DIPLOMARBEIT

Titel der Diplomarbeit

Evaluation of the traditional and the well-established use of Hyperici herba

angestrebter akademischer Grad

Magister/Magistra der Pharmazie (Mag.pharm.)

Verfasserin / Verfasser:	Dagmar Gruber
Matrikel-Nummer:	0203944
Studienrichtung (lt. Studienblatt):	Pharmazie
Betreuerin / Betreuer:	Univ.-Doz. Dr. Reinhard Länger

Wien, am 20. Oktober 2008

Acknowledgments

I want to thank Univ.-Prof. Dr. Verena Dirsch for giving me the chance to work with an external adviser.

My special gratitude goes to Univ.-Doz. Dr. Reinhard Länger for offering me the opportunity to work with Hypericum perforatum at the AGES PharmMed and for supporting me during my diploma thesis. With his enthusiastic way he brought much impetus into this work. It was always interesting to work with him.

I also want to express my sincerest gratitude to AGES PharmMed for the financial support and for giving me the certainty that my diploma thesis will be of further use.

My sincere gratitude goes to my family and friends, who always were ready to listen to me and motivated me during my studies and my diploma thesis. In particular I want to thank my mother for her financial support and for her understanding.

Especially I want to thank my friend Michael for lending me his laptop, answering my questions about English vocabulary and of course for his emotional support.

Table of Content

Introduction

The aim of this work is to draw up a first draft of an assessment report of Hypericum perforatum L. with well-established use (based on article 10A of directive 2001/83/EC) and/or traditional use (based on article 16D(1) and article 16H of directive 2001/83/EC as amended by directive 2004/24/EC) for the Committee for Herbal Medicinal Products at the European Medicines Agency (EMEA), where Austria is the assigned rapporteur for this project.

Background:
With the publication of the directive 2004/24/EC traditional herbal medicinal products were defined as a new class of medicinal products. In order to facilitate the harmonisation process a new committee has been established at the EMEA, the Committee for Herbal Medicinal Products (HMPC). This committee should elaborate so called community monographs and community list entries for herbal preparations. These monographs should be relevant for the registration or authorisation of herbal medicinal products. The main content of such a monograph is the information concerning traditional use and/or well-established use of certain herbal preparations.
Well-established use is defined in directive 2001/83/EC:

By way of derogation from Article 8(3)(i), and without prejudice to the law relating to the protection of industrial and commercial property, the applicant shall not be required to provide the results of pre-clinical tests or clinical trials if he can demonstrate that the active substances of the medicinal product have been in well-established medicinal use within the Community for at least ten years, with recognised efficacy and an acceptable level of safety in terms of the conditions set out in the Annex. In that event, the test and trial results shall be replaced by appropriate scientific literature.

The definition of "Traditional use" is according directive 2004/24/EC:

> Article 16a
> A simplified registration procedure is hereby established for herbal medicinal products which fulfil all of the following criteria:
> a) they have indications exclusively appropriate to traditional herbal medicinal products which, by virtue of their composition and purpose, are intended and designed for use without the supervision of a medical practitioner for diagnostic purposes or for prescription or monitoring of treatment;
> b) they are exclusively for administration in accordance with a specified strenght and posology;
> c) they are an oral, external and/or inhalation preparation;
> d) the period of traditional use as laid down in Article 16c(1)(c) has elapsed (= at least 30 years of medicinal use, with 15 years within the EU);
> e) the data on the traditional use of the medicinal product are sufficient; in particular the product proves not to be harmful in the specified conditions of use and the pharmacological effects of efficacy of the medicinal product are palusible on the basis of long-standing use and experience.

Aim of the diploma thesis:
The monograph must be supported by an assessment report reflecting the current scientific knowledge and presenting data on the traditional use. Therefore in the first step a comprehensive literature search should provide an overview on published data. In a second

step papers of interest should be obtained in full text. The assessment report should summarize the published data on topics like traditional use, pharmacology, pharmacokinetics, clinical trials, adverse effects followed by an interpretation by the assessor. The final outcome should be a draft monograph on Hypericum perforatum, which should present information on indication, dosage, duration of use, contraindications, interactions, use during pregnancy and lactation amd adverse effects separately for certain herbal preparations for well-established use or traditional use. The draft monograph will be published on the EMEA webside for public consultation for three months. The final monograph will be prepared by the rapporteur considering the comments received by interested parties.

Methodology

Literature search:
Period: March 2007 to June 2008.

Electronic data search:
Search engine: Pubmed (http://www.ncbi.nlm.nih.gov/sites/entrez);
Keywords: Hypericum perforatum, Hypericin, Hyperforin
Hager CD-Rom

Manual search:
University of Vienna, Pharmacy and Nutritional Sciences Library
Medical University of Vienna, main library

The Pharmacy and Nutritional Sciences Library of the University of Vienna is a valuable source of older handbooks of pharmacy, old pharmacopoeias and old handbooks of phytotherapy, which are not available from electronic sources, but are essential for the documentation of the traditional use.

For the evaluation of the literature and elaboration of the assessment report for traditional and well-established use of Hypericum perforatum L I followed the guidelines of the EMEA:

- Guideline on non-clinical documentation for herbal medicinal products applications for marketing authorisation (bibliographical and mixed applications) and in applications for simplified registration (EMEA/HMPC/32116/2005)
- Guideline on the assessment of clinical safety and efficacy in the preparation of community herbal monographs for well-established and of community herbal monographs/entries to the community list for traditional herbal medicinal products/substances/preparations (EMEA/HMPC/104613/2005)
- Procedure for the preparation of community monograph for traditional herbal medicinal products (EMEA/HMPC/182320/2005 rev 1)
- Procedure for the preparation of community monographs for herbal medicinal products with well-established use (EMEA/HMPC/182352/2005 rev 2).

The structure of the assessment report and of the monograph are defined in the template EMEA/HMPC/418902/2005

All guidelines and templates are available at the website of the EMEA (http://www.emea.europa.eu/htms/human/hmpc/index.htm).

Results

I Assessment report for herbal substance(s), herbal preparation(s) or combinations thereof with well-established use and/or traditional use

II Monograph on Hypericum perforatum L., herba

I.	ASSESSMENT REPORT FOR HERBAL SUBSTANCE(S), HERBAL PREPARATION(S) OR COMBINATIONS THEREOF WITH WELL-ESTABLISHED USE AND/OR TRADITIONAL USE

Hypericum perforatum L., herba

BASED ON ARTICLE 10A OF DIRECTIVE 2001/83/EC AS AMENDED

(WELL-ESTABLISHED USE)

BASED ON ARTICLE 16D(1) AND ARTICLE 16F AND 16H OF DIRECTIVE 2001/83/EC AS AMENDED

(TRADITIONAL USE)

Herbal substance(s) (binomial scientific name of the plant, including plant part)	Whole or cut, dried flowering tops of Hypericum perforatum L., harvested during flowering time.
Herbal preparation(s)	
Pharmaceutical forms	Herbal substance or comminuted herbal substance for liquid dosage forms for cutaneous use or liquid and solid dosage forms for oral use.
Rapporteur	Austria
Assessors	Reinhard Länger

I.1 INTRODUCTION

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

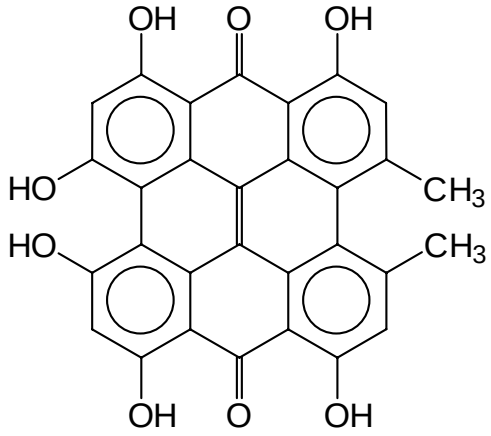
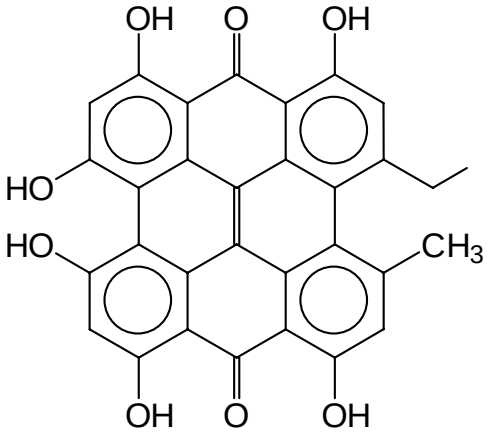
I.1.1.1 Herbal substance(s) [1]:

Hyperici herba (European Pharmacopoeia)

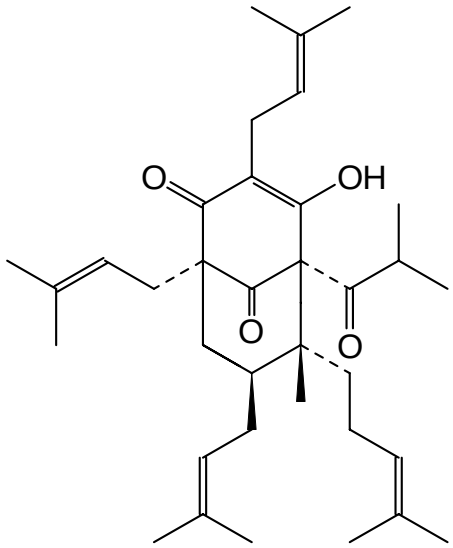
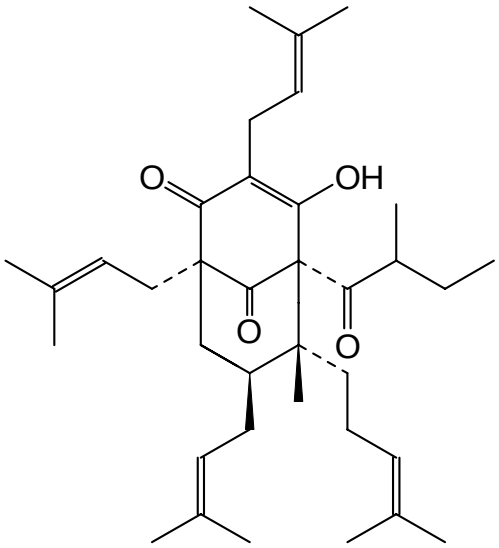
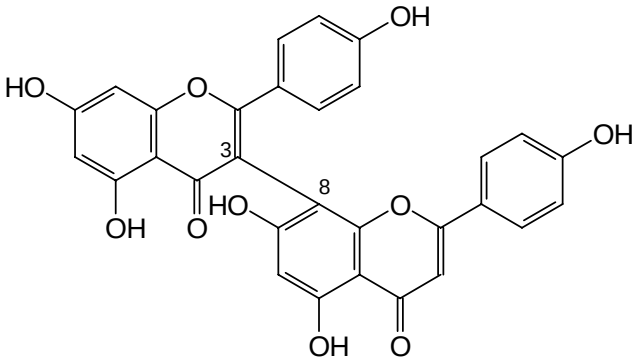
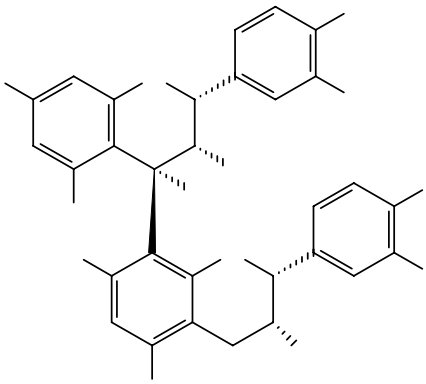
Hyperici herba consist of the whole or cut, dried flowering tops of Hypericum perforatum L., harvested during flowering time. It contains not less than 0.08 % of total hypericins expressed as hypericins calculated with reference to the dried drug.

Constituents (Wichtl 2002; Bradley 2006, Hänsel et al 2007)

- Phloroglucinol derivates: 0,2-4 %, depending on the age of the herbal drug, mainly hyperforin and its homologue adhyperforin, furanohyperforin
- Naphtodianthrones: 0,06-0,4 %, mainly pseudohypericin and hypericin, protohypericin, protopseudohypericin, cyclopseudohypericin, skyrinderivatives. The amount of pseudohypericin is about 2-4 times higher than that of hypericin.
- Flavonoids: 2-4 %, mainly glycosides of the flavonol quercetin: hyperoside, rutin, isoquercitrin, quercitrin; also biflavones (I3,II8-Biapigenin, Amentoflavone)
- Procyanidines: e.g. procyanidine B_2, tannins with catechin skeletal (6-15 %)
- Xanthones: in trace amounts
- Essential oil: 0.1-0.25 %; the essential oil of dried flowering tops contains as main compounds 2-methyloctane (16 %) and α-pinene (10.6 %). In the essential oil of leaves of Indian origin 58 components were identified, α-pinene (67 %) being dominant; the other components included caryophyllene, geranyl acetate and nonane (each about 5 %)
- Other constituents: include small amounts of chloregenic acid and other caffeoylquinic and p-coumaroylquinic acids, and also free amino acids.

	
Hypericin	Pseudohypericin

3 According to the 'Procedure for the preparation of Community monographs for traditional herbal medicinal products' (EMEA/HMPC/182320/2005 Rev.2) and the 'Procedure for the preparation of Community monographs for herbal medicinal products with well-established medicinal use (EMEA/HMPC/182352/2005 Rev.2)

	
Hyperforin	Adhyperforin
	
Biapigenin	Procyanidin B_2

I.1.1.2 Herbal preparation(s):

St. John's wort dry extract, quantified extract (Pharm. Eur. ref. 07/2008:1874)
Extraction solvents ethanol (50-80% v/v) or methanol (50-80% v/v). Content of total hypericins (expressed as Hypericin) 0.10-0.30%, content of flavonoids (expressed as rutin) minimum 6.0%, content of hyperforin maximum 6.0% and not more than the content stated on the label.

Herbal preparations with evidence of tradition according Dir. 2004/24:

A) Dry extract, DER 4,6-6,5:1, extraction solvent ethanol 38% v/v. This type of extract is in medicinal use since more than 30 years.
B) Dry extract (DER 3.5-6:1), extraction solvent ethanol 60% (m/m): on the market in DE at least since 1976
C) Dry extract (DER 5-7:1), extraction solvent ethanol 70% (m/m): on the market in DE at least since 1976
D) Liquid extract (DER 1:4-20), extraction solvent vegetable oil (e.g. olive oil, sunflower oil, linseed oil, wheat germ oil)
Preparation:
According to the German "Ergänzungsbuch" to the German Pharmacopoeia 6 (Erg.-B6. 1941): The crushed fresh flowers of Hypericum perforatum (25 parts) are doused with olive oil (100 parts) in a white glass. The mixture is

allowed to ferment at a warm place. After completion of the fermentation the glass has to be sealed. It is then stored at a sunny place for about 6 weeks until the oil is bright red. The herbal substance has to be pressed out, the oil is dried with sodium sulfate (6 parts).

According to Swiss Pharmacopoeia (1999, Pharm. Helv. 8): The comminuted fresh flowering tops of Hypericum perforatum are overflowed with 40.0 g refined sun flower oil. The mixture is reshuffled frequently; extraction and fermentation take place at a temperature of 15-30 °C. After 50-80 days the herbal substance is pressed out, the water layer is removed. It is allowed to dilute the oil to a maximum of 80 g of sunflower oil, the content of hypericin is at least 0.001% (spectrophotometric determination).

According to the company 'Caelo', Germany: the dried herbal substance (according to Pharm. Eur.) is macerated with olive oil in a DER of 1:20. The mixture is agitated under light exposure for at least 40 hours. The content of hypericin is at least 0.005% (spectrophotometric determination).

Constituents of Hyperici oleum

Hyperici Oleum does not contain hypericin. By using the sunlight maceration method described in the supplement to DAB 6 (EB 6), lipophilic breakdown products of this compound are obtained which lend the oil its red colour. The stability of hyperforin is limited; sufficient shelf-life could only be achieved by hot maceration of dried flowers with eutanol G and storage in the absence of air. The action of light during preparation of the oil led to a rise in the content of flavonoids (Maisenbacher et al 1992).
As a consequence the spectrophotometric determination used for the specification of the St. John's wort oils mentioned above detects primarily artifacts of hypericin.
Schempp et al (2000) used for the test of the influence of Hypericum extracts on skin sensitivity Hypericum oil containing 110 µg/ml Hypericin. It is not transparent, whether the authors determined hypericin or the oil derivatives.

E) Liquid extract (DER 1: 13), extraction solvent maize oil: on the market in DE at least since 1976

F) Tincture (DER 1:10), extraction solvent ethanol 45-50% v/v: The external use of a tincture (no further details on DER and extraction solvent) is mentioned in Irion (1955) as an equivalent to hypericum oil. Also Madaus (1938) mentions the use of a tincture, details about the DER are lacking. A tincture with a DER of 1:10 (extraction solvent ethanol 45-50% v/v) is mentioned as traditional herbal preparation in Barnes et al (2002) and Gruenwald et al (2004).

G) Tincture (DER 1:5), extraction solvent ethanol 50% v/v: mentioned in Bradly (2006). The evidence of tradition of this tincture has to be proved.

H) Liquid extract DER 1:2, extraction solvent ethanol 50%
The product 'Hyperforat-drops', contains a liquid extract with DER of 1:2, extraction solvent ethanol 50% (no details v/v or m/m). This is in contrast to details given in the review of Linde (2007) with respect to the DER: Hyperforat is cited in the study with a DER of 5-7:1, which would indicate a dry extract. This difference is due to obvious incorrect data in the German "Rote Liste". The first clinical study with this liquid extract is published 1979, therefore when the monograph on hypericum will be published the extract is in medicinal use at least 30 years.

I) Liquid extract 1: 5-7, ethanol 50%. Trade name Psychotonin, since 1963 in medicinal use.
J) Expressed juice from the fresh herb (DER 1.25-2.5:1): on the market in DE at least since 1976
K) Comminuted herbal substance: cut herbal substance used for tea preparation; powdered herbal substance in solid dosage forms for oral use on the market in DE at least since 1976

Further liquid extracts:

Since the edition 1993 of Hager's Handbuch (Hänsel et al 1993) a liquid extract with a DER 1:6, extraction solvent ethanol 70% is mentioned. This type of extract is not mentioned in previous literature. The period of 30 years of medicinal use is therefore not fulfilled.

The request for marketed products in the EU revealed that numerous further extracts are on the market. However, they neither do fulfil the criteria for traditional use nor are supported by clinical evidence.

Influence of the extraction solvent on the composition of the extract:
When using extraction solvents containing more than 50% of ethanol or methanol in water the content of hypericin in the extract seems to be very similar independent of the actual concentration of the extraction solvent. In contrast the extraction of hyperforin and adhyperforin depends strongly on the concentration of the extraction solvent. Best yield (60% of the hyperforin in the herbal substance, 45% of adhyperforin in the herbal substance) is achieved with 70% (e.g. ethanol), while with 50% ethanol only 20% of hyperforin and no adhyperforin are extracted (Meier 1999).

I.1.1.3 Combinations of herbal substance(s) and/or herbal preparation(s)[2]

Not applicable

I.1.1.4 Vitamin(s)[3]

Not applicable

I.1.1.5 Mineral(s)[3]

Not applicable

I.1.2 Information on period of medicinal use in the Community regarding the specified indication

I.1.2.1 Type of tradition, where relevant

European tradition

I.1.2.2 Bibliographic/expert evidence on the medicinal use

[2] According to the 'Guideline on the clinical assessment of fixed combinations of herbal substances/herbal preparations' (EMEA/HMPC/166326/2005)
[3] Only applicable to traditional use

I.1.2.2.1 Evidence regarding the indication/traditional use

Traditional indications for oral use of herbal teas liquid extracts (extraction solvent ethanol) and dry extracts

Nervous system disorders, psychiatric disorders:

Indication	References
Psychovegetative disturbances	Hamacher et al (2003), Hänsel (1993), Wichtl (2004), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (1998), Gruenwald et al (2004), Commission E monographs (1998), Dingermann et al (2004), DAC (1991-1999)
Mood depression	Hamacher et al (2003), Hänsel (1993), Bradley (2006), Irion (1955), Blaschek et al (2006), Hänsel et al (1993), Auster et al (1958), Gruenwald et al (1998), Gruenwald et al (2004), Commission E monograpsh (1998), Dingermann et al (2004), Hänsel et al (1988), DAC (1991-1999), Flamm et al (1940)
Mild depression	Escop 2003
Anxiety and/or nervous restlessness	Hänsel (1993), Wichtl (2004), Bradley 2006 (including menopausal anxiety), Blaschek et al (2006), Hänsel et al (1993), Madaus (1938), Gruenwald et al (1998), Gruenwald et al (2004), Barnes et al (2002), Commission E monographs (1998), Dingermann et al (2004), Hänsel et al (1988), DAC (1991-1999), Martindale (2002) (particularly if associated with the menopause)
Insomnia	Gerlach (2008), Irion (1955), List et al (1976), Madaus (1938), Barnes et al (2002), Hänsel et al (1988), Martindale (2002)
Support of emotional balance	Gerlach (2008), ESCOP (2003)
'Weak nerves', vegetative dystonia	Gerlach (2008), Irion (1955),
Agitation, excitability, irritability	List et al (1976), Barnes et al (2002), Upton (1997)
After mental efforts	Madaus (1938)
Neuralgia, Neurasthenia	List et al (1976), Madaus (1938), Barnes et al (2002), Flamm et al (1940), WHO monographs (2002)
Traumatic damages of nerves, paralysis	Madaus (1938)
Migraine, headache	Flamm et al (1940), WHO monographs (2002)
Sciatica	Barnes et al (2002), WHO monographs (2002)

Gastrointestinal disorders

Indication	References
Gastritis	Wichtl (2002), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (2004), Böhme (2006)
Ulcers, dyspepsia	WHO monographs (2002)
Unspecific catarrhs of the gastro-intestinal tact	Gerlach (2008), Irion (1955), Flamm et al (1940), Hänsel et al (1988)
Nervous gastric diseases	Gerlach (2008), Blaschek et al (2006), Hänsel (1993)

Abdominal pain	Madaus (1938)
Diarrhoea	Wichtl (1984), List et al (1976), Blaschek et al (2006), Hänsel et al (1993), Madaus (1938), Gruenwald et al (2004), Flamm et al (1940), Karsten et al (1962)
Haemorrhoids	Irion (1955), List et al (1976), Flamm et al (1940), WHO monographs (2002)

Hepatobiliary disorders

Indication	References
Gallbladder diseases	Wichtl (2004), Irion (1955), List et al (1976), Blaschek et al (2006), Hänsel et al (1993), Madaus (1938), Gruenwald et al (2004), Böhme (2006), WHO monographs (2002)
As a choleretic	Blaschek et al (2006), Hänsel et al (1993)

Renal and urinary disorders

Indication	References
Nocturnal enuresis	Gerlach (2008), Wichtl (1984), Irion (1955), List et al (1976), Blaschek et al (2006), Hänsel et al (1993), Madaus (1938), Gruenwald et al (2004), Flamm et al (1940), Dingermann et al (2004), Schneider (1990)
Kidney stones, bladder stones, inflammations of the urogenital tract	Irion (1955), Flamm et al (1940), WHO monographs (2002)
Irritable bladder, incontinence	Hamacher et al (2003), WHO monographs (2002)
As a diuretic	Wichtl (1984), List et al (1976), WHO monographs (2002)

Respiratory, thoracic and mediastinal disorders

Indication	References
Cough, bronchitis, asthma	Irion (1955), Blaschek et al (2006) Hänsel et al (1993), Madaus (1938), Gruenwald et al (2004), Flamm et al (1940), WHO monographs (2002)
Common cold	WHO monographs (2002)

Infections and infestations

Indication	References
Worm infestations	Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (2004), Flamm et al (1940), Zörnig (1911)
As an antimalaria agent	WHO monographs (2002)

Endocrine disorders

Indication	References
Polymenorrhoea, oligomenorrhoea, dysmenorrhoea, endometritis	Madaus (1938), Irion (1955), Flamm et al (1940), Heinrich et al (2004)
As emmenagogue	WHO monographs (2002)
Diabetes	Irion (1955), Auster et al (1958), WHO monographs (2002)

Musculoskeletal and connective tissue disorders

Indication	References
Rheumatism	Wichtl (1984), Blaschek et al (2006), Hänsel et al (1993), Auster et al (1958), Gruenwald et al (2004)

Metabolism and nutrition disorders

Indication	References
Gout	Wichtl (1984), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (2004)
Metabolic disorders	List et al (1976)

Traditional indications for oral use of liquid extracts (extraction solvent vegetable oil):

Indication	References
Dyspepsia	Hamacher et al (2001), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (1998), Gruenwald et al (2004), Commission E monographs (1998), DAC (1991)
Nervous gastric diseases	Gerlach (2008), Blaschek et al (2006), Hänsel et al (1993)
As a choleretic	Blaschek et al (2006), Hänsel et al (1993)
Abdominal pain	Madaus (1938)

Discussion and assessment of traditional indications for oral use:

Since the hypericum oil differs considerably in the nature of the constituents from aqueous and ethanolic liquid extracts these types of herbal preparations are discussed separately.

a) Aqueous extracts (herbal teas), liquid extracts prepared with ethanol, dry extracts:

Infusions prepared with water are widely used in the traditional medicine at least in Central Europe (Gerlach 2008). The indication ‘depression’ is unknown in traditional medicine; Hypericum is used in order to ‘strengthen the nerves’, to restore emotional balance. The wording which is found in literature reflects this fact, although put into different words. This traditional indication is plausible because of the pharmacological and clinical data which are available for isolated compounds and alcoholic extracts of Hypericum perforatum.

The numerous further traditional indications mentioned in the literature for these kinds of extracts are not plausible.

Since for the indication of a traditional herbal medicinal product terms like ‘depression’ or ‘depressed mood’ are not suitable, special care is taken to the wording.

Proposals:

Somatoform disorders (ICD-10 F45.0):

The main feature is repeated presentation of physical symptoms together with persistent requests for medical investigations, in spite of repeated negative findings and reassurances by doctors that the symptoms have no physical basis. If any physical disorders are present, they do not explain the nature and extent of the symptoms or the distress and preoccupation of the patient.

Neurasthenia (ICD-10 F48.0)

Considerable cultural variations occur in the presentation of this disorder, and two main types occur, with substantial overlap. In one type, the main feature is a complaint of increased fatigue after mental effort, often associated with some decrease in occupational performance or coping efficiency in daily tasks. The mental fatiguability is typically described as an unpleasant intrusion of

distracting associations or recollections, difficulty in concentrating, and generally inefficient thinking. In the other type, the emphasis is on feelings of bodily or physical weakness and exhaustion after only minimal effort, accompanied by a feeling of muscular aches and pains and inability to relax. In both types a variety of other unpleasant physical feelings is common, such as dizziness, tension headaches, and feelings of general instability. Worry about decreasing mental and bodily well-being, irritability, anhedonia, and varying minor degrees of both depression and anxiety are all common. Sleep is often disturbed in its initial and middle phases but hypersomnia may also be prominent.

Adjustment disorders (ICD-10 F43.2)

States of subjective distress and emotional disturbance, usually interfering with social functioning and performance, arising in the period of adaptation to a significant life change or a stressful life event. The stressor may have affected the integrity of an individual's social network (bereavement, separation experiences) or the wider system of social supports and values (migration, refugee status), or represented a major developmental transition or crisis (going to school, becoming a parent, failure to attain a cherished personal goal, retirement). Individual predisposition or vulnerability plays an important role in the risk of occurrence and the shaping of the manifestations of adjustment disorders, but it is nevertheless assumed that the condition would not have arisen without the stressor. The manifestations vary and include depressed mood, anxiety or worry (or mixture of these), a feeling of inability to cope, plan ahead, or continue in the present situation, as well as some degree of disability in the performance of daily routine. Conduct disorders may be an associated feature, particularly in adolescents. The predominant feature may be a brief or prolonged depressive reaction, or a disturbance of other emotions and conduct.

All these three types of disorders reflect partly the traditional oral use Hypericum. The definition of the somatoform disorders includes characteristics which are not suitable for THMPs. Particularly the persistent request for medical investigation is in contrast to the concept that the products are intended for use without medical supervision.
The traditional use seems to be covered in a most suitable way by the definition of neurasthenia.
The plausibility of the efficacy in this traditional indication is supported by an observational study (Grube et al 1996). Hypericum dry exctract LI 160 was administered corresponding to 900 µg Hypericine daily (= app. 540 mg extract) to patients with mild temporary depressed mood.
Additionally the indication should be clearly different from the proposed health claims for food supplements (contributes to emotial balance and general wellbeing; contributes to optimal relaxation; helps to support relaxation and mental and physical wellbeing; helps to maintain a healthy sleep; helps maintain a positive mood).

Proposed traditional indication (oral use, aqueous extracts, liquid ethanolic extracts, dry extracts)
Indication 1)
Traditional herbal medicinal product for the symptomatic relief of temporary neurasthenia (e.g. difficulty in concentrating, inefficient thinking, feeling of physical weakness and exhaustion after minimal effort, gastrointestinal discomfort).

The product is a traditional herbal medicinal product for use in the specified indication exclusively based upon long-standing use.

b) Liquid extracts prepared with vegetable oil (hypericum oil):

The indications mentioned in the references cannot be explained by the constituents of the hypericum oil, data from pharmacological experiments are lacking. Therefore the mentioned indications are not plausible. There is no traditional indication for the oral use of hypericum oil.

Traditional indications for cutaneous use of liquid extracts (extraction solvent vegetable oil):

Skin and subcutaneous tissue disorders

Indication	References
First degree burns, sunburn	Gerlach (2008), Wichtl (1984), Wichtl (2004), Bradley (2006), Irion (1955), Flamm et al (1940), Schneider (2004), Wagner et al (1999), Böhme (2006), WHO monographs (2002), Hamacher et al (2001), Hamacher et al (2006), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (1998), Gruenwald et al (2004), Comission E monographs (1998), DAC (1991), DAC (1998)
Wound healing	Gerlach (2008), Wichtl (1984), Wichtl (2004), Bradley (2006), Irion (1955), Hager (1878), Fischer et al (1902), Frerichs et al (1938), List et al (1976), Auster et al (1958), Flamm et al (1940), Wagner et al (1999), Schneider (1990), Karsten et al (1962), WHO monographs (2002), Hamacher et al (2001), Hamacher et al (2006), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (1998), Gruenwald et al (2004), Comission E monographs (1998), DAC (1991), DAC (1998), Madaus (1938)
Gingivits, stomatitis	Gerlach (2008), Hänsel et al (1972)
Fibrositis	Barnes et al (2002)
Neurodermatitis	Wagner et al (1999)
Eczema	Irion (1955)
Prevention of decubitus	Hänsel et al (1988), Hänsel et al (2007)
Shingles, viral infections	Bradley (2006), WHO monographs (2002)

Musculoskeletal and connective tissue disorders

Indication	References
Myalgia	Hamacher et al (2001), Hamacher et al (2006), Länger et al (2001), Blaschek et al (2006), Hänsel et al (1993), Gruenwald et al (1998), Gruenwald et al (2004), Comission E monographs (1998), DAC (1991), DAC (1998), Flamm et al (1940), Wagner et al (1999)
Rheumatism	Gerlach (2008), Irion (1955), Flamm et al (1940), Hager (1878), Blaschek et al (2006), Hänsel et al (1993), Wagner et al (1999), Auster et al (1958)
Lumbago	Blaschek et al (2006), Hänsel et al (1993), Flamm et al (1940), Madaus (1938)
Ischialigia, neuralgia	Irion (1955), Flamm et al (1940), Böhme (2006)

Articular pain	Gerlach (2008)
Sprains	Flamm et al (1940), Madaus (1938)
Bruises	Bradley (2006), Irion (1955), Flamm et al (1940)
Swellings	Bradley (2006), Flamm et al (1940), Madaus (1938)

Metabolism and nutrition disorders

Indication	References
Gout	Gerlach (2008), Irion (1955), Flamm et al (1940), Madaus (1938)

Traditional indications for cutaneous use of liquid extracts (extraction solvent ethanol):
Skin and subcutaneous tissue disorders

Wound healing	Gerlach (2008), Irion (1955)

Traditional indications for cutaneous use of liquid extracts (extraction solvent water):
Skin and subcutaneous tissue disorders

Wound healing	WHO monographs (2002)

Discussion and assessment of traditional indications for cutaneous use:
The traditional use of liquid preparations of hypericum for wound healing is supported by pharmacological data. Anti-inflammatory activity, analgesic activity, adstringent activity and antibacterial activity are documented, in vivo-data are poor, clinical data are lacking. In contrast the traditional use for the treatment of symptoms caused by an injury or related to rheumatism is not yet plausible. Antiviral effects are documented for several types of viruses, but not for Varicella zoster. Therefore the traditional use in the treatment of shingles cannot be supported.

