



EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

12 July 2011
EMA/HMPC/232100/2011
Committee on Herbal Medicinal Products (HMPC)

Assessment report on *Rhodiola rosea* L., rhizoma et radix

Based on Article 16d(1), Article 16f and Article 16h of Directive 2001/83/EC as amended (traditional use)

Draft

Herbal substance(s) (binomial scientific name of the plant, including plant part)	<i>Rhodiola rosea</i> L., rhizoma et radix
Herbal preparation(s)	Dry extract (DER 1.5-5:1), extraction solvent ethanol 67-70% v/v
Pharmaceutical forms	Herbal preparations in solid dosage forms for oral use.

Note: This Assessment Report is published to support the release for public consultation of the draft Community herbal monograph on *Rhodiola rosea* L., rhizoma et radix. It should be noted that this document is a working document, not yet fully edited, and which shall be further developed after the release for consultation of the monograph. Interested parties are welcome to submit comments to the HMPC secretariat, which the Rapporteur and the MLWP will take into consideration but no 'overview of comments received during the public consultation' will be prepared in relation to the comments that will be received on this assessment report. The publication of this draft assessment report has been agreed to facilitate the understanding by Interested Parties of the assessment that has been carried out so far and led to the preparation of the draft monograph.



Table of contents

Table of contents	2
1. Introduction	3
1.1. Description of the herbal substance(s), herbal preparation(s) or combinations thereof .	3
1.2. Information about products on the market in the Member States	5
Regulatory status overview	5
1.3. Search and assessment methodology.....	6
2. Historical data on medicinal use	6
2.1. Information on period of medicinal use in the Community	6
2.2. Information on traditional/current indications and specified substances/preparations ...	6
2.3. Specified strength/posology/route of administration/duration of use for relevant preparations and indications.....	7
3. Non-Clinical Data	7
3.1. Overview of available pharmacological data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof	7
Pharmacological data regarding the herbal preparation:	7
3.2. Overview of available pharmacokinetic data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof	22
3.3. Overview of available toxicological data regarding the herbal substance(s)/herbal preparation(s) and constituents thereof	23
3.4. Overall conclusions on non-clinical data	23
4. Clinical Data	24
4.1. Clinical Pharmacology	24
4.1.1. Overview of pharmacodynamic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents	24
4.1.2. Overview of pharmacokinetic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents	25
4.2. Clinical Efficacy	25
4.2.1. Dose response studies.....	25
4.2.2. Clinical studies (case studies and clinical trials).....	25
4.2.3. Clinical studies in special populations (e.g. elderly and children).....	30
4.3. Overall conclusions on clinical pharmacology and efficacy	30
5. Clinical Safety/Pharmacovigilance	31
5.1. Overview of toxicological/safety data from clinical trials in humans.....	31
5.2. Patient exposure	31
5.3. Adverse events and serious adverse events and deaths	31
5.4. Laboratory findings	31
5.5. Safety in special populations and situations	31
5.6. Overall conclusions on clinical safety	32
6. Overall conclusions	32
Annex	32
List of references.....	32

1. Introduction

1.1. Description of the herbal substance(s), herbal preparation(s) or combinations thereof

- Herbal substance(s)

The herbal substance consists of the dried roots and rhizomes of *Rhodiola rosea* L., *Crassulaceae*. Although the plant part in use is commonly referred to as the root, the anatomy of these parts reveals that these parts are underground stems (rhizomes) with irregular secondary thickening. The plant is native throughout the mountains in the Northern Hemisphere. A main source of commercially available rhizomes is the Altai region in Asia (Panossion *et al.* 2010).

Common names are: Roseroot, Rosenroot, Golden Root, Arctic Root, Rosenwurz. The name refers to the rose-like fragrance of the fresh cut underground organs.

Constituents (according to Panossion *et al.* 2010, Ali *et al.* 2008, Tolonen *et al.* 2003):

Phenylalkanoïds: Phenylethanoids (e.g. salidroside [Syn. rhodioloside]: p-hydroxyphenylethyl-O-β-D-glucopyranoside), phenylpropanoids (e.g. rosin: cinnamyl-O-β-D-glucopyranoside; rosarin: (cinnamyl-(6'-O-α-L-arabinofuranosyl)-O-β-D-glucopyranoside; rosavin: (cinnamyl-(6'-O-α-L-arabinopyranosyl)-O-β-D-glucopyranoside), phenylpropanes (e.g. tyrosol). Only limited data are available regarding the quantitative composition. Hellum *et al.* 2010 report a considerable variability between clones of *Rhodiola rosea* in Norway based on data obtained from ethanolic extracts (primary extraction solvent ethanol 96%). Irrespective of the plant origin (cultivated in Lithuania or naturally occurring in Altai Mountains) Kucinskaite *et al.* (2007) found in aqueous-ethanolic extracts 1.35-1.62 mg/ml of salidroside, while the profile of rosavins differed considerably. Ethanol 70% v/v yields extracts with a low content of salidroside compared to ethanol 40% v/v; in contrast rosavins are more efficiently extracted by ethanol 70% (Kucinskaite 2007).

Essential oil: The dried rhizome contains approximately 0.05% of essential oil. Main components are α-pinene, geraniol, limonene, β-phellandrene, linalool, n-octanol, n-decanol, dodecanol, 1,4-p-menthadien-7-ol and cuminalcohol (Rohloff 2002). Evstatieva *et al.* (2010) found considerable differences in the composition of the essential oil of different origin. In a sample from Bulgaria, myrtenol and geraniol counted for more than 75%; in an Indian sample phenethylalcohol was the main component (56%); in a sample from China, geraniol (57%) and n-octanol were the dominating components.

Monoterpene derivatives: rosiridol, rosiridin, rhodiolosides A-E.

Cyanogenic glycosides: rhodiocyanoside A, lotaustralin

Proanthocyanidines: prodelfinidine-gallate esters

Flavonolignans: rhodiolin

Flavonoids: *Rhodiola*-specific flavonoids like rhodionidin, rhodiogin, rhodalidin, rhodionin, rhodiogidin, rhodalin, rhodiosin; tricetin and kaempferol derivatives.

Phenolic acids: chlorogenic acid, hydroxycinnamic acid

- Herbal preparation(s)

Dry extract (DER 1.5-5:1), extraction solvent ethanol 67-70% v/v

- Combinations of herbal substance(s) and/or herbal preparation(s) including a description of vitamin(s) and/or mineral(s) as ingredients of traditional combination herbal medicinal products assessed, where applicable.

Not applicable.

1.2. Information about products on the market in the Member States

Regulatory status overview

Member State	Regulatory Status				Comments
Austria	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Belgium	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Food supplements
Bulgaria	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Cyprus	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Czech Republic	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Denmark	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Estonia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Finland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
France	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Germany	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Greece	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Hungary	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Iceland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Ireland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Italy	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Latvia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Liechtenstein	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Lithuania	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Luxemburg	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Malta	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
The Netherlands	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Norway	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No product
Poland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Portugal	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Romania	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Slovak Republic	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Slovenia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Spain	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Sweden	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
United Kingdom	<input type="checkbox"/> MA	<input checked="" type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	

MA: Marketing Authorisation

TRAD: Traditional Use Registration

Other TRAD: Other national Traditional systems of registration

Other: If known, it should be specified or otherwise add 'Not Known'

This regulatory overview is not legally binding and does not necessarily reflect the legal status of the products in the MSs concerned.

1.3. Search and assessment methodology

Search term: *Rhodiola rosea*

Databases: Pubmed, Medline and Toxnet.

Libraries: University Vienna, centre of pharmacy; Medical University Vienna, central library.

2. Historical data on medicinal use

2.1. Information on period of medicinal use in the Community

Herbal preparations in medicinal use reported from the member states:

A. Liquid extract, DER 1:1, extraction solvent ethanol 40%

Saratikov (1974) reports that in 1969, the Ministry of Health of the former USSR has recommended to allow the medical utilisation and industrial production of the liquid extract of the *Rhodiola*. In this paper, a herbal tea and a tincture prepared with ethanol 40% (or vodka) are mentioned as traditional herbal preparations. In 1975 *Rhodiola* fluid extract was accepted in the former USSR as a 'Temporary Pharmacopoeial Article' which allowed the large-scale industrial production of a liquid extract (DER 1:1, extraction solvent ethanol 40%). The products were in use also in those parts of the former USSR which now belong to the European Union (Estonia, Latvia, Lithuania), which was confirmed by the National Medicines Agency of Lithuania.

B. Dry extract of root and rhizome, DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water. On the market in Sweden since 1987 (in Panossian et al. 2010, the year 1985 is mentioned). The herbal preparation is named 'SHR-5' in the literature.

C. Dry extract of root and rhizome, DER 1.5-5:1, extraction solvent ethanol 60% m/m. On the market as traditional herbal medicinal product according Dir. 2004/24 in Austria and UK since 2008, in The Netherlands since 2009, in Sweden and Spain since 2010, in Italy since 2011. The registrations were granted based on the traditional medicinal use of the herbal preparations A and B.

Based on this information on the medicinal use of more than 30 years within the European Union, the criteria laid down in Dir. 2004/24 are fulfilled.

The herbal preparations B and C are at the moment in medicinal use in the European Union, while there are no reports on the use of the herbal preparation A. Therefore the following specification for the herbal preparation is proposed for the monograph: Dry extract (DER 1.5-5:1), extraction solvent ethanol 67-70% v/v.

2.2. Information on traditional/current indications and specified substances/preparations

Herbal preparation A (Liquid extract, DER 1:1, extraction solvent ethanol 40%): Asthenia, fatigue, dystonia

Herbal preparation B (Dry extract of root and rhizome, DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water): Traditional herbal medicinal product used as an adaptogen at decreased performance, such as fatigue and weakness

Herbal preparation C (Dry extract of root and rhizome, DER 1.5-5:1, extraction solvent ethanol 60% m/m): Traditional herbal medicinal product for relief of mental and physical symptoms of stress, such as fatigue, weakness, exhaustion, irritability and slight anxiety.

Panosian *et al.* (2010) report traditional use in folk medicine in the indications like headaches, as astringent, to strengthen the head, to enhance the intellect, for restoration of weak nerves; external use for hair growth, relieve of swellings, decrease of back pain, pain in joints. It is said that Lapps chewed on bits of *Rhodiola* on long journeys.

2.3. Specified strength/posology/route of administration/duration of use for relevant preparations and indications

Herbal preparation A (Liquid extract, DER 1:1, extraction solvent ethanol 40%): single dose 5-10 drops, 15-30 minutes before the meal; 2-3 times daily. Duration of use: 10-20 days.

Herbal preparation B (Dry extract of root and rhizome, DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water): One tablet corresponds to ca 500 mg dry root. Dosage: 1 tablet 1-2 times daily. Not recommended to children under 12 years. Oral use.

Herbal preparation C (Dry extract of root and rhizome, DER 1.5-5:1, extraction solvent ethanol 60% m/m): One tablet corresponds to ca 600 mg dried root. Dosage: 1 tablet 2 times daily. Not recommended to children under 12 years. Oral use. Duration of use: 2 weeks.

3. Non-Clinical Data

3.1. Overview of available pharmacological data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

Pharmacological data regarding the herbal preparation:

Oxidative stress, antioxidative effects:

Battistelli *et al.* (2005): An aqueous extract (100 mg herbal substance in 2.4 ml water, 3-fold extraction) was applied *in vitro* to human erythrocytes which were exposed to hypochlorous acid (HOCl)-oxidative stress. The evaluation of the antioxidant capacity of the *Rhodiola* extract has been carried out by means of scanning electron microscopy and of haemolytic behaviour in the presence of increasing doses of the aqueous extract in different experimental environments (co-incubation and subsequent incubations). The results obtained are consistent with a significant protection of the extract in presence of the oxidative agent.

De Sanctis *et al.* (2004): An aqueous extract (12 mg dried herbal substance in 2.4 ml final volume) was tested for its ability to counteract some of the main damages induced by HOCl to human erythrocytes. Ascorbic acid was used as a reference substance because of its physiological HOCl-scavenging ability. The extract was able to significantly protect, in a dose-dependent manner, human red blood cells from glutathione depletion, glyceraldehyde-3-phosphate dehydrogenase inactivation and haemolysis induced by the oxidant. It was demonstrated that the extract acts from the inside of the erythrocyte suggesting a probable involving of cell components.

