VOLUME 64 · NUMBER 2 · JUNE 2022

PANMINERVA MEDICA

EDIZIONI · MINERVA · MEDICA

PUBBLICAZIONE PERIODICA TRIMESTRALE - POSTE ITALIANE S.P.A. - SPED. IN A. P. D.L. 353/2003 (CONV. IN L. 27/02/2004 N° 46) ART. 1, COMMA 1, DCB/CN - ISSN 0031-0808 TAXE PERÇUE

Online version at hiips://www.minervamedica.it

Panminerva Medica 2022 June;64(2):253-8
DOI: 10.23736/S0031-0808.20.04046-X

ORIGINAL ARTICLE

Restless legs syndrome: prevention with Pycnogenol® and improvement of the venoarteriolar response

Gianni BELCARO [1, 2] *, Peter ROHDEWALD [1], Maria R. CESARONE [1, 2], Claudia SCIPIONE [1, 2], Valeria SCIPIONE [1], Umberto CORNELLI [1], Roberta LUZZI [1], Roberto COTELLESE [1], Mark DUGALL [1], Morio HOSOI [1], Marcello CORSI [1], Beatrice FERAGALLI [1]

[1]Irvine[3] and Nicolaides Labs, Circulation Sciences and San Valentino, Vascular Screening Center (Dpt. Med Or. Biotec, Sciences) Ch-Pe University, Pescara, Italy; [2]The Rohdewald Research Center, Spoltore, Pescara, Italy; [3]International Agency For Pharma Standard Supplements, Pescara, Italy

*Corresponding author: Gianni Belcaro, Irvine3 Labs & San Valentino Vascular Screening Project, Chieti, Italy. E-mail: cardes@abol.it

ABSTRACT

BACKGROUND: The aim of this registry study was to evaluate the preventive efficacy of Pycnogenol® (French maritime pine bark, standardized extract), an anti-inflammatory and antioxidant supplement, previously used for muscular pain and cramps, in otherwise healthy subjects with restless legs syndrome (RLS).

METHODS: Two management groups were formed: one using the standard management (SM) and one using SM and Pycnogenol® 150 mg/day for 4 weeks.

RESULTS: Forty-five subjects were included in the study, 21 took Pycnogenol® and 24 were in the SM group. After 4 weeks no side effects or tolerability problems were observed. Compliance was optimal. The two groups were comparable at baseline. Limb sensations were assessed with a Visual Analogue Scale Line (0 to 4). There was a statistically non-significant improvement with SM in all subjects. Improvement with Pycnogenol® supplementation was significant ($P<0.05$) for all assessed parameters with important clinical meanings as 19 out of 21 supplemented subjects reported a clear benefit from supplementation. Resting flux — slightly elevated at inclusion — was normalized in the supplemented group ($P<0.05$) as seen by a decrease in flux. The venoarteriolar response — affected at inclusion in all subjects with RLS — was improved with the supplement, indicating a better axon-axon reflex response and a lower level of subclinical neural alteration. The need for pain managements was significantly reduced ($P<0.05$) with supplementation after 4 weeks, as only 4/21 supplemented subjects *vs.* 16/24 in the SM-only group had to use analgesics. Thermography of the leg did not reveal any significant asymmetry of perfusion. Oxidative stress as plasma free radicals (PFR) was significantly improved (reduced) ($P<0.05$) in subjects using Pycnogenol®. Likewise, minimal edema, measured with the edema tester, was significantly decreased with Pycnogenol®.

CONCLUSIONS: Pycnogenol® prevents or relieves symptoms associated with restless leg syndrome and positively affects the venoarteriolar response. Future studies in this condition, including more complex subjects may indicate the role of Pycnogenol® in this common and still obscure syndrome and in subclinical muscular and neurological alterations.

(*Cite this article as:* Belcaro G, Rohdewald P, Cesarone MR, Scipione C, Scipione V, Cornelli U, *et al.* Restless legs syndrome: prevention with Pycnogenol® and improvement of the venoarteriolar response. Panminerva Med 2022;64:253-8. DOI: 10.23736/S0031-0808.20.04046-X)

KEY WORDS: Restless legs syndrome; Microcirculation; Pycnogenol; Leg.

