FILM COATED TABLET COMPRISING AN EXTRACT OF RED VINE LEAVES

BACKGROUND OF THE INVENTION

1. TECHNICAL FIELD

The invention relates to a film coated tablet comprising a dried extract of red vine leaves, an excipient and a tablet film and the use thereof for the improvement of the blood circulation and/or the oxygen supply of the lower extremities.

2. BACKGROUND INFORMATION

Chronic venous insufficiency (CVI) is a progredient disease and will lead in many patients – especially if untreated – to oedema, coronal phlebectasia (Widmer stage I), hyperpigmentation, induration, lipodermatosclerosis, white atrophy (Widmer stage II), or varicose leg ulcers (Widmer stage III). Chronically disturbed haemodynamics of deep or superficial veins due to obstructed venous segments or valvular incompetence lead usually to skin diseases in the inner ankle area of the lower limbs.1 Disturbances in the microcirculation of the skin have been considered to be major contributors for skin changes associated with chronic venous hypervolaemia and venous hypertension. (e.g. Fagrell B Vital microscopy and the pathophysiology of deep venous insufficiency. Int Angiol 1995;14:18-22.; Jünger M, Klyscz T, Hahn M, Rassner G. Disturbed blood flow regulation in venous leg ulcers. Int J Microcirc 1996;16:259-265).

Obviously, cutaneous microangiopathy of clinical relevance such as enlarged, tortuous capillaries surrounded by micro-oedema contributes to the skin alterations in the lower limbs and determines the course of CVI (Fagrell B, *loc. cit.* and Jünger M et al., *loc. cit.*).

The application of the laser Doppler technique in venous disorders is well illustrated. (e.g. Tulevski II, Ubbink DT, Jacobs MJHM. Red and green laser Doppler compared with capillary microscopy to assess skin microcirculation in the feet of healthy subjects. Microvasc Res 1999;58(2):83-88 and Bollinger A, Jäger K, Jünger M, Seifert H. The vascular laboratory: advances in non-invasive techniques. World J Surg 1988;12:724-731).

Different techniques have been developed to investigate microcirculation in both functionally different layers of the skin: the deeper, mainly thermoregulatory layer and the superficial, nutritive layer. Microcirculatory disturbances in the superficial nutritive layer are of utmost relevance for trophical skin changes. (Jünger M et al., *loc. cit.* and Gschwandtner ME, Ambrozy E, Fasching S, Willfort A, Schneider B, Böhler K, et al. Microcirculation in venous ulcers and surrounding skin: findings with capillary microscopy and laser Doppler imager. Eur J Clin Invest 1999;29:708-716).

The British patent GB 934,554 discloses that the capillary resistance of guniea pigs deficient in a vitamin can be enhanced by intraperitonally administration of an alcoholic extract of vine leaves.

The International patent application WO 01/28363 discloses a method for preventing or alleviating the discomfort associated with mild-to-moderate chronic venous insufficiency of the lower extremities with the aid of an aqueous extract of red vine leaves. In addition a daily dosage regimen of 80 to 1000 mg divided up in 1 to 3 capsules is suggested.

The problem underlying the present invention was to provide a dosage form which allows to administer such high amounts of aqueous extract of red vine leaves in the suggested regimen. In view of patient compliance there is the requirement that such dosage forms shall not be too big, in order to facilitate swallowing. On the other hand the dosage form must have a high stability to ensure long shelf storage times. Moreover, high bioavailability is but a prerequisite for the therapeutic and/or preventive success of such a dosage form.

BRIEF SUMMARY OF THE INVENTION

It has been surprisingly found that a film coated tablet comprising:

(a) at least 50 % by weight of a dried extract of red vine leaves, which is obtainable by extraction of red vine leaves with water and drying;

(b) up to 50 % by weight of an excipient consisting essentially of
- at least one binder,
- at least one disintegrant,
- at least one filler, and
- a lubricant; and

(c) a tablet film consisting essentially of a film former, a plastiziser, a coating agent and optionally a coloring agent;

fulfils these requirements and can be used to significantly enhance the microcirculation and the oxygen supply at the predominantly affected perimalleolar area of the leg in CVI patients.

Accordingly, a first aspect of the invention provides a film coated tablet comprising the following constituents:

(a) at least 50 % by weight of a dried extract of red vine leaves, which is obtainable by extraction of red vine leaves with water , drying and addition of 0.5 to 10% by weight of silica relating to the total amount of component (a);

(b) up to 50 % by weight of an excipient consisting essentially of

- at least one binder,
- at least one disintegrant,
- at least one filler, and
- a lubricant; and

(c) a tablet film consisting essentially of a film former, a plastiziser, a coating agent and optionally a coloring agent.

A second aspect of the present invention provides a process for preparing such a film coated tablet comprising the steps of:

(A) mixing the dried aqueous extract of red vine leaves (a) with the excipients (b), optionally in the presence of a volatile diluent;

(B) optionally screening the mixture obtained;

(C) compressing the mixture with a suitable tablet press; and

(D) coating the resulting tablet with the tablet film (c).