Calcabrini *et al.* (2010): The authors investigated the protection afforded by an aqueous extract (DER 1:1) to reduce glutathione levels, glyceraldehyde-3-phosphate dehydrogenase activity, and thiobarbituric acid reactive substances levels in cultured human keratinocytes (NCTC 2544) exposed to different oxidative insults (Fe(II)/ascorbate, Fe(II)/H₂O₂, and tert-butyl-hydroperoxide) as well as the influence of the extract on the production of intracellular reactive oxygen species and on the activity of antioxidant enzymes (catalase, superoxide dismutase, glutathione peroxidase, and glutathione reductase). It could be demonstrated that the extract was able to increase in a time- and dose-dependent manner the activity of the trans-plasma membrane oxidoreductase activity as an

indirect evaluation of the intracellular redox status. Keratinocytes of the type NCTC 2544 are able to better counteract several oxidative insults if incubated with a *Rhodiola rosea* aqueous extract.

Chen et al. (2008, only abstract available): This study investigated the antioxidant potential of 3 adaptogen extracts, *Rhodiola rosea* (golden root), *Eleutherococcus senticosus* (Siberian ginseng) and *Embllica officinalis* (Indian gooseberry, Amla). The results of this study showed that *Rhodiola rosea* had the highest potential for singlet oxygen scavenging, hydrogen peroxide scavenging, ferric reducing, ferrous chelating and protein thiol protection than either of the other two extracts. In addition, the polyphenol content in the three adaptogen extracts followed the order: *Rhodiola rosea*, *Eleutherococcus officinalis* and *Eleutherococcus senticosus*. The data suggest that the antioxidant potential of the three adaptogen extracts was proportional to their respective polyphenol content.

Anti-fatigue effects:

Lee et al. (2009): An extract of *Rhodiola rosea* (no details on extraction solvent and DER, approximately 1.4% salidroside, 0.4% rosin, 0.4% rosarin, 1% rosavin) was tested on swimming-induced fatigue in rats. The supplementation (dosages from 5 to 125 mg/kg) in water for 2-4 weeks was evaluated in male Wistar rats with 90-minutes unloaded swimming exercise and 5% body weight loaded swimming up to fatigue. Blood urea nitrogen (BUN), glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT), lactate dehydrogenase (LDH), hepatic glycogen content, the activity of fat metabolism enzymes, sterol regulatory element-binding protein-1 (SREBP-1) and fatty acid synthase (FAS), the tissue oxygen content and ratio of red and white skeletal muscle fibres in rats were measured. The extract significantly increased liver glycogen, SREBP-1, FAS, heat shock protein 70 expression, B-cell lymphoma 2/Bax protein ratio and oxygen content before swimming. *Rhodiola rosea* supplementation significantly increased the swimming time in a dose-dependent manner and reduced swimming-enhanced serum BUN, GOT and GPT levels. The ratio of red and white muscle fibres was not altered after chronic *Rhodiola rosea* extract supplementation. Chronic *Rhodiola rosea* supplementation significantly improved exhaustive swimming-induced fatigue by the increased glycogen content, energy supply of lipogenic enzyme expressions and protective defence mechanisms.

Abidov et al. (2003): The effects of oral treatment with an ethanolic extract (ethanol 40%) from *Rhodiola rosea* (50 mg/kg) and *Rhodiola crenulata* (50 mg/kg) roots on the duration of exhaustive swimming and ATP content in mitochondria of skeletal muscles in rats was investigated. Treatment with *Rhodiola rosea* extract significantly (by 24.6%) prolonged the duration of exhaustive swimming in comparison with control rats and rats treated with *Rhodiola crenulata*. *Rhodiola rosea* extract activated the synthesis or resynthesis of ATP in mitochondria and stimulated reparative energy processes after intense exercise.

Stress-protective effects:

Mattioli & Perfumi (2007): The aim of this work was to determine whether in rats a hydroalcoholic *Rhodiola rosea* extract (no details on the strength of the extraction solvent published) standardised in 3% rosavin and 1% salidroside reverses hypophagia induced by (1) physical stress due to 60 minutes immobilisation; (2) intracerebroventricular injection of corticotrophin-releasing factor (CRF, 0.2 µg/rat), the major mediator of stress responses in mammals; (3) intraperitoneal injection of *Escherichia coli* Lipopolysaccharide (LPS, 100 µg/kg); (4) intraperitoneal administration of fluoxetine (FLU, 8 mg/kg). The effect of the same doses of the plant extract was also tested in freely-feeding and in 20 hours food-deprived rats. The extract was administered acutely by gavage to male Wistar rats 1 hour before the experiments. The results show that at doses of 15 and 20 mg/kg the extract reversed the anorectic effects induced both by immobilisation and by intracerebroventricular CRF injection. Moreover, at the same doses, the extract failed to reduce the anorectic effect induced both

by LPS and FLU, and did not modify food intake in both freely-feeding and food-deprived rats. The authors interpret the results that *Rhodiola* extract is able selectively to attenuate stress-induced anorexia, providing functional evidence of claimed adaptogen and anti-stress properties of *Rhodiola rosea* L.

Zhu et al. (2003): Noise is one of the factors that induce critical stress in animals. The contents of glycogen, lactic acid and cholesterol in the liver of noise-stressed rats were analysed in order to investigate the alleviation of noise-stress-induced physiological damages by a decoction of the underground organs of *Rhodiola rosea*. More than 95 dB noise ranging from 2 to 4 kHz reduced the contents of these compounds in the liver of rats not injected with the extract, but did not change the contents in the liver of rats treated with the extract. The results indicate that *Rhodiola* extract improved the ability for rats to resist noise stress.

Boon-Niermeijer et al. (2000): The authors examined whether *Rhodiola* extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) is able to exert a protective action against stress-induced death of embryos of the pond snail *Lymnaea stagnalis*, and whether a possible protective action by the extract can be explained by the induction of heat shock proteins. The extract was applied for a period of 20 hours to 3-day old larvae of the pond snail. Subsequently they were exposed to a high and toxic dose of different environmental stressors (heat shock: 43° C for 4 minutes, an oxidative stress condition (superoxide radicals induced by menadione 600 µM for 2 hours) and heavy metal-induced stress (copper 150 µM for 1 hour or cadmium 20 µM during 1 hour). The *Rhodiola* extract exerted a strong protective action against a lethal heat shock. It also significantly protected against the negative effect of superoxide radicals as induced by menadione. With respect to the protective action against exposure to heavy metals a small but significant protection was observed against intoxication with copper or cadmium. Although the degree to which resistance is enhanced appears to depend on the type of stressor applied, the results confirm in the opinion of the authors the definition of phyto-adaptogens as being universal enhancers of non-specific resistance against different kinds of stress conditions. The extract did not induce the synthesis of any of the heat shock proteins, nor did it modulate the normal heat shock induced synthesis of these stress proteins. Therefore it is concluded that it is unlikely that heat shock proteins play a major role in obtaining an enhanced state of resistance provided by *Rhodiola* extracts.

Mattioli et al. (2009): The aim of this study was to determine whether chronic treatment with a hydroalcoholic *Rhodiola rosea* extract (no details on the strength of the extraction solvent published) containing 3% rosavin and 1% salidroside can prevent alterations induced in female rats following 6 weeks of a chronic mild stress (CMS) procedure. This was analysed through the behavioural and physiological parameters of consumption of 1% sucrose solution, locomotor and exploratory activities, body weight gain and oestrous cycle length. After the first 3 weeks of stress, the extract was administered daily by gavage at doses of 10, 15 and 20 mg/kg for the remaining 3 weeks. In addition, fluoxetine (10 mg/kg orally), which has been shown to reverse CMS-induced disruptions, was used as the reference treatment. Rats subjected to the CMS procedure demonstrated decreased sucrose intake, reduced moving behaviour, minimised weight gain and dysregulation of their oestrous cycle. Treatment with the *Rhodiola* extract completely reverted all of these changes. The effects of the extract were comparable to those of fluoxetine. The authors are of the opinion that chronic administration of *Rhodiola* extract results in potent inhibition of the behavioural and physiological changes induced by chronic exposure to mild stressors.

Chen et al. (2008a, only abstract available): Aim of the study was to explore the effects of *Rhodiola rosea* on the body weight and the intake of sucrose and water in depressive rats induced by chronic mild stress. A total of 70 male SD rats were divided into seven groups, including normal control group (treated with 0.5% sodium carboxymethylcellulose), untreated group, negative control group (treated with 0.5% sodium carboxymethylcellulose), positive control group (treated with fluoxetine), low-,

medium- and high-dose *Rhodiola rosea* group (treated with 1.5, 3, 6 g/kg *Rhodiola rosea* respectively, no details on the type of herbal preparation available). Except for rats in normal control group, the other sixty rats endured chronic stress for 4 weeks to establish the depression model. After that, rats were administered *Rhodiola rosea* for 3 weeks. After the termination of the stress regime, compared with the normal control group, the body weight and 1% sucrose intake in depressive rats were decreased. After 3-week *Rhodiola rosea* treatment, the body weight and 1% sucrose intake increased in rats of the low-dose *Rhodiola rosea* group and recovered to the level of the normal control group.

Cifani et al. (2010): Stress is a key determinant of binge eating (BE). BE for highly palatable food (HPF) was evoked in female rats by three 8-day cycles of food restriction/re-feeding (for 4 days 66% of the usual chow intake; for 4 days food ad libitum) and acute stress on the test day (day 25). *Rhodiola rosea* dry extract (3% rosavin, 3.12% salidroside no details on extraction solvent) or its active principles were given by gavage 1 hour before access to HPF. Only rats exposed to both food restrictions and stress exhibited BE in the first 15-60min after the stressful procedure. *Rhodiola rosea* extract 10mg/kg significantly reduced and 20 mg/kg abolished the BE episode. *Rhodiola rosea* extract 20mg/kg abolished also stress-induced increase in serum corticosterone levels. The *Rhodiola rosea* active principle salidroside, but not rosavin, at doses present in the extract, dose-dependently reduced or abolished BE for the period in which it was elicited. In conclusion results indicate that *Rhodiola rosea* extracts may have therapeutic properties in bingeing-related eating disorders and that salidroside is the active principle responsible for this effect.

Effects on nervous system

Qu et al. (2009): The authors investigated the pre-treatment effects of *Rhodiola rosea* extract (no details on extraction solvent published) on cognitive dysfunction, oxidative stress in hippocampus and hippocampal neuron injury in a rat model of Alzheimer's disease. Male Sprague-Dawley rats were pre-treated with *Rhodiola rosea* extract at doses of 1.5, 3, and 6 g/kg for 3 weeks by gavage twice daily, followed by bilateral intracerebroventricular injection with streptozotocin (1.5 mg/kg) on day 1 and 3. Behavioural alterations were monitored after 2 weeks from the lesion using Morris water maze task. Three weeks after the lesion, the rats were sacrificed for measuring the malondialdehyde (MDA), glutathione reductase (GR) and reduced glutathione (GSH) levels in hippocampus and histopathology of hippocampal neurons. The MDA level was significantly increased while the GR and GSH levels were significantly decreased with striking impairments in spatial learning and memory and severe damage to hippocampal neurons in the model rat induced by intracerebroventricular injection of streptozotocin. These abnormalities were significantly improved by pre-treatment with *Rhodiola rosea* extract (3 g/kg).

Perfumi & Mattioli (2007): the purpose of the present study was to re-investigate the effects produced by a single oral administration of a *Rhodiola rosea* hydroalcoholic extract (containing 3% rosavin and 1% salidroside, no information on the extraction solvent published) on the central nervous system in mice. The extract was tested on antidepressant, adaptogenic, anxiolytic, nociceptive and locomotor activities at doses of 10, 15 and 20 mg/kg, using predictive behavioural tests and animal models. The results show that this *Rhodiola rosea* extract significantly, but not dose-dependently, induced antidepressant-like, adaptogenic, anxiolytic-like and stimulating effects in mice. This study thus provides evidence of the efficacy of *Rhodiola rosea* extracts after a single administration.

Van Diermen et al. (2009): In order to investigate the influence of *Rhodiola rosea* L. roots on mood disorders, three extracts were tested against monoamine oxidases (MAOs A and B) in a microtitre plate bioassay. Twelve compounds were then isolated by bioassay-guided fractionation using chromatographic methods. The methanol and water extracts exhibited respectively inhibitions of 92.5% and 84.3% on MAO A and 81.8% and 88.9% on MAO B, at a concentration of 100 µg/ml. The

most active compound (rosiridin) presented an inhibition over 80% on MAO B at a concentration of 10^{-5} M ($pIC_{50}=5.38\pm 0.05$).