Proposed traditional indication (cutaneous use, liquid extracts, extraction solvents vegetable oil, ethanol or water)
Indication 2)
Traditional herbal medicinal product for the symptomatic treatment of minor inflammations of the skin (such as sunburn) and as an aid in healing of minor wounds.
The product is a traditional herbal medicinal product for use in the specified indication exclusively based upon long-standing use.

I.1.2.2.2 Evidence regarding the specified posology

Oral administration

Comminuted herbal substance
The single dose for tea preparation is 1 teaspoon per cup, which is equivalent to 1.8 to 2 g of herbal substance. The recommendation of the daily dose is 1-2 cups equivalent to 1.8 to 4 g of herbal substance (Wichtl 2004, Hänsel et al 1993, ESCOP 2003), only few references recommend higher daily dosages (Duke 2002: up to 8 g; Madaus 1983: up to 7 g; Irion 1955 and Barnes et al 2002: up to 12 g).

Proposal for the posology of the comminuted herbal substance for tea preparation:
Adults, elderly: single dose 2 g; daily dose 4 g
Children and adolescents: Since no data on the safe traditional use in children are available the use in children and adolescents is not recommended.

The powdered herbal substance is in medicinal use in solid dosage forms longer than 30 years. The proposed posology (single dose 300-500 mg, daily dose 900-1000 mg) reflects the posology of the authorized products.

Herbal preparations:
Dry extracts: traditionally dry extracts are administered in considerably lower doses than those used for the treatment of mild to moderate depression. The herbal preparation type B was traditionally administered in a single dose of 120 mg and a daily dose of 360 mg. A daily dose of 252 mg showed no beneficial effects in the treatment of mild major depression compared to placebo. Therefore a single dose of 60-180 mg and a daily dose of 180-360 mg of dry extracts are proposed for traditional use.
Tincture (DER 1:5): 3-4.5 ml daily dose (Bradley 2006)
Tincture (DER 1:5) (= ESCOP 2003, posology with reference to a product from Steigerwald): 3-4.5 ml daily = equivalent to 0.6 – 0.9 g herbal substance.
Tincture (DER 1:10), 45% ethanol: 2-4 ml 3 x daily (Barnes et al 2006, Duke 2002) = equivalent to 0.6 – 1.2 g herbal substance.
Tincture (DER 1:2): Single dose 0.8-1.2 ml, daily dose 2.4-3.6 ml.
Tincture (DER not given): 10-15 drops (= app. 0.5-0.75 ml), 2-3 x daily (= 1-2.25 ml) (Madaus 1938). Because of the lack of data of the DER this posology cannot be considered.
The posology of the further mentioned herbal preparations reflects the posology of the authorized products.

Children and adolescents: Since no data on the safe traditional use in children are available the use in children and adolescents is not recommended.

Cutaneous administration
Strength of the preparations:
Hypericum oil is administered undiluted.
Hypericum tea prepared with a DER of 1:5 (WHO monographs 2002)

Children and adolescents: Since no data on the safe traditional use in children are available the use in children and adolescents is not recommended.

I.2 NON-CLINICAL DATA

I.2.1 Pharmacology

I.2.1.1 Overview of available data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

I.2.1.1.1 Effects associated with depression

The mechanisms of action as well as the responsible compounds of hypericum extracts are still under discussion. Several actions contributing to a clinical efficacy are reported: blockade of the reuptake of serotonin, noradrenalin and dopamine; up regulation of postsynaptic 5-HT_1 and 5-HT_2 receptors and of dopaminergic receptors; increased affinity for GABAergic receptors. Constituents which contribute to the activity are hypericin, pseudohypericin, flavonoids, and oligomeric procyanidins. The relevance of hyperforin is discussed controversially. As a consequence the entire extract has to be considered as the active substance.

Effects of constituents (primarily according to a review from Butteries et al 2003**)**

Flavonoids, Bioflavonoid

In-vitro the flavonols inhibit the monoaminooxidase (MAO), a fraction rich in flavonols also inhibited the catechol-O-methyltransferase (COMT). The concentrations which were necessary to achieve the results were considerably higher than concentrations which could be expected in therapeutic dosages.

Amentoflavon, which occurs only in traces in the flowers, inhibits in very low concentrations the binding of flumazenil on the benzodiazepine binding sites of the GABA receptor (Baureithel et al 1997) as well as to the δ-subunit of the opioid receptor. I3,II8 Biapigenin, which is present in much higher concentrations, is not investigated up to now, because it is commercially not available.

A flavonoid fraction, free of hypericins and hyperforin, induced a reduction of the β-receptors in the frontal cortex of the rat brain in similar manner like imipramin.

It is not known whether the flavonoids or their metabolites are responsible for these effects.

Hyperosid directly stimulates the endocytosis of β1-receptors and reduces their lateral mobility (Häberlein et al 2008).

Quercetin (10 – 40 mg/kg) altered dose-dependently the pattern in an electropharmacogram of rats, the changes resembled those of the MAO A inhibitor moclobemide and the MAO B inhibitor selegiline (Dimpfel 2008).

Naphthodianthrones

Pure naphthodianthrones are only poorly soluble in water. The solubility is increased by compounds of the extract. For example procyanidines are able to increase solubility in water 10-fold. Rutin and hyperosid may also contribute to the better solubility of hypericin in extracts.

Hyperosid and the procyanidines increase the serum level of hypericin considerably.

Isolated hypericin has no influence on the MAO A and B. In-vitro it inhibits the dopamine-β-hydroxylase, it interacts with β-adrenergic receptors and possesses a high affinity to the σ-receptor as well as to the D_3-receptor.

Hypericin alters the concentration of several neurotransmitters after 8 weeks of treatment comparable to imipramine. 8 weeks treatment also increased the serotonin concentration in the hypothalamus, while the concentration of metabolites of dopamine is reduced. Hypericin induces a reduction of the β-receptors in he frontal cortex of the rat brain, after 8 weeks of treatment a significant change could be observed.

Hypericin reduced, similar to imipramine, the expression of mRNA of the corticotrophin releasing hormone in the hypothalamus as well as the ACTH and corticosterone levels in the plasma of rats. An overview presenting chemical and biological properties of hypericin is published by Lavie et al (1995).

Phloroglucinol derivatives

Müller et al (1998) and Chatterjee et al (1998) revealed that hyperforin plays an important role in the inhibition of neurotransmitter reuptake. In vitro the IC_{50} value for the inhibition of the synaptosomal reuptake of serotonin, dopamine, noradrenaline, GABA and glutamate caused by hyperforin is in the range of 80 to 1234 nM. After a daily intake of 900 mg of hypericum extract a blood level of 180 nM of hyperforin was detected. Therefore it could be concluded that sufficient amounts of hyperforin for the inhibition of the reuptake of some neurotransmitters could be achieved during therapy.

Several mechanisms for these effects are in discussion: inhibition of calcium channels of the P-type, a reserpin-like action, increase of the intracellular sodium concentration.

Isolated hyperforin did not influence the density of β-receptors in rats, while an extract rich in hyperforin induced a down-regulation of the β-receptor density.

Hyperforin directly causes the endocytosis of β1-receptors and reduces their lateral mobility (Häberlein et al 2008).

Xanthones
Due to the low content of xanthones in the herbal substance (about 0.0004%) it is not likely that the experimentally documented inhibition of MAO A and B is of clinically relevance.

Effects of extracts (examples of publications)
The results on MAO inhibition are controversial. At concentration of 10^{-4} to 10^{-3} mol/l of a methanolic extract (Bladt et al 1994) and of a methanolic extract (Thiede et al 1994) MAO could be inhibited up to 82%. In a later study a methanolic extract exhibited a rather weak potency as MAO inhibitor in vitro (Müller et al 1997).

The inhibition of synaptosomal reuptake of several neurotransmitters could be demonstrated for different kinds of extracts (different extraction solvent, different amounts of hypericin, hyperforin).

Long-term treatment of rats with a methanolic extract (500 mg/kg p.o.) significantly increased the 5-HT levels in the hypothalamus of rats; the 5-HT turnover was significantly lowered in hippocampus and hypothalamus (Butterweck et al 2002).
Misane et al (2001) found that an ethanolic extract given in high doses affects the neuronal 5-HT uptake more like tricyclic antidepressants than SSRIs.
Calapai et al (2001) investigated an extract standardised to 50% flavonoids, 0.3% hypericin and 4.5% hyperforin. After acute oral administration (250 – 500 mg/kg) dose-dependently the contents of 5-HT and 5-hydroxyindolacetic acid (5-HIAA) were significantly enhanced in all brain regions examined. Noradrenalin and dopamine levels were significantly increased in the diencephalon; in the brainstem only noradrenalin was significantly enhanced.
The down regulation of β-receptors could be demonstrated with an ethanolic extract (0.2% hypericin) dose dependently (Kientsch et al 2001).
A methanolic extract (4.5% hyperforin) interacted with a GABA A receptor, an extract rich in hyperforin did not show an interaction. Data on the inhibition of specific bindings to the dopamine transporters indicate that the hyperforin content cannot explain effects of extracts on receptors (Gobbi et al 2001).
A methanolic extract could also inhibit the binding of flumazenil to the benzodiazepine binding site of the GABA A receptor in vitro.
Simbrey et al (2004) used quantitative radioligand receptor binding studies to examine the effects of short-term (2 wks) and long-term (8 wks) administration of different hypericum extracts and constituents on β-adrenergic binding in rat frontal cortex. The effects were compared to those of the standard antidepressants imipramine and fluoxetine. A lipophilic CO2 extract decreased beta-AR-binding (13%) after two weeks and slightly increased the number of β-receptors after 8 weeks (9%). Short-term treatment with the methanolic hypericum extract decreased β-receptor-binding (14%), no effects for this extract were observed after 8 weeks. Treatment with hypericin led to a significant down-regulation (13%) of β-receptors in the frontal cortex after 8-weeks, but not after 2 weeks, while hyperforin (used as trimethoxybenzoate), and hyperoside were ineffective in both treatment paradigms. Compared to the hypericum extracts and single compounds the effect of imipramine on β-receptor-binding was more pronounced in both treatment paradigms.
Teufel-Mayer et al (1997) could demonstrate that a methanolic extract significantly increased the numbers of 5-HT receptors, whereas the affinity of these receptors remained unchanged.
Hypericum extract inhibited the binding of naloxone to the μ- and κ-opioid receptor (IC_{50} values 25 and 90 μg/ml). Isolated flavonoids like quercetin, kaempferol as well as quercitrin

did not inhibit naloxone binding. The authors suggest that this inhibition may contribute to the anti-depressant activity (Simmen et al 1998).

I.2.1.1.2 Antidepressant activity in animal models

Forced swimming test

In the FST pure naphthodianthrones were active in high concentrations only, after addition of a fraction rich in procyanidines (which is itself inactive in the FST) was hypericin active in a concentration of 0.009 mg/kg (Butterweck et al 1998).
Hyperosid, Isoquercitrin and miquellianin showed activity in low concentrations (0.6 mg/kg) in the FST. Rutin, when administered as isolated compound, has no activity in the FST. However, extracts with a low content of rutin show only a weak activity in the FST, but when pure rutin is added to the extract, an activity could be demonstrated. Unfortunately the contents of the naphthodianthrones were not presented in the study (Butterweck et al 2000).
Daily administration of 3 mg/kg isorhamnetin, a main metabolite from quercetin, for 9 days induced a statistically significant decrease in the immobility time in the forced swimming test (Paulke et al 2008).
Noldner et al (2002) found that an ethanolic extract (doses 30-300 mg/kg, 7 days oral treatment) shortened the spent time immobile in a dose dependent manner, while a methanolic extract was inactive. After addition of rutin to the methanolic extract a strong effect comparable to the ethanolic extract could be demonstrated.
Beijamini et al (2003) investigated the same methanolic extract. The rats received 150 to 500 mg/kg extract in 3 portions starting 24 hours before the experiment. All doses significantly reduced the immobility time.
Two different hydroethanolic extracts (4.5% and 0.5% hyperforin) and a stable salt of hyperforin were tested in the forced swimming test by Cervo et al (2002). All test substances caused a significant reduction of immobility; the effect was more pronounced in the extract containing more hyperforin. The authors therefore conclude that hyperforin plays an important role in the antidepressant-like activity.
Calapai et al (2001) found that an extract standardised to 50% flavonoids, 0.3% hypericin and 4.5% hyperforin was able to reduce the immobility time by about 30-40% compared to control. The effect was antagonised by the coadministration of sulpiride and metergoline. The effect was also significantly ower in animals pre-treated with 6-hydroxydopamine, which destroys noradrenergic neurons. The authors suggest that the action is probably mediated by serotonergic, noradrenergic and dopaminergic system activation.

Learned-helplessness paradigm

Chatterjee et al (1998) compared an ethanolic extract (4.5% hyperforin) and a supercritical CO2 extract (38.8% hyperforin). Oral doses of 300 mg/kg/day of the ethanolic extract and 30 mg/kg/day of the CO2 extract were almost equieffective to 10 mg/kg/day of imipramine (52% reduction).

Model of escape deficit

Usai et al (2003) compared 2 hydroethanolic extracts (4.5% hyperforin, 7.47% hyperforin). Both extracts exhibit a strong protective effect to prevent the development of escape deficit in rats. The extract with 4.5% hyperforin was effective in doses 8 times lower than of the other extract.

I.2.1.1.3 Anxiolytic effects

Kumar et al (2000) reported anxiolytic effects of a hypericum extract (ethanol 50%) of Indian origin in animal models, the dosage was relatively high (100 and 200 mg / kg p.o.).

Flausino et al (2002) investigated the effects of acute and chronic oral treatment with Hypericum perforatum L. (HP LI 160, 62.5-500 mg/kg) in rats submitted to different anxiety models: the elevated T-maze (for inhibitory avoidance and escape measurements), the light/dark transition, and the cat odor test. These models were selected for their presumed capacity of evidencing specific subtypes of anxiety disorders as recognized in clinical practice. The results showed that acute HP (125 mg/kg) impaired elevated T-maze inhibitory avoidance, an anxiolytic effect, without altering escape performance. Chronic HP (250 mg/kg) enhanced avoidance latencies only in animals that were preexposed to the open arms of the maze. Preexposure shortens escape latency, improving it as an escape index. Differently from the reference drug imipramine (IMP, 15 mg/kg), chronic HP did not impair escape from the open arms of the maze. On the other hand, similarly to IMP, the extract increased the number of transitions between the two compartments in the light/dark transition model. Treatment regimens with HP and IMP did not alter behavioral responses of rats to a cloth impregnated with cat odor. These observations suggest that HP LI 160 exerts anxiolytic-like effects in a specific subset of defensive behaviors, particularly those related to generalized anxiety.

The aim of a study from Beijamini et al (2003) was to evaluate the putative antipanic/anxiolytic effect of standardised H. perforatum extract (LI 160) on rats tested in the elevated T-maze, an animal model of innate (panic) and learned (generalised) anxiety, at doses that exhibit antidepressant-like activity. H. perforatum (150, 300 and 500 mg/kg, administered orally 24, 18 and 1h before the test) decreased the immobility time in the forced swim test. Rats were treated orally with H. perforatum (150 or 300 mg/kg) or paroxetine (5mg/kg) 24, 18, and 1h before being tested in the elevated T-maze (subacute treatment). Immediately after this test, the animals were submitted to the open field to evaluate locomotor activity. Paroxetine was used as a positive control. Other groups of animals were submitted to the same drug treatment for 7 days (subchronic treatment). Paroxetine (5mg/kg) impaired inhibitory avoidance after subacute treatment, while subchronic administration increased one-way escape latency. Subacute treatment with H. perforatum (300 mg/kg) exerts a partial anxiolytic-like effect in the inhibitory avoidance task. Repeated administration of H. perforatum (300 mg/kg) induced an anxiolytic effect (decreased inhibitory avoidance) and an antipanic effect (increased one-way escape). No effect on locomotor activity was found with any treatment. Thus, the results suggest that H. perforatum extract could exert an anxiolytic and antipanic effect.

Grundmann et al (2006) used exposure to an open field (OF) as inescapable stressor to mice. Exposure of male BL6/C57J mice to OF stress significantly increased body temperature (DeltaT = 1.8 +/- 0.13 degrees C, $p < 0.05$). Anxiolytic drugs (the benzodiazepine diazepam; 5 mg/kg, and the 5HT (1A) receptor agonist buspirone; 10 mg/kg) significantly reduced DeltaT, whereas antidepressants (imipramine and fluoxetine) had no effect on DeltaT. Oral administration of hypericum extract significantly reduced DeltaT in doses of 250 and 500 mg/kg. Higher (750 and 1000 mg/kg) as well as a lower dose (125 mg/kg) did not affect DeltaT after stress, indicating a U-shaped dose-response curve. Hypericin (0.1 mg/kg, p. o.) administered 60 min prior to testing significantly decreased DeltaT ($p < 0.05$) whereas hyperforin (1 - 10 mg/kg, p. o.) had no effect in this test paradigm. The flavonoids hyperoside, isoquercitrin and quercitrin (all at 0.6 mg/kg, p. o.) and rutin (1 mg/kg, p. o.) only partially blocked OF-induced hyperthermia. If compared to all other flavonoids, the quercetin 3-O-glucuronide miquelianin (1.2 mg/kg, p. o.) was the most potent compound tested in this experimental design. From the biflavonoids in hypericum, only amentoflavone decreased SIH-induced hyperthermia in a dosage of 0.1 mg/kg. These findings could be interpreted as putative anxiolytic effects of hypericum extract and single consituents.

I.2.1.1.4 Neuroprotection, memory impairment, nootropic effects

Kaltschmidt et al (2002) analyzed the effect of hypericin on NF-κB activation. Hypericin alone was able to induce short-time activation of NF-κB, which declined to basal levels after 24 h. Cell death was induced by hypericin at a concentration of 10 µM. A profound synergistic action in inducing apoptosis was detected in co-treatment of hypericin together with $FeSO_4$. In contrast, hypericin in low concentrations was able to partly prevent cell death induced by amyloid-beta-peptide (Abeta). Hypericin (10 µM) synergistically enhanced Abeta neurotoxicity. The data describe NF-κB in the same primary neuronal culture as stimulus-dependent, anti-apoptotic, or pro-apoptotic factor.

Extracts of Hypericum perforatum exhibited upgrading and significant protective effects on the trauma of PC12 cells induced by 200 µM H_2O_2 in a dose-dependent manner within 24-hour treatment (Lu et al 2004). Cell viability was assessed by the MTT method, and in situ cellular hydrogen peroxide (H_2O_2)-induced oxidative stress was examined by measurement of reactive oxygen species (ROS) formation using CDCFH procedures. Intra- and extra-cellular ROS levels decreased significantly to 71.9% and 50.0% of the control at a moderate concentration of 20 µg/ml, respectively, suggesting that hypericum extract could easily enter the cells and play important roles in reducing ROS levels. Our results were proved by detection of DNA fragmentation and inspection of cell morphology of PC12 cells. Hypericum extract can obviously block DNA fragmentation and prevent the cells from shrinking and turning round of H_2O_2-induced apoptosis in PC12 cells at concentrations of 10 approximately 100 µg/ml. The authors conclude that hypericum extracts may be a candidate for application in neurodegenerative diseases such as Alzheimer's disease or Parkinson's disease.

Genovese et al (2006) evaluated the effect of H. perforatum (given at 30 mg / kg) in an experimental animal model of spinal cord injury, which was induced by the application of vascular clips to the dura via a four-level T5 through T8 laminectomy. The degree of (a) spinal cord inflammation and tissue injury (histological score), (b) nitrotyrosine, (c) poly(adenosine diphosphate-ribose), (d) neutrophils infiltration, and (e) the activation of signal transducer and activator transcription 3 was markedly reduced in spinal cord tissue obtained from H. perforatum extract-treated mice. It could be demonstrated that H. perforatum extract significantly ameliorated the recovery of limb function.

Froestl et al (2003) studied the effect of hyperforin on the processing of the amyloid precursor protein (APP) in rat pheochromocytoma PC12 cells, stably transfected with human wildtype APP. The authors observed transiently increased release of secretory APP fragments upon hyperforin treatment. Unique features, like a strong reduction of intracellular APP and the time course of soluble APP release, distinguished the effects of hyperforin from those of alkalizing agents and phorbol esters, well known activators of secretory processing of APP. Carbonyl cyanide 4-(trifluoromethoxy)phenylhydrazone (FCCP), a protonophore, induced an almost identical decrease in intracellular pH in PC12 cells as does hyperforin. Despite this, FCCP induced a less pronounced release of soluble APP fragments and only slightly reduced intracellular APP levels. These results suggest that hyperforin is an activator of secretory processing of APP.

Silva et al (2004) assessed the neuroprotective role of a Hypericum perforatum ethanolic extract and obtained fractions in amyloid-beta peptide (Abeta)((25-35))-induced cell death in rat cultured hippocampal neurons. Lipid peroxidation was used as a marker of oxidative stress by following the formation of TBARS in rat cortical synaptosomes, after incubation with ascorbate/Fe2+, alone or in the presence of EC97 effective concentrations of H. perforatum fractions. Induced lipid peroxidation was significantly inhibited by fractions containing

flavonol glycosides, flavonol and biflavone aglycones, and by a fraction containing several phenols, mainly chlorogenic acid-type phenolics (21%, 77% and 98%, respectively). Lipid peroxidation evaluated after incubation with 25 μM Abeta(25-35), was significantly inhibited by H. perforatum extract. Cell viability was assessed by use of the Syto-13/PI assay. The total ethanolic extract (TE) and fractions containing flavonol glycosides, flavonol and biflavone aglycones, reduced Abeta(25-35)-induced cell death (65%, 58% and 59%, respectively). These results were further supported by morphological analysis of cells stained with cresyl violet. Peptide beta-amyloid(25-35) induced a decrease in cell volume, chromatin condensation and nuclear fragmentation, alterations not evident in the presence of the TE and fractions containing hypericins (hypericin concentration = 11.02 μM), or fractions containing flavonoids (quercetin concentration = 21.13 μM). Dendritic lesion, an evidence of neurodegeneration, was observed by neuronal staining with cobalt following insult with Abeta(25-35), but prevented after exposure to the peptide plus the fractions referred above. The results suggest that H. perforatum extracts may be endowed with neuroprotective compounds able to prevent Abeta(25-35)-induced toxicity.

The major protein constituent of amyloid deposits in Alzheimer's disease (AD) is the amyloid beta-peptide (Abeta). Dinamarca et al (2006) have determined the effect of hyperforin on Abeta-induced spatial memory impairments and on Abeta neurotoxicity. Hyperforin decreases amyloid deposit formation in rats injected with amyloid fibrils in the hippocampus, decreases the neuropathological changes and behavioral impairments in a rat model of amyloidosis, and prevents Abeta-induced neurotoxicity in hippocampal neurons both from amyloid fibrils and Abeta oligomers, avoiding the increase in reactive oxidative species associated with amyloid toxicity. Both effects could be explained by the capacity of hyperforin to disaggregate amyloid deposits in a dose and time-dependent manner and to decrease Abeta aggregation and amyloid formation.

Acute administration of Hypericum extract (4.0, 8.0, 12.0, and 25.0 mg/kg i.p.) in mice before retrieval testing increased the step-down latency during the test session. The same doses of Hypericum extract, on the other hand, failed to reverse scopolamine-induced amnesia of a two-trial passive avoidance task. Pretreatment of the animals with serotonergic 5-HT1A receptor antagonist (-)-pindolol (0.3, 1.0, and 3.0 mg/kg), serotonergic 5-HT2A receptor blocker spiperone (0.01, 0.03, and 0.1 mg/kg), alpha adrenoceptor antagonist phentolamine (1, 5, and 10 mg/kg), beta receptor antagonist propranolol (5, 7.5, and 10 mg/kg), dopaminergic D1 receptor antagonist SCH 23390 (0.01, 0.05, and 0.1 mg/kg), and dopaminergic D2 receptor antagonist sulpiride (5, 7.5, and 10 mg/kg) revealed the involvement of adrenergic and serotonergic 5-HT1A receptors in the facilitatory effect of Hypericum extract on retrieval memory. It is concluded that Hypericum extract may be a better alternative for treatment of depression commonly associated with dementia than other antidepressants known to have anticholinergic side effects causing delirium, sedation and even exacerbating already existing impaired cognition (Khalifa 2001).

The effects of a Hypericum extract and hyperforin sodium salt were evaluated in rat and mouse avoidance tests by Klusa et al (2001). In a conditioned avoidance response (CAR) test on the rat, oral daily administration of hyperforin (1.25 mg/kg/day) or of the extract (50 mg/kg/day) before the training sessions considerably improved learning ability from the second day onwards until the day 7. In addition, the memory of the learned responses acquired during 7 consecutive days of administration and training was largely retained even after 9 days without further treatment or training. The observations made using different doses indicate that these learning-facilitating and/or memory-consolidating effects by the agents follow inverse U-shaped dose-response curves in dose ranges lower than (for hyperforin) or equal to (for Hypericum extract) their effective dose in the behavioral despair test for

antidepressants. In a passive avoidance response test on the mouse, a single oral dose (1.25 mg/kg) of hyperforin not only improved memory acquisition and consolidation, but also almost completely reversed scopolamine-induced amnesia. The single Hypericum extract dose tested (25 mg/kg) did not reveal any significant effects in the passive avoidance response (PAR) test on the mouse. These observations suggest that the Hypericum extract could be a novel type of antidepressant with memory enhancing properties, and indicate that hyperforin is involved in its cognitive effects. Pure hyperforin seems to be a more potent antidementia agent than an antidepressant.

Kumar et al (2000, 2002) found that orally administered extracts of Hypericum perforatum of Indian origin (doses 100 mg and 200 mg /kg) to rats showed effects which could be interpreted as possible nootropic action.

Widy-Tyszkiewicz et al (2002) investigated the effects of long-term Hypericum perforatum treatment on spatial learning and memory in rats. A hypericum powder (HP) standardized to 0.3% hypericin content was administered orally for 9 weeks in doses of 4.3 and 13 µg/kg corresponding to therapeutic dosages in humans of 0.3 and 0.9 mg of total hypericins daily. A Morris water maze paradigm was used. The mean escape latency over 4 d for the Control group (21.9 s) and HP 4.3 group (21.7 s) was significantly greater than the latency of the HP 13 group (15.8s). In the probe trial on day 5, the HP 13 group crossed the correct annulus in the SE quadrant more often (4.5) than the other groups: Con (2.4) and HP 4.3 (3.1). After completion of the behavioural experiment, the regional brain concentrations of monoamines and metabolites were estimated in selected brain regions, i.e. prefrontal cortex, hippocampus and hypothalamus. Significant differences in the content of monoamines and metabolites between the treatment groups compared to the control were detected. The increased 5-hydroxytryptamine (5-HT) levels in the prefrontal cortex correlated positively with the retention of spatial memory. These findings show that the long-term administration of Hypericum perforatum can improve learning and spatial memory with significant changes in the content of monoamines in several brain regions.

Administration of Hypericum perforatum (350 mg / kg daily for 21 days) significantly enhanced recall of passive avoidance behavior (PAB), but had no effect on the acquisition of conditioned avoidance responses (CARs). Rats stressed chronically (2 h daily for 21 days) displayed diminished recall of the PAB and this effect was abolished by St John's wort. Chronic administration of the "equivalent" to the stress dose of exogenous corticosterone (5 mg / kg daily for 21 days) also impaired recall of PAB, and this effect was also reversed by Hypericum perforatum. None of the treatments produced significant motor coordination impairments as tested in a 'chimney' test. It appears that H. perforatum prevents stress-induced deterioration of memory in rats (Trofimiuk et al 2005, 2006).

Mohanasundari et al (2006, 2007) tested the effect of an extract of Hypericum perforatum in chemically induced Parkinson’s disease in mice. Treatment with Hypericum perforatum extract resulted in an inhibition of monoamine oxidase-B activity and reduced astrocyte activation in striatal area. The effects were more pronounced when hypericum was combined with bromocriptine.

Sanchez Reus et al (2007) designed a study to investigate the pro-oxidant activity of rotenone, the protective role of standardized extract of Hypericum perforatum (SHP), as well as the mRNA levels of antioxidant enzymes, in brain homogenates of rats following exposure to rotenone and SHP extract. Quercetin in liposomes, one active constituent, was tested in the same experimental conditions to serve as a positive control. The animals received

pretreatment with SHP (4 mg/kg) or quercetin liposomes (25 and 100 mg/kg) 60 min before of rotenone injection (2 mg/kg). All treatments were given intraperitoneally in a volume of 0.5 ml/kg body weight, for 45 days. SHP extract exerted an antioxidant action which was related to a decrease of MnSOD activity and mRNA levels of some antioxidant enzymes evaluated. One possible mechanism of action of SHP extract may be related to quercetin in protecting neurons from oxidative damage.

El-Sherbiny et al (2003) investigated the effect of acute administration of Hypericum perforatum extract (4.0, 8.0, 12.0, and 25.0 mg/kg ip) on the brain oxidative status of naive rats treated with an amnestic dose of scopolamine. The results showed that the administration of 1.4 mg/kg of scopolamine impaired the retrieval memory of rats. Pretreatment of the animals with Hypericum extract (4, 8, and 12 mg/kg) resulted in an antioxidant effect through altering brain malondialdehyde, glutathione peroxidase, and/or glutathione level/activity.

I.2.1.1.5 Support in smoking cessation

Catania et al (2003) investigated the effects of an extract of Hypericum perforatum (Ph-50) on withdrawal signs produced by nicotine abstinence in mice. Nicotine (2 mg/kg, four injections daily) was administered for 14 days to mice. Different doses of Ph-50 (125-500 mg/kg) were administered orally immediately after the last nicotine injection. In another experiment, Ph-50 (500 mg/kg) was orally administered in combination with nicotine, i) starting from day 8 until the end of the nicotine treatment period, or ii) during nicotine treatment and after nicotine withdrawal, or iii) immediately after the last nicotine injection. The locomotor activity reduction induced by nicotine withdrawal was abolished by Ph-50, which also significantly and dose-dependently reduced the total nicotine abstinence score when injected after nicotine withdrawal.

Mannucci et al (2007) investigated the possible involvement of 5-HT in the beneficial effects of H. perforatum on nicotine withdrawal signs. With the aim to induce nicotine dependence, nicotine (2 mg/kg, four intraperitoneal injections daily) was administered for 14 days to mice (NM). After nicotine withdrawal the measurement of 5-HT metabolism in the cortex showed a reduction of the 5-HT content while animals treated only with Hypericum extract showed a significant reduction of total abstinence score compared to controls. A selective 5-HT receptor antagonist inhibited the reduction of total abstinence score induced by H. perforatum. Moreover, 5-HT1A expression has been evaluated 30 days after nicotine withdrawal. The results show a significant increase of cortical 5-HT content in NM treated with H. perforatum, with a concomitant significant increase of 5-HT1A receptor.

I.2.1.1.6 Treatment of alcoholism

Several studies were performed in order to investigate the influence of hypericum extracts on alcohol intake in animals (Rezvani et al 1999, Perfumi et al 1999, Perfumi et al 2005, Perfumi et al 2005a, Coskun et al 2006). All studies report beneficial effects on ethanol withdrawal symptoms, additionally a reduction of alcohol intake in alcohol preferring animals was observed. Hypericum extracts and opioid receptor antagonists act synergistically (Perfumi et al 2003).

The changes in the alcohol drinking behaviour may be caused by hyperforin (Perfumi et al 2001, Wright et al 2003). In contrast, De Vry et al (1999) reported a reduction in alcohol

preference after administration of an extract with very low hyperforin content. Clinical data are missing (Uzbay 2008).

I.2.1.1.7 Antibacterial activity

Gibbons et al (2002) screened extracts of 34 species and varieties of the genus Hypericum for activity against a clinical isolate of methicillin-resistant Staphylococcus aureus, which in addition possessed a multidrug efflux mechanism conferring a high level of resistance to therapeutically useful antibiotics. Thirty-three of the 34 chloroform extracts showed significant activity in a disk diffusion assay, and five extracts had minimum inhibitory concentrations of 64 µg/ml.

I.2.1.1.8 Antiinflammatory activity

Schempp (2000) investigated the alloantigen presenting function of human epidermal cells (EC) exposed to Hypericum ointment in vivo in a mixed EC lymphocyte reaction (MECLR). The effect of Hypericum ointment was compared with the immunosuppressive effect of solar-simulated radiation (SSR). Subsequently, the authors tested purified hyperforin in vivo and in vitro in a MECLR to evaluate its possible contribution to the effect of the Hypericum ointment. Furthermore, the effect of hyperforin on the proliferation of peripheral blood mononuclear cells (PBMC) in vitro was assessed. Compared with untreated skin, treatment with Hypericum ointment resulted in a significant suppression of the MECLR (P </= 0.001) that was similar to the effect of SSR. The combination of Hypericum ointment plus SSR was not significantly different from either treatment alone. EC isolated from skin treated with the hyperforin containing ointment also showed a reduced capacity to stimulate the proliferation of allogeneic T cells (P </= 0.001). Similarly, in vitro incubation of EC with hyperforin suppressed the proliferation of alloreactive T cells (P </= 0.001). Furthermore, hyperforin inhibited the proliferation of PBMC in a dose-dependent manner, without displaying pronounced toxic effects as determined by Trypan blue staining. The results demonstrate an inhibitory effect of Hypericum extract and of its metabolite hyperforin on the MECLR and on the proliferation of T lymphocytes that may provide a rationale for the traditional treatment of inflammatory skin disorders with Hypericum extracts.