A third aspect of the present invention provides a use of a film coated tablet as defined above for preparing a pharmaceutical or dietary composition for the treatment or prevention of the discomfort, disorder and/or disease associated with chronic venous hypervolaemia and venous hypertension.

A fourth aspect of the present invention provides an aqueous extract of red vine leaves, which is obtainable by a method comprising the steps of:

(a) collecting red vine leaves at a point of time when the content in flavonoids has reached an optimum;

(b) drying and crushing the leaves;

(c) cutting the leaves to pieces;

(d) extracting the leaves with water at elevated temperatures for 6 to 10 hours;

(e) concentrating and drying the obtained extract, and

(f) addition of 0.5 to 10 % by weight of silica relating to the final total amount of the resulting extract.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the schematic design of the clinical study which has been carried out to prove the efficacy of the film coated tablet according to the present invention.

Figure 2 shows the influence of the vine leaf extract containing film coated tablet

—●— AS195 360 mg compared with

—O— placebo

on the microcirculation measured with Laser Doppler flowmetry (LDF 10-37 kHz).

Figure 3 shows the influence of the vine leaf extract containing film coated tablet

—●— AS195 360 mg compared with

—O— placebo

on Transcutaneous oxygen partial pressure (tpO_2)

DETAILED DESCRIPTION OF THE INVENTION

The film coated tablet of the present invention consists of herbal ingredients derived by an aqueous extraction from red vine leaves (*folia vitis viniferae*; Extractum Vitis viniferae e folium spissum et siccum) and drying (a), excipients (b) and the tablet film (c). This extract contains flavon(ol)-glycosides, -glucuronides and flavonoids, with quercetin-3-O-ß-D-glucuronide and isoquercitrin (quercetin-3-O-ß-glucoside) as its main active ingredients, . The range of their pharmacological actions has not yet been fully elucidated, but in-vitro studies indicate that they have antioxidant and anti-inflammatory properties and that they inhibit platelet aggregation and hyaluronidase and reduce oedema, possibly by reducing capillary permeability. Preclinical in-vivo experiments demonstrated anti-inflammatory and capillary wall thickening effects.

The film coated tablet according to the present invention comprises 50 to 70% of a dried aqueous red vine leaf extract with a high flavonoid content of 2-15%.

As a rule the relation by weight between the dried extract and the excipients used to produce the core of the tablet is between 1 : 1 to 2 : 1, preferably between 1.1 : 1 and 1.8 : 1, in particular between 1.25 : 1 and 1.75 : 1.

A film coated tablet comprising

(a) 50 to 70 % by weight of said dried extract of red vine leaves;

(b) 25 to 49 % by weight of said excipient, and

(c) 1 to 5 % by weight of said tablet film,

based on the total mass of the film coated tablet, is preferred.

More preferred is a film coated tablet comprising

(a) 51 to 59 %, in particular about 55 % by weight of said dried extract of red vine leaves;

(b) 38 to 48 %, in particular about 43 % by weight of said excipient, and

(c) 1 to 3 %, in particular about 2.7 % by weight of said tablet film,

based on the total mass of the film coated tablet.

Another preferred embodiment is a film coated tablet according, wherein the excipient (b) essentially consists of

- 70 to 85 % by weight of at least one binder,
- 0,5 to 12,5 % by weight of at least one disintegrant,
- 5 to 15 % by weight of at least one filler, and
- 1 to 5 % by weight of at least one lubricant,

based on the total mass of the combined excipients.

The term "binder" a used hereinbefore and hereinafter denotes an excipient which is suitable for binding other components to one another. Preferred binders according to the invention are selected from among:

powdered cellulose, microcrystalline cellulose, sorbitol, starch, polyvinylpyrrolidone (povidone), copolymers of vinylpyrrolidone with other vinyl derivatives (copovidone), cellulose derivatives, particularly methylhydroxypropylcellulose, e.g. Methocel A 15 LV, and mixtures of these compounds. The preferred binders are powdered cellulose, particularly microcrystalline cellulose and/or copovidone. If the abovementioned binders are used, the amount by weight, based on the total mass of the tablet according to the invention, is preferably in a range of 15 - 45 wt.%, more

preferably 25 – 40 wt.%, most preferably about 33 wt.%. Thanks to the particularly preferred binder microcrystalline cellulose, tablets are obtained having a high stability and good compliance for the patients to whom the aqueous extract of red vine leaves has to be aministered.

The tablet according to the invention also contains disintegrants in addition to the abovementioned ingredients. Within the scope of the present invention these disintegrants may optionally also be known as breakdown agents. These are preferably selected, according to the invention, from among sodium starch glycolate, crosslinked polyvinylpyrrolidone (crospovidone), croscarmellose sodium salt (sodium salt of cellulose carboxymethyl ether, crosslinked), sodium-carboxymethylcellulose, dried maize starch, colloidal anhydrous silica and mixtures thereof. Within the scope of the present invention it is particularly preferred to use sodium starch glycolate, crospovidone and, preferably, the sodium salt of crospovidone or croscarmellose and colloidal anhydrous silica. Most preferred is a mixture of croscarmellose sodium, colloidal anhydrous silica and optionally crospovidone. If the abovementioned disintegrants are used, the amount by weight, based on the total mass of the tablet according to the invention, is preferably in a range of about 0.5 - 10 wt.%, more preferably about 1.5 – 7.5 wt.%. Thanks to the particularly preferred combination of disintegrants, tablets are obtained having a high stability and provide high bioavailability to the aqueous extract of red vine leaves.