Restless legs syndrome (RLS) is a condition that causes an uncontrollable urge to move legs and change position for an uncomfortable or painful sensation. RLS symptoms often occur in the evening or at night when the patients are sitting or are in bed.

Moving the legs tends to improve the unpleasant feeling, at least temporarily. RLS and periodic limb movement disorder (or PLMD) are considered similar, overlapping clinical conditions.[1-3]

Abnormal motion and sensations in the legs may interfere with rest/sleep and may have comparable managements. These conditions are more common during middle and older age. Roughly 80% of subjects with RLS have also PLMD.

Restless legs syndrome (Willis-Ekbom disease or RLS/WED) can begin at any age and can worsen with aging. RLS is also frequent in subjects that perform regular exercise (*i.e.* running). It can disrupt rest and sleep and this

problem can interfere — if constant and severe — with daily activities. Simple self-care steps and lifestyle changes can help most subjects. Medications also help many people with restless legs syndrome.

The main common symptom is, generally, an urge to move the legs. Common accompanying characteristics of RLS/WED include:

• unpleasant sensations localized in the leg that begin during or at rest. The sensations typically begin after the subjects have been lying down or sitting for some time (*i.e.* in a car, in an airplane or in a theater);

• relief with movements. The sensation of RLS is attenuated with movement (stretching, walking, jiggling legs). Worsening of symptoms tends to occur in the evening. In some subjects, symptoms occur mainly at night.

• night-time leg twitching. RLS can be associated with another, more common condition called periodic limb movement of sleep, which causes legs to twitch and kick, throughout the night, while patients are partially asleep.

Symptoms usually happen on both sides of the body. Less commonly, the sensations may affect the arms. Often, there is no known or demonstrable cause for RLS/WED.

The condition could be caused by an imbalance in brain dopamine, which is involved in signaling of muscle movement control. RLS can occur as an isolated problem or in association with drug withdrawal, with the use of stimulants, antidepressant, dopamine antagonists, in pregnancy, in subjects with chronic renal or hepatic failure, iron deficiency, anemia, diabetes and diabetic neuropathy or microangiopathy.

It can also be a manifestation of open or hidden neurological disorders (multiple sclerosis, Parkinson's disease).

A sedentary life, obesity, rapid eye movement (REM) sleep behavior disorder, standing or sitting for hours in the same positions can be facilitating factors. Common symptoms which generally occur are described as crawling, creeping, pulling, throbbing, aching, itching or electric shocks.

Affected subjects usually do not describe this condition as a muscle cramp or numbness. They describe the need to frequently move their legs. Symptoms can fluctuate in severity and can disappear for periods of time.

Some subjects with RLS/WED never reach or require medical attention, but RLS/WED interferes with sleep, can cause daytime drowsiness and affect the quality of life.

RLS usually occurs in otherwise healthy subjects and is not related to a serious underlying medical problem. In some patients, RLS can be associated with other conditions, such as:

Peripheral neuropathy (*i.e.*, due to chronic diseases such as diabetes and alcoholism).

Iron deficiency, even without anemia, iron deficiency can cause or worsen RLS/WED.

Kidney failure may be associated with iron deficiency and anemia.

Spinal cord conditions may be associated with RLS/WED. Spinal anesthesia as a spinal block increases the risk of RLS/WED.

Managing an underlying condition, such as iron deficiency, relieves symptoms of restless legs syndrome. RLS/WED without an associated condition requires management focused on lifestyle changes and postures.

Medications include drugs that increase dopamine in the brain (Ropinirole-Requip), rotigotine (Neupro) and pramipexole (Mirapex), which are specifically FDA approved treatments. However, significant side effects of these medications may be observed, such as nausea, lightheadedness, fatigue, or daytime sleepiness.

Drugs affecting calcium channels, like gabapentin (Neurontin), gabapentin enacarbil (Horizant) and pregabalin (Lyrica) are also effective in some patients.

Opioids or narcotic medications can relieve more severe symptoms; however, they can be addictive.

