothiazines by Exposure to Laser Radiation. *Recent Patents on Anti-Infective Drug Discovery,* Vol.6, No.2, (May 2011), pp. 147-157, ISSN1574-891X

PubChem. (2011). Polidocanol, In: Compund, August, 2011, Available from: http://pubchem.ncbi.nlm.nih.gov/

Querry, M. R.; Wieliczka, D. M. & Segelstein, D. J. (1998). Water: H_2O, In: *Handbook of Optical Constants of Solids II*, E. D.Palik, (Ed.), 1059–1077, Academic Press 1998, ISBN 0-12-5444422-2, San Diego, CA, USA.

Rabe, E.; Otto, J.; Schliephake, D. & Pannier, F. (2008). Efficacy and Safety of Great Saphenous Vein Sclerotherapy Using Standardised Polidocanol Foam (ESAF): A Randomised Controlled Multicentre Clinical Trial, *Eur J Vasc Endovasc Surg,* Vol.35, No.2, (February 2008), pp. 238-245, ISSN 1078-5885

Rabe, E. & Pannier, F. (2010). Sclerotherapy of Varicose Veins with Polidocanol Based on the Guidelines of the German Society of Phlebology. *Dermatol Surg*, Vol.36, Suppl.2, (June 2010), pp. 968–975, ISSN 1076-0512 (print), 1524-4725 (electronic)

Railan, D.; Parlette, E.; Uebelhoer, N. & Rohrer, T. (2006). Laser treatment of vascular lesions. *Clinics in Dermatology*, Vol.24, No.1, (January-February 2006), pp. 8-15, ISSN 0738-081X

Rao, J. & Goldman, M.P. (2005). Stability of foam in sclerotherapy: differences between sodium tetradecyl sulfate and polidocanol and the type of connector used in the

double-syringe system technique. *Dermatol Surg,* Vol.31, No.1, (January 2005), pp. 19–22, ISSN 1076-0512 (print), 1524-4725 (electronic)

Redondo, P. & Cabrera, J. (2005). Microfoam Sclerotherapy. *Seminars in Cutaneous Medicine and Surgery*, Vol.24, No.4, (December 2005), pp. 175-183, ISSN 1085-5629

Rogachefsky, A.S.; Silapunt, S. & Goldberg, D.J. (2002). Nd:YAG laser (1064nm) irradiation for lower extremity telangiectases and small reticular veins: efficacy as measured by vessel color size. *Dermatol Surg*, Vol.28, No.3, (March 2002), pp. 220 -223, ISSN 1524-4725 (electronic)

Ross, E.V. & Domankevitz, Y. (2005). Laser treatment of leg veins: physical mechanisms and theoretical considerations. *Lasers Surg Med*, Vol. 36, No.2, (February 2005), pp. 105-116, ISSN 1096-9101 (electronic)

Sadick, N.S.; Prieto, V.G.; Shea, C.R.; Nicholson, J. & McCaffrey, T. Clinical and Pathophysiologic Correlates of 1064-nm Nd:YAG Laser Treatment of Reticular Veins and Venulectasias. *Arch Dermatol,* Vol.137, No.5, (May 2001), pp. 613-617, ISSN 1538-3652

Sadoun, S. & Benigni, J.P. (1998). The treatment of varicosities and telangiectases with TDS and Lauromacrogol foam. *XIII World Congress of Phlebology 1998, abstract book*, pp. 327, Sydney, Australia, September 6-11, 1998.

Santos, P.; Watkinson, A.C.; Hadgraft, J. & Lane, M.E. (2008). Application of Microemulsions in Dermal and Transdermal Drug Delivery, *Skin Pharmacol Physiol,* Vol.21, No.5, (2008), pp. 246-259

Smarandache, A.; Trelles, M. & Pascu, M.L. (2010). Measurement of the modifications of Polidocanol absorption spectra after exposure to NIR laser radiation. *J Optoelectronics Advanced Materials,* Vol.12, No.9, pp. 1942 – 1945, (2010) ISSN 1454-4164

Smarandache, A.; Nastasa, V.; Militaru, A.; Staicu, A.; Trelles, M.; Moreno-Moraga, J. & Pascu, M.L. (June 2011). Comparison of the experimental techniques used to obtain foams out of medicines solutions, *ISWLA 2011*, May 31 – June 4, 2011, Bran, Romania, Available from: http://iswla.inflpr.ro/CompleteProgram.pdf