The tablet according to the invention also contains a filler. As a rule fillers are inert compounds such as inorganic metal oxides or inorganic phosphate or hydrogen phosphate. Preferably the filler is anhydrous calcium hydrogen phosphate. If the abovementioned fillers are used, the amount by weight, based on the total mass of the tablet according to the invention, is preferably in a range of about 1 - 10 wt.%, more preferably about 2 – 8 wt.%.

The tablet according to the invention also contains flow agents or flow regulators and also lubricants, as additional ingredients. These include, within the scope of the present invention, for example, silicon dioxide, talc, stearic acid, sodium stearylfumarate, magnesium stearate and glycerol tribehenate. According to the invention magnesium stearate is preferably used. If the abovementioned preferred lubricants are used, the amount by weight thereof, based on the total mass of the tablet according to the invention, is preferably in a range of about 0.1 - 10 wt.%, preferably about 0.5 – 5 wt.%, more preferably between 0.6 and 1.5 wt.%.

Furthermore preferred is a film coated tablet, wherein the tablet film (c) essentially consists of

- 50 to 85 % by weight of at least one film former,
- 5 to 10 % by weight of at least one plasticizer,
- 10 to 20 % by weight of at least one coating agent such as talc, and
- 0 to 15 % by weight of at least one colorant.

based on the total mass of the tablet film (c).

The tablet film according to the invention may also contain one or more synthetic or natural, pharmaceutically acceptable colorant, preferably one or more inorganic metal oxides such as titanium dioxide (E 171) and/or ferric oxide (E 172). If the abovementioned preferred colorants are used the amount by weight thereof based on the total mass of the tablet according to the invention is 0.01 to 0.5 wt.%.

It is a further object of the present invention to provide a film coated tablet for preventing and/or alleviating the discomfort associated with mild-to-moderate chronic venous insufficiency of the lower extremities comprising herbal ingredients, wherein the tablet is manufactured pursuant to a controlled process that preserves the herbal curing qualities of the ingredients.

It is still a further object of the present invention to provide a film coated tablet which is effective in preventing and/or alleviating the discomfort associated with mild-to-moderate chronic venous insufficiency of the lower extremities.

It is still a further object of the present invention to provide a film coated tablet for preventing and/or alleviating the discomfort associated with mild-to-moderate chronic venous insufficiency of the lower extremities comprising herbal ingredients and having minimal or no side effects and thus being safe for internal consumption and a high stability and good patient compliance.

The aqueous extract prepared from dried red vine leaves is characterised by a high content of 2 to 20%, preferably 2 to 10 % of biologically active flavonoids.

The term "a person in need thereof" or "patient" as used hereinabove and hereinbelow relates to a female or male person who suffers from clinically not relevant early stages of chronic venous insufficiency (CVI) from proven CVI stage I and II according to Widmer. As a rule such patients are elderly people with an age of between 30 and 80, preferably between 32 and 76 years having

an mean age (± standard deviation) of 55.2 ± 7.7 years. As a rule CVI is more expressed in female than in male patients.

In order that this invention be more fully understood, the following examples are set forth. These examples are for the purpose of illustrating embodiments of this invention, and are not to be construed as limiting the scope of the invention in any way.

The examples which follow are illustrative and, as recognised by one skilled in the art, particular conditions could be modified as needed for individual compositions. Materials used in tests below are either commercially available or easily prepared from commercially available materials by those skilled in the art.

The basis of the tablet is the aqueous extract of red vine leaves (*foliae vitis viniferae L.*). The starting material for the preparation of the extract are red vine leaves collected at a point of time where the content in flavonoids has reached an optimum. This is usually the case around the harvesting time of the grapes. The leaves are carefully dried and crushed. For extraction the leaves are cut to pieces of preferably 5 to 10 mm. To achieve a high content in flavonoids the extraction is done at elevated temperature, preferably at a temperature in the range of 60° to 80°C, over a time of at least 6 up to 10 hours. The preferred method is that of an exhaustive percolation.

The so-called fluid extract obtained in the course of the extraction is concentrated by use of a suitable evaporator. The thick extract obtained in this step is dried, for instance by use of a vacuum drying oven or a vacuum drying conveyer.

All or some of the excipients may be added during drying to facilitate further processing of the extract. As a rule up to 10 % of one or more constituents of the excipients can be added during the drying process.

Preferably a part of the flow regulator such as colloidal, anhydrous silica is added to the extract during drying or before admixing with the other constituents. Preferably the resulting extract composition contains 0.5 to 10 % by weight, in particular 2.5 to 7.5 % by weight, most preferably about 4 % by weight of colloidal, anhydrous silica.

Surprisingly, tablets obtained from an extract to which a part of the excipients have been added during the drying process show an enhanced stability.