Three preparations of Hypericum perforatum L. (a hydroalcoholic extract, a lipophilic extract and an ethylacetic fraction) and the pure compounds hypericin, adhyperforin, amentoflavone, hyperoside, isoquercitrin, hyperforin dicyclohexylammonium (DHCA) salt and dicyclohexylamine were evaluated for their topical anti-inflammatory activity (Sosa et al 2007). H. perforatum preparations provoked a dose-dependent reduction of Croton-oil-induced ear oedema in mice, showing the following rank order of activity: lipophilic extract > ethylacetic fraction > hydroalcoholic extract (ID50 220, 267 and >1000 microg cm^{-2}, respectively). Amentoflavone (ID50 0.16 micromol cm^{-2}), hypericin (ID50 0.25 micromol cm^{-2}), hyperforin DHCA salt (ID50 0.25 micromol cm^{-2}) and adhyperofrin (ID50 0.30 micromol cm^{-2}) had anti-inflammatory activity that was more potent or comparable to that of indometacin (ID50 0.26 micromol cm^{-2}), whereas isoquercitrin and hyperoside were less active (ID50 about 1 micromol cm^{-2}). As dicyclohexylamine alone was inactive, the effect of hyperforin DHCA salt can be attributed completely to the phloroglucinol moiety. The pharmacological activity and phytochemical profile of the tested extracts and fraction suggest that different constituents are involved in the topical antiphlogistic property of H. perforatum in-vivo.

Eckert et al (2001) tested the effects of hyperforin on the fluidity of crude brain membranes from young guinea pigs. Hyperforin modifies specific membrane structures in different ways. It decreases the flexibility of fatty acids in the membrane hydrocarbon core, but fluidizes the hydrophilic region of membrane phospholipids. Relatively low concentrations of hyperforin (0.3 µmol/l) significantly decreased the annular fluidity of lipids close to membrane proteins. These findings are remarkable, as inhibition of several neurotransmitter-uptake systems and modulation of several ionic conductance mechanisms by hyperforin occur in the same concentration range. However, bulk fluidity was unchanged by this low hyperforin concentration. The results emphasise a physicochemical interaction of hyperforin with specific membrane structures that probably contribute to its pharmacological properties.

I.2.1.1.9 Wound healing

The wound-healing effect of St. John's Wort (Hypericum perforatum L.) extract was evaluated by comparing with dexpanthenol and titrated extract of Centella asiatica (TECA) on cultured chicken embryonic fibroblasts (Ozturk et al 2006). Chicken embryonic fibroblasts from fertilized eggs were incubated with the plant extract, dexpanthenol and TECA. The wound-healing activity of Hypericum perforatum extract seems to be mainly due to the increase in the stimulation of fibroblast collagen production and the activation of fibroblast cells in polygonal shape, which plays a role in wound repair by closing damaged area. The findings demonstrated the wound-healing activity of Hypericum perforatum, which has previously been based on ethnomedical data.

I.2.1.1.10 Pregnancy, lactation

Rayburn et al (2001) investigated the cognitive impact of prenatal exposure to hypericum in CD-1 mice. Hypericum (182 mg/kg/day) or a placebo was consumed in food bars for 2 weeks before mating and throughout gestation. The hypericin content in the hypericum formulation was in the middle range of standardized hypericum products. One offspring per gender from each litter (hypericum 13, placebo 12) was tested on each of the following tasks: juvenile runway with adult memory, adult Morris maze, adult passive avoidance, or adult straight water runway followed by a dry Cincinnati maze. Learning occurred in both genders in all tasks ($P<.003$) with no significant differences between treatments at the final trial. Female offspring exposed to hypericum, rather than to a placebo, required more time to learn the Morris maze task ($P<.05$). Postlearning sessions did not show any significant differences. In conclusion, prenatal exposure to a therapeutic dose of hypericum did not have a major impact on certain cognitive tasks in mice offspring.

I.2.1.1.11 Photodynamic therapy

Hypericin is considered as potential agent in the photodynamic therapy of cancer (Agostinis et al 2002). However, since only isolated hypericin and not extracts have been tested this approach is out of the scope of this assessment.

I.2.1.1.12 Other effects

Capasso et al (2005) evaluated the effect of hypericum on rat and human vas deferens contractility. Hypericum extract (1 to 300 microM) decreased in a concentration dependent

manner the amplitude of electrical field stimulation and agonist induced contractions with the same potency, suggesting direct inhibition of rat vas deferens smooth muscle. Of the chemical constituents of Hypericum extract tested hyperforin but not hypericin or the flavonoids quercitrin, rutin and kaempferol inhibited phenylephrine induced contractions. Hypericum extract and hyperforin also inhibited phenylephrine induced contractions in human vas deferens. These results might explain delayed ejaculation described in patients receiving Hypericum extract.

Effects of different extracts of Hypericum perforatum L. on the kindling epileptic discharges were analyzed by Ivetic et al (2002). The experiment was carried out on Chinchilla rabbits with chronically implanted electrodes in cortical structures and hippocampus. Water, n-butanol and ether fractions (mass concentrations 0.1 g/ml) of crude ethanol extract of Hypericum perforatum were used. The particular extracts were given intramuscularly in single dose of 1 ml/kg BW. The bioelectric activity was registered before and after applications of each extracts. The obtained results show that the effect depends on the constituents present in particular fractions. Most polar constituents that remained in water fraction exerted highest antiepileptic activity in all (100%) animals tested. Substances present in butanol fraction repressed the epileptic manifestations in 40% of animals with kindling epilepsy, whereas lipid-soluble constituents in ether fraction potentiated the epileptic activity.

Hypericin, pseudohypericin and hyperforin at doses as low as 2.5 µmol/l are potent antioxidants in the LDL oxidation systems used. These results indicate that these derivatives have possible antiatherogenic potential (Laggner et al 2007). An extract rich in flavonoids lowered serum levels of total cholesterol, total triglycerides and LDL (Zou et al 2005).

Free radical scavenging and antioxidant activities of a standardized extract of Hypericum perforatum were examined for inhibition of lipid peroxidation, for hydroxyl radical scavenging activity and interaction with 1,1-diphenyl-2-picrylhydrazyl stable free radical (DPPH) by Benedi et al (2004). The results suggest that hypericum extracts shows relevant antioxidant activity both in vitro and in a cell system, by means of inhibiting free radical generation and lipid peroxidation.

Antioxidative and radical scavenging effects are also reported for the flavonoid fraction of Hypericum perforatum (Zou et al 2004) and for several commercially available formulations (Hunt et al 2001). The antioxidative properties protect human neuroblastoma cells against induced apoptosis (Jang et al 2002).

Genovese et al (2006) evaluated the effect of Hypericum perforatum extract on acute pancreatitis induced by cerulein administration in male CD mice. The degree of pancreatic inflammation and tissue injury (histological score), expression of ICAM-1, the staining for nitrotyrosine and PAR, and myeloperoxidase activity was markedly reduced in pancreatic tissue sections obtained from cerulein-treated mice administered Hypericum perforatum extract (30 mg/kg, suspended in 0.2 mL of saline solution, o.s.). Moreover, the treatment with Hypericum perforatum extract significantly reduced the mortality rate at 5 days after cerulein administration.

Following the traditional use of Hypericum against viruses of the Herpes family in Greece Axarlis et al (1998) found antiviral activity of a methanolic extract against Human Cytomegalovirus.

Panossian et al (1996) concluded that the antiviral, antiinflammatory and antitumroal effects of hypericin may result from the inhibition of the PKC-mediated signalling pathway demonstrated in human leukocytes, which influences the arachidonic acid metabolism and the interleukin-1-alpha production resulting in an immunosuppressive effect.

I.2.1.2 Assessor's overall conclusions on pharmacology

There are numerous pharmacological findings published which propose a similar pharmacology to established synthetic antidepressant drugs. However, the discussion on the responsible compounds in the extract is still ongoing. Since several hydroethanolic and hydromethanolic extracts with different contents of hypericin and hyperforin are positively tested it could be concluded that the naphthodianthrones and the phloroglucine derivatives are of minor relevance for the antidepressant activity.

I.2.2 Pharmacokinetics

I.2.2.1 Overview of available data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

Wurglics et al (2006) published a review on pharmacokinetic data of compounds of hypericum extracts. The hyperforin plasma concentration in humans was investigated in a small number of studies. The results of these studies indicate a relevant plasma concentration, comparable with that used in in-vitro tests. Furthermore, hyperforin is the only ingredient of H. perforatum that could be determined in the brain of rodents after oral administration of alcoholic extracts. The plasma concentrations of the hypericins were, compared with hyperforin, only one-tenth and, until now, the hypericins could not be found in the brain after oral administration of alcoholic H. perforatum extracts or pure hypericin.

The intestinal absorption characteristics of protohypericin were studied and compared with those of hypericin by Kamuhab et al (1999). The Caco-2 model was used as a model of the intestinal mucosa to assess transepithelial transport and cell uptake. Following application of the individual compounds (80-200 microM) to the apical side of the monolayers, the appearance in the basolateral compartment was found to be very low (<0.5%/5 h), but was comparable for both compounds. A lag-time of 2-3 h was observed, suggesting gradual saturation of binding sites on the membrane or inside the cells. Uptake experiments of protohypericin and hypericin by Caco-2 cells revealed a very significant cellular accumulation (4-8%); uptake was characterised by saturation after 3 h. The findings of this study suggest that protohypericin has comparable absorption characteristics as hypericin.

Investigations by Sattler et al (1997) indicate that a significant accumulation of hypericin in the cell membrane and the cell nucleus membrane of Caco-2-cells takes place. The authors conclude that hypericin is absorbed through the intestinal epithelium by passive transcellular diffusion and that increasing its solubility by cyclodextrin appears as a promising approach to increase its oral bioavailability for pharmaceutical formulations.

Plasma levels of hypericin in rats in the presence and absence of procyanidin B2 or hyperoside were determined by reversed phase HPLC using fluorimetric detection (Butterweck et al 2003). Both compounds increased the oral bioavailability of hypericin by ca. 58 % (B2) and 34 % (hyperoside). Procyanidin B2 and hyperoside had a different influence on the plasma kinetics of hypericin; median maximal plasma levels of hypericin were detected after 360 min (C max : 8.6 ng/mL) for B2, and after 150 min (C max : 8.8

ng/mL) for hyperoside. It can be speculated that, when administered together with these compounds, a significant accumulation of hypericin in rat plasma in the presence of both polyphenols might be responsible for the observed increased in vivo activity.

Juergenliemk et al (2003) examined the pathway of miquelianin (quercetin 3-O-beta-D-glucuronopyranoside), a flavonoid with antidepressant acitivity, from the small intestine to the central nervous system. The permeability coefficient of miquelianin (Pc = 0.4 +/- 0.19 x 10(-6) cm/sec) was in the range of orally available drugs assuming sufficient absorption from the small intestine. The permeability coefficients (blood-brain-barrier: Pc = 1.34 +/- 0.05 x 10(-6) cm/sec; blood-CSF-barrier: Pc = 2.0 +/- 0.33 x 10(-6) cm/sec) indicate the ability of miquelianin to cross both barriers to finally reach the CNS.

Paulke et al (2008) suggested that not the genuine flavonoid glycosides reach the plasma. After oral uptake they are deglycosylated in the small intestine, after absorption the quercetin aglycone is glucuronidated (yielding e.g. miquelianin). Further methylation is possibly leading to isorhamnetin and tamarixetin. These metabolites have the ability to penetrate the blood-brain-barrier. Moreover, a significant accumulation of these metabolites in the CNS tissue is observed. After a single dose of a hypericum extract (1600 mg/kg rat) the quercetin plasma level increased rapidly and reached the maximum of about 700 ng/ml after 4 hours. After 24 hours, 50% of the Cmax was still measurable. In contrast the concentration level of isorhamnetin/Tamarixetin increase much slower, the maximum was reached after 24 hours with a Cmax of 903 ng/ml. Repeated doses of 1600 mg/kg rat yielded a continuous increase in the plasma levels of quercetin and isorhamnetin for 5 days, after that time the concentration ermained constant.

I.2.2.2 Assessor's overall conclusions on pharmacokinetics

The few data on pharmacokinetics do not allow a final conclusion about absorption, distribution, metabolism and excretion of constituents. Additionally it is not clear whether putative active constituents of hypericum extracts reach the brain tissue in sufficient concentration.

I.2.3 Toxicology

I.2.3.1 Overview of available data regarding the herbal substance(s)/herbal preparation(s) and constituents thereof

I.2.3.1.1 Single-dose toxicity

For a methanolic extract (DER 3-6:1) the following data are published in the SPC:

Species	Application	LD_{50} (mg / kg BW)
Mouse	oral	≥ 5000
Rat	oral	≥ 5000
Mouse	intraperitoneal	1780
Rat	intraperitoneal	1000

Intravenous application of hypericin was well tolerated by rhesus monkeys in a dose of 2 mg / kg, at 5 mg/kg transient severe photosensitivity rash occurred (Fox et al 2001). The amount of hypericin administered daily in usual therapeutic dosages is not more than 3 mg for adults (= 0.04 mg / kg).

I.2.3.1.2 Repeated-dose toxicity

Studies on long-term toxicity (900 mg/kg and 2700 mg/kg extract per day (LI 160), 26 weeks treatment) in rats and dogs revealed only minor non-specific symptoms (weight loss, minor pathological changes in liver and kidney). All changes reverted to normal when treatment was stopped (Leuschner 1996, Greeson et al 2001). The dosages were approximately 70 and 200 times the mean therapeutic dosage. Therefore the therapeutic dosage of 13 mg/kg per day (= 900 mg extract) can be regarded as safe from that perspective.

I.2.3.1.3 Mutagenicity

The genotoxicity testing of ethanolic extracts showed weak positive results in the AMES-test (Salmonella typhimurium TA 98 and TA 100, with and without metabolic activation). After chromatographic separation the effect could be assigned to quercetin (Poginsky et al 1988, Schimmer 1988).
Okpanyi et al (1990) tested an ethanolic extract (DER 1:5-7, 0.2-0.3% hypericin, 0.35 mg/g quercetin) in several in-vivo (mammalian spot test in mice, chromosome aberration test in Chinese hamsters) and in-vitro test systems (HGPRT hypoxanthine-guanine-phosphoribosyl-transferase etst, UDS unscheduled DNA synthesis test, cell transformation test). The authors could not detect signs of a mutagenic potential of the extract.

I.2.3.1.4 Carcinogenicity

No data available

I.2.3.1.5 Phototoxicity

In order to estimate the potential risk of phototoxic skin damage during antidepressive therapy, Bernd et al (1999) investigated the phototoxic activity of hypericin extract using cultures of human keratinocytes and compared it with the effect of the well-known phototoxic agent psoralen. The absorbance spectrum of the Hypericum extract revealed maxima in the whole UV range and in parts of the visible range. Human keratinocytes were cultivated in the presence of different Hypericum concentrations. The determination of the bromodeoxyuridine incorporation rate showed a concentration- and light-dependent decrease in DNA synthesis with high hypericin concentrations ($\geq$ 50 µg/mL) combined with UVA or visible light radiation. In the case of UVB irradiation a clear phototoxic cell reaction was not detected. The authors found phototoxic effects even with 10 ng/mL psoralen using UVA with the same study design as in the case of the Hypericum extract. These results confirm the phototoxic activity of Hypericum extract on human keratinocytes. However, the blood levels that are to be expected during antidepressive therapy are presumably too low to induce phototoxic skin reactions.

Schempp et al (2002) assessed the phototoxic and apoptosis-inducing capacity of pseudohypericin (PH) compared to hypericin (H) in a cell culture model with human leukemic lymphoma cells (Jurkat). Treatment with both photoactivated H and PH resulted in a dose-dependent inhibition of cell proliferation, whereas not photoactivated H and PH had no effect at the concentrations tested. The half-maximal inhibitory concentration (IC50) of H was lower (100 ng/mL) as compared to PH (200 ng/mL) ($p < 0.05$). In an apoptosis assay the authors found a dose-dependent increase of DNA fragmentation after treatment with both photoactivated H and PH. The cytotoxic potential of PH should be taken into account during systemic therapy with Hypericum extracts, since PH is about two times more abundant than H in Hypericum perforatum.

Wilhelm et al (2001) investigated the phototoxic potential of 3 Hypericum perforatum extracts from different sources as well as some of its main constituents. Hypericum perforatum extracts demonstrated cytotoxicity and photocytotoxicity in a dose and UVA-dose dependent manner. Hypericine itself also evoked severe phototoxic effects and was thus identified as the main phototoxic constituent. Among the tested flavonoids quercitrin was found to be cytotoxic, while rutin unexpectedly demonstrated phototoxicity whereas quercitrin was effective to control the phototoxic activity of Hypericum perforatum extracts.

Clinical evidence suggests that administration of Hypericum perforatum (Hp) extracts containing the photo-activated hypericin compounds may cause fewer skin photosensitization reactions than administration of pure hypericin. Schmitt et al (2006) conducted a study to determine whether the phototoxicity of hypericin in HaCaT keratinocytes could be attenuated by H. perforatum extracts and constituents. Two extracts, when supplemented with 20 microM hypericin: (1) an ethanol re-extraction of residue following a chloroform extraction (denoted ethanol(-chloroform)) (3.35 microM hypericin and 124.0 microM total flavonoids); and (2) a chloroform extract (hypericin and flavonoids not detected), showed 25% and 50% ($p<0.0001$) less phototoxicity than 20 microM hypericin alone. Two H. perforatum constituents, when supplemented with 20 microM hypericin: (1) 10 microM chlorogenic acid; and (2) 0.25 microM pyropheophorbide, exhibited 24% ($p<0.05$) and 40% ($p<0.05$) less phototoxicity than 20 microM hypericin alone. The peroxidation of arachidonic acid was assessed as a measure of oxidative damage by photo-activated hypericin, but this parameter of lipid peroxidation was not influenced by the extracts or constituents. However alpha-tocopherol, a known antioxidant also did not influence the amount of lipid peroxidation induced in this system. These observations indicate that hypericin combined with H. perforatum extracts or constituents may exert less phototoxicity than pure hypericin, but possibly not through a reduction in arachidonic acid peroxidation.

Schmitt et al (2006a) examined the cytotoxicity of Hp extracts prepared in solvents ranging in polarity, fractions of one extract, and purified compounds in three cell lines. All extracts exhibited significant cytotoxicity; those prepared in ethanol (no hyperforin, 3.6 microM hypericin, and 134.6 microM flavonoids) showed between 7.7 and 77.4% cell survival ($p < 0.0001$ and 0.01), whereas the chloroform and hexane extracts (hyperforin, hypericin, and flavonoids not detected) showed approximately 9.0 ($p < 0.0001$) and 4.0% ($p < 0.0001$) survival. Light-sensitive toxicity was observed primarily with the ethanol extracts sequentially extracted following removal of material extracted in either chloroform or hexane. The absence of light-sensitive toxicity with the Hp extracts suggests that the hypericins were not playing a prominent role in the toxicity of the extracts.

A study of Traynor et al (2005) used HaCaT keratinocytes to investigate the photoclastogenic ability of hypericin on irradiation with UVA. The results show that although the combination of hypericin and UVA light increased the genotoxic burden, when all factors are taken into account, the risk of significant photogenotoxic damage incurred by the combination of Hypericum extracts and UVA phototherapy may be low in the majority of individuals.

Phototoxicity to the eye

To determine if hypercin could be phototoxic to the eye, He et al (2004) exposed human lens epithelial cells to 0.1-10 microM hypericin and irradiated them with 4 J/cm2 UV-A or 0.9 J/cm2 visible light. Neither hypericin exposure alone nor light exposure alone reduced cell viability. In contrast, cells exposed to hypericin in combination with UV-A or visible light underwent necrosis and apoptosis. The ocular antioxidants lutein and N-acetyl cysteine did

not prevent damage. Thus, ingested Hypericum extract is potentially phototoxic to the eye and could contribute to early cataractogenesis.

Lens alpha-crystallin, isolated from calf lenses, was irradiated in the presence of hypericin (5 x 10(-5) M, 10 mM ammonium bicarbonate, pH 7.0) and in the presence and absence of light (> 300 nm, 24 mW/cm2). Hypericin-induced photosensitized photopolymerization as assessed by sodium dodecylsulfate-polyacrylamide gel electrophoresis. Further analysis of the oxidative changes occurring in alpha-crystallin using mass spectrometry showed specific oxidation of methionine, tryptophan and histidine residues, which increased with irradiation time. Hypericin did not damage the lens protein in the dark. Damage to alpha-crystallin could undermine the integrity of the lens directly by protein denaturation and indirectly by disturbing chaperone function. Therefore, in the presence of light, hypericin can induce changes in lens protein that could lead to the formation of cataracts. The authors conclude that appropriate precautions should be taken to protect the eye from intense sunlight while on this antidepressant medication (Schey et al 2000).

Wahlman et al (2003) found that the total accumulated protein leakage was positively correlated (r = 0.9) with variability in focal length. Lenses treated with hypericin and irradiated with UVB had an increase in focal length variability as compared with the lenses that were only UVB-irradiated. Lenses without UVB irradiation had much lower focal length variability than irradiated lenses. For non-hypericin-treated lenses, UVB-irradiated lenses had a larger variability (4.58 mm) than the unirradiated lenses (1.78 mm). The lenses incubated in elevated glucose concentrations had a focal length variability (3.23 mm) equivalent to that of the unirradiated hypericin-treated lenses (3.54 mm). The authors conclude that photooxidative damage by hypericin results in changes in the optical properties of the lens, protein leakage and finally cataract formation. In contrast to this, high concentrations of glucose induced protein leakage but not changes in optical properties or the opacity associated with a cataract. This work provides further evidence that people should protect their eyes from intense sunlight when taking St. John's Wort.

To determine if hypericin might be phototoxic to the human retina, Wielgus et al (2006) exposed human retinal epithelial cells to 10(-7) to 10(-5) M hypericin. Fluorescence emission detected from the cells (lambda(exc) = 488 nm; lambda(em) = 505 nm) confirmed hypericin uptake by human RPE. Neither hypericin exposure alone nor visible light exposure alone reduced cell viability. However when irradiated with 0.7 J/cm(2) of visible light (lambda > 400 nm) there was loss of cell viability as measured by MTS and LDH assays. The presence of hypericin in irradiated hRPE cells significantly changed the redox equilibrium of glutathione and a decrease in the activity of glutathione reductase. Increased lipid peroxidation as measured by the TBARS assay correlated to hypericin concentration in hRPE cells and visible light radiation. Thus, ingested Hypericum extract is potentially phototoxic to retina and could contribute to retinal or early macular degeneration.

I.2.3.1.6 Reproduction Toxicity

Ondrizek et al (1999) incubated zona-free hamster oocytes for 1 hour in St. John's wort (Hypericum perforatum), or control medium before sperm-oocyte interaction. The DNA of herb-treated sperm was analyzed with denaturing gradient gel electrophoresis. Pre-treatment of oocytes with 0.6 mg/mL of St. John's wort resulted in zero penetration. A lower concentration (0.06 mg/mL) had no effect. Exposure of sperm to St. John's wort resulted in DNA denaturation. Sperm exposed to 0.6 mg/mL of St. John's wort showed mutation of the BRCA1 exon 11 gene. The data suggested in the view of the authors that St. John's wort at

high concentrations damage reproductive cells. St. John's wort was mutagenic to sperm cells. Since the concentrations used were several orders of magnitude higher than considered as therapeutically relevant these effects should be considered with caution (Greeson et al 2001).

Rayburn et al (2001) examined the influence of 180 mg / kg daily of a hypericum extract on long-term growth and physical maturation of mouse offspring for 2 weeks before conception and throughout gestation. No differences between the hypericum group and the placebo group could be detected.

Assessor's comment:
Details on the type of extract are missing.

The purpose of a study of Gregoretti et al (2004) was to investigate the effects of a treatment with hypericum administered prenatally and during breastfeeding (from 2 weeks before mating to 21 days after delivery) in Wistar rats. Two doses of the extract were chosen, 100 mg/kg per day, which, based on surface area, is comparable to the dose administered to humans, and 1000 mg/kg per day. A microscopical analysis of livers, kidneys, hearts, lungs, brains, and small bowels was performed. A severe damage was observed in the livers and kidneys of animals euthanized postnatally on days 0 and 21. The lesions were more severe with the higher dose and in animals that were breastfed for 21 days; however, an important renal and hepatic damage was evident also with the dose of 100 mg/kg per day. In addition, similar serious hepatic and renal lesions were evident also in animals that were exposed to hypericum only during breastfeeding. In particular, a focal hepatic damage, with vacuolization, lobular fibrosis, and disorganization of hepatic arrays was evident; in the kidney, a reduction in glomerular size, disappearance of Bowman's space, and hyaline tubular degeneration were found. The results obtained in this study indicate that further, appropriate histological studies should be performed in other animal species to better evaluate the safety of hypericum extracts taken during pregnancy and breastfeeding.

Borges et al (2005) treated inseminated rats orally with a methanilc extract of Hypericum perforatum. A dosage of 36 mg/kg was given on days 9-15 of pregnancy (= period of organogensis). No clinical signs of maternal toxicity were found. In the administered dose Hypericum extract did not interfere with the progress of gestation during organogenesis in rats.

I.2.3.2 Assessor's overall conclusions on toxicology

The few data available on acute and subchronic toxicity do not reveal signs of a risk to the patient. The weak positive outcome of tests on mutagenicity of ethanolic extracts is explained with the presence of quercetin in the extracts. Tests on reproduction toxicity demonstrated no differences between Hypericum extract (108 mg/kg) and placebo in mice. Numerous publications deal with the potential phototoxicity of hypericin and Hypericum extracts. Extracts exert less phototoxicity than pure hypericin. Considering the outcome of clinical test on phototoxicity herbal preparations of Hypericum perforatum can be considered as safe when administered in the proposed dosage. The data on reproduction toxicology are contradictory. For safety reasons the oral use of Hypericum during pregnancy and lactation should not be recommended.

I.3 CLINICAL DATA

I.3.1 Clinical Pharmacology

I.3.1.1 Pharmacodynamics

I.3.1.1.1 Overview of available data regarding the herbal substance(s)/herbal preparation(s) including data on constituents with known therapeutic activity.

I.3.1.1.2 Assessor's overall conclusions on Pharmacodynamics

To be completed

I.3.1.2 Pharmacokinetics

I.3.1.2.1 Overview of available data regarding the herbal substance(s)/herbal preparation(s) including data on constituents with known therapeutic activity.

Biber et al (1998) investigated the pharmacokinetics of hyperforin after oral administration of 300, 600 and 1200 mg of two different ethanolic extracts (5% and 0.5% hyperforin). Maximum plasma levels of hyperforin were reached after 2.8 to 3.6 hours. The 5% extract yielded a total $AUC_{0-\infty}$ of 1336, 2215 and 3378 h x ng/ml. The hyperforin pharmacokinetics were linear up to 600 mg of the extract, higher dosages resulted in a lower concentration than linear extrapolation would expect. The plasma concentrations of hyperforin after administration of the other extract were considerably lower. In a repeated dose study no accumulation of hyperforin could be detected, the steady state plasma concentration after 3 x 300 mg/day of the extract was approximately 100 ng/ml.

The objective of two open phase I clinical trials (Schulz et al 2005) was to obtain pharmacokinetic data of hypericins, hyperforin and flavonoids from a hypericum extract containing tablet. Each trial included 18 healthy male volunteers who received the test preparation, containing 900 mg dry extract of St John's wort (STW 3-VI, Laif 900), either as a single oral dose or as a multiple once daily dose over a period of 14 days. After single dose intake, the key pharmacokinetic parameters were determined as follows: Hypericin: Area under the curve (AUC(0-infinity)) = 78.33 h x ng/ml, maximum plasma concentration (Cmax) = 3.8 ng/ml, time to reach Cmax (tmax) = 7.9 h, and elimination half-life (t1/2) = 18.71 h; pseudohypericin: AUC(0-infinity) = 97.28 h x ng/ml, Cmax = 10.2 ng/ml, tmax = 2.7 h, t1/2 = 17.19 h; hyperforin: AUC(0-infinity) = 1550.4 h x ng/ml, Cmax = 122.0 ng/ml, tmax = 4.5 h, t1/2 = 17.47 h. Quercetin and isorhamnetin showed two peaks of maximum plasma concentration separated by about 3-3.5 h. Quercetin: AUC(0-infinity) = 417.38 h x ng/ml, Cmax (1) = 89.5 ng/ml, tmax (1) = 1.0 h, Cma (2) = 79.1 ng/ml, tmax (2) = 4.4 h, t1/2 = 2.6 h; isorhamnetin: AUC(0-infinity) = 155.72 h x ng/ml, Cmax (1) = 12.5 ng/ml, tmax (1) = 1.4 h, Cmax (2) = 14.6 ng/ml, tmax (2) = 4.5 h, t1/2 = 5.61 h. Under steady state conditions reached during multiple dose administration similar results were obtained.

A study from Johne et al (2004) evaluated the influence of cimetidine and carbamazepine on the pharmacokinetics of hypericin and pseudohypericin. In a placebo-controlled, double blind study, 33 healthy volunteers were randomized into three treatment groups that received Hypericum extract extract (LI160) with different comedications (placebo, cimetidine, and carbamazepine) for 7 days after a run-in period of 11 days with Hypericum extract alone. Hypericin and pseudohypericin pharmacokinetics were measured on days 10 and 17. Between-group comparisons showed no statistically significant differences in AUC(0-24), C(max), and t(max) values for hypericin and pseudohypericin. Within-group comparisons, however, revealed a statistically significant increase in hypericin AUC(0-24) from a median of 119 (range 82-163 microg h/l) to 149 microg h/l (61-202 microg h/l) with cimetidine comedication and a decrease in pseudohypericin AUC(0-24) from a median of 51.0 (16.4-

102.9 microg h/l) to 36.4 microg h/l (14.0-102.0 microg h/l) with carbamazepine comedication compared to the baseline pharmacokinetics in each group. Hypericin and pseudohypericin pharmacokinetics were only marginally influenced by comedication with the enzyme inhibitors and inducers cimetidine and carbamazepine.

I.3.1.2.2 Assessor's overall conclusions on pharmacokinetics

Pharmacokinetic data on the most compounds which are considered to contribute to the activity are available. The long elimination lalf-life has to be considered after overdose. The possible role of metabolites is not addressed yet.

I.3.2 Clinical Efficacy[4]

I.3.2.1 Dose response studies

I.3.2.2 Clinical studies (case studies and clinical trials)

I.3.2.2.1 Studies on the treatment of depression

Extract LI 160

Extract	LI 160
Extraction solvent	80 % methanol
DER	3-6:1, initially 4-7:1
Hypericin	0,12-0,28 %
Hyperforin	App. 4.5% (Mueller et al 2006)
Flavonoids	App. 8.3% (Mueller et al 2006)

From several notes in publications it can be assumed that the content of hyperforin is in the range 3-6%.

Study	**Bjerkenstedt et al 2005**	
Indication	mild to moderate major Depression (DSM-IV: 296.31, 296.32); minimum of a total score of 21 on the 21-item Hamilton Depression scale	
Duration of use	4 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	20 mg fluoxetine
	multicentre	n=15
	number of patients	163
Outcome	HAMD reduction:	

[4] In case of traditional use the long-standing use and experience should be assessed.

	LI 160: from 24.9±4.5 to 15.0±8.4 (-9.9±8.1) Fluoxetine: from 23.8±3.7 to 14.9±8.4 (-8.9±8.0) Placebo: from 25.2±4.6 to 15.5±6.7 (-9.7±7.0) Conclusion: LI 160 and fluoxetine are not more effective in short-term treatment in mild to moderate depression than placebo. Hypericum is better tolerated than fluoxetine
Comment	Short time of treatment; high number of drop-outs in all groups (Hypericum start n=59, end n=38; fluoxetine start n=57, end n=37; placebo start n=58, end n=40)

Study	**Fava et al 2005**	
Indication	major depressive disorder (HAMD-17 ≥ 16)	
Duration of use	12 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	20 mg/d fluoxetine
	multicentre	n=2
	number of patients	135
Outcome	HAMD reduction (ITT-analysis): LI 160: -38 %, from 19.6± 3.5 to 10.2 ± 6.6 Fluoxetine: -30 %, from 19.6 ± 3.1 to 13.3 ± 7.3 Placebo: -21 %, from 19.9 ± 2.9 to 12.6 ± 6.4 Conclusion: LI 160 is significantly more effective than fluoxetine and superior to placebo. It is well tolerated and safe.	
Comment	Sample smaller than originally planned; the lack of efficacy of fluoxetine is explained by the fixed-dose approach.	