Chen *et al.* (2009): The purpose of this study was to investigate the effects of *Rhodiola rosea* extract (ethanol 70%) on the serotonin (5-HT) level, cell proliferation and quantity of neurons in the cerebral hippocampus of depressive rats induced by Chronic Mild Stress. After 3 weeks of oral administration of 1.5 g up to 6 g/kg per day 5-HT level had recovered to normal status. In the low dose group the extract induced neural stem cell proliferation in the hippocampus to return to normal level, repairing the injured neurons in the hippocampus.

Qin *et al.* (2008, only abstract available): The purpose of this study was to investigate the effects of *Rhodiola rosea* (no details on the type of the herbal preparation available) on the level of 5-HT, cell proliferation and differentiation, and number of neurons in the cerebral hippocampus of rats with depression induced by chronic mild stress. After 3 weeks of administration of the preparation all measured parameters like 5-HT content, number of bromodeoxyuridine positive cells, percentage of bromodeoxyuridine and beta-tubulin III double labelled cells and number of neurons in the cerebral hippocampus returned to normal level.

Panossian *et al.* (2008): The antidepressant-like activity of an extract of the roots of *Rhodiola rosea* (DER 2.5-5:1, no information on the extraction solvent, containing 2.7% rhodioloside, 6% rosavin, 0.8% tyrosol), its combination with piperine, isolated constituents from *Rhodiola*, such as rhodioloside, rosavin, rosin, rosarin, tyrosol, cinnamic alcohol, cinnamaldehyde and cinnamic acid has been assessed in laboratory animals through application of the Porsolt behavioural despair assay. The orally administered extract reduced in dosages of 10, 20 and 50 mg/kg dose-dependently the immobility time more strongly compared to Hypericum extract LI160 (20 mg/kg), amitriptyline (3 mg/kg) or imipramine (30 mg/kg). The combination with piperine was also active in a similar degree, but no dose-dependency was detectable. Rhodioloside and tyrosol contributed considerably to this effect. A fixed combination of rhodioloside, rosavin, rosarin and rosin was more active than any of the individual components alone.

Mattioli & Perfumi (2011): The aim of the present study was to investigate the effects of a *Rhodiola rosea* hydroalcoholic extract (no details on extraction solvent published, 3% total rosavins, 1% salidroside) on the prevention of the development of nicotine dependence and for the reduction of abstinence suffering following nicotine cessation in mice. Dependence was induced in mice by subcutaneous injections of nicotine (2 mg/kg, 4 times daily) for 8 days. Spontaneous abstinence syndrome was evaluated 20 hours after the last nicotine administration, by analysis of withdrawal signs, as affective (anxiety-like behaviour) and physical (somatic signs and locomotor activity). The extract was administered orally during nicotine treatment (10, 15 and 20 mg/kg) or during nicotine withdrawal (20 mg/kg). Both affective and somatic signs (head shaking, paw tremors, body tremors, ptosis, jumping, piloerection and chewing) induced by nicotine withdrawal were abolished by administration of the extract in a dose-dependent fashion, during both nicotine exposure and nicotine cessation.

Mattioli & Perfumi (2011a): The same extract, as in Mattioli & Perfumi (2011) was investigated for effects on acquisition and expression of morphine tolerance and dependence in mice. Therefore animals were injected with repeated administration of morphine (10mg/kg, subcutaneous) twice daily for 5 or 6 days, in order to make them tolerant or dependent. The extract (0, 10, 15 and 20mg/kg) was administered by the intragastric route 60 minutes prior to each morphine injection (for acquisition) or prior the last injection of morphine or naloxone on test day (for tolerance or dependence expression, respectively). Morphine tolerance was evaluated by testing its analgesic effect in the tail flick test at the 1st and 5th days. Morphine dependence was evaluated by counting the number of withdrawal signs (jumping, rearing, forepaw tremor, teeth chatter) after naloxone injection (5mg/kg; intraperitoneal) on

the test day (day 6). The extract significantly reduced the expression of morphine tolerance, while it was ineffective in modulating its acquisition. Conversely, *Rhodiola rosea* L. extract significantly and dose-dependently attenuated both development and expression of morphine dependence after chronic or acute administration.

Life-span increasing effects:

Schriner *et al.* (2009): The extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) could extend both mean (24% in both sexes) and maximum (16% in males and 31% in females) life span in *Drosophila melanogaster* when compared to controls. It lowered mitochondrial superoxide levels and afforded elevated protection against the superoxide generator paraquat in both sexes. The extract did not alter the activities of the major antioxidant enzymes, the superoxide dismutases or catalase, nor did it afford protection against H₂O₂ or soluble iron.

Schriner *et al.* (2009a): A *Rhodiola rosea* extract (no details published) could protect cultured cells against ultraviolet light, paraquat, and H₂O₂. However, it did not alter the levels of the major antioxidant defences nor did it markedly activate the antioxidant response element or modulate heme-oxygenase-1 expression levels at relevant concentrations. In addition, *Rhodiola rosea* extract was not able to significantly degrade H₂O₂ *in vitro*. These results suggest that in human cultured cells *Rhodiola rosea* does not act as an antioxidant and that its mode of action cannot be sufficiently explained through a pro-oxidant hormetic mechanism.

Jafari *et al.* (2007): Using the fruit fly, *Drosophila melanogaster*, the effects of *Rhodiola* extract (no details published) on life-span was investigated. *Rhodiola* supplied every other day at 30 mg/ml significantly increased the lifespan of *Drosophila melanogaster*. When comparing the distribution of deaths between *Rhodiola*-supplemented and control flies, *Rhodiola*-fed flies exhibited decelerated aging. Although the observed extension in lifespan was associated with statistically insignificant reductions in fecundity, correcting for a possible dietary restriction effect still did not eliminate the difference between supplemented and control flies, nor does the effect of *Rhodiola* depend on dietary manipulation, strongly suggesting that *Rhodiola* is not a mere dietary restriction mimetic.

Wiegant *et al.* (2009): Extracts of *Rhodiola* (SHR-5, DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) and of *Eleutherococcus* increased the mean lifespan of the nematode *C. elegans* in a dose-dependent way. In at least four independent experiments, 250 µg/ml *Eleutherococcus* and 10-25 µg/ml *Rhodiola* (SHR-5) significantly increased life span between 10 and 20% (P < 0.001), increased the maximum lifespan with 2-3 days and postponed the moment when the first individuals in a population die, suggesting a modulation of the ageing process. With higher concentrations, less effect was observed, whereas at the highest concentrations tested (2500 µg/ml *Eleutherococcus* and 250 µg/ml *Rhodiola*) a lifespan shortening effect was observed of 15-25% (P < 0.001). Both extracts were also able to increase stress resistance in *C. elegans* against a relatively short heat shock (35° C during 3 hours) as well as chronic heat treatment at 26° C. An increase against chronic oxidative stress conditions was observed in mev-1 mutants, and during exposure of the wild type nematode to paraquat (10 mM) or UV stress, be it less efficiently. Both extracts induced the translocation of the DAF-16 transcription factor from the cytoplasm into the nucleus, suggesting a reprogramming of transcriptional activities favouring the synthesis of proteins involved in stress resistance (such as the chaperone HSP-16) and longevity.

Cardio protective effects, effects on the vascular system:

Maslov *et al.* (2009, only abstract available): A course of treatment (16 mg/kg orally during 5 days) by *Aralia mandshurica* or *Rhodiola rosea* extracts (no details available) reduced the incidence of ischemic and reperfusion ventricular arrhythmias during 10-min ischemia and 10-min reperfusion in rats.

Chronic treatment by *Aralia*, *Rhodiola*, and *Eleutherococcus* elevated the ventricular fibrillation threshold in rats with postinfarction cardiosclerosis.

Li *et al.* (2005, only abstract available): On the basis of successful establishment of myocardial infarction rat model, the experimental animals were divided into the model group, the *Rhodiola* group (no details on the type of the herbal preparation and on posology available), the positive control group and the sham-operated group, they were sacrificed after 6 weeks feeding. The expressions of Flt-1 and angiotensin receptor (Tie-2) in myocardial tissue were significantly increased in the *Rhodiola* treated group after treatment, showing significant difference as compared with those in the positive control group and the model group ($P < 0.05$). The expression of the growth factor KDR in myocardium after *Rhodiola* intervention was higher than that in the sham-operated and nonintervened group ($P < 0.05$), but insignificantly different to that in the positive control group and model group. Therefore it was concluded that *Rhodiola* could improve angiogenesis to ameliorate myocardial ischemia by regulating the expression of Flt-1 and Tie-2 in ischemic myocardium.

Shen *et al.* (2008, only abstract available): Thirty male New Zealand rabbits were randomly divided into three groups equally, i. e. the control group (A) fed with common diet and treated with distilled water, the high fat diet group (B) and the *Rhodiola* group (C, no details on the type of the herbal preparation and on dosage available) fed with diet containing 1.5% cholesterol and treated respectively with distilled water and *Rhodiola* (1 ml/kg per day), all the treatments were administered via gastrogavage once daily for 9 successive weeks. Levels of blood lipids in various groups was determined and compared at the end of the experiment. Meanwhile, the tissue sample of aorta was taken for observation through HE and Sudan red staining, for detecting the CD34 positive response intensity by immunohistochemical staining and the vascular endothelial cell growth factor VEGF expression by Real-time fluorescent quantitative PCR and Western blot. The determination of blood lipids showed that in Group C, TC was 42.01 +/- 1.99 mmol/l, TG 4.83 +/- 0.75 mmol/l and LDL-C 38.40 +/- 0.74 mmol/l, all lower than those in Group B (70.74 +/- 2.66 mmol/l, 8.75 +/- 0.78 mmol/l and 51.05 +/- 0.34 mmol/l, respectively), showing statistical difference between groups ($P < 0.05$). The intima/media tunica thickness ratio and the CD34 positive area of plaque in Group C were all lower than those in Group B (0.35 +/- 0.03 vs 0.43 +/- 0.03 and 29.12 +/- 2.56% vs 39.28 +/- 3.48%, $P < 0.05$). Besides, the VEGF expression in atherosclerotic plaque was also lower in Group C than that in Group B.

Maslov & Lishmanov (2007, only abstract available): The chronic administration of *Rhodiola rosea* extract (no details available) in a single daily dose of 1 ml/kg (p.o.) during 8 days increased the resistance of myocardium with respect to the cardiotoxic action of isoproterenol and the arrhythmogenic action of epinephrine in rats. Pre-treatment with the extract prevented the stressor cardiac damages, as measured by ^{99m}Tc-pyrophosphate accumulation in the heart. The cardioprotective action of RRE was highest after 5-day administration. The antiarrhythmic effect of the adaptogen was at a maximum after 8-day administration. It was found that p-tyrosol also exhibited antiarrhythmic and cardioprotective properties. Pre-treatment with the extract decreased the infarction size/risk area ratio during the coronary artery occlusion and reperfusion *in vivo*. The chronic administration of the extract increased the tolerance of the isolated perfused rat heart to the pathogenic action of global ischemia and reperfusion. Pre-treatment not only prevented the occurrence of arrhythmias, but also abolished cardiac electrical instability in rats with postinfarction cardiac sclerosis. It has been found that the chronic administration of the extract (1 ml/kg, p.o., over 8 days) increased the level beta-endorphin in rat blood plasma and the content of leu-enkephalin in myocardial tissue. Naloxone (2 mg/kg) abolished the cardioprotective and antiarrhythmic effect of the extract.

Effects on the blood system:

Provalova *et al.* (2002, only abstract available): Extracts of *Rhodiola rosea* did not modulate granulocytopoiesis (no details available on type of preparation and methods).

Anti-inflammatory effects:

Pooja *et al.* (2009): The present study was undertaken to evaluate the anti-inflammatory effects of a liquid extract of *Rhodiola rosea* underground organs (extraction solvent ethanol 40%). The anti-inflammatory activity was determined through carrageenan-induced paw oedema, formaldehyde-induced arthritis and nystatin-induced paw oedema in rat model. The liquid extract exhibited inhibitory effect against acute and subacute inflammation at a dose of 250 mg/kg body weight. Inhibition of nystatin-induced oedema was also observed in a dose-dependent manner. The extract showed varying inhibitory activities against enzymes related to inflammation depending on the concentrations. A potent inhibition was observed against Cox-2 and phospholipase A₂. Inhibition of nystatin induced oedema and phospholipase A₂ suggested that membrane stabilisation could be the most probable mechanism of action of the extract in anti-inflammation.