Most preferably the film coated tablet according to this invention consists of

- 300 to 500 mg, preferably 320 to 400 mg, in particular about 355 to 380 mg of dry aqueous extract of red vine leaf (4-6 : 1) (*extractum vitis viniferae foliae aquosum siccum*), which may contain up to 10 % weight of a flow regulator, in particular colloidal, anhydrous silica;
- the following excipients of the tablet core:

  microcrystalline cellulose, croscarmellose sodium, calcium hydrogen phosphate (anhydrous), colloidal silica (anhydrous), magnesium stearate, and optionally crospovidone, and
- a tablet film consisting of:

  hypromellose, glyceryl tristearate, titanium dioxide (E 171), talc, ferric oxide, red (E 172).

Film coated tablets were prepared with the ingredients listed in the following tables A and B:

Table A

Name of ingredient	Quantity per film coated tablet [mg / 658.000 mg]	Function
Tablet Core		
Vitis viniferae folium dry extract aqueous (4 - 6 : 1)	360.000	Active ingredient
Microcrystalline cellulose	219.000	Binder, disintegrant
Croscarmellose sodium	18.000	Disintegrant
Calcium hydrogen phosphate, anhydrous	30.000	Filler
Silica, colloidal,anhydrous	4.000	Flow regulator, disintegration accelerator

Magnesium stearate	9.000	Lubricant
Tablet Film		
Hypromellose	11.383	Film former
Glyceryl tristearate	1.138	Plasticizer
Titanium dioxide (E 171)	0.783	Colouring agent
Talc	3.131	Coating agent
Ferric oxide, red (E 172)	1.565	Colouring agent

The extract is mixed with the excipients of the tablet core and compressed on a suitable tablet press.

Table B

Name of ingredient	Quantity per film coated tablet [mg / 658.000 mg]	Function
Tablet Core		
Vitis viniferae folium dry extract aqueous (4 - 6 : 1)	360.000	Active ingredient
Silica, colloidal,anhydrous	15.000	binder
Microcrystalline cellulose	214.000	Binder, disintegrant
Croscarmellose sodium	18.000	Disintegrant
Calcium hydrogen phosphate, anhydrous	30.000	Filler
Silica, colloidal,anhydrous	6.000	Flow regulator, disintegration accelerator
Magnesium stearate	9.000	Lubricant
Crospovidone	18.000	Disintegrant
Tablet Film		
Hypromellose	11.383	Film former
Glyceryl tristearate	1.138	Plasticizer
Titanium dioxide (E 171)	0.783	Colouring agent
Talc	3.131	Coating agent
Ferric oxide, red (E 172)	1.565	Colouring agent

The extract is mixed with 15.000 mg silica during the drying process, which yields an extract consisting of 96 % by weight of the ingredients of the extract and 4 % of silica. This resulting mixture is mixed with the remaining excipients of the tablet core and compressed on a suitable tablet press.

The compression forces which are needed to produce tablets of suitable breaking resistance and hence with the required breakdown times are dependent on the shapes and sizes of the punching tools used. Compression forces in the range from 2 - 20 kN are preferred. Higher compression

forces may lead to tablets with a delayed released of active substance. Lower compression forces may produce mechanically unstable tablets. The tablet cores may have different shapes; the preferred shapes are round biplanar or biconvex and oval or oblong forms.

The coating solution is prepared by mixing the film-forming agent with the colouring materials and a plasticizer in water. Using a suitable coating pan the film-coating solution is applied on to the tablet cores.

Preferably the tablets have an oblong shape to facilitate swallowing. In the case of a film-coated tablet containing 360 mg of extract and an extract to excipient ratio as indicated before, an oblong tablet may be about 17-18 mm long and have a width of about 8 to 9 mm. These film coated tablets of Table A are hereinbelow coded "AS 195".

To enhance the blood circulation and/or the oxygen supply of the lower extremities, the tablet should be taken in dosages corresponding to 150 and 1000 mg of extract, preferably 300-800 mg, in particular 350-750 mg daily. The total amount of extract may be divided up in 1 to 3 film coated tablets a day. The daily dose should be taken at once, preferably in the morning.

Impressive improvement of the symptoms can be expected within 6 weeks of continuous use. The optimum effect is maintained or amplified on longer use.

Methods

Participants

Male and female patients, age 18 years or more, with proven CVI I or CVI according to Widmer, with diagnosis confirmed and present for at least one year were enrolled. Medically relevant concomitant diseases have to be absent. Patients who used drugs to alleviate their CVI symptoms within 4 weeks or were treated with theophyllin, diuretics, cardiac glycosides, ACE inhibitors or calcium antagonists within 8 days prior to the first examination were not allowed to be enrolled. Compression bandages or concomitant therapy for venous problems were forbidden during the participation in the trial.

Design and procedures

The double-blind, randomised, placebo-controlled cross-over trial was run according to the principles of the declaration of Helsinki and the International Conference of Harmonisation of Good Clinical Practice.

Each patient participated for 17 weeks in the trial: for a one-week wash-out (placebo-treated), for a 6-week treatment period (Group_1 starting with AS 195, Group_2 starting with placebo), for a 4-week wash-out (placebo-treated), and for a second 6-week treatment period (Group_1 continuing with placebo, Group_2 continuing with the).