Study	**Brenner et al 2000**	
Indication	mild to moderate depression (HAMD: ≥ 17, according to DSM IV)	
Duration of use	7 weeks	
Daily dosage	600 mg (1st week) 900 mg (6 weeks)	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no?
	reference-controlled	50 mg sertraline (1st week) 75 mg sertraline (6 weeks)
	multicentre	no
	number of patients	30

Outcome	HAMD reduction: LI 160: -40 % ± 30 %; from 21.3 ± 3.2 to 12.7 ± 6.7) Sertraline: -42 % ± 24 %, from 21.7 ± 2.7 to 12.5 ± 5.6) Conclusion: LI 160 is as effective as sertraline in the treatment of mild to moderate depression.
Comment	Small number of patients, relatively high drop out rate. In the chapter 'Study medication' placebos are mentioned. However, no placebo group in table of results.

Study	**HDTSG 2002**	
Indication	moderately severe major depressive disorder (according to DSM-IV; HAM-D ≥ 20; GAF ≤ 60)	
Duration of use	8 weeks	
Daily dosage	900-1800 mg (3 x 300- 3 x 600 mg)	
Single dosage	300 – 600 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	50-150 mg sertraline
	multicentre	n=12
	number of patients	340
Outcome	HAMD reduction/response rate: Placebo: -9.20/-31,9 % LI 160: -8,68/-23,9 % Sertraline: -10.53/-24,8 % Conclusion: No efficacy of LI 160 and of sertraline in treatment of moderately severe major depression. Possible reason: low assay sensitivity.	
Comment	No efficacy despite of increase of dosage during study	

Study	**Montgomery et al 2000**	
Indication	mild to moderate depression (DSM-IV)	
Duration of use	12 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=18
	number of patients	248
Outcome	Conclusion: There is no statistically significant difference between placebo	

	and LI 160. (HAMD-score after 6 weeks). Negative Outcome

Study	**Vorbach et al 1997**	
Indication	severe mild to moderate major depression according to ICD-10 F 33.2, recurrent, without psychotic symptoms	
Duration of use	6 weeks	
Daily dosage	1800 mg	
Single dosage	600 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	
	reference-controlled	150 mg/d imipramine
	multicentre	n=20
	number of patients	209
Outcome	HAMD (17 items) reduction: LI 160:from 25.3±4.7 to 14.4±6.1 Imipramine: from 26.1±4.8 to 13.4±5.9 Conclusion: Efficacy not statistically equivalent, equivalence only in subgroups with more than 33% and 50% reduction of the HAMD total score. Superiority for LI 160 in adverse events.	

Study	**Wheatley 1997**	
Indication	mild to moderate major depression (HAMD-17 score: 17-24; according to DSM-IV)	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	75 mg/d amitriptyline
	multicentre	n=19
	number of patients	165
Outcome	HAMD reduction: LI 160: from 20 to 10 Amitriptyline: from 21 to 6 Conclusion: No statistically significant difference between LI 160 and amitriptyline; Hypericum is better tolerated.	

Study	**Harrer et al 1994**
Indication	moderately severe depressive episodes, according to ICD-10, F 32.1 (HAMD 17-items >- 16)

Duration of use	4 weeks	
Daily dosage	900 mg/d	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	Maprotiline (3 x 25 mg)
	multicentre	n=6
	number of patients	102
Outcome	HAMD reduction: LI 160: 20.5 -> 12.2, no statistically significant difference compared to maprotiline. Maprotiline: 21.5 -> 10.5 Responder rate: 61% in hypericum, 67% in maprotiline Onset of effects up to the second week of treatment. After 2 weeks of treatment more pronounced effect with maprotiline, after 4 weeks both groups similar.	

Study	**Shelton et al 2001**	
Indication	major depression (HAMD: ≥ 20 for more than 2 years, according to DSM-IV: major depression disorder, single episode or recurrent, without psychotic features)	
Duration of use	8 weeks	
Daily dosage	900 mg/d for 4 weeks, in case of not adequate response increase to 1200 mg/d	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=11
	number of patients	200
Outcome	Response rate in the ITT-analysis: LI 160: 26,5 % (for statistical significance 36,1 % is needed) Placebo: 18,6 % In Hypericum group a significant greater proportion of remissions. Conclusion: LI160 is not effective in treatment of major depression; good compliance: only adverse effect: headache.	
Comment	High number of patients with chronic depression. Sponsoring by Pfilzer	

Study	**Hänsgen et al 1996**
Indication	mild to moderate major depression, according to DSM-III-R (HAMD >- 16)
Duration of use	4 weeks (+2 weeks)

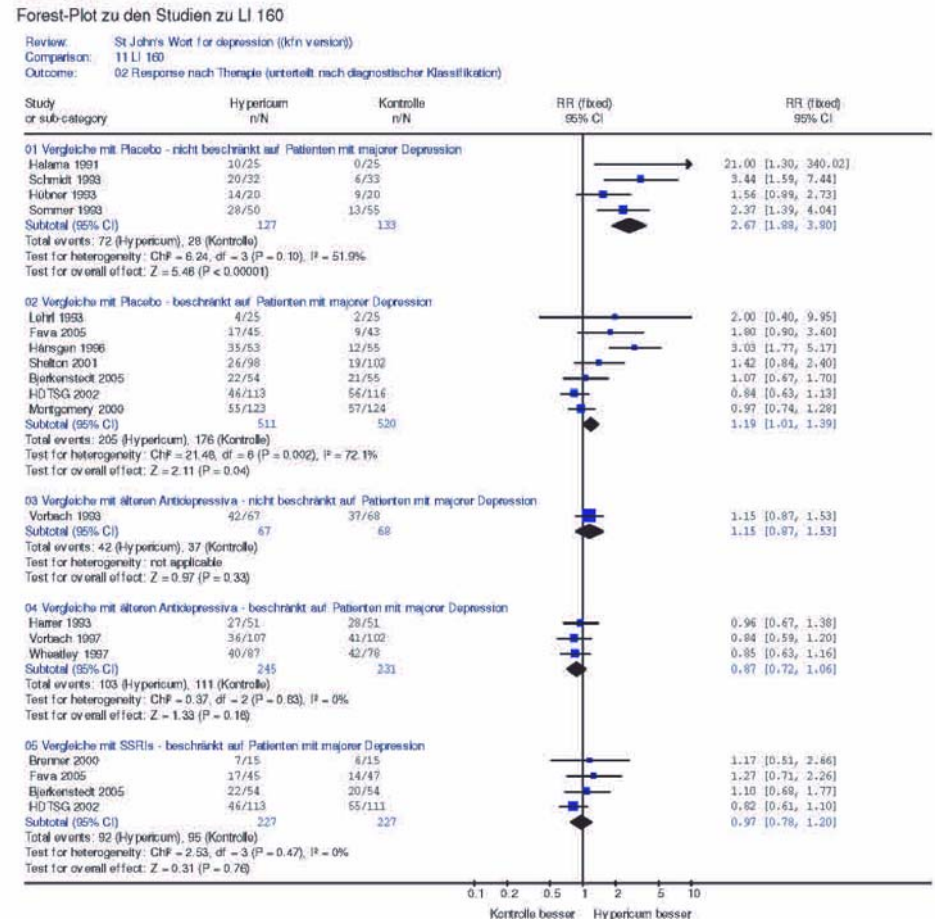
Daily dosage	900 mg/d	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=17
	number of patients	102
Outcome	HAMD reduction: LI 160: 21.0 -> 8.9, statistically significant compared to placebo. Placebo: 20.4 -> 14.4 Responder rate: 70% in hypericum, 24% in placebo Further 2 weeks of verum-treatment in both groups: similar improvement in the former placebo-group like in the first 2 weeks of treatment in the verum group.	

Study	**Hänsgen et al 1994**	
Indication	mild to moderate major depression, according to DSM-III-R (HAMD >- 16)	
Duration of use	4 weeks (+2 weeks)	
Daily dosage	900 mg/d	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=11
	number of patients	67
Outcome	HAMD reduction: LI 160: 21.8 -> 9.2, statistically significant compared to placebo. Placebo: 20.4 -> 14.7 Responder rate: 81% in hypericum, 26% in placebo Further 2 weeks of verum-treatment in both groups: similar improvement in the former placebo-group like in the first 2 weeks of treatment in the verum group.	

Study	**Sommer et al 1994**	
Indication	Depressive symptoms according ICD-09 300.4 (neurotic depression) and 309.0 (brief depressive reaction).	
Duration of use	4 weeks	
Daily dosage	900 mg/d	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes

	reference-controlled	no
	multicentre	n=3
	number of patients	105
Outcome	Only graphical presentation of data; improvement under hypericum after 4 weeks significant at a 1% level compared to placebo. Responder rate: 67% in hypericum, 28% in placebo	

Metanalaysis of clinical studies with LI 160 (Linde 2007)



Conclusion:
Reference controlled studies: all studies included patients with major depression; similar or insignificant less efficacy compared to standard antidepressants; in some studies no difference between hypericum, reference medication and placebo. Only one study (Vorbach et al 1997) was designed for proof of equivalence.
Placebo controlled studies: tendency that in older studies (published before 2000) better outcome for hypericum; modern studies also with negative outcome.
Studies including patients with more severe depressive episodes (daily dosage up to 1800 mg extract) do not show sufficient efficacy.

Extract WS 5570:

Extract	WS 5570
Extraction solvent	80 % methanol

DER	4-7:1
Hypericin	0,12-0,28 %
Hyperforin	3-6 %

The extraction solvent and the declared amount of Hypericine of this extract are identical with that of LI 160.

Study	**Anghelescu et al 2006**	
Indication	moderate to severe depression according to DSM-IV criteria: 296.22, 296.23, 296.32 and 296.33 (HAMD 17-item: ≥ 22)	
Duration of use	6 weeks of acute treatment	
Daily dosage	900 mg or 1800 mg; dose increase in week 2 in patients with HAMD improvement <20%	
Single dosage	300 mg or 600 mg	
Relapse	Patients with a HAM-D total score decrease of >=50% during the 6 weeks of acute treatment were asked to continue the treatment for another 16 weeks (n=133)	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	20/40 mg/d paroxetine; dose increase in week 2 in patients with HAMD improvement <20%
	multicentre	n=21
	number of patients	133
Outcome	HAMD reduction: WS 5570: .from 25.3+/-2.5 to 4.3+/-6.2 Paroxetine: from 25.3+/-2.6 to 5.2+/-5.5 During maintenance treatment alone 61.6% of the hypericum group and 54.6% of the paroxetine group showed additional reduction of HAMD score. Remission occurred 81.6% in the hypericum group and 71.4% in the paroxetine group. 3 Patients under Hypericum and 2 patients under paroxetine showed an increase in HAMD score of >5 during continuation treatment Conclusion: WS 5580 an paroxetine are similarly effective in preventing relapse in a continuation treatment after recovery from an episode of modeate to severe depresssion	

Study	**Szegedi et al 2005**	
Indication	moderate to severe major depression (HAMD 17-item: ≥ 22; DSM-IV: 296.22, 296.23, 296.32, 296.33)	
Duration of use	6 weeks	
Daily dosage	900 mg-1800 mg; dose increase in week 2 in patients with HAMD improvement <20%	
Single dosage	300 mg-600 mg	
Relapse	Responders (decrease in total HAMD score ≥ 50%) were invited to participate in a four month double blind maintenance phase	
Studydesign	*randomized*	yes

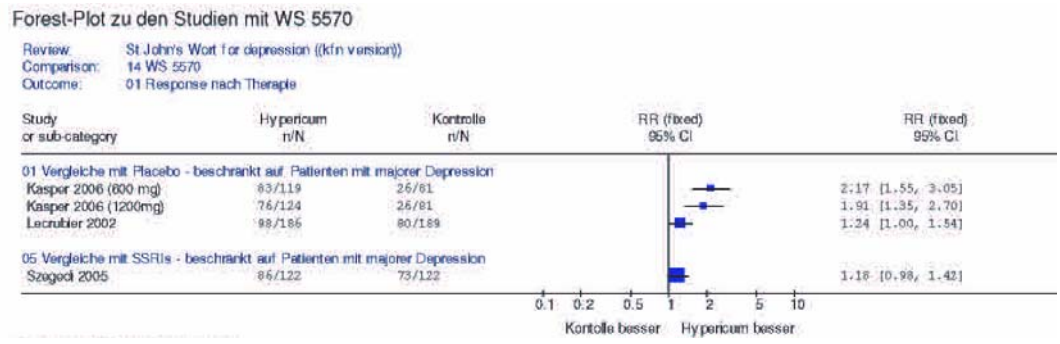
	double blind	yes
	placebo-controlled	no
	reference-controlled	20 mg-40 mg/d paroxetine; dose increase in week 2 in patients with HAMD improvement <20%
	multicentre	n=21
	number of patients	251
Outcome	HAMD recuction/ % responder: WS 5570: -14.4 / 71% Paroxetine: -11.4 / 60% Conclusion: WS 5570 is as effective as paroxetin in the treatment of moderate to severe major depression	
Comment	The results of the maintenance phase will be published eslewhere.	

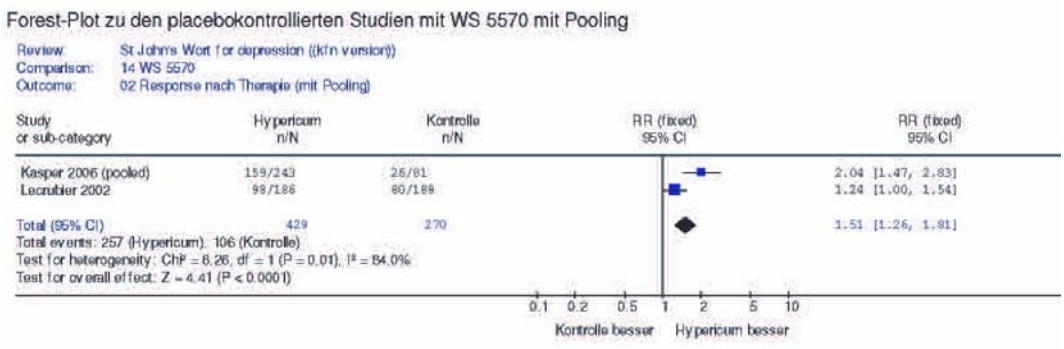
Study	**Lecrubier et al 2002**	
Indication	mild to moderate major depression (single or recurrent episode, DSM-IV code: 296.21, 296.22, 296.32, HAMD 17-item: 18-25)	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=26
	number of patients	375
Outcome	HAMD reduction/Responders WS 5570: -9,9/52,7 % Placebo: -8,1/42,3 % (The difference is significant) Conculusion: WS 5570 is safe and more effective than placebo in the treatment of mild to moderate major depression	

Study	**Kasper et al 2006**	
Indication	mild or moderate major depressive episode (single or recurrent episode, DSM-IV criteria: 296.21, 296.31, 296.21, 296.22, 296.31, 296.32; HAMD 17-item: ≥ 18, "depressive mood" ≥ 2)	
Duration of use	6 weeks	
Daily dosage	600-1200 mg	
Single dosage	600 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no

	multicentre	n=16
	number of patients	332
Outcome	More patients in the WS 5570 1200 mg group met the criterion of remission (HAMD: ≤ 7 at treatment end) HAMD reduction: WS 5570 600 mg: -11.6 ± 6,4 WS 5570 1200 mg: -10.8 ± 7,3 Placebo: -6.0 ± 8,1 Conclusion: WS 5570 is safe and more effective than placebo in treatment of mild to moderate depression.	

Study	**Kasper et al 2008**	
Indication	recurrent episode of moderate major depression; HAMD 17-item: ≥ 20, ≥ 3 previous episodes in 5 years (ICD-10 F33.0. F33.1, DSM-IV 296.3)	
Duration of use	6 weeks single blind acute treatment, then 26 weeks double blind continuation treatment, then 52 weeks double blind maintenance treatment	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	+	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	yes
	number of patients	426
Outcome	Relapse rates: Hypericum 18.1%; placebo 25.7% Average time to relapse: Hypericum 177 days; placebo 163 days Conclusion: WS 5570 showed a beneficial effect in preventing relapse after recovery from acute depression. Tolerability in continuation and long-term maintenance treatment was on the placebo level..	





Conclusion: All studies are well designed. All studies report superiority compared to placebo or non-inferiority compared to standard medication.

Comparison with LI 160:
In contrast to the recent studies published for LI 160 all modern studies for WS 5570 demonstrated a positive outcome for hypericum. Since the extracts LI 160 and WS 5570 are very similar in their key parameters, it seems to be justified to combine the results. It can be concluded that the efficacy of this type of extract in the treatment of mild to moderate severe depression is well documented.

Extract Ze 117:

Extract	Ze 117
Extraction solvent	There are differing informations on the extraction solvent: 50 % ethanol or ethanol 49% m/m : 2-propanol (97.3 : 2.7)
DER	4-7:1
Hypericin	0,2 %
Hyperforin	nearly free of hyperforin

Information on the refinement of the extract in order to reduce the content of hyperforin are not available.

Study	**Schrader 2000 (=Friede et al 2001?)**	
Indication	mild to moderate depression (ICD-10; F 32-0 and F 32-1, HAMD scale (21-item) 16-24)	
Duration of use	6 weeks	
Daily dosage	500 mg	
Single dosage	250 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	20 mg fluoxetine
	multicentre	n=7

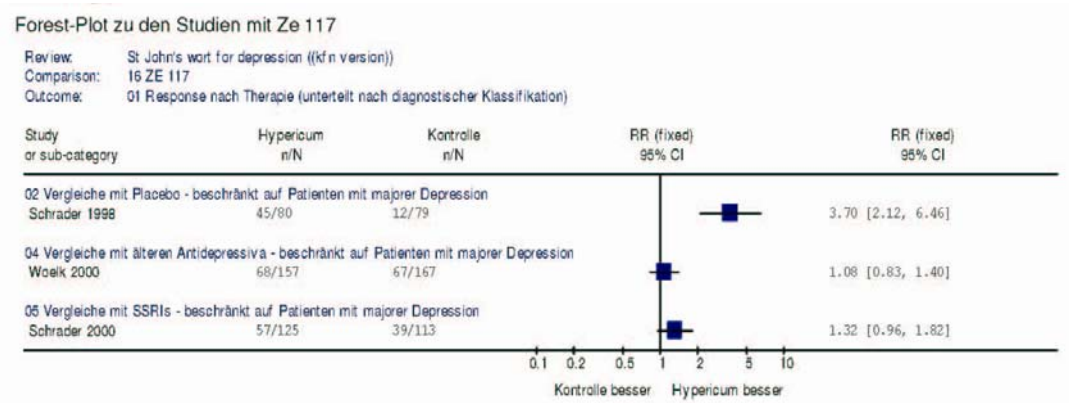
	number of patients	240
Outcome	HAMD reduction: Ze 117: from 19.65 to 11.54/-7,25 Fluoxetine: from 19.50 to 12.20/-8,11 Conclusion: Hypericum and fluoxetine are equipotent. Ze 117 safety was superior to fluoxetine.	
Comment	Contact of patients with investigators only at the beginning of the study and after 6 weeks in order to minimize the placebo effect. Nearly identical data in the publication compared to Friede et al 2001. However, no coincidence of the authors and the sponsor.	

Study	**Friede et al 2001 (=Schrader 2000?)**	
Indication	mild to moderate depressive episodes (ICD-10: F 32.0, F 32.1; HAMD range: 16-24)	
Duration of use	6 weeks	
Daily dosage	500 mg	
Single dosage	250 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	20 mg fluoxetine
	multicentre	n=7
	number of patients	240
Outcome	HAMD reduction/responder rate: Ze 117: from 19.7 to 11.5/60 % Fluoxetine: from 19.5 to 12.2/40 % Conclusion: Ze 117 is equipotent to fluoxetine in the treatment of mild to moderate depression	
Comment	There was a marked decrease of depressive agitation (pre-post comparison: 46%) and anxiety symptoms (44%) during the therapy with St. John's wort. Depressive obstruction (44%) and sleep disorders (43%) were reduced during the treatment. Nearly identical data in the publication compared to Schrader et al 2000. However, no coincidence of the authors and the sponsor.	

Study	**Woelk 2000**	
Indication	mild to moderate depression (ICD-10 codes F32.0, F33.0, F32.1, F 33.1; HAMD score (17-item) > 17)	
Duration of use	6 weeks	
Daily dosage	500 mg	
Single dosage	250 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes

	placebo-controlled	no
	reference-controlled	150 mg/d imipramine
	multicentre	n=40
	number of patients	324
Outcome	HAMD reduction: Ze 117: from 22.4 to 12.0 Imipramine: from 22.1 to 12,75 Conclusion: Ze 117 and imipramine are therapeutically equivalent in the treatment of mild to moderate depression. In the treatment of depression with anxiety hypericum has more benefit. There are fewer adverse events in the Ze 117 group.	
Comment	The dosage of imipramine is relatively high, which could be the reason the high number of drop outs in the reference group.	

Study	**Schrader et al 1998**	
Indication	mild to moderate depression (ICD-10; F 32-0 and F 32-1)	
Duration of use	6 weeks	
Daily dosage	500 mg (corresponding to 1 mg hypericin daily)	
Single dosage	250 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=16
	number of patients	162
Outcome	HAMD (21-item) reduction / % responder: Ze 117: from 20,13 to 10,53 / 56% Placebo: from 18,76 to 17,89 / 15% Conclusion: ZE 117 is significantly superior compared to placebo and safe in treatment of mild to moderate depression	
Comment	Contact of patients with investigators only at the beginning of the study and after 6 weeks in order to minimize the placebo effect.	

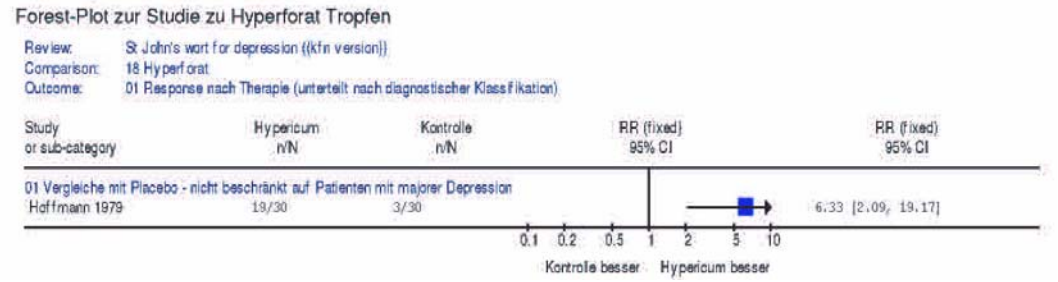


Conclusion: The superiorioty of the extract ZE 117 against placebo and the non-inferiority against imipramin and fluoxetin could be demonstrated. Compared with the results obtained with the extracts LI 160 and WS 5570 it can be concluded that the content of hyperforin in the extract is not correlated with clinical efficacy. The extract is according to the manufacturer at least since 1996 in Germany on the market. Therefore the minimum of 10 years of medicinal use is fulfilled.

Liquid extract

Extract	Hyperforat drops
Extraction solvent	50 % ethanol
DER	0.5:1
Hypericin	2 mg / ml
Hyperforin	not specified

Study	**Hoffmann et al 1979**	
Indication	mild to severe forms of depression	
Duration of use	6 weeks	
Daily dosage	90 drops (= 3.6 ml = 7.2 mg hypericin)	
Single dosage	30 drops	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	-
	multicentre	-
	number of patients	60
Outcome	Not standardised symptom score with 47 items; improvement under hypericum after 6 weeks of 61.4%, in placebo group of 16.8%. Responder: Hypericum: 80% Placebo: 33%	
Comment	Lack of statistical evaluation; inadequate study design	

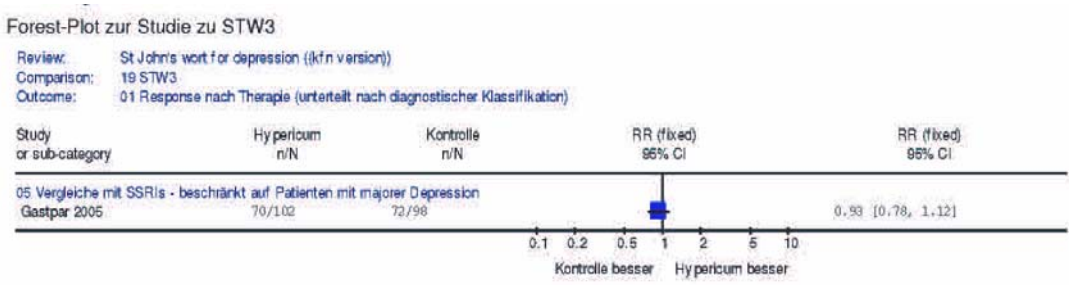


Conclusion: An old study (inadequate study design) shows superiority against placebo. No data are available on comparison to synthetic antidepressants.

Extract STW 3

Extract	STW 3
Extraction solvent	50 % ethanol
DER	5-8:1
Hypericin	mean 0.21%
Hyperforin	mean 3,3%
Flavonoids	mean 7.11%

Study	**Gastpar et al 2005**	
Indication	moderate depressive disorder (according to ICD-10 criteria: F32.1 or F33.1; HAMD 17-items: 20-24)	
Duration of use	12 weeks	
Daily dosage	612 mg	
Single dosage	612 mg	
Relapse	after 12 weeks additional treatment for 12 weeks of n=161	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	50 mg sertraline
	multicentre	n=18
	number of patients	241
Outcome	HAMD reduction: (week 12/week 24) STW 3: from 22.0 to 8.3/5.7 Sertraline: from 22.1 to 8.1/7.1 Conclusion: STW 3 is therapeutically equivalent to sertraline in moderate depression and it is well tolerated.	
Comment	Single daily dose	



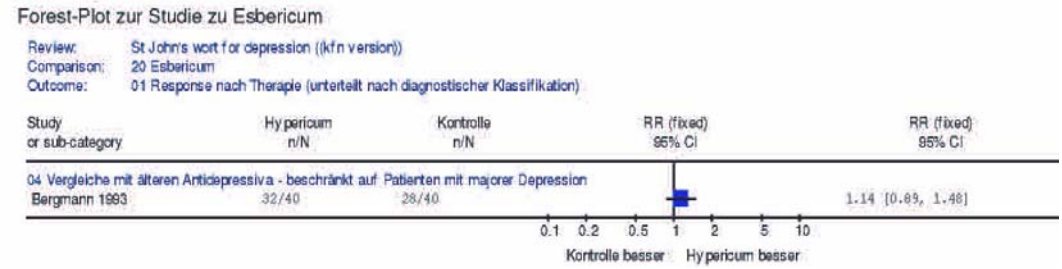
Conclusion: an adequate study demonstrates non-inferiority compared to sertraline (50 mg).

Esbericum capsules:
Daily dosage: 213-252 mg extract;

Extract	Esbericum
Extraction solvent	60 % ethanol
DER	2-5,5:1
Hypericin	0.1%

Hyperforin	not specified
Flavonoids	not specified

Study	**Bergmann et al 1993**	
Indication	mild to moderate depression (ICD-10 F32.0, F32.1, F33.0, F33.1)	
Duration of use	6 weeks	
Daily dosage	213-252 mg extract (Corresponding to 0.75 mg hypericin)	
Single dosage		
Relapse	-	
Studydesign	*randomized*	yes
	placebo-controlled	yes
	reference-controlled	30 mg amitriptylin/day
	multicentre	n=1
	number of patients	80
Outcome	HAMD reduction / % responder: Esbericum: from 15.82 to 6.43 / 84.2% Amitriptylin: from 15.26 to 6.65 / 73.7% From the fact that the low dosage of amitriptylin was effective it can be assumed that only patients with mild depression were included.	
Comment	Low dosage of amitriptylin; study design insufficient	



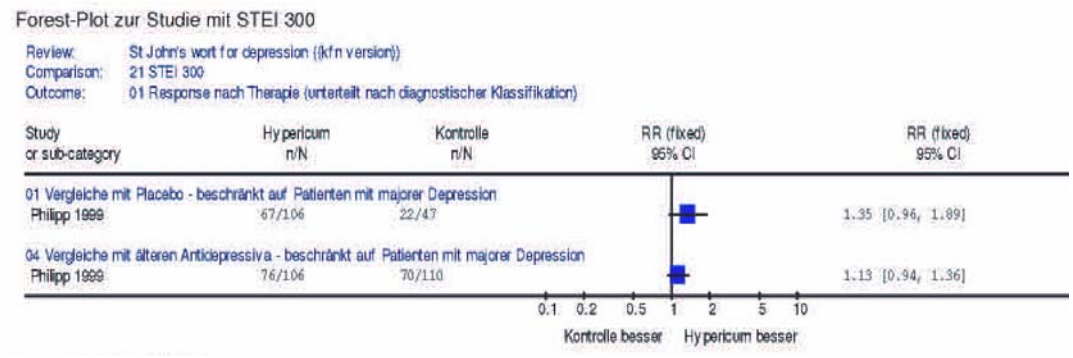
Conclusion: The results do not allow the conclusion, that this type of extract in the chosen posology is suitable for the treatment of mild or moderate depression.

Extract STEI 300:

Extract	STEI 300
Extraction solvent	60 % ethanol
DER	5-7:1
Hypericin	0.2-0.3%
Hyperforin	2-3%
Flavonoids	not specified

Study	**Philipp et al 1999**
Indication	moderate depression according to ICD-10 (codes F32. 1 and F33.1) (HAMA score > 18)
Duration of use	8 weeks
Daily dosage	1050 mg

Single dosage	350 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	100 mg/d imipramine (titrated within 4 days from 50 mg)
	multicentre	n=18
	number of patients	263
Outcome	HAMD reduction / % responder: STEI 300: 22.7-> 7.3 / 76% Placebo: 22.7-> 10.6 / 63% Imipramine: 22.2-> 8.0 / 66.7% Conclusion: STEI 300 is as effective as imipramine and more effective than placebo in the treatment of moderate depression and it is safe.	
Comment	Study of high methodological quality	



Conclusion: superiority against placebo and non-inferiority against imipramine (100 mg) could be demonstrated.

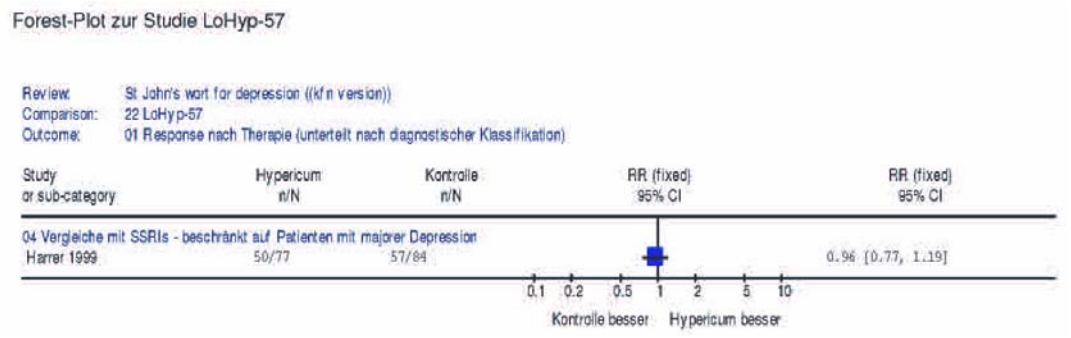
Extract LoHyp-57:

Extract	LoHyp-57
Extraction solvent	60 % ethanol
DER	5-7:1
Hypericin	0.2-0.3%
Hyperforin	2-3%
Flavonoids	not specified

Extract identical to STEI 300, but different dosage form and posology.

Study	**Harrer et al 1999**
Indication	mild to moderate major depression according to ICD 10 (F32.0, F32.1)
Duration of use	6 weeks
Daily dosage	800 mg
Single dosage	400 mg

Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	20 mg fluoxetine (= 22.4 mg fluoxetine HCl)
	multicentre	n=17
	number of patients	149
Outcome	HAMD (17-items) reduction: LoHyp 57: from 16.60 to 7.91 (mild: from 14.21 to 6.21; moderate: from 18.73 to 9.43) Fluoxetine: from 17.18 to 8.11 (mild: from 15.21 to 7.46; moderate: from 19.10 to 8.75) Responder rate: LoHyp 57: 71.4% (mild subgroup: 81.8%; moderate subgroup: 62.2%) Fluoxetine: 72.2% (mild subgroup: 76.9%; moderate subgroup: 67.5%) Conclusion: LoHyp 57 is equivalent to fluoxetine in treatment of mild to moderate major depression particularly in elderly patients.	
Comment	No confidence intervals shown in the publication, therefore the non-inferiority is formally not demonstrated; patients 60-80 years	



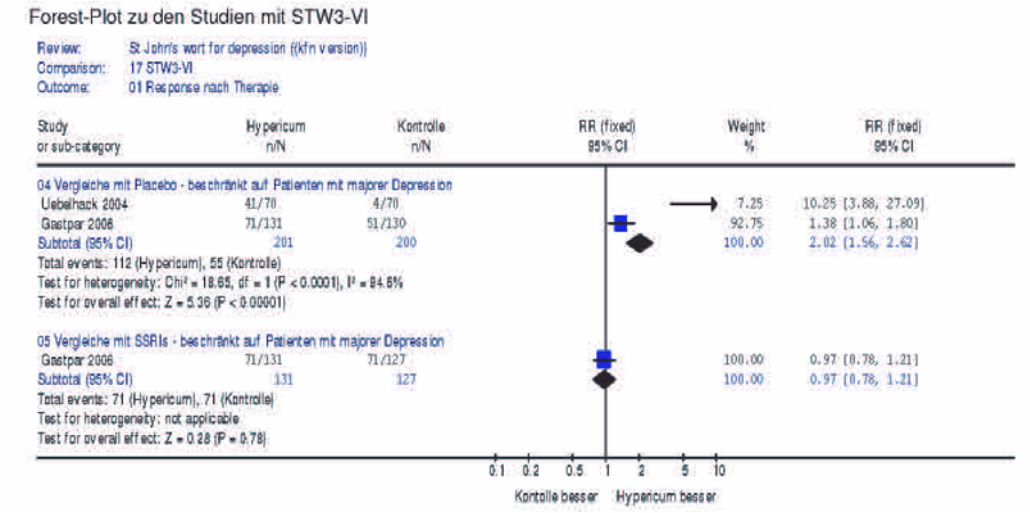
Conclusion: 800 mg of LoHyp-57 is equivalent to 20 mg fluoxetine in the treatment of mild to moderate major depression.