Effects on metabolism:

Kim *et al.* (2006): This study was designed to examine the effects of *Cinnamomum cassia* and *Rhodiola rosea* extracts (dry extract DER 10:1, extraction solvent ethanol 85%) on blood glucose, lipid peroxidation, the level of reduced glutathione and its related enzymes (glutathione reductase, glutathione S-transferase), and the activity of the antioxidant enzymes (catalase, superoxide dismutase and glutathione peroxidase) in the liver of db/db mice. Diabetic C57BL/Ks db/db mice were used as experimental models. Mice were divided into control (n=10), *Cinnamomi cassiae* (200 mg/kg/day, n=10), and *Rhodiola rosea* (200 mg/kg/day, n=10) treated groups for 12 weeks of oral treatment. Both extracts significantly decreased blood glucose, increased levels of reduced glutathione and the activities of glutathione reductase, glutathione S-transferase, glutathione peroxidase, catalase and superoxide dismutase in the liver. Extract treatment also significantly decreased lipid peroxidation.

Effects on the endocrinous system:

Kwon *et al.* (2006): Two species of the genus *Rhodiola* (*Rhodiola crenulata* and *Rhodiola rosea*) were investigated for the inhibition of α -amylase, α -glucosidase and angiotensin converting enzyme (ACE) inhibitory activity. Water extracts of *Rhodiola crenulata* had the highest alpha-amylase inhibitory activity (IC₅₀, 98.1 μ g total phenolic/ml) followed by ethanol extract of *Rhodiola crenulata* (IC₅₀, 120.9 μ g total phenolic/ml) and ethanol extract (ethanol 12%) of *Rhodiola rosea* (IC₅₀, 173.4 μ g total phenolic/ml). Ethanol extract of *Rhodiola rosea* (IC₅₀, 44.7 μ g total phenolic/ml), water extract (macerate DER 1:10) of *Rhodiola rosea* (IC₅₀, 52.3 μ g total phenolic/ml), water extract of *Rhodiola crenulata* (IC₅₀, 60.3 μ g total phenolic/ml) and ethanol extract of *Rhodiola crenulata* (IC₅₀, 60.2 μ g total phenolic/ml) also showed significant α -glucosidase inhibitory activity. The α -glucosidase inhibitory activity of the extracts was compared to standard tyrosol, which was significantly detected in the extracts using HPLC. Tyrosol had strong α -glucosidase inhibitory activity (IC₅₀, 70.8 μ g total phenolic/ml) but did not have any inhibitory effect on the α -amylase activity. Results suggested that α -glucosidase inhibitory activities of both *Rhodiola* extracts correlated to the phenolic content, antioxidant activity and phenolic profile of the extracts. The ability of the above *Rhodiola* extracts to inhibit rabbit lung angiotensin I-converting enzyme (ACE) was investigated. The ethanol extracts of *Rhodiola rosea* had the highest ACE inhibitory activity (38.5%) followed by water extract of *Rhodiola rosea* (36.2%) and *Rhodiola crenulata* (15.4%).

Ip et al. (2001): Chronic hypoxia significantly increased the mRNA expression for angiotensinogen II receptor subtypes AT1b and AT2 in the pancreatic renin-angiotensin system. The activation of the renin-angiotensin system may play an important role in cellular pathophysiological processes. Angiotensin II enhances the formation of reactive oxygen species via the activation of xanthine oxidase or NAD(P)H oxidase. The reactive oxygen species can cause oxidative damage in the pancreas and other tissues either directly or indirectly via the formation of other radicals such as reactive nitrogen species. *Rhodiola* therapy may protect hypoxia-induced pancreatic injury in two ways. It prevents hypoxia-induced biological changes by increasing intracellular oxygen diffusion and efficiency of oxygen utilisation. Alternatively, it reduces hypoxia-induced oxidative damage by its antioxidant activities. Additional experimental data are required to fully elucidate the mode of action of this herbal drug.

Anti-cancer effects:

Majewska et al. (2006): It has been found that the extract of *Rhodiola rosea* rhizomes (extraction solvent ethanol 96%) inhibits division of HL-60 cells, which is preceded by an accumulation of cells at the prophase stage. This leads to induction of apoptosis and necrosis in HL-60 cells, and to marked reduction of their survival. The cells enter apoptosis from phase G2/M of the cell cycle. After treatment with the extract, no chromosome aberrations or micronuclei were observed, which indicates the mild action of the extract.

Pharmacological data regarding isolated constituents:

Salidroside and other phenylalkaloids:

Oxidative stress, antioxidative effects:

Huang et al. (2009, only abstract available): Wistar rats received 5, 25, 125 mg of a *Rhodiola* extract per day for 4 weeks. Salidroside, rosin, rosavin and rosarin scavenged O₂(-)*, H₂O₂, and HOCl activity in a dose-dependent manner. The 90 minutes swimming exercise increased the O₂(-)* production in the order: liver > skeletal muscle > blood, indicating that liver is the most sensitive target organ. The level of plasma malonedialdehyde, a lipid peroxidation product, was also increased after exercise. Treatment of 4 weeks of *Rhodiola rosea* extracts significantly inhibited swimming exercise-enhanced O₂(-)* production in the blood, liver and skeletal muscle and plasma malonedialdehyde concentration. The expression of Mn-superoxide dismutase Cu/Zn-superoxide dismutase and catalase in livers were all enhanced after 4 weeks of *Rhodiola rosea* supplementation especially at the dose of 125 mg/day/rat. Treatment of *Rhodiola rosea* extracts for 4 weeks significantly increased swimming performance.

Li et al. (2011): In the present study the protective activity of salidroside against 1-methyl-4-phenylpyridinium (MPP(+)) -induced apoptosis in PC12 cells was investigated. The incubation of PC12 cells with salidroside prior to MPP(+) exposure significantly reduced cell apoptosis and attenuated collapse of the mitochondrial membrane potential (MMP). Furthermore, salidroside inhibited the MPP(+)-induced nitric oxide (NO) increase and overexpression of nNOS and iNOS, and suppressed accumulation of reactive oxygen species (ROS) and intracellular free Ca(2+). The results show that the protective effects of salidroside on PC12 cells are mediated, at least in part, by inhibition of the NO pathway.

Yu et al. (2010): The aim of this study was to investigate the effects of salidroside on hydrogen peroxide (H₂O₂)-induced cell apoptosis in nerve growth factor (NGF)-differentiated PC12 cells and the possible involvement of the extracellular signal-related protein kinase 1/2 (ERK1/2) signaling pathway. MTT assay, Hoechst 33342 staining, and TdT-mediated dUTP-biotin nick end labelling assay collectively showed that the pre-treatment with salidroside alleviated, in a dose-dependent manner, cell viability loss and apoptotic cell death induced by H₂O₂ stimulation in cultured NGF-differentiated PC12 cells.

According to Western blot analysis, pre-treatment with salidroside transiently caused the activation of the ERK1/2 pathway; a selective inhibitor of the mitogen-activated protein kinase (MAPKK, MEK) blocked the salidroside-activated ERK pathway and thus attenuated the influences of salidroside on H₂O₂-induced increase in the level of cleaved caspase-3, a chief executant of apoptosis cascades. Morphological analysis further indicated that in the presence of the MEK inhibitor, the neuroprotective effect of salidroside against H₂O₂-evoked cell apoptosis was significantly abrogated. Taken together, the results suggest that the neuroprotective effects of salidroside might be modulated by the ERK signalling pathway, especially at the level or upstream of the caspase-3 activation.

Tan *et al.* (2009): The protective effects of salidroside on endothelial cells apoptosis induced by the hypoxia mimicking agent cobalt chloride was investigated. After challenge with cobalt chloride for 24 hours, loss of cell viability and excessive apoptotic cell death were observed in EA.hy926 endothelial cells, and the level of intracellular ROS increased concentration-dependently. However, the endothelial cell apoptosis and excessive ROS generation were attenuated markedly by salidroside pre-treatment. In addition, salidroside inhibited activation of caspase-3 and cleavage of poly(ADP-ribose) polymerase (PARP) induced by cobalt chloride, decreased expression of Bax and rescued the balance of pro- and anti-apoptotic proteins. These findings suggest that salidroside protects endothelial cells from cobalt chloride-induced apoptosis as an antioxidant and by regulating, the Bcl-2 family.

Chen *et al.* (2009a): This study investigated whether salidroside was able to extend its unique neuroprotection to primary cultured rat hippocampal neurons against hydrogen peroxide (H₂O₂)-induced cell damage. Cell viability tests and cell apoptosis assays confirmed that salidroside pre-treatment attenuated H₂O₂-stimulated apoptotic cell death in primary culture of hippocampal neurons in a concentration-dependent manner. The measurements of caspase-3 activity, NO production, and NO synthase (NOS) activity suggest that the protection of salidroside, shown in this study, might be mediated by inhibiting caspase-3 activity, and antagonising NO production and NOS activity during H₂O₂ stimulation.

Mao *et al.* (2010): Salidroside considerably reversed senescence-like phenotypes in the oxidant challenged model, including alterations of morphology, cell cycle, SA-β-gal staining, DNA damage, as well as related molecules expression such as p53, p21 and p16. The protection occurred in a dose-dependent manner, with 5μM offering best efficacy. The proposed antioxidant property of the compound was confirmed in this cellular system, and thus at least partially accounted for the protection of the compound against premature senescence. Similar protection of salidroside against replicative senescence was observed as well. The regulation of senescence-related molecules by salidroside involved ROS-irrelevant mechanisms in both models.

Effects on the endocrinous system:

Wang *et al.* (2009, only abstract available): The effects of salidroside on the function and ultramicro-pathological change of the hypothalamic-pituitary-gonadal (HPG) axis of male rats in experimental navigation and intensive exercise was investigated. Six-week SD rats were randomised into three groups: non-stress control (NC, n = 10), training control (TC, n = 12) and salidroside treatment (ST, n = 12) group. Blood samples were collected from the NC rats that did not receive any stimulus after a 7-day intragastric administration of saline. The TC rats underwent a 10-day running training with increasing load on the treadmill followed by a 7-day intragastric administration of saline. The ST rats were subjected to the same process of running training as the TC group and received intragastric administration of salidroside. Then blood samples were immediately obtained. The serum testosterone level was significantly lower in the TC than in the NC group, but showed no significant difference between the ST and NC groups. HE staining revealed no significant difference in testis histopathology among the three groups. Ultramicro-pathology showed that the secretory granules of the pituitary cells

were significantly reduced in the TC rats compared with the NC ones; the number of the granules significantly increased in the ST group compared with the TC rats; mitochondrial swelling, increase of electron density and decrease/disappearance of mitochondrial cristae were observed in the Leydig cells of the TC rats. No significant differences were found in the testicular cells between the ST and NC groups. It is concluded that negative psychological stress and intensive exercise can significantly suppress the function of the HPG axis in rats. Salidroside therapy may have protective effects on the HPG axis.

Neuroprotective effects:

Chen *et al.* (2008b): This study aimed to evaluate the inhibitory effects of salidroside on glutamate-induced cell death in a primary culture of rat hippocampal neurons as compared to brain-derived neurotrophic factor (BDNF) as positive control. MTT and LDH assays, together with Hoechst 33342 staining, terminal deoxynucleotidyl transferase dUTP-mediated nicked end labelling (TUNEL) assay and flow cytometric analysis using annexin-V and propidium (PI) label, indicated that salidroside pre-treatment attenuated glutamate-induced apoptotic cell death in primary cultured hippocampal neurons, showing a dose-dependent pattern. Furthermore, caspase-3 activity assay and calcium measurements with Fluo 4-AM, respectively, revealed that salidroside pre-treatment antagonised activation of caspase-3 and elevation of intracellular calcium level, both of which were induced by glutamate stimulation.

Cao *et al.* (2006): Salidroside could protect PC12 cell against injuries caused by exposure of PC12 cells to 2 mmol/l glutamate for 15 minutes followed by incubation with serum-free medium for 24 hours, which resembled the excitotoxin *in vivo* system. Furthermore, salidroside could decrease the cytosolic free calcium concentration $[Ca^{2+}]_i$ of PC12 cells in Mg^{2+} -free Hanks' solution and D-Hanks' solution but there was no effect on basal $[Ca^{2+}]_i$ in Hanks' solution. The studies also indicated that salidroside inhibited the increases of $[Ca^{2+}]_i$ induced by KCl and glutamate. In conclusion, salidroside may protect PC12 cells against glutamate excitotoxic damage through suppressing the excessive entry of Ca^{2+} and the release of the calcium stores.