AS 195 (film-coated tablets containing 360 mg dry extract of red vine leaves) or placebo tablets were taken according to the randomisation schedule as single dose in the morning. Both tablets were identical with respect to size, shape, weight, inner appearance, and taste.

For laser Doppler flowmetry the equipment was provided by LMTB, Berlin, Germany (e.g. Doerschel K, Mueller G. Velocity resolved laser Doppler flow measurement in skin. Lasermedizin 1996;12:163-171.). The equipment is a computer-based mobile unit using a laser frequency of 785 nm. The laser probe was fixed 3.5 cm distal to the inner ankle of the more affected leg. After 30 minutes sitting for adaptation to room temperature, measurement started after 10 minutes standing (256 points of measurement, duration of measurement: approx. 0.4 seconds). The back-scattered light was retrieved by two diodes in the range of frequencies between 0.2 to 37.2 kHz. The data were processed using a Fast Fourier Transformation. Finally, the output referred to the range of frequencies between 0.2 to 10.0 kHz for vessels in the reticular venous plexus (larger mainly thermoregulative vessels, diameter more than 30 micrometer) and to the range of frequencies between 10.1 to 37.2 kHz for capillaries in the subpapillary venous plexus (superficial small nutritive vessels, diameter 6 to 30 micrometer).

Transcutaneous oxygen pressure (tcPO2) was measured using modified Clark-type polarographic electrodes containing noble metal cathodes and silver/sliver chloride anodes (TCM 3, Radiometer Copenhagen, Brønshøj, Denmark). A heating element adjacent to the anode maintained skin temperature at 43° Celsius. At this temperature the arterioles are maximally dilated, tcPO2 approximates the PO2 of arterial blood (e.g. Bollinger A, Jäger K, Jünger M, Seifert H. The vascular laboratory: advances in non-invasive techniques. World J Surg 1988;12:724-731.).

The electrode was attached to the skin surface by an adhesive ring device which was filled with physiological saline, 3.5 cm anteriolateral from the Laser Doppler probe. After 30 minutes sitting for adaptation to room temperature, measurement started after 10 minutes of standing. A measurement lasted approx. 15 minutes. The $tcPO_2$ values are expressed in millimeter mercury column (mmHg). Normal values available for the dorsum of the foot of patients without CVI are ranged between 40 and 80 mmHg.

Local skin temperature was measured with a thermistor fixed adjacent to the oxygen electrode in the perimalleolar region. In order to minimise effects on the skin perfusion, LDF and $tcPO_2$ measurements were conducted between 28 and 32° C local skin temperature.

Calf and ankle circumference were measured using a measuring tape. Measurements were carried out at the lateral and medial ankle and at the middle of the calf.

Subjective symptoms of CVI (tired heavy legs, sensation of tension, tingling sensation, and pain) were measured by using a 10-cm visual analogue scale with zero as "none at all" and 10 cm as "very strong".

Overall treatment efficacy was rated by patients and investigators on a 4-point verbal rating scale (good, satisfactory, not satisfactory, and bad) at the end of each treatment period.

Overall tolerability was rated by patients and investigators on a 4-point verbal rating scale (good, satisfactory, not satisfactory, and bad). The patients were questioned about their well-being in general terms at each visit.

Results

Seventy-one women and men aged between 32 and 76 years with proven CVI stage I and II according to Widmer were included. The mean age (± standard deviation) was 55.2 ± 7.7 years; 55 were women, 16 men. The phlebological status revealed moderate or severe intensity of varicosis in 47 (67.1%), pigmentation in 27 (38.6%), ankle oedema in 26 (37.1%), and lower leg oedema in 25 (35.7%) patients. Mild signs of atrophy were present in 13 patients (18.6%), of eczema in none (Table 1).

Table 1: **Demographics and baseline characteristics of CVI**

	AS 195 / Placebo (n=36)	Placebo / AS195 (n=35)
Continuous variates (median (range))		
Age [years]	66 (32-76)	66 (37-76)
Height [cm]	168 (150-186)	165 (150-191)
Weight [kg]	76.5 (48-97)	73 (55-120)
Body mass index [kg/m^2]	27.6 (20.6-32.0)	26.7 (20.1-42.5)
Systolic blood pressure [mmHg]	130 (100-150)	135 (120-140)
Diastolic blood pressure [mmHg]	80 (60-90)	80 (65-90)
Categorical variates (n (%))		
Female	24 (66.7)	31 (88.6)
Current smoker	4 (11.1)	1 (2.9)
CVI stage		
Stage I	26 (72.2)	23 (65.7)
Stage II	10 (27.8)	12 (34.3)
Phlebological status of moderate to severe intensity		
Varicosis	26 (72.2)	22 (62.9)
Pigmentation	11 (30.6)	17 (48.6)
Atrophy	0 (0.0)	0 (0.0)
Eczema	0 (0.0)	0 (0.0)
Ankle oedema	13 (36.1)	14 (40.0)
Lower leg oedema	12 (33.3)	14 (40.0)

Protocol violations did not occur in the remaining patients. Therefore, all patients remained in the intention to treat analyses (Figure 1). Patient characteristics were homogenously distributed across the two treatment sequences (Group_1, Group_2) except for the sex ratio (12 men in Group_1, 4 men in Group_2) (Table 1). Baseline values for the laser Doppler parameters, transcutaneous oximetry, ankle and calf circumferences, and subjective symptoms were comparable for Group_1 and Group_2 (Table 2). Compliance with the film coated tablet according to the invention was approximately 100% in both treatment sequences.