Extract STW3-VI:

Extract	STW3-VI
Extraction solvent	80 % ethanol
DER	3-6:1
Hypericin	mean 0.26%
Hyperforin	mean 3.0%
Flavonoids	mean 7.17%

Study	**Gastpar et al 2006**	
Indication	moderate depression (HAMD 17-items score: 20-24, ICD-10. F32.1, F33.1, according to DSM-IV major depressive episode and recurrent major depression)	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	900 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	20 mg citalopram
	multicentre	n=21
	number of patients	388
Outcome	HAMD reduction / % responder: Hypericum: 21.9 -> 10.3 / 54.2% Citalopram: 21.8-> 10.3 / 55.9% Placebo: 22.0 -> 13.0 / 39.2% Conclusion: The hypericum group was statistically non-inferior to citalopram and significantly superior to the placebo.	
Comment	Study of high methodological quality	

Study	**Uebelhack et al 2004**	
Indication	moderate depressive disorders (ICD-10 F32.1, F33.1) and HAMD (17-items) score : 20-24	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	900 mg	
Relapse		
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=1
	number of patients	140
Outcome	HAMD reduction / % responder: STW3-VI: 22.8 -> 11.8 / 58.6% Placebo: 22.6 -> 19.2 / 5.7% Conclusion: STW3-VI in a single daily dose is superior to placebo.	
Comment	Conspicuously low responder rate under placebo. This is explained by the authors that primarily patients with moderate depression were included, while other studies included also a higher number of patients with mild depression.	



Conclusions: superiority against placebo and non-inferiority against citalopram (20 mg) could be demonstrated.

Extract WS 5572:

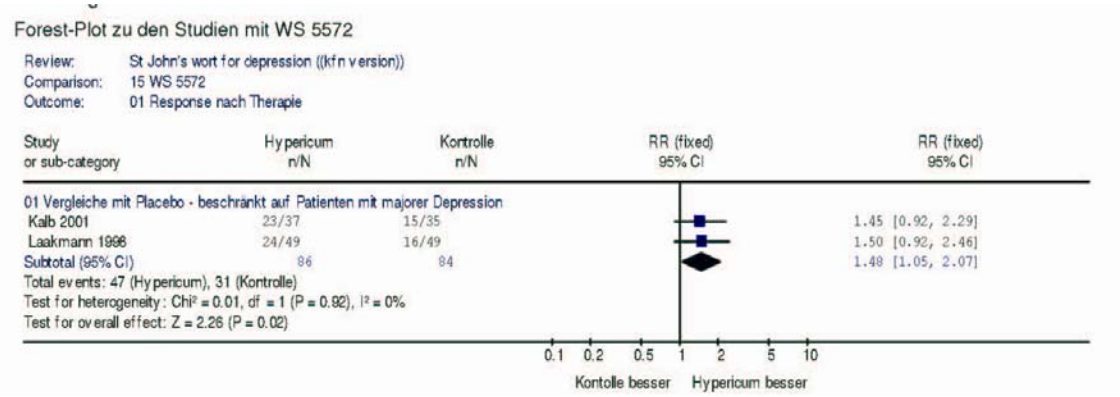
Extract	WS 5572
Extraction solvent	60 % ethanol
DER	2,5-5:1
Hypericin	not specified
Hyperforin	4-5 % (Rychlik et al 2001, Lackmann et al 1998) 1,5 % (Kalb et al 2001)

Study	**Rychlik et al 2001**	
Indication	mild to moderate depression (based on CGI)	
Duration of use	7 weeks	
Daily dosage	600 mg/1200 mg	
Single dosage	600 mg	
Relapse	-	
Studydesign	*randomized*	no
	double blind	no
	placebo-controlled	no
	reference-controlled	comparison of 600 mg and 1200 mg daily
	multicentre	n=446
	number of patients	2166
Outcome	Responder: 600 mg: 83,7 % 1200 mg: 86,9 % Conclusion: Good effectiveness and tolerability of WS 5572	
Comment	Observational study	

Study	**Kalb et al 2001**	
Indication	mild to moderate major depressive disorder (according to DSM-IV criteria) (DSM-IV code: 296.21, 296.31, 296.22, 296.32, HAMD (17-items): ≥ 16)	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	no
	multicentre	n=11
	number of patients	72
Outcome	HAMD reduction / % responder: WS 5572: 19.7 -> 8.9 / 62.2% Placebo: 20.1 -> 14.4 / 42.9% Conclusion: superior compared to placebo	
Comment	Efficacy was already statistically significant at day 28 Adaptive 2-stage design	

Study	**Laakmann et al 1998**	
Indication	mild or moderate depression according to DSM-IV criteria, HAMD ≥ 17	
Duration of use	6 weeks	
Daily dosage	900 mg	
Single dosage	300 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	yes
	reference-controlled	900 mg WS 5573
	multicentre	n=11
	number of patients	147
Outcome	HAMD reduction / % responder: WS 5572: 20.9 -> 10.7 / 49.0% WS 5573: 20.3 -> 11.8 / 38.8% Placebo: 21.2 -> 13.3 / 32.7% Conclusion: WS 5572 (5% hyperforin) was superior to placebo WS 5573 (0.5% hyperforin) and placebo were descriptively comparable The therapeutic effect depends on the content of hyperforin.	
Comment	WS 5572 and WS 5573 are produced with the identical manufacturing process (identical DER, extraction solvent), the only difference between the extracts relates to the content of	

	hyperforin. The fingerprint chromatograms of the two extracts are except for hyperforin identical. It is not mentioned how the differences in the conbtent of hyperforin are achieved. The negative outcome for the extract with low content of hyperforin is in contrast to the positive findings with the extract ZE 117, which is said to be nearly free of hyperforin.



Conclusion: WS 5572 is superior to placebo in the treatment of mild to moderate major depression.

Extract Calmigen:

Extract	Calmigen
Extraction solvent	not specified
DER	not specified
Hypericin	0.3%
Hyperforin	not specified

Study	**Behnke et al 2002**	
Indication	mild to moderate depression (ICD-10 F32), HAMD (17-items) score 16-24	
Duration of use	6 weeks	
Daily dosage	300 mg	
Single dosage	150 mg	
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	40 mg fluoxetin
	multicentre	n=446
	number of patients	70
Outcome	HAMD reduction / % responder; Calmigen: 20.0 -> 10.0 / 55% Fluoxetin: 20.7 -> 8.7 / 66%	
Comment	Number of patients too small for comparison of efficacy. Not to be confused with Calmigen capsules (300 mg Hypericum	

	extract, extraction solvent methanol 80%, 0.3% hypericin, authorized in DK)

Extract Hyperiforce:

Extract	Hyperiforce
Extraction solvent	not specified
DER	4-5:1 (shoot tips)
Hypericin	0.5%
Hyperforin	not specified

Study	**Lenoir et al 1999**	
Indication	mild to moderate depression (ICD-10)	
Duration of use	6 weeks	
Daily dosage	corresponding 0.17 mg, 0.33 mg or 1 mg hypericin	
Single dosage		
Relapse	-	
Studydesign	*randomized*	yes
	double blind	yes
	placebo-controlled	no
	reference-controlled	comparison of different dosaged
	multicentre	n=38
	number of patients	348
Outcome	Reduction in HAMD score from initially 16-17 to 8-9 in all groups. Resonder rate 62%-68%. The extract was effective on all three dosages.	
Comment	No placebo group	

Psychotonin (Liquid extract, DER 1: 5-7, extraction solvent ethanol 50%)
All studies performed with this type of extract (Harrer et al 1991, Osterheider et al 1992, Quandt et al 1993, Schlich et al 1987, Schmidt et al 1989) are not convincing from the current point of view. The methodology is inadequate, the number of included patients is small, the drop-out-rate is considerably high. The studies do not fulfil the criteria for well-established use.

Overall metanalysis
Roder et al (2004) published a meta-analysis of effectiveness and tolerability of treatment of mild to moderate depression with hypericum extracts.
The results demonstrate a significant superiority of hypericum extracts over placebo (mean response: Hypericum: 53.3 % and placebo: 32.7 %). Compared to standard antidepressive hypericum is similar effective for the treatment of depression (mean response: Hypericum: 53.2 %, synthetic antidepressive: 51.3 %). In the sub group of mild to modeate depression hypericum showed better results against the standard antidepressive groupe (mean response: 59.5 %/52.9 %) and a better side-effect profile. The fail-safe-N-test indicates that 423 studies with no effect would be needed to negate the presented result for placebo studies.

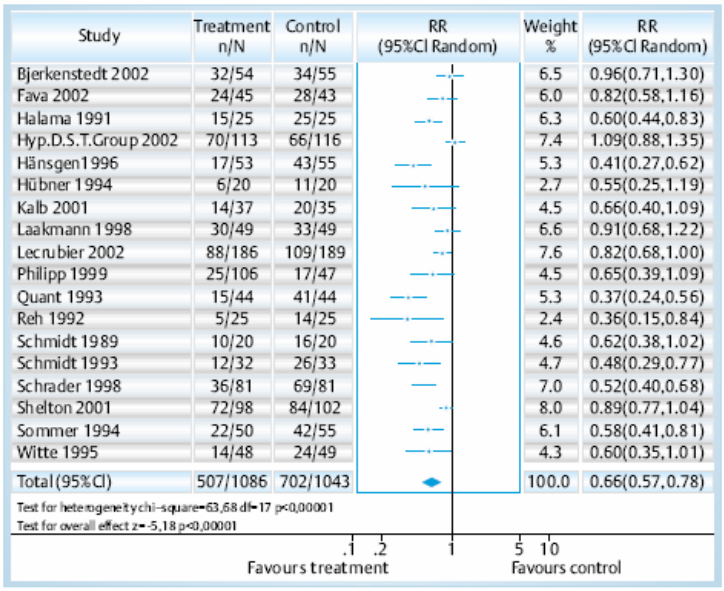


Abb. 2 Relatives Risiko der Non-Response nach Gabe von Johanniskraut (treatment) bzw. Plazebo (control).

Werneke et al (2004) came to similar results. They found that the effect sizes in recent studies were smaller than those resulted form earlier studies.

Linde et al (2005) concluded that the available data for major depression is confusing. While hypericum has minimal benefical effects over placebo, other trials suggest that hypericum and standard antidepressives are equal.

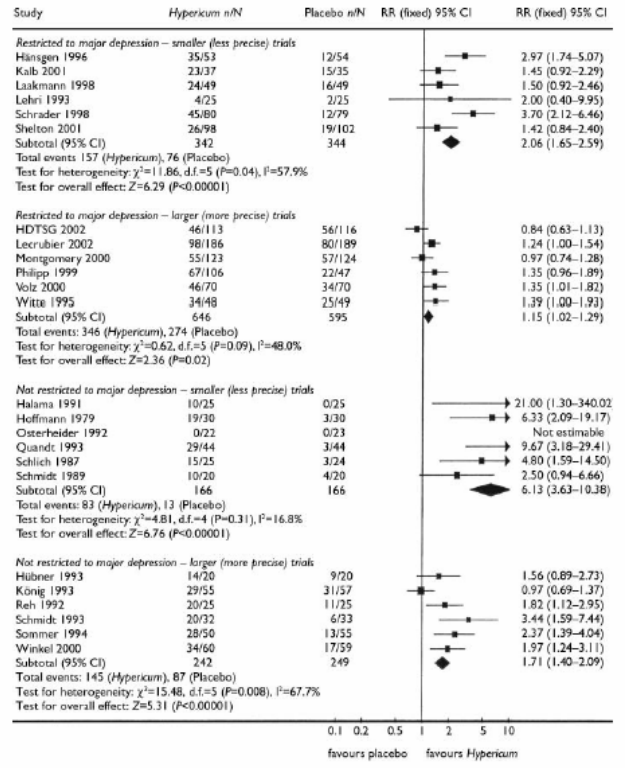


Fig. 3 Response to *Hypericum* extracts in depression. Results (fixed-effects model) from placebo-controlled trials stratified by type of depression (major and other) and study size (above and below median of variance). Studies identified by first author and year (HDTSG, Hypericum Depression Trial Study Group; *n*, number of responders; *N*, number of patients per group; RR, response rate ratio).

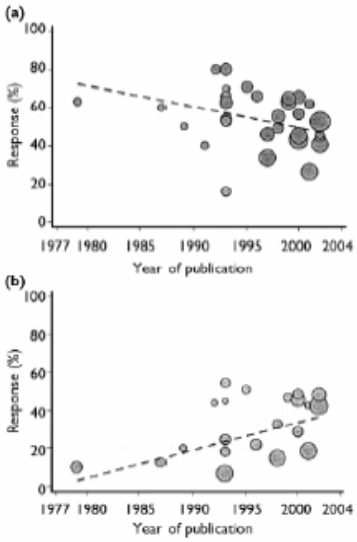


Fig. 4 Response rates over time to (a) *Hypericum perforatum* extracts and (b) placebo, from 34 active and 22 placebo trial arms.

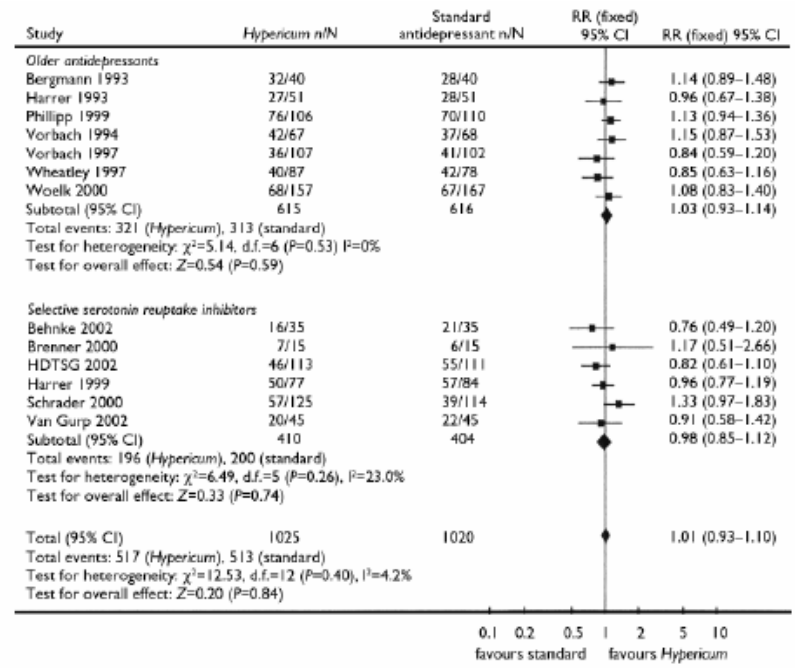


Fig. 5 Response to *Hypericum perforatum* extracts in depression: results from controlled trials stratified by type of comparison drug. Studies identified by first author and year (HDTSG, Hypericum Depression Trial Study Group; *n*, number of responders; *N*, number of patients per group; RR, response rate ratio).

Assessor's conclusion:

Overview of extracts with predominately positive study outcome (Superiority against placebo, equivalence to reference medication):

ICD-10 F32.0: mild depressive episode
ICD-10 F32.1: moderate depressive episode
ICD-10 F33.0: recurrent depressive disorder, current episode mild
ICD-10 F33.1: recurrent depressive disorder, current episode moderate
DSM-IV 296.21: major depressive disorder, single episode, mild
DSM-IV 296.22: major depressive disorder, single episode, moderate
DSM-IV 296.23: major depressive disorder, single episode, severe without psychotic features
DSM-IV 296.31: major depressive disorder, recurrent, mild
DSM-IV 296.32: major depressive disorder, recurrent, moderate
DSM-IV 296.33: major depressive disorder, recurrent, severe without psychotic features

	single episode				recurrent				single	recurrent
	mild		moderate		mild		moderate		severe	severe
	ICD-10 F32.0	DSM-IV 296.21	ICD-10 F32.1	DSM-IV 296.22	ICD-10 F33.0	DSM-IV 296.31	ICD-10 F33.1	DSM-IV 296.32	DSM-IV 296.23	DSM-IV 296.33
A: LI 160			x			x		x		
B: WS 5570				x				x	x	x
C: STW3-VI			x				x			
D: STEI 300			x				x			
E: LoHyp-57	x		x							
F: WS 5572		x		x		x		x		
G: STW3			x				x			
H: ZE 117	x		x		x		x			

	DER	extraction solvent	% hypericin	% hyperforin	% flavonoids	daily dosage	duration
A: LI 160	3-6:1	methanol 80%	0.12-0.28	app. 4.5%	app. 8.3%	900 mg	4-12 weeks
B: WS 5570	4-7:1	methanol 80%	0.12-0.28	3-6%	not specified	600-1800 mg	6/26 weeks relapse +
C: STW3-VI	3-6:1	ethanol 80%	0.26% (mean)	3.0% (mean)	7.17% (mean)	900 mg	6 weeks
D: STEI 300	5-7:1	ethanol 60%	0.2-0.3%	2-3%	not specified	1050 mg	8 weeks
E: LoHyp-57	5-7:1	ethanol 60%	0.2-0.3%	2-3%	not specified	800 mg	6 weeks
F: WS 5572	2.5-5:1	ethanol 60%	not specified	4-5%	not specified	600-1200 mg	6-7 weeks
G: STW3	5-8:1	ethanol 50%	0.21% (mean)	3.3% (mean)	7.11% (mean)	612 mg	12 weeks
H: ZE 117	4-7:1	ethanol 50%	0.2%	nearly free	not specified	500 mg	6 weeks
Pharm.Eur.	not specified	ethanol 50-80% methanol 50-80%	0.10-0.30%	< 6.0%	> 6.0%		

It is proposed to include the extracts types A-G into the monograph under well-established use with the indication 'mild to moderate depressive episodes'. Since the informations on the extraction solvent of the extract of type H are inconsistent, it could not be included in the monograph.

I.3.2.2.2 Somatoform disorders

Volz et al (2002) conducted a multicentre, randomised, placebo controlled, 6-week trial comparing the efficacy of LI 160 (600 mg/day) and placebo in 151 out-patients suffering from somatization disorder (ICD-10: F45.0), undifferentiated somatoform disorder (F45.1), or somatoform autonomic dysfunctions (F45.3). The primary outcome measure was the decrease of the Hamilton Anxiety Scale, subfactor somatic anxiety (HAMA-SOM), during the trial period. The hypericum extract was superior effective concerning the primary outcome criterion HAMA-SOM [decrease from 15.39 (SD 2.68) to 6.64 (4.32) in the Hypericum group and from 15.55 (2.94) to 11.97 (5.58) in the placebo group (statistically significant difference, P=0.001)]. This was corroborated by the result of a statistically significant superior efficacy in the outcome criteria additionally used such as Clinical Global Impression, HAMA-total score, HAMA, subscore psychic anxiety, Hamilton Depression Scale, Self-Report Symptom Inventory 90 items - revised (SCL-90-R), and SCL-90-R, subscore somatic anxiety. The efficacy of LI 160 was preserved after splitting the population in those with and those without mild depressive symptoms [corrected]. Tolerability of LI 160 was excellent. The efficacy is independent of an existing depressive mood.

In a prospective, randomized, placebo-controlled, and double-blind parallel group study, 184 outpatients with somatization disorder (ICD-10 F45.0), undifferentiated somatoform disorder (F45.1), and somatoform autonomic dysfunction (F45.3), but not major depression, received either 300 mg of Hypericum extract extract LI 160 twice daily or matching placebo for 6 weeks (Muller et al 2004). Six outcome measures were evaluated as a combined measure by means of the Wei Lachin test: Somatoform Disorders Screening Instrument--7 days (SOMS-7), somatic subscore of the HAMA, somatic subscore of the SCL-90-R, subscores "improvement" and "efficacy" of the CGI, and the global judgment of efficacy by the patient. In the intention to treat population (N=173), for each of the six primary efficacy measures as well as for the combined test, statistically significant medium to large-sized superiority of Hypericum extract treatment over placebo was demonstrated (p <.0001). Of the Hypericum extract patients, 45.4% were classified as responders compared with 20.9% with placebo (p =.0006). Tolerability of Hypericum extract treatment was equivalent to placebo.

I.3.2.2.3 Schizophrenia

Hypericum extract LI160 has demonstrated a ketamine-antagonising effect. Therefore Murck et al (2006) examined whether LI160 reverses changes of a low dose ketamine on auditory evoked potentials (AEP) in healthy subjects. The authors performed a double-blind

randomized treatment with either 2 x 750 mg LI 160 or placebo given one week, using a crossover design, in 16 healthy subjects. A test-battery including AEPs, the oculodynamic test (ODT) and a cognitive test were performed before and after an infusion with 4 mg of S-ketamine over a period of 1 hour. S-ketamine led to a significant decrease in the N100-P200 peak to peak (ptp) amplitude after the placebo treatment, whereas ptp was significantly increased by S-ketamine infusion in the LI160 treated subjects. The ODT and the cognitive testing revealed no significant effect of ketamine-infusion and therefore no interaction between treatment groups. Provided that ketamine mimics cognitive deficits in schizophrenia, LI160 might be effective to treat these symptoms.

I.3.2.2.4 Nootropic effects

A study from Elis et al (2001) aimed to examine whether acute administration of standardized hypericum extract could exert a nootropic effect in normal human subjects. The study employed a double-blind, crossover, repeated-measures design. Twelve healthy young subjects completed the Cognitive Drug Research (CDR) memory battery, following administration of placebo, 900 mg and 1800 mg hypericum (Blackmore's Hyperiforte). The findings suggested that hypericum does not have an acute nootropic effect in healthy humans at these doses. However, there was some evidence for an impairing effect on accuracy of numeric working memory and delayed picture recognition at the higher dose. This observed impairment could be due to a sensitivity of these specific tasks to modulation by neurotransmitters that have been noted to have memory-impairing effects (e.g. y-aminobutyric acid (GABA), serotonin).

In a randomized, double-blind, cross over study of 12 healthy male volunteers received capsules with 255-285 mg St John's wort extract (900 µg hypericin content), 25 mg amitriptyline and placebo three times daily for periods of 14 days each with at least 14 days between (Siepmann et al 2002). The doses of amitriptyline and St John's wort extract are comparable with respect to their antidepressant activity. Neither St John's wort extract nor amitriptyline had an influence on cognitive performance such as choice reaction, psychomotor coordination, short-term memory and responsiveness to distractive stimuli. Amitriptyline but not St John's wort extract decreased self rated activity ($P < 0.05$). Both drugs caused significant qEEG changes. St John's wort extract increased theta power density. Amitriptyline increased theta as well as fast alpha power density.

I.3.2.2.5 Cutaneous treatment

In a half-side comparison study Schempp et al (2003) assessed the efficacy of a cream containing Hypericum: extract standardised to 1.5% hyperforin (verum) in comparison to the corresponding vehicle (placebo) for the treatment of subacute Atopic Dermatitis. The study design was a prospective randomised placebo-controlled double-blind monocentric study. In twenty one patients suffering from mild to moderate Atopic Dermatitis (mean SCORAD 44.5) the treatment with verum or placebo was randomly allocated to the left or right site of the body, respectively. The patients were treated twice daily over a period of four weeks. Eighteen patients completed the study. The severity of the skin lesions on the left and right site was determined by means of a modified SCORAD-index (primary endpoint). The intensity of the eczematous lesions improved on both sites of treatment. However, the hypericum-cream was significantly superior to the vehicle at all clinical visits (days 7, 14, 28) ($p < 0.05$). Skin colonisation with Staphylococcus aureus was reduced by both verum and placebo, showing a trend to better antibacterial activity of the hypericum-cream ($p = 0.064$). Skin tolerance and cosmetic acceptability was good or excellent with both the hypericum-cream and the vehicle (secondary endpoints).

I.3.2.2.6 Premenstrual syndrome

19 women with premenstrual syndrome who were in otherwise good physical and mental health and not taking other treatments for premenstrual syndrome were investigated in a prospective, open, uncontrolled, observational pilot study (Stevinson et al 2000). The participants took hypericum tablets for two complete menstrual cycles (1 x 300 mg hypericum extract per day standardised to 900 µg hypericin). Symptoms were rated daily throughout the trial using a validated measure. The Hospital Anxiety and Depression scale and modified Social Adjustment Scale were administered at baseline and after one and two cycles of treatment. There were significant reductions in all outcome measures. The degree of improvement in overall premenstrual syndrome scores between baseline and the end of the trial was 51%, with over two-thirds of the sample demonstrating at least a 50% decrease in symptom severity. Tolerance and compliance with the treatment were encouraging.

Hicks et al (2004) performed a randomized, double-blinded, placebo-controlled trial, with two parallel treatment groups. After a no-treatment baseline cycle, volunteers were randomized to either Hypericum extract or placebo for a further two menstrual cycles. 169 normally menstruating women who experienced recurrent premenstrual symptoms were recruited onto the study. 125 completed the protocol and were included in the analysis. Study medication: 600 mg of Hypericum extract (standardized to contain 1800 µg of hypericin) or placebo (containing lactose and cellulose). A menstrual diary was used to assess changes in premenstrual symptoms. The anxiety-related subgroup of symptoms of this instrument was used as the primary outcome measure. After averaging the effects of treatment over both treatment cycles it was found that there was a trend for Hypericum extract to be superior to placebo. However, this finding was not statistically significant.

I.3.2.2.7 Menopausal symptoms

In a drug-monitoring study Grube et al (1999) investigated 12 weeks of treatment with St. John's Wort, one tablet three times daily (900 mg Hypericum, Kira), in 111 women from a general medical practice. The patients, who were between 43 and 65 years old, had climacteric symptoms characteristic of the pre- and postmenopausal state. Treatment outcome was evaluated by the Menopause Rating Scale, a self-designed questionnaire for assessing sexuality, and the Clinical Global Impression scale. The incidence and severity of typical psychological, psychosomatic, and vasomotor symptoms were recorded at baseline and after 5, 8, and 12 weeks of treatment. Substantial improvement in psychological and psychosomatic symptoms was observed. Climacteric complaints diminished or disappeared completely in the majority of women (76.4% by patient evaluation and 79.2% by physician evaluation). Of note, sexual well-being also improved after treatment with St. John's Wort extract.

In a double-blind randomized, placebo-controlled, multicenter study (Chung et al 2007), 89 peri- or postmenopausal women experiencing climacteric symptoms were treated with St. John's wort and black cohosh extract (Gynoplus), Jin-Yang Pharm., Seoul, Korea) or a matched placebo for 12 weeks. Climacteric complaints were evaluated by the Kupperman Index (KI) initially and at 4 and 12 weeks following treatment. Vaginal maturation indices, serum estradiol, FSH, LH, total cholesterol, HDL- cholesterol, LDL-cholesterol, and triglyceride levels were measured before and after treatment. From the initial 89 participants, 77 completed the trial (42 in the Gynoplus group, 35 in the placebo group). Results: Baseline characteristics were not significantly different between the two groups. Mean KI scores and hot flushes after 4 and 12 weeks were significantly lower in the Gynoplus group. Differences

in superficial cell proportion were not statistically significant. HDL levels decreased in the control group from 60.20 +/- 16.37 to 56.63 +/- 12.67, and increased in the Gynoplus group from 58.32 +/- 11.64 to 59.74 +/- 10.54; this was statistically significant (p=0.04).
Conclusion: Black cohosh and St. John's wort combination was found to be effective in alleviating climacteric symptoms and might provide benefits to lipid metabolism.

I.3.2.3 Clinical studies in special populations (e.g. elderly and children)

Depression in children

Hübner et al (2001) investigated a hypericum extract in children under 12 years with symptoms of depression and psychovegetative disturbances.
Study design: multi-center, post-marketing surveillance study; n=101 children under 12 years, dosage: 300 to 1800 mg per day.
Based on the data available for analysis, the number of physicians rating effectiveness as 'good' or 'excellent' was 72% after 2 weeks, 97% after 4 weeks and 100% after 6 weeks. The ratings by parents were very similar. There was, however, an increasing amount of missing data at each assessment point with the final evaluation including only 76% of the initial sample. Tolerability was good and no adverse events were reported. The results of this study suggest that Hypericum is a potentially safe and effective treatment for children with symptoms of depression.

Findling et al (2003) conducted an open-label prospective outpatient pilot study of St. John's wort in juvenile depression.
Youths 6 to 16 years of age meeting DSM-IV criteria for major depressive disorder, prospective, open-label, outpatient study; dosage: 3 x 150-300 mg. The extract is not further specified.
33 children with a mean age of 10.5 (2.9) years were enrolled. After 4 weeks of St. John's wort therapy, 22 youths had their dose increased to 900 mg/day. Twenty-five of the patients met response criteria after 8 weeks of treatment. Overall, St. John's wort was well tolerated. The authors conclude that hypericum may be an effective treatment for youths diagnosed with major depressive disorder.

Attention deficit, hyperactivity disorder
In a randomized, double-blind, placebo controlled study (Weber et al 2008) 54 children aged 6 to 17 years received 300 mg Hypericum extract (containing 0.3% Hypericine) 3 times daily for 8 weeks. All participants met the diagnostic criteria for ADHD. Hypericum did not improve the symptoms.

Assessor's conclusion on the use in the paedriatic population:
The only relevant study is that performed by Hübner et al (2001) with the extract LI 160. However, this study was performed as post-marketing surveillance study and not as prospective phase III clinical trial. The efficacy and safety of the medication were assessed using very coarse rating scales but not generally accepted scales like HAMD-score. The data support the safety of a potential use in children but not the efficacy which is necessary for well-established use. Therefore the use in children and adolescents is not recommended.

I.3.2.4 Assessor's overall conclusions on clinical efficacy

Dry extracts prepared with ethanol (50-80%) or methanol (50%) demonstrated clinical efficacy in the treatment of depression. Since relapse studies have been performed with

extracts containing more than 2% hyperforin these extracts are suitable for the treatment of mild to moderate major depression. Extracts containing less hyperforin should be used for the treatment of mild depression only.
There are several new therapeutical approaches (smoking cessation, treatment of alcolohism, menopausal symptoms …) published. However, the clinical evidence is at the moment insufficient to be considered in a community monograph.
The clinical data of the hyperforin cream demonstrate a great potential. However, the herbal preparation is less than 10 years in medicinal use; therefore well-established use is impossible in the monograph for now.