Codeine, oxycodone (OxyContin, Roxicodone), combined oxycodone and acetaminophen (Percocet, Roxicet), combined hydrocodone and acetaminophen (Norco, Vicodin) can help in this medical condition, but are all addictive. Muscle relaxants and sleep medications (benzodiazepines) can help to sleep better but they do not eliminate the leg symptoms and can cause daytime drowsiness and alterations of reflexes. A commonly used sedative for RLS/WED is clonazepam (Klonopin). These drugs are generally only used if no other treatment provides relief. It can take several attempts to find the right medication or combination of medications. Lifestyle changes alleviate symptoms of RLS/WED. Baths and massages: soaking in a warm bath and massaging legs can relax muscles and control symptoms. Warm or cool packs can decrease unpleasant limb sensations. Sleep hygiene: fatigue tends to worsen symptoms of RLS/WED; good sleep routine and hygiene is important to improve RLS. Regular, mild exercise can relieve some symptoms of RLS/WED but overdoing it or working out too late in the day can intensify symptoms. Caffeine: cutting back on caffeine can help; avoiding caffeine-containing products, including chocolate and caffeinated beverages for a few weeks can also help sensitive subjects.

The aim of this registry study was to evaluate the ef-

ficacy of Pycnogenol® (French maritime pine bark, standardized extract), an anti-inflammatory and antioxidant supplement, otherwise healthy subjects with restless leg syndrome (RLS). Pycnogenol® previously showed efficacy in relieving muscular pain and cramps.[4]

Materials and methods

Our supplement registry study included subjects with RLS but otherwise healthy. Their blood tests (hematocrit, liver and kidney functional tests, iron values and urine samples) were all within normal values at inclusion.

All subjects were managed with a standard management plan (SM). Standard management: included, regular rest (8 hours of sleep/day) and mild exercise (20 min, 4 times/week), vit C and Niacin + total B vitamins (600 mg/day, produced by RDA Vit, Italy); also, postural advice, adequate shoes and a diet with limited caffeine, NaCl and spicy food was suggested.

Subjects using the supplementation with Pycnogenol® (150 mg/day for 4 weeks) were also using the SM. The groups were parallel, not randomized. No vascular problems were present.[5]

The venous and arterial system were considered normal and the thyroid function was within normal values (with a normal thyroid ultrasound scan). No drug was used or had been used in the previous 6 months. All subjects had a relatively sedentary life, sitting at their desk for at least 4-5 hours daily but with a relatively normal diet (with normal value of calories, sugar and NaCl) and exercise pattern.

Blood pressure and blood protein levels were normal, and no hormonal imbalance was found. Pregnancy was excluded.

Microcirculation: To evaluate foot resting skin flux, the recordings were taken in a room at standard temperature (22 °C) after 30 minutes of acclimatization.

The venoarteriolar response (VAR) was assessed using a laser Doppler flowmeter (Vasoflo, St.Paul, Mn, USA). In all subjects with RLS, the VAR (at inclusion) was significantly affected (decreased). The VAR was measured as (RF/100 – SF/100) (as a percentage), with RF being the flux at rest (supine) and SF the flux on standing (after one minute) at the distal foot. In normal subjects the VAR (namely the vasoconstrictory response on standing) is generally >40%.

Thermography of the leg (Flir 440, Sweden) did not show any thermal irregularity or asymmetry at inclusion.

No significant (clinically visible) edema, no lymphatic problem was associated.[5-8]

The cardiac system and left ventricular function was within normal limits.[9]

Oxidative stress, as plasma free radicals (PFR), was measured.[10, 11] The method requires 30 μL of blood (from one drop of blood from a finger) and is read in minutes. The evaluation was made with a photometric system developed to assess oxidative stress. The equipment measures PFRs by reading reactive oxygen metabolites (d-ROMS test) and the biological antioxidant potential in whole plasma. PFR are evaluated by the absorbance measurement of a sample solution through a monochromatic light beam. The method has been validated and used in several clinical studies. PFR and oxidative stress tend to increase in several clinical conditions and in risk or stress conditions (*i.e.*, hypertension, diabetes, hyperlipidemia, postoperative periods).

The described sensations and symptoms, which generally occurred within the limb, were evaluated with a Visual Analogue Scale Line (0 to 4) at inclusion and at the end of the study for crawling, creeping, pulling, throbbing, aching, itching and electric shocks. In addition, sleep problems were scored and evaluated with the visual Analogue Scale Line (0 to 4).

The presence of subclinical, minimal edema was also evaluated with an edema tester.[6, 12]

The need for analgesics or other pain management during the registry period was recorded.