Zhang *et al.* (2007): In this paper, the neuroprotective effects of salidroside on hydrogen peroxide H_2O_2 -induced apoptosis in SH-SY5Y cells were investigated. Pre-treatment with salidroside markedly attenuated H_2O_2 -induced cell viability loss and apoptotic cell death in a dose-dependent manner. The mechanisms by which salidroside protected neuron cells from oxidative stress included the induction of several antioxidant enzymes, thioredoxin, heme oxygenase-1, and peroxiredoxin-I; the down regulation of pro-apoptotic gene Bax and the up regulation of anti-apoptotic genes Bcl-2 and Bcl-X(L). Furthermore, salidroside dose-dependently restored H_2O_2 -induced loss of mitochondrial membrane potential as well as the elevation of intracellular calcium level. These results suggest that salidroside has protective effects against oxidative stress-induced cell apoptosis, which might be a potential therapeutic agent for treating or preventing neurodegenerative diseases implicated with oxidative stress.

Zhang *et al.* (2010): Beta-amyloid ($A\beta$) peptide, the hallmark of Alzheimer's disease (AD), invokes a cascade of oxidative damages to neurons and eventually leads to neuronal death. In this study, salidroside was investigated to assess its protective effects and the underlying mechanisms against $A\beta$ -induced oxidative stress in SH-SY5Y human neuroblastoma cells. $A\beta_{25-35}$ -induced neuronal toxicity was characterised by the decrease of cell viability, the release of LDH, morphological alterations, neuronal DNA condensation, and the cleavage of poly(ADP-ribose) PARP by activated caspase-3. Pre-treatment with salidroside markedly attenuated $A\beta_{25-35}$ -induced loss of cell viability and apoptosis in a dose-dependent manner. The mechanisms of salidroside protection of neurons from oxidative stress included the induction of antioxidant enzymes, thioredoxin, heme oxygenase-1 (HO-1), and

peroxiredoxin-I; the down regulation of pro-apoptotic protein Bax and the upregulation of anti-apoptotic protein Bcl-X(L). Furthermore, salidroside dose-dependently restored A β ₂₅₋₃₅-induced loss of MMP as well as suppressed the elevation of intracellular ROS level. It was also observed that A β ₍₂₅₋₃₅₎ stimulated the phosphorylation of mitogen-activated protein (MAP) kinases, including c-Jun NH(2)-terminal kinase (JNK) and p38 MAP kinase, but not extracellular signal-regulated kinase1/2 (ERK1/2). Salidroside inhibited A β ₂₅₋₃₅-induced phosphorylation of JNK and p38 MAP kinase, but not ERK1/2. In the opinion of the authors these results suggest that salidroside has protective effects against A β ₂₅₋₃₅-induced oxidative stress, which might be a potential therapeutic agent for treating or preventing neurodegenerative diseases.

Yu et al. (2008): The hypoglycaemia and serum limitation-induced cell death in cultured PC12 cells represents a useful *in vitro* model for the study of brain ischemia and neurodegenerative disorders. In this study, MTT assay, Hoechst 33342 staining, and flow cytometry with annexin V/PI staining collectively showed that pre-treatment with salidroside attenuated, in a dose-dependent manner, cell viability loss, and apoptotic cell death in cultured PC12 cells induced by hypoglycaemia and serum limitation. RT-PCR, Western blot analysis, and enzymatic colorimetric assay indicated the changes in expression levels of Bcl-2, Bax, and caspase3 in PC12 cells on exposure to hypoglycaemia and serum limitation with and without salidroside pre-treatment, respectively. Rhodamine 123 staining and flow cytometry with 2',7'-Dichlorofluorescein diacetate staining revealed the changes in the mitochondrial membrane potential and radical ROS production in PC12 cells on exposure to hypoglycaemia and serum limitation with and without salidroside pre-treatment, respectively. The experimental results suggest that salidroside protects the PC12 cells against hypoglycaemia and serum limitation-induced cytotoxicity possibly by the way of the modulation of apoptosis-related gene expression, the restoration of the mitochondrial membrane potential, and the inhibition of the intracellular ROS production.

Cardioprotective effects:

Wu et al. (2009): The modification of proteins with O-linked N-acetylglucosamine (O-GlcNAc) is increasingly recognised as an important posttranslational modification that modulates cellular function. Cardiomyocytes were exposed to 4 hours of ischemia and 16 hours of reperfusion, and then cell viability, apoptosis, glucose uptake, ATP levels and cytosolic Ca²⁺ concentration were determined, and O-GlcNAc levels were assessed by Western blotting. Salidroside (80 μ M) was added 24 hours before ischemia/reperfusion was induced. Treatment with salidroside markedly improved cell viability from 64.7 \pm 4.5% to 85.8 \pm 3.1%, decreased LDH release from 38.5 \pm 2.1% to 21.2 \pm 1.7%, reduced cell apoptosis from 27.2 \pm 3.2% to 12.2 \pm 1.9%, significantly improved cardiomyocytes glucose uptake by 1.7-fold and increased O-GlcNAc levels by 1.6-fold, as well as reducing cytosolic Ca²⁺ concentration compared to untreated cells following ischemia/reperfusion. Furthermore, the improved cell survival and the increase in O-GlcNAc with salidroside were attenuated by alloxan, an inhibitor of O-GlcNAc transferase. These results suggested that salidroside significantly enhances glucose uptake and increases protein O-GlcNAc levels and this is associated with decreased cardiomyocytes injury following ischemia/reperfusion.

Zhang et al. (2009): The study was aimed to investigate the cardioprotective role of salidroside and the underlying mechanisms in hypoxia-induced cardiomyocyte death. Cardiomyocytes pretreated with or without salidroside for 24 hours were exposed to hypoxic condition for 6 hours and then cell viability, necrosis, apoptosis, the expressions of hypoxia inducible factor HIF-1 α and vascular endothelial growth factor (VEGF) were investigated. Pre-treatment with salidroside markedly attenuated hypoxia-induced cell viability loss, cell necrosis and apoptosis in a dose-dependent manner. Mechanistically, pre-treatment with salidroside up-regulated the HIF-1 α protein expression and induced its translocation. Moreover, the level of VEGF, a downstream target of HIF, was significantly

increased in parallel with the level of HIF-1 α following pre-treatment with salidroside. However, 2-methoxyestradiol (2-ME2), a HIF-1 α inhibitor, attenuated the protection of salidroside and blocked the increase of HIF-1 α and VEGF. These data indicated that salidroside has protective effect against hypoxia-induced cardiomyocytes necrosis and apoptosis by increasing HIF-1 α expression and subsequently up-regulating VEGF levels.

Zhong *et al.* (2010): The cardioprotective effects of salidroside, isolated from *Rhodiola rosea* L, on oxygen-glucose deprivation (OGD)-induced cardiomyocyte death and ischemic injury evoked by acute myocardial infarction (AMI) was investigated in rats. Pre-treatment with salidroside notably ameliorated cell viability losses in a dose-dependant manner and in parallel it alleviated morphologic injury detected by electron microscopy. Mechanistically, diminished OGD-induced cardiomyocyte apoptosis was shown in salidroside-pre-treated cardiomyocytes, in accordance with minimal ROS burst. Moreover, salidroside markedly upregulated the Bcl-2/Bax ratio and preserved mitochondrial transmembrane potential. Salidroside administration also inhibited myocardial apoptosis in AMI rats by increasing phosphorylation of AKT and decreasing activation of caspase-3. These findings suggest that salidroside reduced ischemia-mediated myocardial damage.

Effects on the blood system:

Qian *et al.* (2011): This study attempted to examine the potential erythropoiesis-stimulating and anti-oxidative effect of salidroside in TF-1 erythroblasts. The erythropoiesis-promoting effect was determined by treating human TF-1 cells with salidroside in the presence and absence of erythropoietin (EPO) through the measurement of the expression of a series of erythroid markers such as glycophorin A (GPA), transferrin receptor (CD71) and hemoglobin (Hb). The potential protective effect of salidroside against H₂O₂-induced apoptosis and its underlying mechanism in TF-1 erythroblasts were examined by flow cytometry and Western blot analysis. Salidroside promotes erythropoiesis in the EPO-treated cells and it also reduces the number of apoptotic cells in TF-1 erythroblasts after H₂O₂ treatment probably through the up-regulation of protective proteins thioredoxin-1 and glutathione peroxidase-1.

Effects on metabolism

Kobayashi *et al.* (2008): As a methanol extract of the rhizome of *Rhodiola rosea* inhibits the activity of lipase in isolated mouse plasma *in vitro* and in the mouse gastrointestinal tube *in vivo*, the active components in this plant were investigated. After fractionation and separation processes, rhodionin and rhodiosin were isolated as active ingredients. Their IC₅₀ values were 0.093 mM and 0.133 mM *in vitro*, respectively. Both compounds significantly suppressed the elevation of the postprandial blood triglyceride level, e.g., by 45.6% (150 mg/kg, 60 minutes after oral administration) and 57.6% (200 mg/kg, 180 minutes after oral administration), respectively.

Li *et al.* (2008): The metabolic effects of salidroside on skeletal muscle cells were investigated. Salidroside dose-dependently stimulated glucose uptake in differentiated L6 rat myoblast cells. Inhibition of AMP-activated protein kinase (AMPK) by pre-treating the cells with compound C (= dorsomorphin) potently reduced salidroside-stimulated glucose uptake, while inhibition of phosphatidylinositol 3-kinase (PI3K) by wortmannin exhibited no significant inhibitory effect on salidroside-mediated glucose transport activation. Western blotting analyses revealed that salidroside increased the phosphorylation level of AMPK and acetyl-CoA carboxylase (ACC). In addition, salidroside enhanced insulin-mediated AKT activation and glucose uptake, and such enhancement can be specifically inhibited by compound C.

Effects on hepatic tissue:

Ouyang et al. (2010): The present study aimed to investigate whether salidroside can induce differentiation of rat mesenchymal stem cells (rMSCs) towards hepatocytes *in vitro* and the mechanism of hepatic differentiation of rMSCs. rMSCs were subject to hepatic differentiation. One, two and three weeks later, the expression of α -fetoprotein (AFP) and albumin (ALB), cytochrome P450 (CYP450)-dependent activity and inducibility, cellular uptake of low density lipoprotein (LDL) and urea synthesis were assessed and the hepatic differentiation of rMSCs was evaluated. In order to unravel the mechanism of hepatic differentiation of rMSCs *in vitro*, inhibitors of extracellular regulated kinase1/2 (ERK1/2), phosphatidylinositol 3-kinase (PI3K) and p38 were applied. When the process of hepatic differentiation was completed, special proteins of hepatic differentiation were detected and blocking of inhibitors was evaluated. Salidroside significantly induced the differentiation of rMSCs towards hepatocytes. Differentiated rMSCs have typical functional hepatic characteristics. The results also showed that the ERK1/2 and PI3K signalling pathways play important roles in the regulatory effects of salidroside on hepatic differentiation of rMSCs and are involved in cell fate determinations, while the p38 signalling pathway does not.

Wu et al. (2009a): The aim was to investigate the protective effect of salidroside on D - galactosamine/lipopopolysaccharide-induced fulminant hepatic failure. Hepatotoxicity was induced by an intraperitoneal injection of D-galactosamine (700 mg/kg) and lipopolysaccharide (10 μ g/kg); salidroside (20, 50 and 100 mg/kg) was administered intraperitoneally 1 hour before induction of hepatotoxicity. Liver injury was assessed biochemically and histologically. Salidroside attenuated the induced acute increase in serum aspartate aminotransferase and alanine aminotransferase activities, and levels of TNF- α levels and serum nitric oxide. It restored depleted hepatic glutathione, superoxide dismutase, catalase and glutathione peroxidase activities, decreased malondialdehyde levels and considerably reduced histopathological changes. Histopathological, immunohistochemical and Western blot analyses also demonstrated that salidroside could reduce the appearance of necrotic regions and expression of caspase-3 and hypoxia-inducible factor-1 α in liver tissue. The authors conclude that salidroside protected liver tissue from the oxidative stress elicited by D-galactosamine and lipopolysaccharide. The hepatoprotective mechanism of salidroside appears to be related to antioxidant activity and inhibition of hypoxia-inducible factor-1 α .

Wu et al. (2008): The protective effect of salidroside was investigated in the acetaminophen (APAP)-induced hepatic toxicity mouse model in comparison to N-acetylcysteine (NAC). Drug-induced hepatotoxicity was induced by an intraperitoneal injection of 300 mg/kg (sub-lethal dose) of APAP. Salidroside was given orally to mice at a dose of 50 or 100 mg/kg 2 hours before the APAP administration in parallel with NAC. Mice were sacrificed 12 hours after the APAP injection to determine aspartate aminotransferase (AST), alanine aminotransferase (ALT), and TNF- α levels in serum and glutathione (GSH) depletion, malondialdehyde (MDA) accumulation, and caspase-3 expression in liver tissues. Salidroside significantly protected APAP-induced hepatotoxicity, as salidroside improved mouse survival rates better than NAC against a lethal dose of APAP and significantly blocked not only APAP-induced increases of AST, ALT, and TNF- α but also APAP-induced GSH depletion and MDA accumulation. Histopathological and immunohistochemical analyses also demonstrated that salidroside could reduce the appearance of necrosis regions as well as caspase-3 and hypoxia inducible factor-1 α expression in liver tissue. The results indicated that salidroside protected liver tissue from the APAP-induced oxidative damage via preventing or alleviating intracellular GSH depletion and oxidation damage.