I.3.3 Clinical Safety/Pharmacovigilance

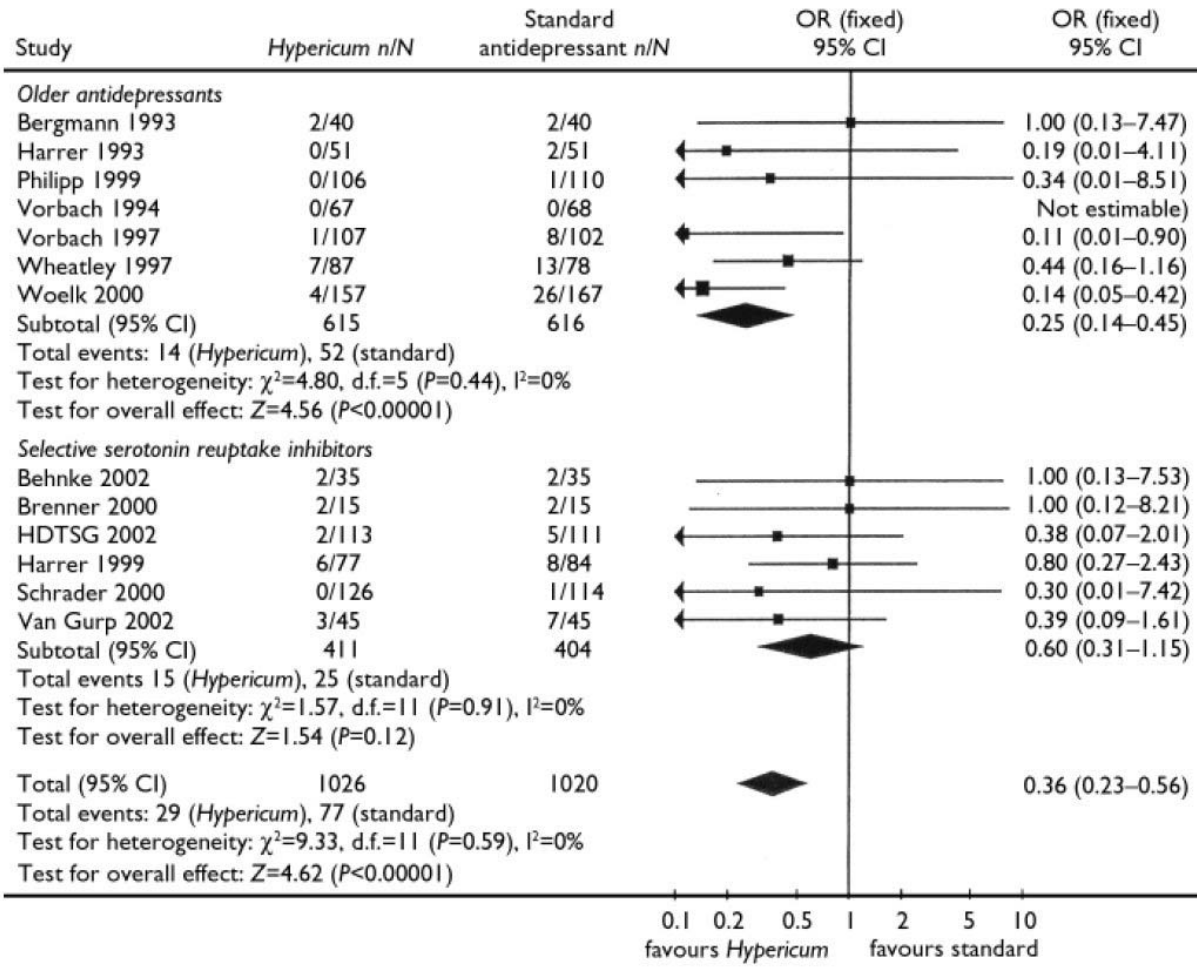
I.3.3.1 Patient exposure

I.3.3.2 Adverse events

A randomized, double-blind, crossover study was performed in healthy male volunteers aged 22 to 31 years (25 +/- 3 years; mean +/- SD) years by Siepmann et al (2004). Subjects orally received capsules with 255 to 285 mg St. John's wort extract (900 µg hypericin content), 25 mg amitriptyline, and placebo 3 times daily for periods of 14 days each with at least 14 days between. Vasoconstrictory response of cutaneous blood flow (VR) and skin conductance response (SR) following a single deep inspiration were employed as parameters of autonomic function. St. John's wort extract had no effect on VR and SR.

Stevinson et al (2004) reviewed systematically the clinical evidence associating Hypericum extract with psychotic events. Seventeen case reports associated the use of Hypericum extract with psychotic events. In 12 instances, the diagnosis was mania or hypomania. Causality is in most cases possible. In no case was a positive rechallenge reported. These case reports raise the possibility that Hypericum extract may trigger episodes of mania in vulnerable patients.

Linde performed in his meta-analysis (Linde et al 2005) also an analysis of safety. There was a trend towards lower probability of dropping out because of adverse effects in the hypericum groups compared to standard therapy.



Adverse events reported from clinical trials:

Study	Extract	Daily dose	Adverse events
Lecrubier et al (2002)	WS 5570	3 x 300 mg	n=21 of 186 Nausea (4.8 %) Headache (1.6 %) Dizziness (2.2 %) Abdominal pain (1.1%) Insomnia (1.6%)
Kalb et al (2001)	WS 5572	3 x 300 mg	Sinusitis Bronchitis Common cold
Schrader et al (1998)	Ze 117	2 x 250 mg	n=6 of 81 (7.4 %) Abdominal pain (2) Moderate Diarrhoea (1) Moderate Melancholia (1) Moderate Acute deterioration (1) Moderate Dry mouth (1) Mild
Schrader (2000)	Ze 117	2 x 250 mg	8 % only GI disturbances (5%) with an incidence greater than 2%
Randlov et al (2006)	PM235, (Cederroth International AB, Sweden)	3 x 270 mg	mild, mainly headache, gastrointestinal symptoms
Anghelescu et al (2006)	WS 5570	900 mg or 1800 mg	26.8 % of 71 no "typical adverse events (except: 1 allergic reaction to sunlight → early study termination)

			0.006 AE/d
Woelk (2000)	Ze 117	2 x 250 mg	62 of 157 (39 %) Dry mouth (13) Headache (3) Sweating (2) Asthnia (2) Nausea (1)
Philipp et al (1999)	STEI 300	3 x 350 mg	0.5 events per patient (22 %) most frequently reported adverse event: Nausea
Gastpar et al (2005)	STW3	612 mg	9.8 % related to study medication Diarrhea (1) Serious adverse events (3): shoulder blade after falling down the stairs, somatic disorder, cerebral hemorrhage)
Bjerkenstedt et al (2005)	LI 160	3x 300 mg	Adverse events: 38 Patients with adverse events: 35.1 % Adverse events possibly related to study medication: 24 Body as a wohole (13) Gastro-intestinal system disorders (6) Autonomic nervous system disorders (10) Central & peripheral nervous system disorers (10) Skin and appendages disorders (9) Psychiatric disorders (2) Others (5)
Kasper et al (2006)	WS 570	600 mg or 1200 mg (2 x 600 mg)	All adverse events. 49 (39.8 %) Serious events 1 (tendon rupture attributable to accidental injury) Ear and labyrinth disorders 3 (2.4 %) Gastrointestinal disorders 24 (19.5 %) General disorders and administraion site conditions 2 (1.6 %) Infection and infestatons 7 (5.7 %) Injury, poisoning and procedural complications 1 (0.8 %) Investigations 1 (0.8 %) Metabolism and nutrition disorders 1 (0.8 %) Musculosceletal and connective tissue disorder 1 (0.8 %) Nervous system disorder 6 (4.9 %) Psychiatric disorders 2 (1.6 %) Renal and unrinary disorders 1 (0.8 %) Reproductive system and breast disorders 1 (0.8 %) Respiratory, thoracic and mediastinal disorders 4 (3.3 %) Skin and subcutaneous disorders4 (3.3 %) Vascular disorders 1 (0.8 %)
Fava et al (2005)	LI 160	3 x 300 mg	n=90 most common adverse events. Headache (42 %) Dry mouth (22 %) Nausea (20 %) Gastrointestinal upset (20 %) Sleepiness (18 %)
Shelton et al (2001)	LI 160	900 mg/d for 4 weeks, after this period no adequat	Headache (41%) Abdominal pain (≥ 10 %)

		response, new dose 1200 mg/d	
Hypericum Depression Trial Study Group (2002)	LI 160	900 to 1500 mg (3-5 x 300 mg)	Diarrhea (21 %) Nausea (19 %) Anorgasmia (25 %) Forgetfulness (25 %) Frequent urination (27 %) Sweating (18 %) Swelling (19 %)
Szegedi et al (2005)	WS 5570	900 mg (3 x 300 mg) – 1800 mg (3 x 600 mg)	adverse events per day WS 5570 900 mg (0,029) WS 5570 1800 mg (0,039) Upper abdominal pain (9.6 %) Diarrhoea (9.6 %) Dry mouth (12.8 %) Nausea (7.2 %) Fatigue (11.2 %) Dizziness (7.2 %) Headache (10.4 %) Sleep disorder (4 %) Increased sweating (7.2 %) highest incidence: Gastrointestinal disorders (59 events in 42 patients) Nervous system disorders (35 events in 29 patients) 2 serious adverse events (psychic decompensation attributable to social problems, hypertensive crisis), both not caused by hypericum
van Gurp et al (2002)	?	900 to 1800 mg/d	Sleep disturbance (54.8 %) Anxiety (42.9 %) Sexual problems (11.9 %) Headaches (42.9 %) Dizziness (11.9 %) Tremor (19.1 %) Sweating (16.7 %) Dry mouth (38.1 %) Muscle spasms (11.9 %) Muscle or joint stiffness (19.1 %) Urinary problems (16.7 %) Difficulty digesting (19.1 %) Nausea or vomiting (9.5 %) Diarrhea (23.8 %) Lack of appetite (23.8 %) Heart palpitations (9.5 %) Fatigue (45.2 %) Pain (11.9 %) Blurred vision (14.3 %) 1 serious adverse reaction (acute manic reaction)
Laakmann et al 1998	WS 5573 WS 5572	3 x 300 mg	WS 5573 (28.6 % of 49 patients) WS 5572 (28.6 % of 49 patients) Bronchitis (3/1) Influenza-like symptoms (2/0) Cough (2/0) Infection (1/0)
Schrader 2000	Ze 117	2 x 250 mg	8 % Hypericum GI disturbances (5 %)

Lenoir et al 1999	Hyperiforce (provided by Biofroce AG, Roggwil, Switzerland)	3 x 1 tablet (standardised to either 0.17 mg, 0.33 mg, or 1 mg total hypericin per day)	There is no difference in AE with possible or probable causality in the three treatment-groups. Probable/Possible realation to study medication: Skin (0/3) Nerves (2/5) Psyche (1/1) Gastrointestinal tract (4/0) Oranism as a whole (0/2)
Harrer et al 1999	LoHyp 57	2 x 400 mg	n=12 (For this reason withdrawn: 6)
Volz et al 2000	D-0496	2 x 250 mg	n=18 at 17 % patients Influenza-like symptoms (7) Gastrointestinal tract (2) Skin (3) Infection of the urinary tract (1) Others (5)
Gastpar et al 2006	STW3-VI	900 mg	17.2 % Total AE´s. 58 Related: 10 Gastrointestinal disorders (6) Ear and labyrinth disorders (1) Skin and subcutaneous tissue disorders (1)
Wheatley 1997	LI 160	3 x 300 mg	37 % of the patients Dry mouth (5 %) Drowsiness (1 %) Sleepiness (2 %) Dizziness (1 %) Lethargy (1%) Nausea/Vomiting (7 %) Headache (7 %) Constipation (5 %) Pruritus (2 %)
Vorbach et al 1997	LI 160	3 x 600 mg	23 % of the patients n=37 Dry mouth (3) Gastric symptoms (5) tiredness/sedation (5) Restlessness (6) Tremor (2) Dizziness (5) Allergic skin reaction (1)
Rychlik et al 2001	WS 5572	600 mg/1200 mg	17 patients n=21 (13 with relation to Hypericum) AE´x frequency < 1 % Skin irritation, pruritus Allergic exanthema Nervousness, restlessness Gastrointestinal disorders (4) Diarrhoe Insomnia

According to the review by Greeson et al (2001) the overall incidence of ADR is in the range of 2%. The most commonly reported side effects were gastrointestinal irritations (0.6%), allergic reactions (0.5%), fatigue (0.4%) and restlessness (0.3%). In comparison, the overall ADR incidence for SSRIs is between 20% and 50%, including more serious side effects.

Additional case reports:

Acute neuropathy after taking 500 mg of Hypericum powder per day and exposure to sunlight (Bove 1998)

I.3.3.3 Serious adverse events and deaths

Several case reports document psychotic side effects like mania and psychosis (review in Hammerness et al 2003). The majority of the affected persons had histories of affected illness, in most cases Hypericum was combined with other psychopharmaceuticals. The symptoms remitted after discontinuation of the medication. As a precaution Hypericum extracts should not be taken by persons with a history of mania or psychosis. Seizures are reported after overdose only (Karalapillai & Bellomo 2007).

I.3.3.4 Laboratory findings

I.3.3.4.1 Phototoxicity

Hypericin

Schempp et al (1999) describe the HPLC detection of hypericin and semiquantitative detection of pseudohypericin in human serum and skin blister fluid after oral single-dose (1 x 6 tablets) or steady-state (3 x 1 tablet/day, for 7 days) administration of the Hypericum extract LI 160 in healthy volunteers (n = 12). Serum levels of hypericin and pseudohypericin were always significantly higher than skin levels (p </= 0.01). After oral single-dose administration of Hypericum extract the mean serum level of total hypericin (hypericin + pseudohypericin) was 43 ng/ml and the mean skin blister fluid level was 5.3 ng/ml. After steady-state administration the mean serum level of total hypericin was 12.5 ng/ml and the mean skin blister fluid level was 2.8 ng/ml. These skin levels are far below hypericin skin levels that are estimated to be phototoxic (>100 ng/ml).

Dry extracts

In a prospective randomized study Schempp et al (2003) investigated the effect of the Hypericum extract LI 160 on skin sensitivity to ultraviolet B (UVB), ultraviolet A (UVA), visible light (VIS) and solar simulated radiation (SIM). Seventy two volunteers of skin types II and III were included and were divided into six groups, each consisting of 12 volunteers. In the single-dose study the volunteers (n = 48) received 6 or 12 coated tablets (5400 or 10 800 microgram hypericin). In the steady-state study the volunteers (n = 24) received an initial dose of 6 tablets (5400 microgram hypericin), and subsequently 3 x 1 tablets (2700 microgram hypericin) per day for 7 days. Phototesting was performed on the volar forearms prior to medication and 6 h after the last administration of Hypericum extract. The erythema-index and melanin-index were evaluated photometrically using a mexameter. After both single-dose and steady-state administration, no significant influence on the erythema-index or melanin-index could be detected, with the exception of a marginal influence on UVB induced pigmentation (p = 0.0471) in the single-dose study. The results do not provide evidence for a phototoxic potential of the Hypericum extract LI 160 in humans when administered orally in typical clinical doses up to 1800 mg daily. This is in accordance with previous pharmacokinetic studies that found hypericin serum and skin levels after oral ingestion of Hypericum extract always to be lower than the assumed phototoxic hypericin threshold level of 1000 ng/mL.

The objective of a study by Schulz et al (2006) was to investigate the effect of two different hypericum extracts (STW 3, STW 3-VI) on photosensitivity with respect to minimal erythema dose (MED) after 14 days treatment. Both open, multiple-dose, one-phase studies were conducted in 20 healthy men, receiving one tablet per day. MED values were determined prior to hypericum extract administration (baseline) and after 14 days treatment using an

erythem tester emitting a light very similar to sun light (main emission spectrum: 285-350 nm). Skin reactions with respect to MED were evaluated 12 h, 24 h (primary endpoint), 48 h and 7 days after irradiation. All volunteers reached steady-state of hypericin/pseudohypericin plasma concentrations before study day 14, when the irradiation under treatment conditions took place. In all subjects MED was measurable under baseline and under hypericum treatment conditions. With respect to the primary endpoint, in both studies, mean MED (24 h) were not significantly different between baseline and after 14 days hypericum treatment. However, individually photosensitivity of the skin could increase under treatment conditions, just as well photosensitivity could decrease or remain unchanged. There were no clinically relevant changes in the laboratory parameters, the vital signs, physical findings and other observations related to safety during the examinations. In one study (STW 3), two adverse events were reported, both described as hypersensitivity to light in mild intensity. The two studies showed that treatment with the two hypericum extracts under steady state and under prescribed conditions were safe medications without significant increases of photosensitivity.

Hypericum oil

Schempp et al (2000) investigated the effects of Hypericum oil (hypericin 110 microg/mL) and Hypericum ointment (hypericin 30 microg/mL) on skin sensitivity to solar simulated radiation. Sixteen volunteers of the skin types II and III were tested on their volar forearms with solar simulated radiation for photosensitizing effects of Hypericum oil (n=8) and Hypericum ointment (n=8). The minimal erythema dose (MED) was determined by visual assessment, and skin erythema was evaluated photometrically. With the visual erythema score, no change of the MED could be detected after application of either Hypericum oil or Hypericum ointment (P>0.05). With the more sensitive photometric measurement, an increase of the erythema-index after treatment with the Hypericum oil could be detected (P< or =0.01). The results do not provide evidence for a severe phototoxic potential of Hypericum oil and Hypericum ointment, detectable by the clinically relevant visual erythema score. However, the trend towards increased photosensitivity detected with the more sensitive photometric measurement could become relevant in fair-skinned individuals, in diseased skin or after extended solar irradiation.

Assessor's comment:
The mentioned content of hypericin is in contrast to investigations from Maisenbacher et al (1992), these authors found only artefacts of hypericin.
From traditional use of hypericum oil it is known that the exposure to sunlight of treated parts of the skin would lead to skin irritations. In traditional medicine it is recommended to protect treated skin from sunlight.

I.3.3.5 Safety in special populations and situations

I.3.3.5.1 Intrinsic (including elderly and children) /extrinsic factors

I.3.3.5.2 Drug interactions

The extent of induction of CYP3A varies among St. John's wort products and depends on hyperforin dose (Mueller et al 2006, Gutmann et al 2006). Products that do not contain substantial amounts of hyperforin (<1%) have not been shown to produce clinically relevant enzyme induction (Madabushi et al 2006). Hypericin may be assumed to be the P-glycoprotein inducing compound, although the available evidence is less convincing (Mannel 2004).
Several meta-analyses are published on the pharmacokinetic interactions of hypericum with the cytochrome-P450 enzyme complex and P-glycoprotein (Pal et al 2006, Madabushi et al

2006, Whitten et al 2006, Zhou et al 2004, Mills et al 2004, Izzo 2004, Henderson et al 2002): Results from clinical studies and case reports indicate that self-administered hypericum extract reduce steady state plasma concentrations of alprazolam, amitriptyline, cyclosporine, tacrolimus, digoxin, fexofenadine, amprenavir, indinavir, lopinavir, ritonavir, saquinavir, benzodiazepines, methadone, nevirapine, simvastatin, theophyline, irinotecan, midazolan, triptans and warfarin. Hypericum has been also reported to cause bleeding and unwanted pregnancies when concomitantly administered with oral contraceptives. When combined with serotonin reuptake inhibitor, antidepressants (e.g. sertaline, paroxetine, nefazodone) or buspirone, hypericum extracts can cause serotonergic syndrome. Whitten et al (2006) concluded that in three studies, where the daily exposure to hyperforin was less than 4 mg, no significant effect on CYP3A4 could be detected. Brattström (2005, unpublished data, cited in Whitten et al 2006) tested the extract ZE117 in 16 females in a dose of 500 mg daily for 14 days. The women started 3 months prior to the study taking 20 µg ethinyl oestradiol and 150 µg desogestrel daily. Pharmacokinetic testing on days 7 and 22 after the treatment with ZE 117 revealed no significant differences in ethinyl estradiol or 3-ketodesgestrel (active metabolite). It is not known whether a longer treatment with ZE117 would induce CYP3A4. Arold et al (2005) report from a pharmacokinetic interaction study with 240 mg Hypericum extract daily containing 3.5 mg Hyperforin. After treatment for 11 days no siginifcant changes in the primary kinetic parameters of alprazolam, caffeine, paraxanthine, tolbutamide, 4-hydroxytolbutamide and digoxin were observed.
CYP1A2 appears to be induced by Hypericum extract only in females, the activities of CYP2D6, NAT2, and XO were not affected by Hypericum extract (Wenk et al 2004).
The causality of some of the published interactions is questioned by Schulz (2005), particularly the combinations with serotonin reuptake inhibitors and digoxin.
The elevated activity of CYP3A returns to basal level approximately 1 week after termination of Hypericum administration (Imai et al 2008). Therefore the warning in the monograph is justified that Hypericum administration should be discontinued at least 10 days prior to elective surgery.

I.3.3.5.3 Use in pregnancy and lactation

Five mothers who were taking 300 mg of St. John's wort 3 times daily (LI 160 [Jarsin]) and their breastfed infants were assessed by Klier at al (2006). Thirty-six breast milk samples (foremilk and hindmilk collected during an 18-hour period) and 5 mothers' and 2 infants' plasma samples were analyzed for hyperforin levels. Hyperforin is excreted into breast milk at low levels. However, the compound was at the limit of quantification in the 2 infants' plasma samples (0.1 ng/mL). Milk/plasma ratios ranged from 0.04 to 0.13. The relative infant doses of 0.9% to 2.5% indicate that infant exposure to hyperforin through milk is comparable to levels reported in most studies assessing anti-depressants or neuroleptics. No side effects were seen in the mothers or infants. The authors conclude that these results add to the evidence of the relative safety of St. John's wort while breast-feeding found in previous observational studies.

In a review Dugoua et al (2006) searched 7 electronic databases and compiled data according to the grade of evidence found. The authors found very weak scientific evidence based on a case report that St Johns wort is of minimal risk when taken during pregnancy. There is in vitro evidence from animal studies that St John's wort during pregnancy does not affect cognitive development nor cause long-term behavioral defects, but may lower offspring birth weight. There is weak scientific evidence that St. John's wort use during lactation does not affect maternal milk production nor affect infant weight, but, in a few cases, may cause colic, drowsiness or lethargy. There is weak scientific evidence that St John's wort induces CYP450 enzymes, which may lower serum medication levels below therapeutic range; this may be of

concern when administering medications during pregnancy and lactation. Caution is warranted with the use of St John's wort during pregnancy until further high quality human research is conducted to determine its safety. St John's wort use during lactation appears to be of minimal risk, but may cause side effects. Caution is warranted when using medications along with St John's wort.

I.3.3.5.4 Overdose

Karalapillai & Bellomo (2007) reported a case of overdose in suicidal intention of a 16-year-old girl. It has been reported that the girl had taken up to 15 tablets per day for 2 weeks and 50 tablets just before hospitalisation. Seizures and confusion were diagnosed, after 6 days the EEG was normal, no further seizures occurred in the following 6 months. The published data on the composition of the tablets are not clear ('300 µg tablets').

Assessor's comment:
In a pesonal communication the author confirmed that there is a typing error in the publication. The product contained 300 mg extract per tablet. These symptoms occurred therefore after ingestion of 4500 mg extract per day over a period of 2 weeks (approximately the 5-fold therapeutic dose) and an additional dose of 15000 mg extract (approximately the 17-fold therapeutic dose).

I.3.3.5.5 Drug abuse

I.3.3.5.6 Withdrawal and rebound

Beckman et al (2000) conducted a telephone survey of 43 subjects who had taken Hypericum extract to assess demographics, psychiatric and medical conditions, dosage, duration of use, reason for use, side effects, concomitant drugs, professional consultation, effectiveness, relapse, and withdrawal effects. Most subjects reported taking Hypericum extract for depression, and 74% did not seek medical advice. Mean dosage was 475.6+/-360 mg/day (range 300-1200 mg/day) and mean duration of therapy was 7.3+/-10.1 weeks (range 1 day-5 yrs). Among 36 (84%) reporting improvement, 18 (50%) had a psychiatric diagnosis. Twenty (47%) reported side effects, resulting in discontinuation in five (12%) and one emergency room visit. Two consumers experienced symptoms of serotonin syndrome and three reported food-drug interactions. Thirteen consumers experienced withdrawal symptoms and two had a depressive relapse. These data suggest the need for greater consumer and provider awareness of the potential risks of Hypericum extract in self-care of depression and related syndromes.

Assessor's comment:
Details on the type of products used are missing. Data from a telephone survey should be assessed reluctantly.

I.3.3.5.7 Effects on ability to drive or operate machinery or impairment of mental ability

I.3.3.6 Assessor's overall conclusions on clinical safety

The adverse events observed in clinical trials, which are most probably linked to the study medication are in general mild, the frequency is, compared to synthetic antidepressants considerably lower. The induction of CYP3A4 is well documented; the amount is directly correlated with the content of hyperforin in the herbal preparation. Pharmacokinetic interactions are documented for several drug substances metabolised via CYP3A4 having with a narrow therapeutic range. Hypericum extracts should not be used concomitantly with

these substances. Adequate studies with extracts with low hyperforin content which could justify exemptions in the wording of contraindications, special warnings and in the interactions section of the monograph are missing. Nevertheless, the risk / benefit assessment favours the benefits of the hypericum dry extracts.
The induction of the CYP3A4 is reversible within approximately 10 days after stopping ingestion of Hypericum extracts. Therefore the administration should be discontinued at least 10 days prior to surgery.
The only risk of the cutaneous application of hypericum oil seems to be the phototoxicity when treated skin is exposed to intense sunlight. A special warning should inform the patient.

I.4 ASSESSOR'S OVERALL CONCLUSIONS

Dry extracts of Hypericum perforatum demonstrated in several controlled clinical trials superiority over placebo and non inferiority against standard medication in mild to moderate major depression. Therefore these types of extracts are proposed for 'well-established use'.
The herbal tea and other mostly liquid extracts orally applied have a long tradition in folk medicine for the treatment of low mood, anxiety, to 'strenghten the nerves'. Provided that the duration of use is restricted to several days the traditional use of such herbal preparations can be granted. The cutaneous application of oil preparations is plausible based on pharmacological tests.

II COMMUNITY HERBAL MONOGRAPH ON HYPERICUM PERFORATUM L., HERBA

1. NAME OF THE MEDICINAL PRODUCT

To be specified for the individual finished product.

2. QUALITATIVE AND QUANTITATIVE COMPOSITION[5] [6]

Well-established use	Traditional use
With regard to the marketing authorisation application of Article 10(a) of Directive 2001/83/EC as amended	With regard to the registration application of Article 16d(1) of Directive 2001/83/EC as amended
Hypericum perforatum L., herba (St. John's Wort)	*Hypericum perforatum* L., herba (St. John's Wort)
i) Herbal substance Not applicable	i) Herbal substance Whole or cut, dried flowering tops, harvested during flowering time.
ii) Herbal preparations A) Dry extract (DER 3-6:1), extraction solvent methanol (80% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0% B) Dry extract (DER 4-7:1), extraction solvent methanol (80% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0% C) Dry extract (DER 3-6:1), extraction solvent ethanol (80% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0% D) Dry extract (DER 5-7:1), extraction solvent ethanol (60% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0% E) Dry extract (DER 5-7:1), extraction solvent ethanol (60% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0%	ii) Herbal preparations[7] A) Dry extract (DER 4-7:1), extraction solvent ethanol 38% (v/v) B) Dry extract (DER 3.5-6:1), extraction solvent ethanol 60% (m/m) C) Dry extract (DER 5-7:1), extraction solvent ethanol 70% (m/m) D) Liquid extract (DER 1:4-20), extraction solvent vegetable oil[8] E) Liquid extract (DER 1: 13), extraction solvent maize oil F) Tincture (DER 1:10), extraction solvent ethanol 45-50% (v/v) G) Tincture (DER 1:5), extraction solvent ethanol 50% (v/v) H) Liquid extract (DER 1:2), extraction solvent ethanol 50% I) Liquid extract (DER 1: 5-7), extraction solvent ethanol 50%. J) Expressed juice from the fresh herb

[5] The material complies with the Pharm. Eur. monograph (ref. 01/2008:1438)

[6] The declaraction of the active substance(s) for an individual finished product should be in accordance with the relevant herbal quality guidance.

[7] For safety reasons, the amount of hyperforin and hypericin should be specified.

[8] Preparation: maceration of the fresh or dried herbal substance with vegetable oil over a period of 2 days to several weeks under sun light exposure.

F) Dry extract (DER 2.5-5:1), extraction solvent ethanol (60% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0% G) Dry extract (DER 5-8:1), extraction solvent ethanol (50% v/v), hypericin 0.10-0.30%, hyperforin > 2%, flavonoids > 6.0%	(DER 1.25-2.5:1) [9] K) Comminuted herbal substance

3. PHARMACEUTICAL FORM

Well-established use	Traditional use
Herbal preparation in solid dosage forms for oral use. The pharmaceutical form should be described by the European Pharmacopoeia full standard term.	Comminuted herbal substance for tea preparation. Herbal preparations A, B, C, E, F, G, H, I, J, K in liquid or solid dosage forms for oral use. Herbal preparations D, F, G, K in liquid or semi solid dosage forms for cutaneous use. The pharmaceutical form should be described by the European Pharmacopoeia full standard term.

4. CLINICAL PARTICULARS

4.1. Therapeutic indications

Well-established use	Traditional use
Herbal medicinal product for the treatment of mild to moderate depressive episodes.	Indication 1: Herbal substance, herbal preparations A, B, C, E, F, G, H, I, J, K: Traditional herbal medicinal product for the symptomatic relief of temporary neurasthenia (e.g. difficulty in concentrating, inefficient thinking, feeling of physical weakness and exhaustion after minimal effort, gastrointestinal discomfort). Indication 2: Herbal preparations D, F, G, K: Traditional herbal medicinal product for the symptomatic treatment of minor inflammations of the skin (such as sunburn) and as an aid in healing of minor wounds.

[9] Fresh material complies with the Pharm. Eur. monograph (ref. 01/2008:1438) when dried.

	The product is a traditional herbal medicinal product for use in specified indication(s) exclusively based upon long-standing use.

4.2. Posology and method of administration

Well-established use	Traditional use
Posology *Adults and elderly*: Herbal preparation A: Single dose: 300 mg Dosage frequency: 3 times daily Daily dose: 900 mg Herbal preparation B: Single dose: 300-600 mg Dosage frequency: 1-3 times daily Daily dose: 600-1800 mg Herbal preparation C: Single dose: 900 mg Dosage frequency: 1 single daily dose Daily dose: 900 mg Herbal preparation D: Single dose: 350 mg Dosage frequency: 3 times daily Daily dose: 1050 mg Herbal preparation E: Single dose: 400 mg Dosage frequency: 2 times daily Daily dose: 800 mg Herbal preparation F: Single dose: 300-600 mg Dosage frequency: 2-3 times daily Daily dose: 900-1200 mg Herbal preparation G: Single dose: 612 mg Dosage frequency: 1 single daily dose Daily dose: 612 mg	**Posology** Indication 1: *Adults, elderly*: Herbal preparation A: Single dose: 60-180 mg Daily dose: 180-360 mg Herbal preparation B: Single dose: 85-300 mg Daily dose: 510 – 600 mg Herbal preparation C: Single dose: 270 mg Daily dose: 540 mg Herbal preparation E: Single dose: 200 mg Daily dose: 600 mg Herbal preparation F: Single dose: 2-4 ml Daily dose: 6-12 ml Herbal preparation G: Single dose: 1-1.5 ml Daily dose: 3-4.5 ml Herbal preparation H: Single dose: 0.8-1.2 ml Daily dose: 2.4-3.6 ml Herbal preparation I: Single dose: 1.3 ml Daily dose: 4.0 ml Herbal preparation J: Single dose: 10-20 ml Daily dose: 10-20 ml

Children and adolescents: The use in children and adolescents under 18 years of age is not recommended (see section 4.4 'Special warnings and precautions for use'). **Duration of use** The onset of the effect can be expected within 4 weeks of treatment. If the symptoms persist during the use of the medicinal product, a doctor should be consulted. **Method of administration** Oral use	Herbal preparation K: For tea preparation: Single dose: 2 g Daily dose: 4 g In solid dosage forms: Single dose: 300-500 mg Daily dose: 900-1000 mg *Children, adolescents:* The use in children and adolescents under 18 years of age is not recommended (see section 4.4 'Special warnings and precautions for use'). Indication 2: *Adolescents, adults, elderly*: Herbal preparations D, K: Cutaneous administration of the undiluted herbal preparation Herbal preparations F, G: Cutaneous administration of the undiluted or diluted herbal preparation *Children:* The use in children and adolescents under 18 years of age is not recommended (see section 4.4 'Special warnings and precautions for use'). **Duration of use** Indication 1: 2 weeks If the symptoms persist longer than 2 weeks during the use of the medicinal product, a doctor or a qualified health care practitioner should be consulted. Indication 2: 1 week If the symptoms persist longer than 1 week during the use of the medicinal product, a doctor or a qualified health care practitioner should be consulted.

	Method of administration Indication 1: Oral use Indication 2: Cutaneous use

4.3. Contraindications

Well-established use	Traditional use
Hypersensitivity to the active substance. Hypericum dry extract must not be used concomitantly with cyclosporine, tacrolimus, digoxin, amprenavir, indinavir and other protease-inhibitors, irinotecan and other cytostatic agents.	Indication 1: Hypersensitivity to the active substance. Hypericum extracts must not be used concomitantly with cyclosporine, tacrolimus, digoxin, amprenavir, indinavir and other protease-inhibitors, irinotecan and other cytostatic agents. Indication 2: Hypersensitivity to the active substance.

4.4. Special warnings and precautions for use

Well-established use	Traditional use
When coadministered with anticoagulants from the coumarin-type the serum concentration of these substances should be controlled regularly. During the treatment intense UV-exposure should be avoided. The product should be discontinued at least 10 days prior to elective surgery due to the potential for interactions with medicinal products used during general and regional anaesthesia. Since no sufficient data on the safe use in children are available the use in children and adolescents under 18 years of age is not recommended.	Indication 1: When coadministered with anticoagulants from the coumarin-type the serum concentration of these substances should be controlled regularly. During the treatment intense UV-exposure should be avoided. The product should be discontinued at least 10 days prior to elective surgery due to the potential for interactions with medicinal products used during general and regional anaesthesia. Since no data on the safe use in children are available the use in children and adolescents under 18 years of age is not recommended.