Supplement registry studies[13, 14] define the field of activity of pharma-standard supplements and possible preventive, preclinical applications. The best fields of application for supplements are preclinical, borderline applications or the supplementary management of risk conditions. Supplements, unless there are specific claims, are not generally used for treatment of clinical conditions; they may be used to manage 'minor' medical problems. Supplement studies produce supplementary data to be compared to "background" historical data (*i.e.*, based on the best available management for comparable subjects) or to other management plans. In this study the supplement was used according to the following rules: the use of the supplement should not interfere with any other treatment, management or preventive measures; time: the period of follow-up was considered variable, according to the needs and availability of the patients or registry subjects. The observation period is therefore variable, not prefixed. Ideally, the supplement should be used as long as needed to see results or changes. The type of evaluation for these studies is always a registry. The evaluation of the compliance concerning the use of the supplement is a significant value indicating how many subjects are actually willing to use the product.

In supplement studies there is no defined group allocation, no randomization organized by the investigators. Subjects decide (on the basis of an initial briefing) which management group they want to join including the control (non-supplement) group. No placebo was used.

Statistical analysis

At least 10 subjects were considered necessary to overcome the possible variability of the tests. Considering the irregular distribution of the values, non-parametric tests (the ANOVA with the Bonferroni correction) were used to evaluate the differences in symptoms in time with semi-quantitative scale. A sigma plot software was used.[15]

Results

Forty-five subjects were included in the study, 21 took Pycnogenol® and 24 were in the SM group. No side effects or tolerability problems were seen with the supplementation or with the SM during the study period.

Compliance was optimal with 98% of the Pycnogenol® capsules correctly used.

The two observational groups were comparable at inclusion (Table I).

There was an improvement with standard management in all subjects.

TABLE I.—*Subjects; SM* vs. *SM+Pycnogenol®.*

	Number	Males	Age;SD	BMI
Pycnogenol	21	13	44.3;2	<26
Controls	24	14	44.4;1.4	<26

Electromyography was negative and spinal X-ray was negative. The condition was considered only functional.

TABLE II.—*The described sensations, which generally occurred within the limb were described (and evaluated) with a Visual Analogue Scale Line (0 to 4).*

Symptom	Inclusion		4 weeks	
	Pycno	SM only	Pycno	SM only
Crawling	3.3;0.2	3.1;0.2	2.2;0.2*	3.0;0.3
Creeping	3.0;0.3	2.9;0.1	2.1;0.2*	2.7;0.2
Pulling	3.2;0.1	3.0;0.2	1.9;0.3*	2.8;0.2
Throbbing	3.2;0.2	3.3;0.2	1.2;0.2*	3.1;0.2
Aching	3.2;0.3	3.3;0.2	0.9;0.2*	3.2;0.2
Itching	2.1;0.4	2.0;0.3	1.1;0.3*	1.8;0.3
Electric shocks	2.3;0.2	2.4;0.2	1.1;0.2*	2.0;0.4
Sleep problems	3.1;0.3	3.2;0.2	1.2;0.3*	2.7;0.3

The scores are averages;SD.
*P<0.05 *vs.* baseline.

The described sensations at the limb were evaluated with the Visual Analogue Scale Line (0 to 4) (Table II). There was a minimal (non-significant) improvement with SM, marginally perceived by the patients.

Improvement with Pycnogenol® supplementation (for all assessed parameters) was significant (P<0.05) with important clinical significance since 19 out of 21 supplemented subjects reported a clearly perceived benefit from the supplementation.

Resting flux (Table III) — slightly elevated at inclusion — was normalized in the supplemented group as seen by a significant decrease in flux (P<0.05).

An elevated flux — even slightly higher, as seen in diabetic microangiopathy — can be associated with a dysregulation of the microcirculation, particularly of the thermo-regulatory layers (associated with minimal micro neuropathy).

The venoarteriolar response — low at inclusion in all subjects with RLS — was significantly increased (improved) with Pycnogenol® indicating a better axon-axon reflex response and a lower level of subclinical neural alteration and/or damage (as seen in diabetic microangiopathy).

The need for analgesics or other pain management (including physical therapy) during the study period, was significantly lower in the Pycnogenol® group (P<0.05) than in the SM-only group (only in 4 subjects out of 21 *vs.* 16 out of 24 in the SM-only group).