Effects on cancer cells:

Hu et al. (2010): To investigate the cytotoxic effects of salidroside on breast cancer cells and in order to reveal possible ER-related differences in response to salidroside, MDA-MB-231 cells and MCF-7 cells (estrogen receptor-positive) were used as models to study possible molecular mechanisms. The effects of salidroside on cell growth characteristics, such as proliferation, cell cycle duration, and apoptosis, and on the expression of apoptosis-related molecules were evaluated. The results demonstrated that salidroside in concentration between 5 µM and 80 µM dose-dependently induces cell-cycle arrest and apoptosis in human breast cancer cells.

Hu et al. (2010a): The present study focused on evaluating the effects of salidroside on the proliferation of various human cancer cell lines derived from different tissues, and further investigating its possible molecular mechanisms. Cell viability assay and [(3)H] thymidine incorporation were used to evaluate the cytotoxic effects of salidroside on cancer cell lines, and flow cytometry analysed the change of cell cycle distribution induced by salidroside. Western immunoblotting further studied the expression changes of cyclins (cyclin D1 and cyclin B1), cyclin-dependent kinases (CDK4 and Cdc2), and cyclin-dependent kinase inhibitors (p21(Cip1) and p27(Kip1)). The results showed that salidroside in concentration between 1 µg/ml and 32 µg/ml dose-dependently inhibited the growth of various human cancer cell lines in concentration- and time-dependent manners, and the sensitivity to salidroside was different in those cancer cell lines. Salidroside could cause G1-phase or G2-phase arrest in different cancer cell lines, meanwhile, salidroside resulted in a decrease of CDK4, cyclin D1, cyclin B1 and Cdc2, and upregulated the levels of p27(Kip1) and p21(Cip1). Taken together, salidroside could inhibit the growth of cancer cells by modulating CDK4-cyclin D1 pathway for G1-phase arrest and/or modulating the Cdc2-cyclin B1 pathway for G2-phase arrest.

Anti-bacterial activity:

Ming et al. (2005): Bioactivity-guided fractionation of a 95% ethanol extract from the stems of *Rhodiola rosea* led to the isolation of five compounds: gossypetin-7-O-L-rhamnopyranoside (1), rhodiolfavonoside (2), gallic acid (3), trans-p-hydroxycinnamic acid (4) and p-tyrosol (5). Compounds 1 and 2 were evaluated for their antibacterial and antiproliferative cancer cell activities. Compounds 1 and 2 exhibited activity against *Staphylococcus aureus* with minimum inhibitory concentrations of 50 µg/ml and 100 µg/ml, respectively. Cytotoxicity studies of 1 and 2 also displayed activity against the prostate cancer cell line with IC₅₀ values of 50 µg/ml and 80 µg/ml, respectively.

Other effects:

Yin et al. (2009): This study aimed to investigate the inhibitory effect of salidroside on high glucose-induced mesangial cell proliferation and its possible mechanism. Salidroside (in concentrations from 1 to approximately 100 µM) dose dependently inhibited high glucose-induced mesangial cell early proliferation. Exposure of mesangial cells to high glucose for 24 hours significantly induced ROS accumulation, ERK1/2 phosphorylation, and p27 (Kip1) expression, and these changes were dramatically inhibited by salidroside in a dose-dependent manner. High glucose-promoted TGF-β1 secretion was also significantly attenuated by treatment of mesangial cells with salidroside.

Wang et al. (2009a): The aim of this study was to investigate the antiviral effects of salidroside. The antiviral effects of salidroside against coxsackievirus B3 (CVB3) were determined *in vitro* and *in vivo*. The effect of salidroside on the mRNA expression of some important cytokines was measured in hearts of infected BALB/c mice by RT-PCR. Salidroside exhibited obvious antiviral effects both in *in vitro* concentrations of 80 and 120 mg/l) and *in vivo* (concentrations between 20 and 80 mg/kg body weight) experiments. Salidroside was found to modulate the mRNA expression of interferon-gamma (IFN-gamma), interleukin-10 (IL-10), tumour necrosis factor-alpha (TNF-α), and interleukin-2 (IL-2).

Flavonoids: effects on neuraminidase

Jeong et al. (2009): The flavonols kaempferol, herbacetin, rhodiolinin, rhodionin and rhodiosin were isolated from *Rhodiola rosea*. The compounds showed neuraminidase inhibitory activities with IC₅₀ values ranging from 1.4 to 56.9 µM. The *in vitro* anti-influenza virus activities were evaluated using two influenza viral strains, H1N1 (A/PR/8/34) and H9N2 (A/Chicken/Korea/MS96/96), testing their ability to reduce virus-induced cytopathic effect (CPE) in MDCK cells. The activity of these compounds ranged from 30.2 to 81.9 µM against H1N1- and 18.5 to 49.6 µM against H9N2-induced CPE. Activity depended on the position and number of hydroxy groups on the flavonoids backbone.

Pharmacological data from combinations:

Panossian et al. (2009): Experiments were carried out with BALB/c mice taking ADAPT-232 forte, a fixed combination of extracts of *Eleutherococcus senticosus*, *Schisandra chinensis* and *Rhodiola rosea* (extract SHR-5), characterised for the content of active markers eleutherosides, schisandrins, salidroside, tyrosol and rosavin and in doses of about 30, 90 and 180 mg/kg for 7 consecutive days followed by forced swimming test to exhaustion. ADAPT-232 forte strongly augments endurance of mice, increasing the time taken to exhaustion (TTE) in a dose-dependent manner from 3.0+/-0.5 to 21.1+/-1.7 min, approximately seven fold. Serum Hsp72 was measured by EIA both in normal and stressful conditions before and after the swimming test. Repeated administration of adaptogen dose dependently increases basal level of Hsp72 in serum of mice from 0.8-1.5 to 5.5-6.3 pg/ml. This effect is even stronger than the effect of stress, including both physical (swimming) and emotional impacts: 3.2+/-1.2 pg/ml. The cumulative effect of stress and adaptogen was clearly observed in groups of animals treated with adaptogen after swimming to exhaustion, when serum Hsp72 increased to 15.1+/-1 pg/ml and remained at almost the same level during the 7 days. The authors conclude that adaptogens induce increase of serum Hsp72, regarded as a defence response to stress, and increase tolerance to stress (in the model combination of physical and emotional stresses). It can be suggested that increased tolerance to stress induced by adaptogen is associated with its stimulation of expression of Hsp70 and particularly with Hsp72 production and release into systemic circulation, which is known as a mediator of stress response involved in reparation of proteins during physical load.

3.2. Overview of available pharmacokinetic data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

Hellum et al. (2010) investigated six clones of *Rhodiola rosea* from different areas in Norway for their *in vitro* inhibitory potential on CYP3A4-mediated metabolism and P-gp efflux transport activity. Extracts were prepared using ethanol 96% as a primary extraction solvent, the dry residue was re-dissolved in ethanol 50%, the supernatant was used for the experiments. C-DNA baculovirus expressed CYP3A4 and Caco-2 cells were used for inhibitory assays, and as positive control inhibitors ketoconazole and verapamil were applied, respectively. Scintillation counting was used to quantify the transport of (3)H-digoxin in Caco-2 cells. All clones showed potent inhibition of CYP3A4 and P-gp activities, with IC₅₀ values ranging from 1.7 to 3.1 µg/ml and from 16.7 to 51.7 µg/ml, respectively. The concentration of presumed biologically active constituents in the different clones varied considerably, but this variation was not related to the clones' inhibitory potential on CYP3A4 or P-gp activities.

Panossian et al. (2009a) investigated whether *Rhodiola rosea* SHR-5 extract (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) interacts with warfarin and theophylline when administered concomitantly. The extract used in the theophylline study contained 2.7% rhodioloside (= salidroside), 6.0% rosavin and 0.8% tyrosol, while that for the warfarin study contained 2.5% rhodioloside, 3.9% rosavin and 0.8% tyrosol. The Wistar rats received orally 50mg/kg daily for 3 days. After the final dose the animals received a single dose of aminophylline or warfarin.

The animals in the placebo group received water by oral gavage. All relevant pharmacokinetic parameters remained unchanged. The authors conclude that the concomitant treatment of rats with theophylline and SHR-5 did not give rise to significant effects on the pharmacokinetics of theophylline. Simultaneous administration of SHR-5 and warfarin did not alter significantly the pharmacokinetics or the anticoagulant activity of warfarin.

3.3. Overview of available toxicological data regarding the herbal substance(s)/herbal preparation(s) and constituents thereof

Herbal preparations:

No data published.

The considerably high dosages which were used in some of the pharmacological tests (e.g. Chen *et al.* 2008a, Qu *et al.* 2009) indicate that dosages up to 6 g herbal preparation per kg body weight per day were well tolerated in rats.

In a Public Assessment Report of the MHRA it is stated that an extract prepared with ethanol 68% v/v did not reveal any mutagenic effect up to a cytotoxic concentration of 3,160 µg/plate in an Ames test, with and without metabolic activation.

Isolated constituents:

Zhu *et al.* (2010) evaluated the potential genotoxicity of salidroside by using the standard battery of tests (i.e., bacterial reverse mutation assay, chromosomal aberrations assay, and mouse micronucleus assay). The results showed that salidroside was not genotoxic under the conditions of the reverse mutation assay, chromosomal aberrations assay, and mouse micronucleus assay conditions. The anticipated clinical dose seems to be smaller compared to doses administered in the genotoxicity assays.

3.4. Overall conclusions on non-clinical data

A high number of pharmacological investigations on herbal preparations and isolated constituents from the underground parts of *Rhodiola rosea* are published. However, in many of the experiments supraphysiological concentrations or dosages far exceeding the proposed dose in humans were applied. Therefore the relevance of the results of such studies must be questioned.

Some of the studies (e.g. in models investigating anti-fatigue effects, stress-protective effects, effects on the nervous system) support the medicinal use of herbal preparations from *Rhodiola rosea* as an adaptogen and make the use in this indication plausible. Results from studies performed with isolated constituents suggest that the phenylethanoids like salidroside contribute to these effects.

Only limited data is published on toxicity of preparations of *Rhodiola*. *In vitro* data concerning metabolic and transporter interactions indicated moderate inhibitory effects by *Rhodiola* extracts, whereas *in vivo* animal studies were negative. No final conclusion can be drawn at the moment.

4. Clinical Data

4.1. Clinical Pharmacology

4.1.1. Overview of pharmacodynamic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents

The purpose of a study by Parisi *et al.* (2010) was to investigate the effects on physical performance as well as on the redox status of a chronic *Rhodiola rosea* supplementation in a group of competitive athletes during endurance exercise. Following a chronic supplementation with *Rhodiola rosea* for 4 weeks (no details on the type of the herbal preparation available), 14 trained male athletes underwent a cardio-pulmonary exhaustion test, additionally blood samples were evaluated for antioxidant status and other biochemical parameters. These data were compared with those coming from the same athletes after an intake of placebo. The supplementation did not affect the maximum heart rate, Borg Scale level (measure for perceived exertion), peak oxygen uptake, blood antioxidant status, inflammatory parameters and blood glucose level. The level of plasma free fatty acids was significantly reduced. Blood lactate and plasma creatine kinase levels were found to be significantly lower ($P < 0.05$) in treated subjects when compared to the placebo treated group.

Evdokimov (2009, only abstract available, original article in Russian language): Seven-hour continuous physical loading test (bicycle ergometry) was used to assess the effects of a cryopowder of *Rhodiola rosea* on the human cardiorespiratory system. Comparing with the control, the preparation facilitated activation of the energy-supplying mechanisms in human organism during physical work. In addition, it increased the efficiency of the cardiovascular and respiration systems and prevented fatigue growth.