	For tinctures the appropriate labelling for ethanol, taken from the 'Guideline on excipients in the label and package leaflet of medicinal products for human use', must be included. Indication 2: During the treatment intense UV-exposure of the respective skin areas should be avoided. Since no data on the safe use in children are available the use in children and adolescents under 18 years of age is not recommended. If signs of skin infections are observed, a doctor or a qualified healthcare practitioner should be consulted.

4.5. Interactions with other medicinal products and other forms of interaction

Well-established use	Traditional use
Contraindicated is the concomitant use of cyclosporine, tacrolimus, digoxin, amprenavir, indinavir and other protease-inhibitors, irinotecan and other cytostatic agents. Special care should be taken with alprazolam, amitriptyline, fexofenadine, benzodiazepines, methadone, simvastatin, theophyline, midazolam, triptans and warfarin, because a reduction of plasma concentrations is possible. The reduction of plasma concentrations of oral contraceptives may lead to bleeding and unwanted pregnancies. Hypericum dry extract may cause a serotonergic syndrome when combined with antidepressants such as serotonine reuptake inhibitors (e.g. sertraline, paroxetine, nefazodone) or with buspirone.	Indication 1: Contraindicated is the concomitant use of cyclosporine, tacrolimus, digoxin, amprenavir, indinavir and other protease-inhibitors, irinotecan and other cytostatic agents. Special care should be taken with alprazolam, amitriptyline, fexofenadine, benzodiazepines, methadone, simvastatin, theophyline, midazolam, triptans and warfarin, because a reduction of plasma concentrations is possible. The reduction of plasma concentrations of oral contraceptives may lead to bleeding and unwanted pregnancies. Hypericum dry extract may cause a serotonergic syndrome when combined with antidepressants such as serotonine reuptake inhibitors (e.g. sertraline, paroxetine, nefazodone) or with buspirone. Patients taking other medicines on prescription should consult a doctor or

	pharmacist before taking Hypericum. Indication 2: None reported

4.6. Pregnancy and lactation

Well-established use	Traditional use
In the absence of sufficient data, the use during pregnancy and lactation is not recommended.	In the absence of sufficient data, the use during pregnancy and lactation is not recommended.

4.7. Effects on ability to drive and use machines

Well-established use	Traditional use
No studies on the effect on the ability to drive and use machines have been performed.	No studies on the effect on the ability to drive and use machines have been performed.

4.8. Undesirable effects

Well-established use	Traditional use
Gastrointestinal disorders, allergic reactions of the skin, fatigue and restlessness may occur. The frequency is not known. Fair-skinned individuals may react with intensified sunburn-like symptoms under intense sunlight. If other adverse reactions not mentioned above occur, a doctor or a pharmacist should be consulted.	Indication 1: Gastrointestinal disorders, allergic reactions of the skin, fatigue and restlessness may occur. The frequency is not known. Fair-skinned individuals may react with intensified sunburn-like symptoms under intense sunlight. If other adverse reactions not mentioned above occur, a doctor or a qualified health care practitioner should be consulted. Indication 2: Skin reactions may occur. The frequency is not known If other adverse reactions not mentioned above occur, a doctor or a qualified health care practitioner should be consulted.

4.9. Overdose

Well-established use	Traditional use
After ingestion of overdoses the patient should be protected from sunlight and other UV-sources for 1-2 weeks.	Indications 1, 2: No case of overdose has been reported.

5. PHARMACOLOGICAL PROPERTIES

5.1. Pharmacodynamic properties

Well-established use	Traditional use
Pharmacotherapeutic group: Other antidepressants ATC code: N06AX Hypericum dry extract inhibits the synaptosomal uptake of the neurotransmitters noradrenaline and serotonine. Subchronic treatment causes a down-regulation of beta-adrenoreceptors; it changes the behaviour of animals in several antidepressant models (e.g., forced swimming test) similar to other antidepressants. Napthodianthrones (e.g. hypericin, pseudohypericin), phloroglucin derivatives (e.g. hyperforin) and flavonoids contribute to the activity.	Not required as per Article 16c(1)(a)(iii) of Directive 2001/83/EC as amended.

5.2. Pharmacokinetic properties

Well-established use	Traditional use
The absorption of hypericin is delayed and starts about 2 hours after administration. The half-life of hypericin is about 37 hours. Maximum hyperforin levels are reached about 3 hours after administration; no accumulation could be detected. Hyperforin and the flavonoid miquelianin can cross the blood-brain-barrier.	Not required as per Article 16c(1)(a)(iii) of Directive 2001/83/EC as amended. Hyperforin is a strong enzyme inducer and may interact with many medicinal products. Therefore, for traditional herbal medicines, the hyperforin content should be kept at a minimum.

5.3. Preclinical safety data

Well-established use	Traditional use
Studies on acute toxicity and repeated dose	

toxicity did not show signs of toxic effects. The weak positive results of an ethanolic extract in the AMES-test (Salmonella typhimurium TA 98 and TA 100, with and without metabolic activation) could be assigned to quercetin. No signs of mutagenicity could be detected in further in vitro and in vivo test systems. No adequate tests on reproduction toxicity have been performed. Tests on the carcinogenic potential have not been performed. Phototoxicity: After oral application of dosages of 1800 mg of an extract per day for 15 days the skin sensitivy against UVA was increased, and the minimum dose for pigmentation was significantly reduced. In the recommended dosage no signs of phototoxicity are reported.	Studies on acute toxicity and repeated dose toxicity did not show signs of toxic effects. The weak positive results of an ethanolic extract in the AMES-test (Salmonella typhimurium TA 98 and TA 100, with and without metabolic activation) could be assigned to quercetin. No signs of mutagenicity could be detected in further in vitro and in vivo test systems. No adequate tests on reproduction toxicity have been performed. Tests on the carcinogenic potential have not been performed. Phototoxicity: After oral application of dosages of 1800 mg of an extract per day for 15 days the skin sensitivy against UVA was increased, and the minimum dose for pigmentation was significantly reduced. In the recommended dosage no signs of phototoxicity are reported.

6. PHARMACEUTICAL PARTICULARS

Well-established use	Traditional use
Extracts should be quantified with respect to hypericin, hyperforin and flavonoids[10].	Not applicable.

7. DATE OF COMPILATION/LAST REVISION

August 2008

[10] Pharm. Eur. monograph (ref. 01/2008:0765) Extracts.

Discussion

Hypericum perforatum L. is an old medicinal herb with a long history of traditional use. In the Austrian folk medicine Hypericum is the most important medicinal plant. Numerous indications can be found in the literature, such as wound healing, gout, rheumatism for external use and dyspeptic complaints and 'to strengthen the nerves' for internal use.
The literature search and the evaluation of the scientific knowledge for the preparation of an assessment report and a community monograph for the European Medicines Agency EMEA were performed under several aspects:

Each indication has strictly to be connected with a well defined herbal preparation and a defined posology. As a consequence scientific literature data bases were searched for clinical data, recent and historical pharmaceutical literature was evaluated with respect to traditional indications and preparations. Additional data on authorized products were collected in the EU member states.

Several liquid extracts, three dry extracts, the expressed juice from the fresh herb and the herbal tea were found to be traditional preparations. Out of the nearly endless lists of traditional indications only neurasthenia for oral use and symptomatic treatment of minor inflammations of the skin were found to be plausible. The posology for all of these preparations can be found in the literature. Although hypericum oil is traditionally used also in children, the use should be limited to persons older than 18 years since no sufficient data on the safe use in the paediatric population could be found. The oral use of hypericum oil was not supported, there are no constituents known which would make this use plausible.

Seven dry extracts were found to fulfil the criteria for well-established use, which means at least one well conducted clinical trial and at least 10 years of broad medicinal use. A further dry extract with sufficient clinical evidence could not be included into the draft monograph: the extract ZE117 is a refined extract, the amount of hyperforin is reduced. Since details of the manufacturing process were not available this extract could not be considered in the monograph.
All of these extracts were clinically tested in patients with single mild to moderate depressive episodes and recurrent mild to moderate depressive episodes. The minimal daily dose is about 600 mg extract. The results of clinical trials where the equivalence with standard medication was examined the fact that synthetic antidepressants usually lead to xerostomia should be considered. Blinding is therefore not really possible.
The constituents of the extracts which could be responsible for the antidepressant activity are still under discussion. Extracts with predominately positive outcome (superiority against placebo, equivalence to reference medication) meet the definition of the monograph of the Pharm. Eur. (Extraction solvent: Ethanol 50-80 % or methanol 50-80 %; Hypericin: 0.10-0.30 %, Hyperforin: < 6,0 %, Flavonoids: 6,0 %).
Most authors suggest that hyperforin is mainly responsible for the antidepressant activity. Since the extract ZE117 with low content of hyperforin exhibited antidepressant activity in several clinical trials other constituents should be considered too. In a recent publication the possible role of the flavonoids is discussed. Unfortunately the content of flavonoids is not declared in most cases.
Hypericum gained interest in clinical pharmacology at least since pharmacokinetic interactions with other substances became evident. Hyperforin is able to induce CYP 3A, therefore numerous warnings and contraindications are included into the draft monograph. The clinical trials with the extract ZE117 were too short. Therefore it is not allowed to conclude that extracts with a low hyperforin content can be used without such restrictions.

A lot of more clinical and not clinical trials about further different effects were published. Some of these studies confirm the above mentioned traditional indications (for example: The wound healing effect activity of Hypericum perforatum (Ozturk et al 2006)), other indications like “support in smoking cessation” or “treatment of alcoholism” might be in context with the antidepressant activity. These activities and indications demonstrate the potency of hypericum extracts, but further data is necessary to confirm these.

A further interesting point is the phototoxicity of Hypericum perforatum L. From traditional use of hypericum oil it is known, that the expose of sunlight to the treated parts of the skin could cause skin irritations. Therefore it is recommended to protect treated skin from sunlight. However, orally administrated hypericum dry extract under therapeutically dose is safe in this respect; no significant increase of photosensitivity was observed (Schempp 2003 and 2006).

Only poor clinical data are available on the use of Hypericum in children as well as during pregnancy and lactation. Therefore the use in this population is not recommended. Surprisingly the published preclinical safety data are not comprehensive.

The here presented review and assessment reveals that Hypericum perforatum is still a valuable herbal substance. Dry extracts can be recommended for the treatment of mild to moderate depressive episodes. Liquid extracts and other preparations are traditionally used orally in neurasthenia, topically for the treatment of small wounds or minor inflammations of the skin. The preparations can be used safely provided that the possible pharmacokinetic interactions are considered during therapy.

Abstract

The aim of this work was to draw up a first draft of an assessment report of Hypericum perforatum L. to evaluate the traditional and the well-established use of preparations of hyperici herba.

Literature was searched in PubMed, the library of pharmacy of the University of Vienna, the library of the Medical University of Vienna and in the AGES PharmMed (Austrian Medicines Agency) from March 2007 to June 2008. The assessment of the literature was performed considering the guidelines of the EMEA (European Medicines Agency).

The data presented in this draft monograph represent the most recent and comprehensive review of the well-established and traditional use of Hypericum perforatum in the European Union.

Seven different dry extracts of Hyperici herba were found to fulfil the criteria of well-established use including clinical evidence of efficacy and broad therapeutic use for more than 10 years. The indication ‘mild to moderate depressive episodes’ is proposed for products containing one of these extracts.

Liquid preparations and some other dry extracts are proposed for traditional use in the indication ‘Neurasthenia’. All these extracts are more than 30 years in medicinal use in the European Union. The topical use of liquid extracts (including hypericum oil) is proposed in the traditional indication ‘treatment of small wounds and minor inflammations of the skin’.

Additionally this present assessment of the literature comprises also aspects like pharmacology, pharmacokinetics, preclinical safety data, use in special populations, side effects and interactions.

Hypericum perforatum L. is an effective medicinal herb in the above mentioned indications. Particularly in the therapy of depression the effects which can be achieved are equal to reference medication. One main advantage of hypericum is the low rate of undesirable effects and the good compliance. In future it could be possible that the above mentioned indications will be extended, for example depression combined with alcoholism or the therapy of the premenstrual syndrome.

Zusammenfassung

Das Ziel meiner Arbeit war die Erstellung eines Vorschlags eines Berichts über Hypericum perforatum L., um die traditonelle Anwendung und die Anwendung nach well-established use von Johanniskrautpräparaten zu evaluieren.

Aus diesem Grund suchte ich nach Literatur in der Pharmaziebibliothek der Universität Wien, in der Bibliothek der Medizin Universität Wien und bei der AGES PharmMed (Austrian Medicines Agency) vom März 2007 bis Juni 2008. Der Bericht wurde nach den Richtlinien der EMEA (European Medicines Agency) erstellt.

Die dargestellten Daten dieses Berichts sind die aktuellsten und umfangsreichsten, die über die traditionelle Anwendung und die Anwendung nach well-established use von Johanniskrautpräparaten in der Europäischen Union dargestellt wurden.

Sieben verschiedene Trockenextrakte von Hyperici herba wurden gefunden, die die Kriterien der Anwendung nach well-established use erfüllen, inklusive klinischer Daten über die Wirksamkeit und die weitreichende therapeutischer Anwendung für mehr als 10 Jahre. Die Indikation ‘mild to moderate depressive episodes’ ist vorgeschlagen für Präparate, die eines dieser Extrakte enthalten.

Flüssige Zubereitungen und einige Trockenextrakte sind indiziert zur traditionellen Anwendung bei der Indikation „Neurasthenia“. Alle diese Extrakte sind für mehr als 30 Jahre in therapeutischer Anwendung in der Europäischen Union. Für die traditionellen topische Indikation ‘treatment of small wounds and minor inflammations of the skin’ werden flüssige Extrakte (inklusive Johanniskrautöl) angewendet.

Außerdem berücksichtigt der erstellte Bericht Punkte wie Pharmakologie, Pharmakokinetik, Präklinische Sicherheitsdaten, die Anwendung für spezielle Bevölkerungsgruppen, Nebenwirkungen und Interaktionen.

Hypericum perforatum L. ist eine sehr wichtige Arzneipflanze bei der Behandlung der erwähnten Indikationen, besonderes wichtig ist die Anwendung bei Depressionen, wo sie gleich wirksam ist wie die Vergleichmedikation. Ihr großer Vorteil ist die geringe Rate unerwünschter Wirkungen und eine gute Compliance. In Zukunft ist es mögliche, dass die Indikationen von Johanniskraut erweitert werden, wie z.B. zur Therapie von Depressionen in Zusammenhang mit Alkoholismus oder zur Therapie des Premenstrualen Syndrom.

List of references

Agostinis, P., Vantieghem, A., Merlevede, W., de Witte P.A.M. Hypericin in cancer treatment: more light on the way. Int. J. Biochem. Cell Biol. 34, 221-241. 2002

Anghelescu, IG., Kohnen, R., Szegedi, A., Klement, S., Kieser, M. Comparison of Hypericum extract WS 5570 and paroxetine in ongoing treatment after recovery from an episode of moderate to severe depression: results from a randomized multicenter study. Pharmacopsychiatry. **39**, 213-219. 2006

Arold, G., Donath, F., Maurer, A., Diefenbach, K., Bauer, S., Henneicke-von Zepelin, H.H., Friede, M., Roots, I. No relevant interaction with alprazolam, caffeine, tolbutamide and digoxin by treatment with a low-hyperforin St John's wort extract. Planta Med 71, 331-337. 2005

Auster, F., Schäfer, J. Arzneipflanzen. Georg Thieme Verlag, Leipzig, 1958

Axarlis, S., Mentis, A., Demetzos, C., Skaltsounis, A.L., Marselos, M., Malamas, M. Antiviral in vitro activity of Hypericum perforatum extract on the Human Cytomegalovirus (HCMV). Phytother Res 12, 507-511. 1998

Barnes, J., Anderson, LA., Phillipson, JD. Herbal medicines. Second Edition. Pharmaceutical Press, London, 2002

Barnes, J., Barber, N., Wheatley, D., Williamson, EM. A pilot randomised, open, uncontrolled, clinical study of two dosages of St John's wort (Hypericum perforatum) herb extract (LI-160) as an aid to motivational/behavioural support in smoking cessation. Planta Med. **72**, 378-382. 2006

Baureithel, K.K., Berger Büter, K., Engesser, A., Burkard, W., Schaffner, W. Inhibition of benzodiazepine binding in vitro by amentoflavone, a constituent of various species of Hypericum. Phar Acta Helv 72, 153-157. 1997

Beckman, SE., Sommi, RW., Switzer, J. Consumer use of St. John's wort: a survey on effectiveness, safety, and tolerability. Pharmacotherapy. **20**, 568-574. 2000

Behnke, K., Jensen, GS., Graubaum, HJ., Gruenwald, J. Hypericum perforatum versus fluoxetine in the treatment of mild to moderate depression. Adv Ther. **19**, 43-52. 2002

Beijamini, V., Andreatini, R. Effects of Hypericum perforatum and paroxetine on rat performance in the elevated T-maze. Pharmacol Res. **48**, 199-207. 2003

Benedi, J., Arroyo, R., Romero, C., Martin-Aragon, S., Villar, AM. Antioxidant properties and protective effects of a standardized extract of Hypericum perforatum on hydrogen peroxide-induced oxidative damage in PC12 cells. Life Sci. **75**, 1263-1276. 2004

Bergmann. R., Nüßner. J., Demling. J. Behandlung leichter bis mittelschwerer Depressionen. TW Neurol Psychiatr. **7**, 235-240. 1993

Bernd, A., Simon, S., Ramirez Bosca, A., Kippenberger, S., Diaz Alperi, J., Miquel, J., Villalba Garcia, JF., Pamies Mira, D., Kaufmann R., Phototoxic effects of Hypericum extract

in cultures of human keratinocytes compared with those of psoralen. Photochem Photobiol. **69**, 218-221. 1999

Biber. A., Fischer. H., Romer. A., Chatterjee. SS. Oral bioavailability of hyperforin from hypericum extracts in rats and human volunteers. Pharmacopsychiatry. **31**, Suppl 1, 36-43. 1998

Bjerkensted, L., Edman, GV., Alken, RG., Mannel, M. Hypericum extract LI 160 and fluoxetine in mild to moderate depression: a randomized, placebo-controlled multi-center study in outpatients. Eur Arch Psychiatry Clin Neurosci. **255**, 40-47. 2005

Bladt. S., Wagner. H. Inhibition of MAO by fractions and constituents of hypericum extract. J Geriatr Psychiatry Neurol. **7**, Suppl 1, 57-59. 1994

Böhme, H. Kommentar zum Europäischen Arzneibuch. Wissenschaftliche Verlagsgesellschaft, Stuttgart, 2006

Borges, L.V., do Carmo Cancino, J.C., Peters, V.M., Las Casas, L., de Oliveira Guerra, M. Development of pregnancy in rats treted with Hypericum perforatum. Phytother. Res. 19, 885-887. 2005

Bove. G.M. Acute neuropathy after exposure to sun in a patient treated with St John's wort. Lancet 352, 1121. 1998

Bradley, P. British Herbal Compendium. Vol 2. British Herbal Medicin Association, 2006

Brenner, R., Azbel, V., Madhuscoodanan, S., Pawlowska, M. Comparison of an extract of hypericum (LI 160) and sertraline in the treatment of depression: a double-blind, randomized pilot study. Clin Ther. **22**, 411-419. 2000

Butterweck. V., Brockens. T., Korte. B., Wittkowski. W., Winterhoff. H. Long-term effects of St. John's wort and hypericin on monoamine levels in rat hypothalamus and hippocampus. Brain Res. **930**, 21-9. 2002

Butterweck. V., Jurgenliemk. G., Nahrstedt. A., Winterhoff. H. Flavonoids from Hypericum perforatum show antidepressant activity in the forced swimming test. Planta Med. **66**, 3-6. 2000

Butterweck. V., Lieflander-Wulf. U., Winterhoff. H., Nahrstedt. A. Plasma levels of hypericin in presence of procyanidin B2 and hyperoside: a pharmacokinetic study in rats. Planta Med. **69**, 189-192.2003

Butterweck. V., Nahrstedt. A. Was ist bekannt über Johanniskraut? Phytochemie und Pharmakologie. Pharmazie in unserer Zeit. **32**, 212. 2003

Butterweck. V., Petereit. F., Winterhoff. H., Nahrstedt. A. Solubilized hypericin and pseudohypericin from Hypericum perforatum exert antidepressant activity in the forced swimming test. Planta Med. **64**, 291-294. 1998

Caelo 2008: personal communication

Capasso, R., Borrelli, F., Montanaro, V., Altieri, V., Capasso, F., Izzo, AA. Effects of the antidepressant St. John's wort (Hypericum perforatum) on rat and human vas deferens contractility. J Urol. **173**, 2194-2197. 2005

Calapai. G., Crupi. A., Firenzuoli. F., Interrera. G., Squadrito. F., Parisi. A., De Sarro. G., Caputi. A. Serotonin, norepinephrine and dopamine involvement in the antidepressant action of hypericum perforatum. Pharmacopsychiatry. **34**, 45-49. 2001

Catania, MA., Firenzuoli, F., Crupi, A., Mannucci, C., Caputi, AP., Calapai, G. Hypericum perforatum attenuates nicotine withdrawal signs in mice. Psychopharmacology. **169**, 186-189. 2003

Cervo. L., Roizo. M., Ekalle-Soppo. CB., Guiso. G., Morazzoni. P., Caccia. S. Role of hyperforin in the antidepressant-like activity of Hypericum perforatum extracts. Psychopharmacology.**164**, 423-428. 2002

Chatterjee. SS., Bhattacharya. SK., Wonnemann. M., Singer. A., Müller. WE. Hyperforin as a possible antidepressant component of hypericum extracts. Life Sci. **63**, 499-510. 1998

Chung, DJ., Kim, HY., Park, KH., Jeong, KA., Lee, SK., Lee, YI., Hur, SE., Cho, MS., Lee, BS., Bai, SW., Kim, CM., Cho, SH., Hwang, JY., Park, JH. Black Cohosh and St. John's Wort (GYNO-Plus(R)) for Climacteric Symptoms. Yonsei Med J. **48**, 289-294. 2007

Commission E monographs. The Complete German Commission E Monographs. American Botanical Council, Austin, 1998

Coskun, I., Tayfun Uzbay, I., Ozturk, N., Ozturk, Y. Attenuation of ethanol withdrawal syndrome by extract of Hypericum perforatum in Wistar rats. Fundam Clin Pharmacol. **20**, 481-488. 2006

DAC, Deutscher Arzneimittel Codex. J010 Hyperici herba. Deutscher Apotheker-Verlag, Stuttgart, 1991-1999

De Vry, J., Maurel, S., Schreiber, R., de Beun, R., Jentzsch, KR. Comparison of hypericum extracts with imipramine and fluoxetine in animal models of depression and alcoholism. Eur Neuropsychopharmacol. **9**, 461-468. 1999

Dimpfel W. Electropharmacogram from quercetin is similar to that of moclobemide and other antidepressant drugs. Z. Phytother. 29, S6. 2008

Dinamarca, MC., Cerpa, W., Garrido, J., Hancke, JL., Inestrosa, NC. Hyperforin prevents beta-amyloid neurotoxicity and spatial memory impairments by disaggregation of Alzheimer's amyloid-beta-deposits. Mol Psychiatry. **11**, 1032-1048. 2006

Dingermann, T., Hiller, K., Schneider, G., Zündorfer, I. Schneider Arzneidrogen. Elesevier, München, 2004

Dugoua, JJ., Mill, E., Perri, D., Koren, G. Safety and efficacy of St. John's wort (hypericum) during pregnancy and lactation. Can J Clin Pharmacol. **13**, 268-276. 2006

Duke, JA. Handbook of Medicinal Herbs. CRP Press LLC, 2002

Eckert, GP., Muller, WE. Effects of hyperforin on the fluidity of brain membranes. Pharmacopsychiatry. **34**, 22-25. 2001

Ellis, KA., Stough, C., Vitetta, L., Heinrich, K., Nathan, PJ. An investigation into the acute nootropic effects of Hypericum perforatum L. (St. John's Wort) in healthy human volunteers. Behav Pharmacol. **12**, 173-182. 2001

El-Sherbiny, DA., Khalifa, AE., Attia, AS., Eldenshary Eel, D. Hypericum perforatum extract demonstrates antioxidant properties against elevated rat brain oxidative status induced by amnestic dose of scopolamine. Pharmacol Biochem Behav. **76**, 525-533. 2003

Ergänzungsbuch zum Deutschen Arzneibuch (Erg.-B6). Deutscher Apotheker-Verlag, Berlin, 1941

ESCOP Monograph. HYPERICI HERBA (St. John´s Wort). ESCOP Monographs Second edition. ESCOP 2003

Fava, M., Alpert, J., Nierenberg, AA., Mischoulon, D., Otto, MW., Zajecka, J., Murck, H., Rosenbaum, JF. A Double-blind, randomized trial of St John's wort, fluoxetine, and placebo in major depressive disorder. J Clin Psychopharmacol. **25**, 441-447. 2005

Findling, RL., McNamara, NK., O´Riordan, MA., Reed, MD., Demeter, CA., Branicky, LA., Blumer, JL. An open-label pilot study of St. John's wort in juvenile depression. J Am Acad Child Adolesc Psychiatry. **42**, 908-914. 2003

Fischer, B., Hartwich, C. Handbuch der pharmazeutischen Praxis. Verlag Julius Springer, Berlin, 1902

Flamm, S., Kroeber, L., Seel, H. Pharmakodynamik Deutscher Heilpflanzen. Hippokrates Verlag, Stuttgart, 1940

Flausino, OA. Jr., Zangrossi, H.Jr., Salgado, JV., Viana, MB. Effects of acute and chronic treatment with Hypericum perforatum L. (LI 160) on different anxiety-related responses in rats. Pharmacol Biochem Behav. **71**, 251-257. 2002

Fox. E., Murphy. RF., McCully. CL., Adamson. PC. Plasma pharmacokinetics and cerebrospinal fluid penetration of hypericin in nonhuman primates. Cancer Chemother Pharmacol. **47**, 41-44. 2001

Frerichs, G., Arends, G., Zörnig, H. Handbuch der pharmazeutischen Praxis. Verlag Julius Springer, Berlin, 1938

Friede, M. Henneicke von Zepelin, HH., Freudenstein, J. Differential therapy of mild to moderate depressive episodes (ICD-10 F 32.0; F 32.1) with St. John's wort. Pharmacopsychiatry. **34**, 38-41. 2001

Froestl, B., Steiner, B., Muller, WE. Enhancement of proteolytic processing of the beta-amyloid precursor protein by hyperforin. Biochem Pharmacol. **66**, 2177-2184. 2003

Gastpar, M., Singer, A., Zeller, K. Comparative efficacy and safety of a once-daily dosage of hypericum extract STW3-VI and citalopram in patients with moderate depression: a double-blind, randomised, multicentre, placebo-controlled study. Pharmacopsychiatry. **39**, 66-75. 2006

Gastpar, M., Singer, A., Zeller, K. Efficacy and tolerability of hypericum extract STW3 in long-term treatment with a once-daily dosage in comparison with sertraline. Pharmacopsychiatry. **38**, 78-86. 2005

Genovese, T., Mazzon, E., Di Paola, R., Muia, C, Crisafulli, C., Menegazzi, M., Malleo, G., Suzuki, H., Cuzzocrea, S. Hypericum perforatum attenuates the development of cerulein-induced acute pancreatitis in mice. Shock. **25**, 161-167. 2006

Genovese, T., Mazzon, E., Menegazzi, M., Di Paola, R., Muia, C., Crisafulli, C., Bramanti, P., Suzuki, H., Cuzzocrea, S. Neuroprotection and enhanced recovery with hypericum perforatum extract after experimental spinal cord injury in mice. Shock. **25**, 608-617. 2006

Gerlach, S. Hypericum. PhD Thesis, University Vienna, 2008

Gibbons, S., Ohlendorf, B., Johnsen, I. The genus Hypericum--a valuable resource of anti-Staphylococcal leads. Fitoterapia. **73**, 300-304. 2002

Gobbi. M., Moia. M., Pirona. L., Morizzoni. P., Minnini. T. In vitro binding studies with two hypericum perforatum extracts--hyperforin, hypericin and biapigenin--on 5-HT6, 5-HT7, GABA(A)/benzodiazepine, sigma, NPY-Y1/Y2 receptors and dopamine transporters.Pharmacopsychiatry. **34**, Suppl 1, 45-48. 2001

Greeson. JM., Sanford. B., Monti. DA. St. John's wort (Hypericum perforatum): a review of the current pharmacological, toxicological, and clinical literature. Psychopharmacology. **153**, 402-414. 2001

Gregoretti, B., Stebel, M., Candussio, L., Crivellato, E., Bartoli, F., Decorti, G. Toxicity of Hypericum perforatum (St. John's wort) administered during pregnancy and lactation in rats. Toxicol Appl Pharmacol. **200**, 201-205. 2004

Grube, B, Grünwald, J., Walper, A. Hopfenmüller, W. Johanniskraut bei leichten temporären Verstimmungen. naturamed 11, 21-27. 1996

Grube, B., Walper, A., Wheatley, D. St. John's Wort extract: efficacy for menopausal symptoms of psychological origin. Adv Ther. **16**, 177-186. 1999

Gruenwald, J., Brendler, T., Jaenicke, C. PDR for Herbal Medicines. Medical Economics Company, Montvale, 1998

Gruenwald, J., Brendler, T., Jaenicke, C. PDR for Herbal Medicines. Medical Economics Company, Montvale, 2004

Grundmann, O., Kelber, O., Butterweck, V. Effects of St. John's wort extract and single constituents on stress-induced hyperthermia in mice. Planta Med. **72**, 1366-1371. 2006

Gutmann, H., Poller, B., Berger Büter, K., Pfrunder, A., Schaffner, W., Drewe, J. Hypericum perforatum: which constituents may induce intestinal MDR1 and CYP3A4 mRNA expression? Planta Med 72, 685-690: 2006

Häberlein H., Prenner L., Zeller K. Einfluss von Hyperosid und Hyperforin auf die Regulation und laterale Mobilität von beta1-adrenergen Rezeptoren an lebenden C6-Glioblastomzellen: ein postsynaptischer Ansatz zum Wirkmechanismus von Johanniskraut. Z. Phytother. 29, S8, 2008

Hager CD-Rom, Blaschek, W., Ebel, S., Hackenthal, E., Holzgrabe, U., Keller, K., Reichluing, J., Schulz, V. HagerROM. Springer Verlag, Berlin, Heidelberg, 2006

Hager, H. Hager´s Handbuch der pharmazeutischen Praxis. Verlag Julius Springer, Berlin, 1878

Hamacher, H., Wahl, MA. Selbstmedikation. 10. Ergänzungslieferung, Deutscher Apotheker Verlag Stuttgart, 2006

Hamacher, H., Wahl, MA. Selbstmedikation. 8. Ergänzungslieferung, Deutscher Apotheker Verlag Stuttgart, 2001

Hamacher, H., Wahl, MA. Selbstmedikation. 9. Ergänzungslieferung, Deutscher Apotheker Verlag Stuttgart, 2003

Hammerness, P., Basch, E., Ulbricht, C., Barrette, E.P., Foppa, I., Basch, S., Bent, S., Boon, H., Ernst, E. St. John's wort: A systematic review of adverse effects and drug interactions for the consultation psychiatrist. Psychosomatics 44(4), 271-282. 2003

Hänsel, R. Kapitel 10, Teedrogen. Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart, 1993

Hänsel, R., Keller, K., Rimpler, H., Schneider, G. Drogen E-O. Springer Verlag, Berlin, 1993

Hänsel, R., Sticher, O. Pharmakognosie – Phytopharmazie. Springer Medizin Verlag, Heidelberg, 2007

Hänsel, R., Sticher, O. Pharmakognosie – Phytopharmazie. Springer Medizin Verlag, Heidelberg, 1972

Hänsgen, KD, Vesper, J, Antidepressive Wirksamkeit eines hochdosierten Hypericum-Extrakts. Münchner Medizinische Wochenschrift **138**, 29-33, 1996

Hänsgen, KD, Vesper, J, Ploch, M, Multicenter double-blind study examining the antidepressant effectiveness of the hypericum extract LI 160. J Geriatr Psychiatr Neurol **7**, S15-S18. 1994

Harrer, G, Hübner WD, Podzuweit H, Effectiveness and tolerance of the hypericum extract LI 160 compared to maprotiline: a multicenter double-blind study. J Geriatr Psychiatr Neurol **7**, S24-S28. 1994

Harrer, G., Sohmidt, K., Kuhn, K., Biller, A., Comparison of equivalence between the St. John's wort extract LoHyp-57 and fluoxetine. Arzneim-Forsch/Drug Res. **49**, 289-296. 1999

He, YY., Chignell, CF., Miller, DS., Andley, UP., Roberts, JE. Phototoxicity in human lens epithelial cells promoted by St. John's Wort. Photochem Photobiol. **80**, 583-586. 2004

Heinrich, M., Barnes, J., Gibbons, S., Williamson, E. Fundamentals of Pharmacognosy and Phytotherapie. Elsevier Science Limited, 2004