Thermography of the leg was inconclusive and did not reveal any significant asymmetry of perfusion or altered flow and before-after images were comparable.

Oxidative stress (PFR) was significantly reduced (P<0.05) only in subjects using the supplementation.

Minimal edema (as assessed by the edema tester) was observed in all subjects at inclusion (Table IV); it was significantly decreased (in score and in number of subjects affected) in the Pycnogenol® group.

TABLE III.—*Microcirculation.*

	Inclusion		4 weeks	
	Pycno	SM only	Pycno	SM only
FLUX flux units;SD Resting skin Flux	2.23;0.5	2.3;0.2	1.8;0.3*	2.22;0.4 NS
VAR (median/range) Venoarteriolar response (Normal values around 44%;35-55)	26.0% 12-35	26.4% 12-33	42.2%* 22-51	35.1% 14-46
Need for analgesics or other drugs			4/21#	16/24
Thermo	Inconclusive		Inconclusive	

In these patients both sides of the body were affected.
*P<0.05 *vs.* baseline; #P<0.05 *vs.* SM only.

TABLE IV.—*Plasma free radicals (oxidative stress) and minimal (subclinical) edema evaluation. *P<0.05* vs. *baseline.*

	Inclusion		4 weeks	
	Pycno	SM only	Pycno	SM only
Oxidative stress PFR Carr Units;SD	388;22	379;13	327;16*	382;19
Minimal edema score;SD Edema tester (0-4) (values <2 are not clinically visible)	2.1;0.2	2.2;0.3	0.4;0.2*	2.1;0.1
Number of subjects with micro-edema	21/21	24/24	4/21*	22/24

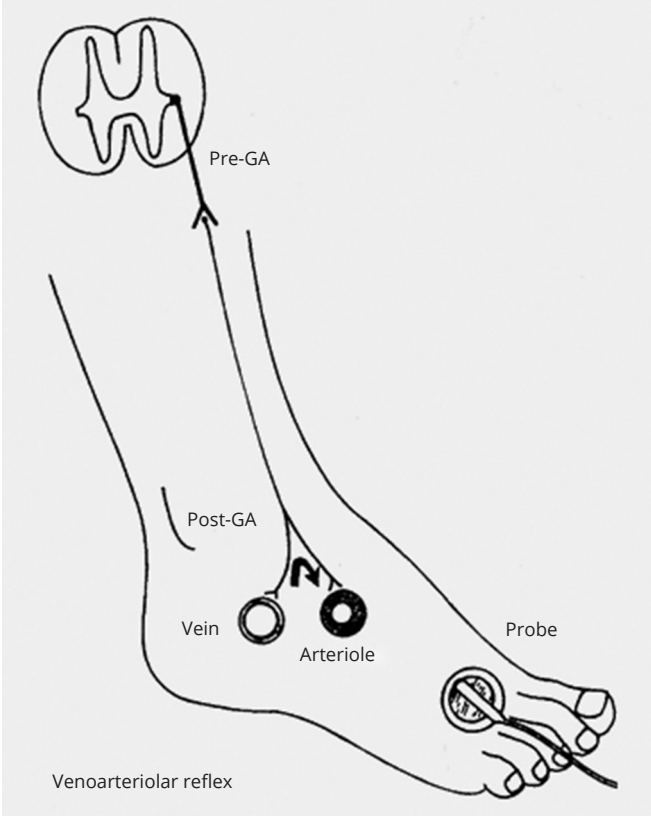

Figure 1.—The venoarteriolar reflex. Pathway of the VAR. The probe of the laser Doppler used to detect the VAR is placed at the distal part of the foot.
Pre-Ga: pre-gangliar axon; post-GA: sympathetic post gangliar axon.

Figure 1 shows in a simplified diagram, the VAR patterns, and pathways.

Figure 2 shows three graphs (laser Doppler flowmetry); Figure 2A shows the basic test flux, the spike (motion artifact) due to the passage to the standing position and the repositioning of the skin flux to a much lower level.[16, 17]

The VAR occurs in seconds.