Table 2: **Mean (±SD) of baseline characteristics of each treatment period**

	Period 1		Period 2	
	AS 195 (n=36)	Placebo (n=34)	AS 195 (n=34)	Placebo (n=36)
Laser Doppler Flowmetry [AU]				
10-37 kHz	303.5 (135.2)	333.5 (153.0)	275.4 (126.4)	293.3 (119.9)
< 10 kHz	352.7 (87.7)	370.8 (120.0)	174.7 (77.0)	189.4 (67.6)
Transcutaneous Oximetry [mmHg]	32.1 (7.0)	32.3 (6.4)	30.1 (6.2)	30.8 (6.4)

Circumference [cm]				
Ankle	20.3 (2.2)	20.4 (2.4)	20.2 (2.6)	20.3 (2.2)
Calf	34.7 (3.1)	34.2 (3.0)	34.0 (3.1)	34.6 (3.2)
Subjective symptoms [cm]				
Tired/heavy legs	4.3 (2.8)	3.7 (2.9)	4.6 (2.9)	5.2 (2.6)
Pain in legs	4.0 (3.2)	3.2 (3.1)	4.5 (2.7)	4.9 (3.1)
Sensation of tension	4.5 (2.9)	4.1 (2.8)	4.5 (2.6)	5.1 (2.5)
Tingling sensation	3.3 (3.1)	2.7 (2.9)	3.7 (2.6)	4.2 (2.8)

Laser Doppler Flow measurements in the frequency range of 10-37 kHz were elected for the primary endpoint. These frequencies are considered to be determined by the number of erythrocytes and their movements (flow velocity) in the capillaries of the superficial layer of the skin of the leg. After 6 weeks the laser Doppler frequencies (10-37 kHz) increased in the AS 195 group (plus 241.8±18.7 AU) but decreased in the placebo group (minus 41.0±18.7 AU, $p<0.0001$) (Table 3). This effect was present as early as 3 weeks after start of treatment ($p<0.0001$) (Table 4, Figure 2).

Table 3: **Mean (±SEM) of change from baseline adjusted for period effects, 95 % confidence interval for treatment contrasts and p value after 3 weeks treatment with 360 mg AS 195 or placebo**

	Treatment		Treatment contrast		
	AS 195 (n=70)	Placebo (n=70)	Difference (n=70)	Confidence interval (n=70)	p value
Week 3					
Laser Doppler Flowmetry [AU]					
10-37 kHz	132.2 (11.9)	-28.2 (11.9)	160.5	127.0 to 194.0	< 0.0001
< 10 kHz	-3.7 (9.2)	-99.9 (9.2)	96.2	70.2 to 122.2	< 0.0001
Transcutaneous Oximetry [mmHg]	0.62 (0.97)	-3.84 (0.97)	4.46	1.72 to 7.20	0.0018
Circumference [cm]					
Ankle	-0.19 (0.09)	0.21 (0.09)	-0.40	-0.65 to -0.15	0.0025
Calf	-0.24 (0.04)	0.04 (0.04)	-0.28	-0.40 to -0.17	< 0.0001
Subjective symptoms [cm]					
Tired/heavy legs	-0.94 (0.25)	0.21 (0.25)	-0.73	-1.42 to -0.04	0.0396
Pain in legs	-1.17 (0.23)	- 0.24 (0.23)	-0.94	-1.59 to -0.28	0.0061
Sensation of tension	-1.00 (0.24)	- 0.52 (0.24)	-0.49	-1.17 to 0.19	0.1588
Tingling sensation	-0.99 (0.26)	- 0.20 (0.26)	-0.79	-1.52 to - 0.06	0.0335

Table 4: **Mean (±SEM) of change from baseline adjusted for period effects, 95 % confidence interval for treatment contrasts and p value after 6 weeks treatment with 360 mg AS 195 or placebo**

	Treatment		Treatment contrast		
	AS 195 (n=70)	**Placebo (n=70)**	**Difference (n=70)**	**Confidence interval (n=70)**	**p value**
Week 6					
Laser Doppler Flowmetry [AU]					
10-37 kHz **(primary endpoint)**	241.8 (18.7)	-41.0 (18.7)	282.8	229.9 to 335.7	< 0.0001
< 10 kHz	57.0 (12.4)	-107.7 (12.4)	164.7	129.7 to 199.7	< 0.0001
Transcutaneous Oximetry [mmHg]	1.35 (0.97)	-7.27 (0.97)	8.63	5.88 to 11.38	< 0.0001
Circumference [cm]					
Ankle	-0.39 (0.09)	0.29 (0.09)	-0.68	-0.94 to -0.43	< 0.0001
Calf	-0.54 (0.05)	0.14 (0.05)	-0.68	-0.83 to -0.53	< 0.0001
Subjective symptoms [cm]					
Tired/heavy legs	-0.78 (0.33)	-0.94 (0.33)	0.16	-0.76 to 1.09	0.7285
Pain in legs	-0.76 (0.35)	-0.86 (0.35)	0.10	-0.88 to 1.09	0.8323
Sensation of tension	-0.96 (0.35)	- 1.40 (0.35)	0.44	-0.46 to 1.44	0.3819
Tingling sensation	-0.55 (0.30)	- 0.66 (0.30)	0.11	-0.75 to 0.96	0.8044