Smarandache, A.; Trelles, M.; Staicu, A.; Moreno-Moraga, J. & Pascu, M.L. Laser beams interaction with Polidocanol foam: physical bases, *SELMQ 2011- XIX Congreso Sociedad Espanola de Laser Medico Quirurgico*, 8 – 10 July 2011, Jerez de la Frontera, Spain, 2011, Available from: http://congresos.net/frame.php?id= 1015&web =http : //www.selmq.net/

Suthamjariya, K.; Farinelli, W.A.; Koh, W. & Anderson, R,R. (2004). Mechanisms of microvascular response to laser pulses. *J Invest Dermatol*, Vol.122, No.2, (February 2004), pp. 518-525, ISSN 0022-202x

Tessari, L. (2000). Nouvelle technique d'obtention de la sclero-mousse. *Phlebologie*, Vol.53, No.1, (2000), pp. 129-133, ISSN 0031-8280

Tessari, L. (2001). Extemporary sclerosing foam according to personal method: experimental clinical data and catheter usage. *Int Angiol Suppl,* Vol.20, Suppl.1, (2001), pp. 54.

Tessari, L., Cavezzi, A. & Frullini, A. (2001). Preliminary experience with a new sclerosisng foam in the treatment of varicose veins. *Dermatol Surg,* Vol.27, No.1, (January 2001), pp. 58-60, ISSN 1524-4725 (electronic)

Tibbs, D.J.; Sabiston, D.; Davis, M. & Mortimer, P. (1997). *Varicose Veins, venous disorders and lymphatic problems in the lower limb*, Oxford University Press, ISBN 978-0192627629, USA

Trelles, M.; Allones, I.; Alvareza, X,; Veleza, M.; Buila, C.; Luna, R. & Trelles, O. (2005). Long-pulsed Nd:YAG 1064 nm in the treatment of leg veins: Check up of results at 6 months in 100 patients. *Medical Laser Application,* Vol.20, No.4, (December 2005), pp. 255–266, ISSN 1615-1615

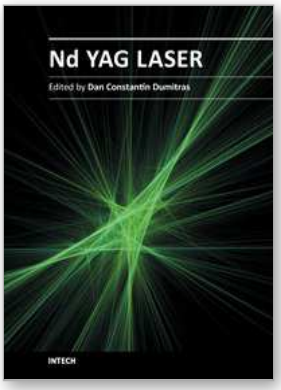
Trelles, M.A.; Weiss, R.; Moreno-Moraga, J.; Romero, C. Velez, M. & Perez, X. (2010). Treatment of legs veins with combined pulsed dye and Nd:YAD lasers: 60 patients assessed at 6 months. *Lasers Surg Med*, Vol.42, No.9, (November 2010), pp.609-614, ISSN 1096-9101 (electronic)

Trelles, M.; Moreno-Moraga, J.; Alcolea, J.; Smarandache, A. & Pascu M.L.. (2011) Laser in leg veins: our personal approach of treatment, In: *Synopsis of Aesthetic Dermatology & Cosmetic Surgery*, M.L. Elsaie, (Ed), Nova Science Publishers Inc, NY, USA, in press

U.S. Department of Health & Human Services, FDA. (2010). FDA Approves Asclera to Treat Small Varicose Veins, In: *Press Announcements*, (September 2011), Available from: http://www.fda.gov/NewsEvents/Newsroom/PressAnnouncements/

Wainwright, M. (2008). Photodynamic therapy: the development of new photosensitisers. *Anticancer Agents Med Chem,* Vol.8, No.3, (April 2008), pp. 280-291, ISSN 1871-5206

Wollmann, J.C. (2004). The history of sclerosing foams. *Dermatol Surg,* Vol.30, No.5, (May 2004), pp. 694-703, ISSN 1524-4725 (electronic)



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Discovered almost fifty years ago at Bell Labs (1964), the Nd:YAG laser has undergone an enormous evolution in the years, being now widely used in both basic research and technological applications. Nd:YAG Laser covers a wide range of topics, from new systems (diode pumping, short pulse generation) and components (a new semiorganic nonlinear crystal) to applications in material processing (coating, welding, polishing, drilling, processing of metallic thin films), medicine (treatment, drug administration) and other various fields (semiconductor nanotechnology, plasma spectroscopy, laser induced breakdown spectroscopy).

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