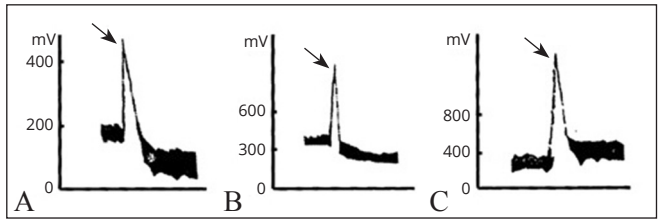

Figure 2.—Patterns of the VAR in different subjects. The three graphs (laser Doppler flowmetry tracings) show the following: A) The basic test flux at rest (supine position), the spike (arrow) due to the passage to the standing position and — the last part of the tracing — the repositioning of the skin flux to a much lower level (-40%). The VAR occurs in seconds; the tracing in the middle (B) shows an impairment of the VAR in subclinical diabetes; the third graph (C) shows the reposition of the flux after the changing position (the resulting flux is higher than in the supine, resting, original position).
The microcirculation — without the protection of the VAR — expands its bed, increasing the flux value, causing micropooling and increasing the exchange surface and the production of fluids resulting in edema.[16, 17]

The tracing in the middle (Figure 2B) shows an impairment of the VAR in subclinical diabetes.

The third graph (Figure 2C) shows the reposition of the flux after the changing in position (the flux is higher than in the supine, resting position). The microcirculation — without the protection of the VAR — expands its bed, increasing the flux value.

Discussion

RLS as a simple, isolated, idiopathic symptom is very common and can be associated with several conditions or treatments.

In many, otherwise healthy subjects, RLS is apparently a primary condition without any evident cause or concomitant, demonstrable disease.

Primary RLS is definitely not associated with clinical conditions or clear diseases of the spinal system or with defined vascular conditions or vertebral problems.

Postural problems, exercise, sedentary life or a very active sport life, lifting weights, working and sitting habits can all be considered to have an impact on RLS.

In subjects with RLS the venoarteriolar response[16-19] is often altered or affected as an early manifestation (*i.e.* as in preclinical diabetes, in diabetic microangiopathy and in other neurological disorders).

RLS is often associated with functional microangiopathy.

Often, as seen in Tooke's work,[16, 19, 20] the alterations in VAR are present before diabetes is clinically evident.

The VAR is an axon-axon reflex; this reflex response decreases skin flow (*i.e.*, flux, as measured by laser Dop-

pler flowmetry, at the foot) when the subject passes from the supine to the standing position.

The VAR protects with vasoconstriction the peripheral microcirculation from overload on standing and prevents edema.

The microvascular dilatation (produced *i.e.*, by ACE inhibitors or nifedipine)[18] alters (decreases its efficacy) the VAR with a lower constriction on standing, resulting in distal edema.

In this study all subjects in the Pycnogenol® group with idiopathic RLS had a significant (>15%) increase in venoarteriolar response (seen by LDF) after 4 weeks.

The role of minimal edema should be better explored in relation to the symptoms of RLS.

Pycnogenol® has a very significant anti-edema efficacy[14, 21-25] both in venous patients and in subjects with diabetic microangiopathy.[25]

Pycnogenol® has also shown a significant activity in controlling symptoms associated with training, running and in attenuating cramps and muscular pain after training in athletes.[4]

This action is quite comparable to the activity on symptoms reported in RSL in the present study.[4]

Conclusions

In conclusion, Pycnogenol® prevents or relieves most symptoms associated with restless leg syndrome and significantly improves the venoarteriolar response.

Future studies in this condition, including more complex subjects, could indicate the role of Pycnogenol® in this common and still obscure syndrome and, possibly, in subclinical muscular and neurological alterations.

References

1. Restless legs syndrome Patient Care & Health Information; [Internet]. Available from: hiips://www.mayoclinic.org/diseases-conditions/restless-legs-syndrome/diagnosis-treatment/ [cited 2020, Jul 15].

2. Cesarone MR, Belcaro G. Mayo Clinic: salute e benessere. Turin: Edizioni Minerva Medica; 2007.

3. Porter RS, Kaplan JL. The Merck Manual of Diagnosis and Therapy. Twelfth edition. Kenilworth, NJ: Merck, Sharp & Dohme Corp.; 2018.

4. Vinciguerra G, Belcaro G, Cesarone MR, Rohdewald P, Stuard S, Ricci A, *et al.* Cramps and muscular pain: prevention with pycnogenol in normal subjects, venous patients, athletes, claudicants and in diabetic microangiopathy. Angiology 2006;57:331–9.