Laser Doppler Flow measurements in the frequency range below 10 kHz are considered to be determined by the number of erythrocytes and their movements (flow velocity) in the capillaries in the deeper mainly thermoregulative layer of the skin of the leg. After 6 weeks the laser Doppler frequencies below 10 kHz) increased in the AS 195 group (plus 57.0±12.4 AU) and decreased in the placebo group (minus 107.7±12.4 AU, p<0.0001) (Table 3). This effect seems to depend on the climatic condition during the treatment period. During the study period of moderate temperatures (April/May) the Laser Doppler measurements (<10 kHz) remained unchanged in the AS 195 treatment group after an initial drop whereas the measurements in the placebo group decreased (p<0.0001). During the study period of higher temperatures (July/August) the laser Doppler measurements (<10 kHz) increased in the AS 195 treatment group and remained constant in the placebo group. (p<0.0001).

The transcutaneous oxygen pressure increased in the AS 195 group (plus 1.35±0.97 mmHg) but decreased in the placebo group (minus 7.27±0.97 mmHg, p<0.0001). This observation was consistent in both treatment periods and would therefore be in line with the Laser Doppler Flow in the nutritive superficial layer of the skin (i.e., 10-37 kHz) (Table 3,4, Figure 3).

The statistically significant and clinically relevant reduction of ankle (after 3 weeks: AS 195 minus 0.19±0.09 cm, placebo plus 0.21±0.09 cm, p=0.0025) and calf circumferences (after 3 weeks: AS 195 minus 0.24±0.04 cm, placebo plus 0.04±0.04 cm, p<0.0001) indicate an onset of action as early as 3 weeks after start of treatment (Table 3). This effect becomes more pronounced after 6 weeks (AS 195 ankle: minus 0.39±0.09 cm, calf: minus 0.54±0.05; placebo ankle: plus 0.29±0.09 cm, calf: plus 0.14±0.05 cm, p<0.0001) (Table 4)

There was no relevant change of the intensity of the subjective symptoms related to CVI after 6 weeks of treatment. This result is in line with those of a previous study where subjective symptoms measured on a visual analogue scale were reduced only after longer treatment periods (12 weeks).

Adverse events occurred rarely in this study. Thirteen of 71 patients experienced at least one adverse event, 12 of them experienced the onset of action while on placebo treatment, one while on AS 195 (bronchitis, moderate intensity, considered not drug related by the investigator). The patient who died from cardiac arrest had been treated with placebo (never received AS 195 in this trial). All patients assessed the overall tolerability as good or satisfactory. The laboratory parameters did not change during the study.

Discussion

It has been shown in a previous study (WO 01/28363) that red vine leaves extract AS 195 reduces lower leg oedema, calf circumference, and ankle circumference in addition to improving subjective symptoms related to chronic venous insufficiency in patients treated once daily for 12 weeks.[7] The present study was designed to provide additional information on the underlying mechanism of action by investigating microcirculation as a clinically relevant surrogate parameter for CVI related leg problems. This study is the first one in CVI patients aimed to investigate in addition to leg oedema reduction further clinical relevant effects related to the therapy with red vine leaves extract. The reduced venous drainage results in impaired cutaneous microcirculation with trophical disturbances of the skin. If CVI remains untreated this condition may even result venous leg ulcers. Laser Doppler flowmetry, as used in the present study, is a valid and sensitive method to measure objective treatment effects which may be related to the subjectively experienced volume reduction after 3 months of treatment.

The study results fit into the clinical data available for AS 195 and add information on the onset of action. The leg volume as an objective parameter will be reduced in a clinically relevant and statistically significant degree after 6 weeks of treatment. This objective effect has also been reported recently with horse chestnut sees extract (e.g. Diehm C, Trampisch HJ, Lange S, Schmidt C. Comparison of leg compression stocking and oral horse-chestnut seed extract therapy in patients with chronic venous insufficiency. Lancet 1996;347:292-294.) and Butchers Broom (e.g. Vanscheidt W, Jost V, Wolna P, et al. Efficacy and safety of a Butcher's Broom preparation (Ruscus aculeatus L. extract) compared to placebo in patients suffering from chronic venous insufficiency. Drug Res 2002;52(4):243-250.).

In the present study it was shown that the laser Doppler flowmetry parameters, the ankle and calf circumferences and the transcutaneous oxygen pressure were affected as early as after 3 weeks of treatment. In contrast, the subjective symptoms of CVI rated on a visual analogue scale were not significantly different from placebo after 6 weeks of treatment as they were in the previous study. A treatment duration of 12 weeks is mandatory for a relevant reduction of subjective CVI symptoms.