Henderson, L., Yue, Q.Y., Bergquist, C., Gerden, B., Arlett, P. St John's wort (Hypericum perforatum): Drug interactions and clinical outcomes. Br. J. Clin. Pharmacol. 54, 349-356. 2002

Hicks, SM., Walker, AF., Gallagher, J., Middleton, RW., Wright, J. The significance of "nonsignificance" in randomized controlled studies: a discussion inspired by a double-blinded study on St. John's wort (Hypericum perforatum L.) for premenstrual symptoms. J Altern Complement Med. **10**, 925-932. 2004

Hoffmann. J., Kühl. ED. Therapie von depressiven Zuständen mit Hypericum. Ztschr Allgmed. **55**, 776-782. 1979

Hübner, WD., Kirste, T. Experience with St John's Wort (Hypericum perforatum) in children under 12 years with symptoms of depression and psychovegetative disturbances. Phytother Res. **15**, 367-370. 2001

Hunt, EJ., Lester, CE., Lester, EA., Tackett, RL. Effect of St. John's wort on free radical production. Life Sci. **69**, 181-190. 2001

Hypericum Depression Trial Study Group. Effect of Hypericum perforatum (St John's wort) in major depressive disorder: a randomized controlled trial. JAMA. **287**, 1807-1814. 2002

Imai, H., Kotegawa, T., Tsutsumi, K., Morimoto, T., Eshima, N., Nakan, S., Ohashi, K. The recovery time-course of CYP3A after induction by St John's wort administration. Br J Clin Pharmacol 65, 701-707. 2008

Irion, H.Drogisten-Lexikon. Springer Verlag, Berlin/Göttingen/Heidelberg, 1955

Ivetic, V., Popovic, M., Mimica-Dukic, N., Barak, O., Pilija, V. St. John's wort (Hypericum perforatum L.) and kindling(auslösend) epilepsy in rabbit. Phytomedicine. **9**, 496-499. 2002

Izzo, AA. Drug interactions with St. John's Wort (Hypericum perforatum): a review of the clinical evidence. Int J Clin Pharmacol Ther. **42**, 139-148. 2004

Jang, MH., Lee, TH, Shin, MC., Bahn, GH., Kim, JW., Shin, DH., Kim, EH., Kim, CJ. Protective effect of Hypericum perforatum Linn (St. John's wort) against hydrogen peroxide-induced apoptosis on human neuroblastoma cells. Neurosic Lett. **329**. 177-180. 2002

Johne. A., Perloff. ES., Bauer. S., Schmider. J., Mai. I., Brockmoller. J., Roots. I. Impact of cytochrome P-450 inhibition by cimetidine and induction by carbamazepine on the kinetics of hypericin and pseudohypericin in healthy volunteers. Eur J Clin Pharmacol. **60**, 617-622. 2004

Juergenliemk. G., Boje. K., Huewel. S., Lohmann. C., Galla. HJ., Nahrstedt. A. In vitro studies indicate that miquelianin (quercetin 3-O-beta-D-glucuronopyranoside) is able to reach the CNS from the small intestine. Planta Med. **69**, 1013-1017. 2003

Kalb, R., Trautmann-Sponsel, RD., Kieser, M. Efficacy and tolerability of hypericum extract WS 5572 versus placebo in mildly to moderately depressed patients. A randomized double-blind multicenter clinical trial. Pharmacopsychiatry. **34**, 96-103. 2001

Kaltschmidt, B., Heinrich, M., Kaltschmidt, C. Stimulus-dependent activation of NF-kappaB specifies apoptosis or neuroprotection in cerebellar granule cells. Neuromolecular Med. **2**, 299-309. 2002

Kamuhabwa. AR., Augustijns. P., de Witte. PA. In vitro transport and uptake of protohypericin and hypericin in the Caco-2 model. Int J Pharm. **188**, 81-86. 1999

Karalapillai, D.,C., Bellomo, R. Convulsions associated with an overdose of St. John's wort. Med J Austr 186(4), 213-214. 2007

Karsten, Weber, Fischer. Lehrbuch der Pharmakognosie. Gustav Fischer Verlag, Stuttgart, 1962

Kasper, S., Anghelescu, IG., Szegedi, A., Dienel, A., Kieser, M. Superior efficacy of St John's wort extract WS 5570 compared to placebo in patients with major depression: a randomized, double-blind, placebo-controlled, multi-center trial. BMC Med. **4**, 14. 2006

Kasper, S., Volz, H.P., Moller, H.J., Dienel, A. Kieser, M. Continuation and long-term maintenance treatment with Hypericum extract WS 5570 after recovery from an acute episode of moderate depresseion – A double-blind, randomized, placebo controlled long-term trial. Eur Neuropsychopharmacol Aug 9. 2008

Khalifa, AE. Hypericum perforatum as a nootropic drug: enhancement of retrieval memory of a passive avoidance conditioning paradigm in mice. J Ethnopharmacol. **76**, 49-57. 2001

Kientsch. U., Burgi. S., Ruedeberg. C., Probst. S., Honegger. UE. St. John's wort extract Ze 117 (Hypericum perforatum) inhibits norepinephrine and serotonin uptake into rat brain slices and reduces 3-adrenoceptor numbers on cultured rat brain cells. Pharmacopsychiatry. **34**, Suppl 1, 56-60. 2001

Klier, CM., Schmid-Siegel, B., Schafer, MR., Lenz, G., Saria, A., Lee, A., Zernig, G. St. John's wort (Hypericum perforatum) and breastfeeding: plasma and breast milk concentrations of hyperforin for 5 mothers and 2 infants. J Clin Psychiatry. **67**, 305-309. 2006

Klusa, V., Germane, S., Noldner, M., Chatterjee, SS. Hypericum extract and hyperforin: memory-enhancing properties in rodents. Pharmacopsychiatry. **34**, 61-69. 2001

Kumar, V., Jaiswal, AK., Singh, PN., Bhattacharya, SK. Anxiolytic activity of Indian Hypericum perforatum Linn: an experimental study. Indian J Exp Biol. **38**, 36-41. 2000

Kumar, V., Khanna, VK., Seth, PK., Singh, PN., Bhattacharya, SK. Brain neurotransmitter receptor binding and nootropic studies on Indian Hypericum perforatum Linn. Phytother Res. **16**, 210-216. 2002

Kumar, V., Singh, PN., Muruganandam, AV., Bhattacharya, SK. Effect of Indian Hypericum perforatum Linn on animal models of cognitive dysfunction. J Ethnopharmacol. **72**, 119-128. 2000

Laakmann, G., Jahn, G., Schule, C. Hypericum perforatum extract in treatment of mild to moderate depression. Clinical and pharmacological aspects. Nervenarzt. **73**, 600-612. 2002

Laakmann, G., Schule, C., Baghai, T., Kieser, M. St. John's wort in mild to moderate depression: the relevance of hyperforin for the clinical efficacy. Pharmacopsychiatry. **31**, 54-59. 1998

Laggner, H., Schreier, S., Hermann, M., Exner, M., Muhl, A., Gmeiner, BM., Kapiotis, S. The main components of St John's Wort inhibit low-density lipoprotein atherogenic modification: a beneficial "side effect" of an OTC antidepressant drug? Free Radic Res. **41**, 234-241. 2007

Länger, R., Kubelka, W., Phytokodex. Krause & Pachernegg GmbH, Gablitz, 2001

Lavie, G., Mazur, Y., Lavie, D., Meruelo, D. The chemical and biological properties of hypericin – a compound with a broad spectrum of biological activities. Med Res Rev 15, 111-119. 1995

Lecrubier, Y., Clerc, G., Didi, R. Kieser, M. Efficacy of St. John's wort extract WS 5570 in major depression: a double-blind, placebo-controlled trial. Am J Psychiatry. **159**, 1361-1366. 2002

Lenoir, S., Degenring, FH., Saller, R. A double-blind randomised trial to investigate three different concentrations of a standardised fresh plant extract obtained from the shoot tips of Hypericum perforatum L. Phytomedicine. **6**, 141-146. 1999

Leuschner, J. Preclinical toxicological profile of Hypericum extract LI 160. Abstracts of the 2nd International Conference on Phytomedicine, Munich. SL 80, 1996

Linde, K., Berner, M., Egger, M., Mulrow, C. St John's wort for depression: meta-analysis of randomised controlled trials. Br J Psychiatry. **186**, 99-107. 2005

Linde, K. Johanniskraut bei Depression – eine Übersicht der randomisierten Studien bezogen auf einzelne Extrakte. http://www.phytotherapie-komitee.de/Forschung/kfn_hyperic_281206.pdf, 2007

List, PH., Hörhammer, L. Hager`s Handbuch der pharmazeutischen Praxis. Springer Verlag, Berlin/Heidelberg, 1976

Lu, YH., Du, CB., Liu, JW., Hong, W., Wei, DZ. Neuroprotective effects of Hypericum perforatum on trauma induced by hydrogen peroxide in PC12 cells. Am J Chin Med. **32**, 397-405. 2004

Madabushi, R., Frank, B., Drewelow, B., Derendorf, H., Butterweck, V. Hyperforin in St. John's wort drug interactions. Eur J Clin Pharmacol. **62**, 225-233. 2006

Madaus, G. Lehrbuch der biologischen Heilmittel. Band II. Georg Thieme Verlag, Leipzig, 1938

Maisenbacher, P., Kovar, KA. Analysis and stability of Hyperici oleum. Planta Med. **58**, 351-354. 1992

Mannel, M. Drug interactions with St John's wort : mechanisms and clinical implications. Drug Saf. **27**, 773-797. 2004

Mannucci, C., Pieratti, A., Firenzuoli, F., Caputi, AP., Calapai, G. Serotonin mediates beneficial effects of Hypericum perforatum on nicotine withdrawal signs. Phytomedicine. **14**, 645-651.2007

Martindale. The complete drug references. Pharmaceutical Press, London, 2002

Meier B. Vergleichbarkeit von Phytopharmaka am Beispiel von Johanniskraut. In: Loew, D., Blume, H., Dingermann Th. Phytopharmaka V, Steinkopff, Darmstadt, 1999

Mills. E., Montori. VM., Wu. P., Gallicano. K., Clarke. M., Guyatt. G. Interaction of St John's wort with conventional drugs: systematic review of clinical trials. BMJ. **329**, 23-30. 2004

Misane. I., Ogren. SO. Effects of Hypericum perforatum (St. John's wort) on passive avoidance in the rat: evaluation of potential neurochemical mechanisms underlying its antidepressant activity. Pharmacopsychiatry. **34**, Suppl 1, 89-97. 2001

Mohanasundari, M., Sabesan, M. Modulating effect of Hypericum perforatum extract on astrocytes in MPTP induced Parkinson's disease in mice. Eur Rev Med Pharmacol Sci. **11**, 17-20. 2007

Mohanasundari. M., Srinivasan. MS., Sethuupathy. S., Sabesan. M. Enhanced neuroprotective effect by combination of bromocriptine and Hypericum perforatum extract against MPTP-induced neurotoxicity in mice. J Neurol Sci. **249**, 140-144. 2006

Montgomery. SA., Hübner. WD., Grigoleit. HG. Efficacy and tolerability of St. John´s wort extract compared with placebo in patients with mild to moderate depressive disorder. Phytomedicine. 7, Suppl 2, 107. 2000

Moreno, RA., Teng, CT., Almeida, KM., Tavares Junior, H. Hypericum perforatum versus fluoxetine in the treatment of mild to moderate depression: a randomized double-blind trial in a Brazilian sample. Rev Bras Psiauiatr. **28**, 29-32. 2006

Mueller, SC., Majcher-Peszynska, J., Uehleke, B., Klammt, S., Mundkowski, RG., Miekisch, W., Sievens, H., Bauer, S., Frank, B., Kundt, G., Drewelow, B. The extent of induction of CYP3A by St. John's wort varies among products and is linked to hyperforin dose. Eur J Clin Pharmacol. **62**, 29-36. 2006

Muller, T., Mannel, M., Murck, H., Rahlfs, VW. Treatment of somatoform disorders with St. John's wort: a randomized, double-blind and placebo-controlled trial. Psychosom Med. **66**, 538-547. 2004

Müller. WE., Rolli. M., Schäfer. C., Hafner. U. Effects of hypericum extract (LI 160) in biochemical models of antidepressant activity. Pharmacopsychiatry. **30**, Suppl 2, 102-107. 1997

Müller, W.E., Singer, A., Wonnemann, M, Hafner, U. Rolli, M., Schäfer, C. Hyperforin represents the neurotransmitter reuptake inhibiting constituenjt of Hypericum extract. Pharmacopsychiatry 31, Suppl.. 16-21. 1998

Murck, H., Fava, M., Alpert, J., Nierenberg, AA., Mischoulon, D., Otto, MW., Zajecka, J., Mannel, M., Rosenbaum, JF. Hypericum extract in patients with MDD and reversed vegetative signs: re-analysis from data of a double-blind, randomized trial of hypericum extract, fluoxetine, and placebo. Int J Neuropsychopharmacol. **8**, 215-221.2005

Murck, H., Spitznagel, H., Ploch, M., Seibel, K., Schaffler, K. Hypericum extract reverses S-ketamine-induced changes in auditory evoked potentials in humans - possible implications for the treatment of schizophrenia. Biol Psychiatry. **59**, 440-445. 2006

Nöldner. M., Schötz. K. Rutin is essential for the antidepressant activity of Hypericum perforatum
extracts in the forced swimming test. Planta Med. **68**, 577-580. 2002

Okpanyi. SN., Lizba. H., Scholl. BC., Miltenburger. HG. Genotoxicity of a standardized Hypericum extract. Arzneimittelforschung. **40**, 851-855. 1990

Ondrizek, RR., chan, PJ., Patton, WC., King, A. An alternative medicine study of herbal effects on the penetration of zona-free hamster oocytes and the integrity of sperm deoxyribonucleic acid. Fertil Steril. **71**, 517-522. 1999

Ozturk, N., Korkmaz, S., Ozturk Y. Wound-healing activity of St. John's Wort (Hypericum perforatum L.) on chicken embryonic fibroblasts. J Ethnopharmacol. **111**, 33-39. 2006

Pal, D., Mitra, AK. MDR- and CYP3A4-mediated drug-herbal interactions. Life Sci. **78**, 2131-2145. 2006

Panossian, A.G,. Gabrielian, E., Manvelian, V., Jurcic, K., Wagner, H. Immunosuppressive effects of hypericin on stimulated human leukocytes: inhibition of the arachidonic acid release, leukotriene B_4 and interleukin-1α production and activation of nitric oxide formation. Phytomed. 3, 19-28. 1996

Paulke, A., Nöldner, M., Schubert-Zsilavecz, M., Wurglics, M. St. John’s wort flavonoids and their metabolites show antidepressant activity and accumulate in brain after mutiple oral dose. Pharmazie 63, 296-302. 2008

Perfumi, M., Ciccocioppo, R., Angeletti, S., Cucculelli, M. Massi, M. Effects of Hypericum perforatum extraction on alcohol intake in Marchigian Sardinian alcohol-preferring rats. Alcohol Alcohol. **34**, 690-698. 1999

Perfumi, M., Mattioli, L., Cucculetti, M., Massi, M. Reduction of ethanol intake by chronic treatment with Hypericum perforatum, alone or combined with naltrexone in rats. J Psychopharmacol. **19**, 448-454. 2005

Perfumi, M., Mattioli, L., Forti, L., Massi, M., Ciccocioppo, R. Effect of Hypericum perforatum CO2 extract on the motivational properties(Wirksamkeit) of ethanol in alcohol-preferring rats. Alcohol Alcohol. **40**, 291-296. 2005

Perfumi, M., Panocka, I., Ciccocioppo, R., Vitali, D., Froldi, R., Massi, M. Effects of a methanolic extract and a hyperforin-enriched CO2 extract of Hypericum perforatum on alcohol intake in rats. Alcohol Alcohol. **36**, 199-206. 2001

Perfumi, M., Santoni, M., Cippitelli, A., Ciccocioppo, R., Froldi, R., Massi, M. Hypericum perforatum CO2 extract and opioid receptor antagonists act synergistically to reduce ethanol intake in alcohol-preferring rats. Alcohol Clin Exp Res. **27**, 1554-1562. 2003

Pharm. Eur.

Philipp, M., Kohnen, R., Hiller, KO. Hypericum extract versus imipramine or placebo in patients with moderate depression: randomised multicentre study of treatment for eight weeks. BMJ. **319**,1534-1538. 1999

Poginsky. B., Westendorf. J., Prosenc. N., Kuppe. M., Marquardt. H. Dtsch Apoth Ztg. **128**, 1364-1366. 1988

Randlov, C., Mehlsen, J., Thomsen, CF., Hedman, C., von Fircks, H., Winther, K. The efficacy of St. John's Wort in patients with minor depressive symptoms or dysthymia--a double-blind placebo-controlled study. Phytomedicin. **13**, 215-221. 2006

Rayburn, WF., Gonzalez, CL., Christensen, HD., Harkins, TL., Kupiec, TC. Impact of hypericum (St.-John's-wort) given prenatally on cognition of mice offspring. Neurotoxicol Teratol. **23**, 629-637. 2001

Rezvani, AH., Overstreet, DH., Yang, Y., Clark, E. Jr. Attenuation of alcohol intake by extract of Hypericum perforatum (St. John's Wort) in two different strains of alcohol-preferring rats. Alcohol Alcohol. **34**, 699-705. 1999

Roder, C., Schaefer, M., Leucht, S. Meta-analysis of effectiveness and tolerability of treatment of mild to moderate depression with St. John's Wort. Fortschr Neurol Psychiatr. **72**, 330-343. 2004

Rychlik, R., Siedentop, H., von den Driesch, V., Kasper, S. St. John's wort extract WS 5572 in minor to moderately severe depression. Effectiveness and tolerance of 600 and 1200 mg active ingredient daily. Fortschr Med Orig. **119**, 119-128. 2001

Sanchesz-Reus, MI., Gomez del Rio MA., Iglesias, I., Elorza, M., Slowing, K., Benedi, J. Standardized Hypericum perforatum reduces oxidative stress and increases gene expression of antioxidant enzymes on rotenone-exposed rats. Neuropharmacology. **52**, 606-616. 2007

Sattler. S., Schaefer. U., Schneider. W., Hoelzl. J., Lehr. CM. Binding, uptake, and transport of hypericin by Caco-2 cell monolayers. J Pharm Sci. **86**, 1120-1126. 1997

Schempp, CM., Ludtke R., Winghofer, B., Simon, JC. Effect of topical application of Hypericum perforatum extract (St. John's wort) on skin sensitivity to solar simulated radiation. Photodermatol Photoimmunol Photomed. **16**, 125-128. 2000

Schempp, CM., Muller, K., Winghofer, B., Schulte-Monting, J., Simon, JC. Single-dose and steady-state administration of Hypericum perforatum extract (St John's Wort) does not influence skin sensitivity to UV radiation, visible light, and solar-simulated radiation. Arch Dermatol. **137**, 512-513. 2001

Schempp, CM., Simon-Haarhaus, B., Simon, JC. Phototoxic and apoptosis-inducing capacity of pseudohypericin. Planta Med. **68**, 171-173. 2002

Schempp, CM., Windeck, T., Hezel, S., Simon, JC. Topical treatment of atopic dermatitis with St. John's wort cream--a randomized, placebo controlled, double blind half-side comparison. Phytomedicine. **10**, 31-37. 2003

Schempp, CM., Winghofer, B., Langheinrich, M., Schopf, E., Simon, JC. Hypericin levels in human serum and interstitial skin blister fluid after oral single-dose and steady-state administration of Hypericum perforatum extract (St. John's wort). Skin Pharmacol Appl Skin Physiol. **12**, 299-304. 1999

Schempp, CM., Winghofer, B., Ludtke, R., Simon-Haarhaus, B., Schopf, E., Simon, JC. Topical application of St John's wort (Hypericum perforatum L.) and of its metabolite hyperforin inhibits the allostimulatory capacity of epidermal cells. Br J Dermatol. 142, 979-984. 2000

Schempp, CM., Winghofer, B., Muller, K., Schulte-Monting, J., Mannel, M., Schopf, E., Simon, JC. Effect of oral administration of Hypericum perforatum extract (St. John's Wort) on skin erythema and pigmentation induced by UVB, UVA, visible light and solar simulated radiation. Phytother Res. **17**, 141-146. 2003

Schey, KL., Patat, S., Chignell, CF., Datillo, M., Wang, RH., Roberts, JE. Photooxidation of lens alpha-crystallin by hypericin (active ingredient in St. John's Wort). Photochem Photobiol. **72**, 200-203. 2000

Schimmer. O., Häfele. E., Krüger. A. The mutagenic potencies of plant extracts containing quercetin in Salmonella typhimurium TA98 and TA100. Mutat Res. **206**, 201-208. 1988

Schmitt, LA., Liu, Y., Murphy, PA., Birt, DF. Evaluation of the light-sensitive cytotoxicity of Hypericum perforatum extracts, fractions, and pure compounds. J Agric Food Chem. **54**, 2881-2890. 2006

Schmitt, LA., Liu, Y., Murphy, PA., Petrich, JW., Dixon, PM., Birt, DF. Reduction in hypericin-induced phototoxicity by Hypericum perforatum extracts and pure compounds. J Photochem Photobiol B. **85**, 118-130. 2006

Schneider, G. Arzneidrogen, Wissenschaftsverlag, Mannheim, 1990

Schrader, E. Equivalence of St John's wort extract (Ze 117) and fluoxetine: a randomized, controlled study in mild-moderate depression. Int Clin Psychopharmacol. **15**, 61-68. 2000

Schrader, E., Meier, B., Brattström, A. Hypericum treatment of mild-moderate depression in a placebo-controlled study. A prospective, double-blind, randomized, placebo-controlled, multicentre study. Human Psychopharmacology. **13**, 163-169. 1998

Schulz, HU., Schurer, M., Bassler, D., Weiser, D. Investigation of the effect on photosensitivity following multiple oral dosing of two different hypericum extracts in healthy men. Arzneimittelforschung. **56**, 212-221. 2006

Schulz. V., Schurer. M., Bassler. B., Weiser. D. Investigation of pharmacokinetic data of hypericin, pseudohypericin, hyperforin and the flavonoids quercetin and isorhamnetin revealed from single and multiple oral dose studies with a hypericum extract containing tablet in healthy male volunteers. Arzneimittelforschung. **55**, 561-568. 2005

Shelton, RC., Keller, MB., Gelenberg, A., Dunner, DL., Herschfeld, R., Thase, ME., Russell, J., Lydiard, RB., Crits-Cristoph, P., Gallop, R., Todd, L., Hellerstein, D., Goodnick, P., Keitner, G., Stahl, SM., Halbreich, U. Effectiveness of St John's wort in major depression: a randomized controlled trial. JAMA. **285**, 1978-1986. 2001

Siepmann, M., Kirch, W., Krause, S., Joraschky, P., Muek-Weymann, M. The effects of St. John's wort extract and amitriptyline on autonomic responses of blood vessels and sweat glands in healthy volunteers. J Clin Psychopharmacol. **24**, 79-82. 2004

Siepmann, M., Krause, S., Joraschky, P., Muck-Weymann, M., Kirch, W. The effects of St John's wort extract on heart rate variability, cognitive function and quantitative EEG: a comparison with amitriptyline and placebo in healthy men. Br J Pharmacol. **54**, 272-282. 2002

Silva, BA., Dias, AC., Ferreres, F., Malva, JO., Oliveira, CR. Neuroprotective effect of H. perforatum extracts on beta-amyloid-induced neurotoxicity. Neurotox Res. **6**, 119-130. 2004

Simbrey. K., Winterhoff. H., Butterweck. V. Extracts of St. John's wort and various constituents affect beta-adrenergic binding in rat frontal cortex. Life Sci. **74**, 1027-1038. 2004

Simmen, U., Schweitzer, C., Burkard, W., Schaffner, W., Lundstrom, K. Hypericum perforatum inhibits the binding of μ- and κ-opioid receptor expressed with the Semliki Forest virus system. Pharm Acta Helv 73, 53-56. 1998

Sommer, H, Harrer, G, Placebo-controlled double-blind study examining the effectiveness of an hypericum preparation in 105 mildly depressed patients. J Geriatr Psychiatr Neurol **7**, S9-S11. 1994

Sosa, S., Pace, R., Bornancin, A., Morazzoni, P., Riva, A., Tubaro, A., Della Loggia, R. Topical anti-inflammatory activity of extracts and compounds from Hypericum perforatum L. J Pharm Pharmacol. **59**, 703-709. 2007

Spiess, PE., Grossmann, HB. Fluorescence cystoscopy: is it ready for use in routine clinical practice? Curr Opin Urol. **16**, 372-376. 2006

Stevinson, C., Ernst, E. A pilot study of Hypericum perforatum for the treatment of premenstrual syndrome. BJOG. **107**, 870-876. 2000

Stevinson, C., Ernst, E. Can St. John's wort trigger psychoses? Int J Clin Pharmacol Ther. **42**, 473-480. 2004

Swiss Pharmacopoeia (Pharm. Helv. 8). Eidgenössisches Department des Inneren, Bern, 1999

Szegedi, A., Kohnen, R., Dienel, A., Kieser, M. Acute treatment of moderate to severe depression with hypericum extract WS 5570 (St John's wort): randomised controlled double blind non-inferiority trial versus paroxetine. BMJ. **330**, 503-506. 2005

Teufel-Mayer. R., Gleitz., J. Effects of long-term administration of hypericum extracts on the affinity and density of the central serotonergic 5-HT1 A and 5-HT2 A receptors. Pharmacopsychiatry. **30**, Suppl 2, 113-113. 1997

Thiede. HM., Walper. A. Inhibition of MAO and COMT by hypericum extracts and hypericin. J Geriatr Psychiatry Neurol. **7**, Suppl 1, 54-56. 1994

Traynor, NJ., Beattie, PE., Ibbotson, SH., Moseley, H., Ferguson, J., Woods, JA. Photogenotoxicity of hypericin in HaCaT keratinocytes: implications(Auswirkung) for St. John's Wort supplements and high dose UVA-1 therapy. Toxicol Lett. **158**, 220-224. 2005

Trofimiuk, E., Walesiuk, A., Braszko, JJ. St john's wort (Hypericum perforatum) counteracts deleterious effects of the chronic restraint stress on recall in rats. Acta Neurobiol Exp. **66**, 129-138. 2006

Trofimiuk, E., Walesiuk, A., Braszko, JJ. St John's wort (Hypericum perforatum) diminishes cognitive impairment caused by the chronic restraint stress in rats. Pharmacol Res. **51**,239-246. 2005

Uebelhack, R., Gruenwald, J., Graubaum, HJ., Busch, R. Efficacy and tolerability of Hypericum extract STW 3-VI in patients with moderate depression: a double-blind, randomized, placebo-controlled clinical trial. Adv Ther. **21**, 265-275. 2004

Upton, R. St. John's Wort, Monograph of the American Herbal Pharmacopoeia, 1997

Usai. M., Leggio. B., Grappi. S., Nanni. G., Gambarana. C., Tolu. P., Giachetti. D., De Mortis. MG. Hypericum perforatum subspecies angustifolium shows a protective activity on the consequences of unavoidable stress exposure at lower doses than Hypericum perforatum perforatum. Pharmacopsychiatry. **36**, 283-287. 2003

Uzbay, T.I. Hypericum perforatum and substance dependence: A review. Phytother Res 22, 578-582. 2008

Van Gurp, G., Meterissian, GB., Haiek, LN., McCusker, J., Bellavance, F. St John's wort or sertraline? Randomized controlled trial in primary care. Can Fam Physician. **48**, 905-912. 2002

Volz, HP., Murck, H., Kasper, S., Moller, HJ. St John's wort extract (LI 160) in somatoform disorders: results of a placebo-controlled trial. Psychopharmacology. **164**, 294-300. 2002

Vorbach. EU., Arnoldt. KH., Hubner. WD. Efficacy and tolerability of St. John's wort extract LI 160 versus imipramine in patients with severe depressive episodes according to ICD-10. Pharmacopsychiatry. **30**, Suppl 2, 81-85. 1997

Volz. HP., Laux. P. Potential treatment for subthreshold and mild depression: a comparison of St. John's wort extracts and fluoxetine. Compr Pschiatry. **4**, Suppl 1, 133-137. 2000

Wagner, H., Bauer, R. Arzneidrogen und ihre Inhaltsstoffe. Band 2. Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart, 1999

Wahlman, J., Hirst, M., Roberts, JE., Prickett, CD., Trevithick, JR. Focal length variability and protein leakage as tools for measuring photooxidative damage to the lens. Photochem Photobiol. **78**, 88-92. 2003

Weber, W., Van der Stoep, A., McCarty, R.L., Weiss, N.S,. Biederman, J., McClellan, J. Hypericum perforatum (St John's wort) for attention-deficit/hyperactivity disorder in children and adolescents. JAMA 299, 2633-2641. 2008

Wenk, M., Todesco, L., Krahenbuhl, S. Effect of St John's wort on the activities of CYP1A2, CYP3A4, CYP2D6, N-acetyltransferase 2, and xanthine oxidase in healthy males and females. Br J Clin Pharmacol. **57**, 495-499. 2004

Werneke, U., Horn, O., Taylor, DM. How effective is St John's wort? The evidence revisited. J Clin Psychiatry. **65**, 611-617. 2004

Wheatley. D. LI 160, an extract of St. John's wort, versus amitriptyline in mildly to moderately depressed outpatients--a controlled 6-week clinical trial. Pharmacopsychiatry. **30**, Suppl 2, 77-80. 1997

Whitten, DL., Myers, SP., Hawrelak, JA., Wohlmuth, H. The effect of St John's wort extracts on CYP3A: a systematic review of prospective clinical trials. Br J Clin Pharmacol. **62**, 512-526. 2006

WHO monographs. WHO-Monographs on selected medical plants. World Health Organization, Geneva, 2002

Wichtl, M. Teedrogen. Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart, 1984

Wichtl, M. Teedrogen. Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart, 4. Auflage 2002

Wichtl, M. Herbal Drugs and Phytopharmaceuticals. Medpharm scientific publishers, Stuttgart 2004

Widy-Tyszkiewicz, E., Piechal, A., Joniec, I., Blecharz-Klin, K. Long term administration of Hypericum perforatum improves spatial learning and memory in the water maze. Biol Pharm Bull. **25**, 1289-1294. 2002

Wielgus, AR., Chignell, CF., Miller, DS., Van Houten, B., Meyer, J., Hu, DN., Roberts, JE. Phototoxicity in Human Retinal Epithelial Cells Promoted by Hypericin, a Component of St. John's Wort. Photochem Photobiol. **83**, 706-713 2006

Wilhelm, KP., Biel, S., Siegers, CP. Role of flavonoids in controlling the phototoxicity of Hypericum perforatum extracts. Phytomedicine. **8**, 306-309. 2001

Woelk, H. Comparison of St John's wort and imipramine for treating depression: randomised controlled trial. BMJ. **321**, 536-539. 2000

Wright, CW., Gott, M., Grayson, B., Hanna, M., Smith, AG., Sunter, A., Neill, JC. Correlation of hyperforin content of Hypericum perforatum (St John's Wort) extracts with their effects on alcohol drinking in C57BL/6J mice: a preliminary study. J Psychopharmacol. **17**, 403-408. 2003

Wurglics. M., Schubert, Zsilavecz. M. Hypericum perforatum: a 'modern' herbal antidepressant: pharmacokinetics ofactive ingredients. Clin Pharmacokinet. **45**, 449-468. 2006

Zhou, S., Chan, E., Pan, SQ., Huang, M., Lee, EJ. Pharmacokinetic interactions of drugs with St John's wort. J Psychopharmacol. **18**, 262-276. 2004

Zörnig, H. Arzneidrogen. Teil II. Verlag Klinkhardt, Leipzig, 1911

Zou, Y, Lu, Y., Wei, D. Antioxidant activity of a flavonoid-rich extract of Hypericum perforatum L. in vitro. J Agric Food Chem. **52**, 5032-5039. 2004

Zou, Y., Lu, Y., Wei, D. Hypocholesterolemic effects of a flavonoid-rich extract of Hypericum perforatum L. in rats fed a cholesterol-rich diet. J Agric Food Chem. **53**, 2462-2466. 2005

Curriculum vitae

Personal data

Name	Dagmar Gruber
Nationality	Austria
Date of birth	27th of September 1982
Place of birth	Oberpullendorf
Parents	Mag. pharm. Ilse Skarka
	Mag. pharm. Johannes Gruber
Family status	unmarried

Education

1989-1993	Elementary school Perchtoldsdorf, Rosseggergasse
1993-1997	AHS Perchtoldsdorf, Rosseggergasse
1997-2002	BHAK Maygasse, 1130 Wien
June 2002	Final exam (Matura)
2002-2008	Study of Pharmacy at the University of Vienna

Part time employment

August 2002-October 2003	Beethoven Apotheke, 1190 Wien
since November 2003	Salvator Apotheke, 2340 Mödling