Skarpanska-Steijnborn *et al.* (2009) investigated the effect of *Rhodiola rosea* supplementation on the balance of oxidants and antioxidants in the serum and erythrocytes of competitive rowers. The study medication contained 100 mg of a *Rhodiola rosea* 'concentrate' (no more details available) and 5 mg zinc and was given twice daily for 4 weeks. This double-blinded study included 22 members of the Polish Rowing Team who were participating in a preparatory camp. At the beginning and end of the study, participants performed a 2,000-m maximum test on a rowing ergometer. Blood samples were taken from the antecubital vein before each exercise test, 1 minute after completing the test, and after a 24-hours restitution period. The following redox parameters were assessed in erythrocytes: superoxide dismutase activity, glutathione peroxidase activity, and thiobarbituric-acid-reactive substances concentrations. In addition, creatine kinase activity and total antioxidant capacity were measured in plasma samples, lactate levels were determined in capillary blood samples, and uric acid concentrations were measured in serum. After supplementation, the total plasma antioxidant capacity was significantly higher ($p = 0.0002$) in the supplemented group than in the placebo group, and superoxide dismutase activity in erythrocytes directly after and 24 hours after the ergometry was significantly ($p = 0.0461$) lower in athletes receiving *Rhodiola rosea* extracts than in the placebo group.

The purpose of the investigation by Walker *et al.* (2007) was to examine the effect of *Rhodiola rosea* ingestion on human skeletal muscle phosphocreatine recovery after exhaustive exercise. The study medication was 1500 mg *Rhodiola rosea* standardised to 3% rosavins (no more details on the herbal preparation available), divided into 3 doses. Twelve resistance-trained men (19 to 39 years of age) completed incremental forearm wrist flexion exercise to volitional fatigue, once after ingesting *Rhodiola rosea* for 4 days, and once after ingesting an equivalent placebo dose. During exercise and recovery from exercise, muscle phosphates were examined using ^{31}P nuclear magnetic resonance spectroscopy. In summary, there were no significant differences between groups for any of the parameters

measured. The authors conclude that *Rhodiola rosea* ingestion does not improve ATP turnover during or immediately after exercise.

Colson *et al.* (2005) examined the effects of a fixed combination of *Cordyceps sinensis* and *Rhodiola rosea* (no details on the type of herbal preparation and on posology available) on circulatory dynamics, specifically muscle tissue oxygen saturation in male subjects during maximal exercise. This study followed a double blind, randomised, placebo-treatment, pre-post test design. Capsules were administered to 8 subjects who were randomly assigned to one of two groups. All subjects performed 2 exercise stress tests to volitional fatigue on a cycle load ergometer. There were no significant ($p \leq 0.05$) differences between or within the treatment or control group. The combination did not affect the muscle tissue oxygen saturation.

In a double-blind, placebo controlled study Abidov *et al.* (2004) studied the effect of 30 mg *Rhodiola rosea* extract (no details available) twice daily for 30 days before and 6 days after exhausting physical exercise on the level of C-reactive protein and creatinine kinase. The study medication reduced the levels of C-reactive protein significantly compared to both placebo and control group.

Wing *et al.* (2003) investigated the effects of a 7-day supplementation with 447 mg *Rhodiola rosea* 4 times daily (no details on the herbal preparation) on hypoxia and oxidative stress at a simulated altitude of 4600 m. Fifteen volunteers (ages 20-33) received 3 separate 60-minute hypoxic exposures by breathing 13.6% oxygen at an ambient barometric pressure of 633 mm Hg (simulating the partial pressure of oxygen at 4600 m elevation). Each subject received, in random order, treatments of a 7-day supply of placebo, *Rhodiola rosea*, and an acute dose of stabilised oxygen dissolved in water. The supplementation did not have a significant effect on blood oxygenation after 60 minutes of sedentary hypoxic exposure. Hypoxia-induced oxidative stress was observed in the control group only. Supplementation appeared not to increase oxidative stress and may decrease free radical formation after hypoxic exposure compared with the control.

4.1.2. Overview of pharmacokinetic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents

No data published

4.2. Clinical Efficacy

4.2.1. Dose response studies

Darbinyan *et al.* (2007): see details in 4.2.2.

Shevtsov *et al.* (2003): see details in 4.2.2.

4.2.2. Clinical studies (case studies and clinical trials)

Clinical trials: Indication mental performance, fatigue

Single preparations:

Olsson *et al.* (2009): The aim of the study was to assess the efficacy of the extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) in the treatment of individuals suffering from stress-related fatigue. The phase III clinical trial took the form of a randomised, double-blind, placebo-controlled study with parallel groups. Participants, males and females aged between 20 and 55 years, were selected according to the Swedish National Board of Health and Welfare diagnostic criteria for fatigue syndrome. A total of 60 individuals were randomised

into two groups, one (N = 30) of which received four tablets daily of SHR-5 extract (576 mg extract/day), while a second (N = 30) received four placebo tablets daily. The effects of the extract with respect to quality of life (SF-36 questionnaire, a validated scale to assess the quality of life), symptoms of fatigue (Pines' burnout scale), depression (Montgomery -Asberg depression rating scale - MADRS), attention (Conners' computerised continuous performance test II - CCPT II), and saliva cortisol response to awakening were assessed on day 1 and after 28 days of medication. Data were analysed by between-within analyses of variance. Significant post-treatment improvements were observed for both groups (placebo effect) in Pines' burnout scale, mental health (SF-36), and MADRS and in several CCPT II indices of attention, namely, omissions, commissions, and standard error of the reaction time (Hit RT SE). When the two groups were compared, however, significant effects of the SHR-5 extract in comparison with the placebo were observed in Pines' burnout scale and the CCPT II indices omissions, Hit RT SE, and variability. Pre- versus post-treatment cortisol responses to awakening stress were significantly different in the treatment group compared with the control group. No serious side effects that could be attributed to the extract were reported. It is concluded that repeated administration of the extract exerts an anti-fatigue effect that increases mental performance, particularly the ability to concentrate, and decreases cortisol response to awakening stress in burnout patients with fatigue syndrome.

Schutgens et al. (2009): In a randomised double blind placebo-controlled study, 30 subjects were randomly assigned to three groups: one group (n = 10) taking placebo pills, one group (n = 10) taking *Rhodiola rosea* (144 mg extract SHR-5, DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) pills and one group (n = 10) taking ADAPT-232 supplements (fixed combination of *Eleutherococcus senticosus*, *Rhodiola rosea* (SHR-5 extract, no details on the amount of extract per tablet) and *Schisandra chinensis*). The study medication was given twice daily for 7 days. All subjects underwent measurements to determine ultra-weak photon emission (UPE) of the dorsal side of their hands using a photon-counting device, both before and after a week of taking the supplements. In addition, the experienced levels of stress and fatigue (tiredness) were evaluated. After 1 week of supplementation, the *Rhodiola* group showed a significant decrease ($p = 0.027$) in photon emission in comparison with the placebo group. Furthermore, after supplementation, a significant decrease ($p = 0.049$) concerning the experienced level of fatigue in the *Rhodiola* group was observed compared with the placebo group. No significant changes were observed between the ADAPT-232 and the placebo group.

Darbinyan et al. (2007): The objective of this study was to assess the efficacy and safety of standardised extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) in patients suffering from a current episode of mild/moderate depression according to DSM-IV. The phase III clinical trial was carried out as a randomised double-blind placebo-controlled study with parallel groups over 6 weeks. The participants were males and females aged 18-70 years. The severity of the depression was determined by scores gained in Beck Depression Inventory and Hamilton Rating Scale for Depression (HAMD) questionnaires. Exclusion criteria: previous attempt to commit suicide, scores on the scales indicating suicidal tendency, total HAMD score above 31, progressive organic or metabolic brain syndrome, compulsive, schizophrenic or other delusive disorders. Patients with initial HAMD scores between 21 and 31 were randomised into three groups, one of which (group A: 31 patients) received two tablets daily of SHR-5 (340 mg/day), a second (group B: 29 patients) received two tablets twice per day of SHR-5 (680 mg/day), and a third (group C: 29 patients) received two placebo tablets daily. The efficacy of SHR-5 extract with respect to depressive complaints was assessed on days 0 and 42 of the study period from total and specific subgroup HAMD scores. For individuals in groups A and B, overall depression, together with insomnia, emotional instability and somatization, but not self-esteem, improved significantly following medication, whilst the placebo group did not show such improvements. In the group A the HAMD score

improved from 24.52 to 15.97, in group B from 23.79 to 16.72, while it remained nearly unchanged in the placebo group (24.17 to 23.41). No serious side-effects were reported in any of the groups A-C.

No difference in the clinical outcome was observed between the different dosages.

Assessors's comment:

The observation of nearly an absent placebo-effect (expressed by a nearly unchanged HAMD score in the placebo group) is untypical for clinical trials with depressant patients. Therefore the significances documented in the publication remain questionable.

De Bock et al. (2004): The purpose of this study was to investigate the effect of acute and 4-week *Rhodiola rosea* extract (no details on DER and extraction solvent, extract contains 3% rosavin and 1% salidroside) intake on physical capacity, muscle strength, speed of limb movement, reaction time, and attention in 24 patients. PHASE I: A double blind placebo-controlled randomised study (n= 24) was performed, consisting of 2 sessions (2 days per session). Day 1: one hour after acute *Rhodiola rosea* intake (R, 200-mg *Rhodiola rosea* extract) or placebo (P, 700 mg starch) speed of limb movement (plate tapping test), aural and visual reaction time, and the ability to sustain attention (Fepsy Vigilance test) were assessed. Day 2: Following the same intake procedure as on day 1, maximal isometric knee-extension torque and endurance exercise capacity were tested. Following a 5-day washout period, the experimental procedure was repeated, with the treatment regimens being switched between groups (session 2). PHASE II: A double blind placebo-controlled study (n = 12) was performed. Subjects underwent sessions 3 and 4, identical to Phase I, separated by a 4-week R/P intake, during which subjects ingested 200 mg R/P per day. RESULTS: PHASE I: Compared with placebo the acute intake of *Rhodiola* extract in Phase I increased (p <.05) time to exhaustion from 16.8 +/- 0.7 min to 17.2 +/- 0.8 min. Accordingly, VO₂peak (p <.05) and VCO₂peak (p<.05) increased during the study medication compared to placebo from 50.9 +/- 1.8ml x min⁽⁻¹⁾ x kg⁽⁻¹⁾ to 52.9 +/- 2.7 ml x min⁽⁻¹⁾ x kg⁽⁻¹⁾ (VO₂peak) and from 60.0 +/- 2.3 ml x min⁽⁻¹⁾ x kg⁽⁻¹⁾ to 63.5 +/- 2.7 ml x min⁽⁻¹⁾ x kg⁽⁻¹⁾ (VCO₂peak). Pulmonary ventilation (p =.07) tended to increase more in the verum group compared to placebo (P: 115.9 +/- 7.7 l/min; R: 124.8 +/- 7.7 l/min). All other parameters remained unchanged. PHASE II: Four-week R intake did not alter any of the variables measured. The authors conclude that acute *Rhodiola rosea* intake can improve the endurance exercise capacity in young healthy volunteers. This response was not altered by prior daily 4-week *Rhodiola* intake.

Shevtsov et al. (2003): A randomised, double-blind, placebo-controlled, parallel-group clinical study with an extra non-treatment group was performed to measure the effect of a single dose of standardised SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) on capacity for mental work against a background of fatigue and stress. Some physiological parameters, e.g. pulse rate, systolic and diastolic blood pressure, were also measured. The study was carried out on a highly uniform population comprising 161 cadets aged from 19 to 21 years. Group 1 (41 subjects) received 370 mg extract per day, group 2 (20 subjects) received 555 mg extract per day. The study showed a pronounced anti-fatigue effect reflected in an anti-fatigue index defined as a ratio called AFI. The verum groups had AFI mean values of 1.0385 and 1.0195, 2 and 3 capsules respectively, whilst the figure for the placebo group was 0.9046. This was statistically highly significant (p < 0.001) for both doses (verum groups), whilst no significant difference between the two dosage groups was observed. There was a possible trend in favour of the lower dose in the psychometric tests. No such trend was found in the physiological tests. Only one case of hypersalivation in the placebo group was reported as undesirable effect.

Ha et al. (2002): The aim of the study was to investigate the changes of sleep architecture and blood oxygen saturation (SaO₂) during sleep in men living at high altitude, and to investigate the effect of

Rhodiola and acetazolamide on these sleep indexes. Twenty-four men aged 18 to 21 years who had stayed at high altitude (5 380 m above sea level) for 1 year were randomly divided into groups A (treated with oral *Rhodiola*, no information on kind of herbal preparation and on posology), B (treated with oral acetazolamide 0.25 g twice daily) and C (treated with *Rhodiola* + acetazolamide). Their sleep architecture and SaO₂ were recorded for 24 days before and after taking the medicines. The authors conclude that both *Rhodiola* and acetazolamide were effective in modulating the sleep architecture and improving the sleep quality in young men living at high altitude, but there was no synergistic effect between *Rhodiola* and acetazolamide.