The present results suggest a major role of red vine leaves extract in prevention of CVI progression and the occurrence of trophical skin lesions and may even prevent or delay the transition from clinically not relevant early stages of CVI to CVI Stage I.

CLAIMS:

1. A film coated tablet comprising the following constituents:

(a) at least 50 % by weight of a dried extract of red vine leaves, which is obtainable by extraction of red vine leaves with water, drying and addition of 0.5 to 10 % by weight of silica relating to the total amount of component (a);

(b) up to 50 % by weight of a excipient consisting essentially of

- at least one binder,
- at least one disintegrant,
- at least one filler, and
- a lubricant; and

(c) a tablet film consisting essentially of a film former, a plastiziser, a coating agent and optionally a coloring agent.

2. A film coated tablet according to claim 1 comprising

(a) 50 to 70 % by weight of said dried extract of red vine leaves;

(b) 25 to 49 % by weight of said excipient, and

(c) 1 to 5 % by weight of said tablet film,

based on the total mass of the film coated tablet.

3. A film coated tablet according to claim 1 or 2 comprising

(a) 51 to 59 % by weight of said dried extract of red vine leaves;

(b) 38 to 48 % by weight of said excipient, and

(c) 1 to 3 % by weight of said tablet film,

based on the total mass of the film coated tablet.

4. A film coated tablet according to any of the preceding claims, wherein the aqueous extract of red vine leaves is obtainable by a method comprising the steps of:

(a) collecting red vine leaves at a point of time when the content in flavonoids has reached an optimum;

(b) drying and crushing the leaves;

(c) cutting the leaves to pieces;

(d) extracting the leaves with water at elevated temperatures for 6 to 10 hours;

(e) concentrating and drying the obtained extract, and

(f) and addition of 0.5 to 10 % by weight of flow regulator relating to the final total amount of the resulting extract during the drying process (e).

5. A method according to claim 4 wherein the leaves in step (iv) are extracted with water at temperatures from 60 to 80 °C.

6. A film coated tablet according to any of the preceding claims, wherein the excipient (b) essentially consists of

- 70 to 85 % by weight of at least one binder,
- 0,5 to 12,5 % by weight of at least one disintegrant,
- 5 to 15 % by weight of at least one filler, and
- 1 to 5 % by weight of at least one lubricant,

based on the total mass of the excipients (b).

7. A film coated tablet according to any of the preceding claims, wherein the binder is selected from among powdered cellulose, microcrystalline cellulose, starch, polyvinylpyrrolidone, copolymers of vinylpyrrolidone with other vinyl derivatives, cellulose derivatives and mixtures of these compounds.

8. A film coated tablet according to any of the preceding claims, wherein the disintegrant is selected from among colloidal silica, sodium starch glycolate, crosslinked polyvinylpyrrolidone (crospovidone), croscarmellose sodium salt (sodium salt of cellulose carboxymethyl ether, crosslinked), sodium-carboxymethylcellulose, dried maize starch and mixtures thereof.

9. A film coated tablet according to any of the preceding claims, wherein the filler is an inorganic phosphate or hydrogen phosphate.

10. A film coated tablet according to any of the preceding claims, wherein the filler is selected among from silicon dioxide, talc, stearic acid, sodium stearylfumarate, magnesium stearate and glycerol tribehenate.

11. A film coated tablet according to any of the preceding claims, wherein the tablet film (c) essentially consists of

- 50 to 85 % by weight of at least one film former,
- 5 to 10 % by weight of at least one plasticizer,
- 10 to 20 % by weight of at least one coating agent, and
- 0 to 15 % by weight of at least one colorant.

based on the total mass of the tablet film (c).

12. Process for preparing a film coated tablet according to one of claims 1 to 11 comprising the steps of:

(A) mixing the dried aqueous extract of red vine leaves (a) with the excipients (b), optionally in the presence of a volatile diluent;

(B) optionally screening the mixture obtained;

(C) compressing the mixture with a suitable tablet press; and

(D) coating the resulting tablet with the tablet film (c).

13. Use of a film coated tablet according to one of claims 1 to 11 for preparing a pharmaceutical or dietary composition for the treatment or prevention of the discomfort, disorder and/or disease associated with chronic venous hypervolaemia and venous hypertension.

14. An aqueous extract of red vine leaves is obtainable by a method comprising the steps of:

(a) collecting red vine leaves at a point of time when the content in flavonoids has reached an optimum;
(b) drying and crushing the leaves;
(c) cutting the leaves to pieces;
(d) extracting the leaves with water at elevated temperatures for 6 to 10 hours;
(e) concentrating and drying the obtained extract, and
(f) addition of 0.5 to 10 % by weight of a flow regulator during the relating to the final total amount of the resulting extract during the drying process (e).

15. The aqueous extract according to claim 14, which comprises 2.5 to 7.5 % by weight of colloidal, anhydrous silica.

16. The aqueous extract according to claim 15, which comprises about 4.0 % by weight of colloidal, anhydrous silica.