5. Belcaro G. The Angiology Bible. London: Imperial College Press, World Scientific Publications; 2018.

6. Cesarone MR, Belcaro G, Nicolaides AN, Arkans E, Laurora G, De Sanctis MT, *et al.* The edema tester in the evaluation of swollen limbs in venous and lymphatic disease. Panminerva Med 1999;41:10–4.

7. Cesarone MR, De Sanctis MT, Laurora G, Incandela L, Belcaro G. [Lymphedema. New non-invasive methods for diagnosis and follow up]. Minerva Cardioangiol 1995;43:211–8. Italian.

8. Belcaro G, Bollinger A, Hoffman U, Nicolaides AN. Laser-Doppler. London: Med-Orion; 1999.

9. Galderisi M. Diastolic dysfunction and diabetic cardiomyopathy: evaluation by Doppler echocardiography. J Am Coll Cardiol 2006;48:1548–51.

10. Cesarone MR, Belcaro G, Carratelli M, Cornelli U, De Sanctis MT, Incandela L, *et al.* A simple test to monitor oxidative stress. Int Angiol 1999;18:127–30.

11. Cornelli U, Belcaro G, Ledda A, Feragalli B. Oxidative stress following administration of levothyroxine in subjects suffering from primary hypothyroidism. Panminerva Med 2011;53(Suppl 1):95–8.

12. Cesarone MR, De Sanctis MT, Incandela L, Belcaro G, Griffin M, Cacchio M. Methods of evaluation and quantification of microangiopathy in high perfusion microangiopathy (chronic venous insufficiency and diabetic microangiopathy). Angiology 2001;52(Suppl 2):S3–7.

13.Belcaro G, Cornelli U, Dugall M, Luzzi R, Hosoi M, Ledda A, *et al.* Panel 2013 Supplements and green drugs studies; New rules 2013. London and Annecy Panel. Angiology online 31.12.2012.

14. Belcaro G. Pharma standard supplements. Clinical use. London: Imperial College Press, World Scientific Publications; 2016.

15. Maxwell C. Clinical research for all. Cambridge: Cambridge Medical Publications Ltd.; 1973.

16. Rayman G, Hassan A, Tooke JE. Blood flow in the skin of the foot related to posture in diabetes mellitus. Br Med J (Clin Res Ed) 1986;292:87–90.

17. Belcaro GV. Venous hypersensitive microangiopathy: evaluation with laser-doppler flowmetry. Thesis: (PhD), Imperial College, St Mar's Campus, University of London. 1998.

18. Salmasi AM, Belcaro G, Nicolaides AN. Impaired venoarteriolar reflex as a possible cause for nifedipine-induced ankle oedema. Int J Cardiol 1991;30:303–7.

19. Rayman G, Williams SA, Spencer PD, Smaje LH, Wise PH, Tooke JE. Impaired microvascular hyperaemic response to minor skin trauma in type I diabetes. Br Med J (Clin Res Ed) 1986;292:1295–8.

20. Flynn MD, Tooke JE. Diabetic neuropathy and the microcirculation. Diabet Med 1995;12:298–301.

21. Cesarone MR, Belcaro G. La medicina alternativa-complementare e la medicina dei supplementi. Turin: Minerva Medica; 2019.

22. Cesarone MR, Belcaro G. Supplementi Pharma standard; informatore Applicazioni cliniche. Turin: Minerva Medica; 2020.

23. Belcaro G, Cesarone MR. The Pharma-standard supplements informator: consensus. Turin: Minerva Medica; 2020.

24. Belcaro G, Cesarone MR, Ricci A, Cornelli U, Rodhewald P, Ledda A, *et al.* Control of edema in hypertensive subjects treated with calcium antagonist (nifedipine) or angiotensin-converting enzyme inhibitors with Pycnogenol. Clin Appl Thromb Hemost 2006;12:440–4.

25. Belcaro G, Cesarone MR, Errichi BM, Ledda A, Di Renzo A, Stuard S, *et al.* Diabetic ulcers: microcirculatory improvement and faster healing with pycnogenol. Clin Appl Thromb Hemost 2006;12:318–23.

Conflicts of interest.—The authors certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

Authors' contributions.—All authors read and approved the final version of the manuscript.

History.—Manuscript accepted: July 10, 2020. - Manuscript received: July 7, 2020.