Assessors's comment:

Because of the lack of any information concerning the type of the herbal preparation and the posology the findings cannot be taken into consideration.

Darbinyan et al. (2000): The aim of this study was to investigate the effect of repeated low-dose treatment with a standardised extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) on fatigue during night duty among a group of 56 young, healthy physicians. The effect was measured as total mental performance calculated as Fatigue Index. The tests chosen reflect an overall level of mental fatigue, involving complex perceptive and cognitive cerebral functions, such as associative thinking, short-term memory, calculation and ability of concentration, and speed of audio-visual perception. These parameters were tested before and after night duty during three periods of two weeks each: a) a test period of one *Rhodiola*/placebo tablet daily, b) a washout period and c) a third period of one placebo/*Rhodiola* tablet daily, in a double-blind cross-over trial. The verum tablets contained 170 mg extract (with approximately 4.5 mg salidroside) and were taken once daily. The perceptive and cognitive cerebral functions mentioned above were investigated using 5 different tests. A statistically significant improvement in these tests was observed in the treatment group during the first two weeks period. No side-effects were reported for either treatment. These results suggest that *Rhodiola* can reduce general fatigue under certain stressful conditions.

Spasov et al. (2000a): The objective was to investigate the stimulating and normalising effect of the adaptogen *Rhodiola rosea* extract SHR-5 (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water) in 40 students during a stressful examination period. The study was performed as a double-blind, randomised and placebo-controlled with low repeated dose regime. One tablet contained 50 mg of the *Rhodiola* extract, taken twice daily for 20 days. The physical and mental performance were assessed before and after the period, based on objective as well as on subjective evaluation. The most significant improvement in the SHR-5 group was seen in physical fitness, mental fatigue and neuro-motoric tests ($p < 0.01$). The self-assessment of the general well-being was also significantly ($p < 0.05$) better in the verum group. No significance was seen in the correction of text tests or a neuro-muscular tapping test. The authors concluded that the study medication gave significant results compared to the placebo group but that the dose level probably was suboptimal.

Spasov et al. (2000b): 60 male students (17-18 years of age) were randomised to three groups; the participants in one group received the study medication (660 mg of *Rhodiola*, no details on the type of the herbal preparation published) for 20 days. The authors observed in the treatment group improvement of health and well-being, activeness level, mood, and work stimulation as well as reduced signs of fatigue. No adverse events in the verum group were observed.

Clinical trials in other indications:

Bystritsky et al. (2008): The goal of this pilot study was to evaluate whether *Rhodiola rosea* is effective in reducing symptoms of generalised anxiety disorder (GAD). Ten participants (age 34-55 years) with a DSM-IV diagnosis of GAD were enrolled in this study. Participants received a total daily dose of 340 mg of *Rhodiola rosea* extract (no details of the type of the herbal preparation published) for 10 weeks. Assessments included the Hamilton Anxiety Rating Scale (HARS), the Four-Dimensional Anxiety and Depression Scale, and the Clinical Global Impressions of Severity/Improvement Scale. Individuals treated with *Rhodiola rosea* showed significant decreases in mean HARS scores at endpoint ($t=3.27$, $p=0.01$). Adverse events were generally mild or moderate in severity, the most common being dizziness and dry mouth.

Reviews:

The current knowledge is summarised in the following review articles: Iovieno et al. (2010), Panossian et al. (2010), Panossian & Wikman (2009), Walker & Robergs (2006), Tharakan & Manyam (2006), Panossian & Wagner (2005).

Opinions:

Based on the published clinical data Zubeldia et al. (2010) propose *Rhodiola rosea* as a viable alternative treatment for the symptoms of short-term hypothyroidism in patients with differentiated thyroid cancer who require hormone withdrawal.

Meta-analyses:

Hung et al. (2011): Eleven RCTs met the inclusion criteria, all were placebo-controlled. (Shevtsov et al. 2003, DeBock et al. 2004, Abodiv et al. 2004, Olsson et al. 2009, Spasov et al. 2000a, Spasoc et al. 2000b, Darbinyan 2007, Darbinyan 2000, Walker 2007, Schutgens 2009, Wing 2003). Six trials investigated the effects of *Rhodiola rosea* on physical performance, four on mental performance, and two in patients diagnosed with mental health condition. The methodological quality of most trials was moderate or good. Only a few mild adverse events were reported. *Rhodiola rosea* may have beneficial effects on physical performance, mental performance, and certain mental health conditions. There is, however, a lack of independent replications of the single different studies. Five of the 11 RCTs reached more than three points on the Jadad score (i.e., good quality). More research seems warranted.

Blomkvist et al. (2009): With a focus on the statistical methods the authors found considerable shortcomings in all but one of the studies that claim significant improvement from roseroot extract. Overall, the study designs have not been well explained. Experimental results have been confused and appear to be in some cases incorrect. Some of the conclusions are based on selected results and contradicting data have not been adequately taken into account. For example it is criticised that Darbinyan et al. (2000) claimed significant results while insignificant results are explained away with unfounded assumptions. In the study of Darbinyan et al. (2007) irrelevant tests and an inappropriate statistical comparison were used. Bystritsky et al. (2008) and Fintelmann & Gruenwald (2007) claimed an effect but did not use a placebo control. In the article of Shevtsov et al. (2003) misprints and mix-ups occur. The authors find it alarming that poorly conceived and performed studies have been published apparently without adequate scientific and editorial scrutiny. The authors conclude that the currently available evidence for the claimed effects is insufficient and that the effect of *Rhodiola rosea* is in need of further investigation before therapeutic claims can be made.

Sarris (2007): This paper reports a critical review of 27 herbal medicines and formulas in treating a broad range of psychiatric disorders (in addition to anxiety and depression), including obsessive-

compulsive, seasonal affective, bipolar depressive, psychotic, phobic and somatoform disorders. The author states that *Rhodiola rosea* might be a promising herb for the treatment of depression. However, most studies are published in the Russian language and could not be located by the author.

Combination products:

Aslanyan et al. (2010) investigated the fixed combination ADAPT-232 containing *Rhodiola rosea* L., *Schisandra chinensis* (Turcz.) Baill., and *Eleutherococcus senticosus* Maxim. The subjects in the ADAPT-232 received a single dose of the study medication. Quickly after intake (two hours after verum was taken) improved attention and increased speed and accuracy during stressful cognitive tasks, in comparison to placebo was observed. There was also a tendency of ADAPT-232 to reduce percentage of errors, which means better accuracy, quality of the work, and degree of care in the volunteers under stressful conditions. No serious side effects were reported, although a few minor adverse events, such as sleepiness and cold extremities, were observed in both treatment groups.

Dieamant et al. (2008): A double-blind comparative study was conducted on 124 volunteers with sensitive skin, who were selected by their reactivity to stinging test. Two randomised groups of 62 each received either a formulation containing 1% of a combination of *Rhodiola* extract (at the moment no details on the type of the extract and on posology) and L-carnosine or placebo, which was applied twice daily for 28 consecutive days. One perceptibility questionnaire was applied at the onset and at the end of the treatment to evaluate the subjective response to test product. Additionally, *in vitro* studies were performed to investigate potential neuroimmunomodulatory mechanisms. The verum treatment produced *in vivo* protective effects in skin barrier function and a positive subjective response of sensitive skin volunteers. *In vitro* treatment promoted the release of proopiomelanocortin peptides and restored to normal the increased levels of neuropeptides and cytokines produced by keratinocytes exposed to ultraviolet radiation. Clinical effectiveness was measured by reduction of transepidermal water loss, positive perceptions of improvements in skin dryness and skin comfort sensation, and reduction of discomfort sensation after stinging test. The protective effect of the combination in skin barrier function and the positive response produced in human subjects with sensitive skin could be partially explained by the *in vitro* results showing a significant increase in opioid peptides release, an inhibitory effect on neuropeptide production, and modulation of cytokines production by keratinocytes under ultraviolet stress.

Fintelmann & Gruenwald (2007) studied a food supplement containing 200 mg *Rhodiola* extract (no more details available), magnesium, vitamin E, vitamin B6, folic acid and vitamin B12 in a 12-week drug monitoring study. The study was neither blinded nor placebo controlled. The authors stated that in the 120 patients a statistically highly significant improvement in physical and cognitive deficiencies was observed. No adverse events occurred during the course of the study.

4.2.3. Clinical studies in special populations (e.g. elderly and children)

No data published.

4.3. Overall conclusions on clinical pharmacology and efficacy

The results from trials on clinical pharmacology are contradictory. The number of clinical trials for clinical efficacy is limited, also the number of included subjects.

Meta-analyses of these clinical trials found considerable shortcomings and deficiencies. Therefore it can be concluded that there is not sufficient evidence for a clinical efficacy of herbal preparations of *Rhodiola rosea* for the treatment of symptoms of fatigue or mental weakness. Therefore 'well-established use' cannot be supported in the monograph.

However the data support the plausibility of the use of traditional herbal medicinal products of *Rhodiola rosea* as adaptogens.

5. Clinical Safety/Pharmacovigilance

5.1. Overview of toxicological/safety data from clinical trials in humans

In the study of Olsson *et al.* (2009) no serious side effects that could be attributed to the extract (DER 2.5-5:1, first extraction solvent ethanol 70%, second extraction solvent water, 567 mg extract/day, duration of use: 28 days) were reported. No data on non-serious side effects are mentioned in the publication.

The same safety profile was observed in the study of Darbinyan *et al.* (2007) in a posology of 340 mg or 680 mg per day over 6 weeks.

In the study of Bystritsky *et al.* (2008) dizziness and dry mouth were observed after oral administration of 340 mg of extract daily (no details on the type of the extract published).

In the SmPC of the registered traditional herbal medicinal products also hypersensitivity reactions and hypoglycaemia are mentioned.

5.2. Patient exposure

No data available.

5.3. Adverse events and serious adverse events and deaths

No additional data compared to those from clinical trials are available.

Assessor's comment: The few reported adverse events may also be caused by the underlying disease. Therefore they should not be included in the monograph.

5.4. Laboratory findings

No data available.

5.5. Safety in special populations and situations

Children and adolescents:

Although the use of registered traditional herbal medicinal products containing *Rhodiola* extract is permitted in adolescents, this age group cannot be considered in the monograph since no safety data from clinical trials in adolescents are available.

Interactions:

Neither clinical data nor case reports on interactions are published.

Pregnancy and lactation:

No data published. The safety during pregnancy and lactation has not been established. In the absence of data the use during pregnancy and lactation is not recommended.

Overdose:

There are no case reports published.

Effects on ability to drive or operate machinery

No studies on the effect on the ability to drive and use machines have been published.

5.6. Overall conclusions on clinical safety

Although experimental data on genotoxicity are missing the published literature does not give reasons for safety concerns. The reported adverse events are mild. The use in children and adolescents as well as during pregnancy and lactation cannot be recommended. Based on the published data it can be concluded that traditional herbal medicinal products containing *Rhodiola rosea* are not harmful when used in the specified conditions.

6. Overall conclusions

Herbal preparations of the underground organs of *Rhodiola rosea* are used in traditional medicine since centuries. In the former USSR (including countries which now belong to the European Union like Estonia, Lithuania and Latvia) a liquid extract (DER 1:1, extraction solvent ethanol 40%) was in medicinal use, in 1975 it was accepted as a so called 'Temporary Pharmacopoeia Article' which allowed the large-scale production. Dry extracts (DER 1.5-5:1), extraction solvent ethanol 67-70% v/v, are in medicinal use within the European Union since 1987. Therefore the criteria for 30 years of medicinal use as defined for traditional herbal medicinal products in Dir. 2004/24 are fulfilled. The traditional use as an adaptogen for symptoms of asthenia such as fatigue and weakness is appropriate for traditional herbal medicinal products.

The published clinical trials exhibit considerable deficiencies in their quality. Therefore 'well-established use' cannot be accepted.

The longstanding use as well as the outcome of the clinical trials supports the plausibility of the use of the mentioned herbal preparation in the proposed indication.

The clinical trials as well as the traditional use do not give reasons for special safety concerns. No serious adverse events are reported. The *in vitro* observed inhibition of CYP3A4 and P-glycoprotein was not confirmed *in vivo*. Additionally no case reports on interactions are published. Because of the limited duration of use of 2 weeks the *in-vitro* data seem to be of minor clinical relevance.

Therefore the overall benefit/risk balance is positive.

Due to the missing published data on genotoxicity the development of a Community List Entry cannot be supported.

Annex

List of references