05 May 2021
EMA/HMPC/625623/2015
Committee on Herbal Medicinal Products (HMPC)

Assessment report on *Saccharomyces cerevisiae* CBS 5926

Draft

Based on Article 10a of Directive 2001/83/EC as amended (well-established use)

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

Herbal substance(s) (binomial scientific name of the plant, including plant part)	*Saccharomyces cerevisiae* CBS 5926
Herbal preparation(s)	Not applicable
Pharmaceutical form(s)	Not applicable
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Note: This draft assessment report is published to support the release for public consultation of the draft public statement on *Saccharomyces cerevisiae* CBS 5926. It is a working document, not yet edited, and shall be further developed after the release for consultation of the public statement. Interested parties are welcome to submit comments to the HMPC secretariat, which will be taken into consideration but no 'overview of comments received during the public consultation' will be prepared in relation to the comments that will be received on this assessment report. The publication of this draft assessment report has been agreed to facilitate the understanding by Interested Parties of the assessment that has been carried out so far and led to the preparation of the draft public statement.

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1. Introduction

Disclaimer

Due to the initial decision of the HMPC to conduct a scientific assessment in order to possibly establish a monograph on Saccharomyces cerevisiae, a draft assessment report and draft EU monograph supporting a traditional and well-established use on herbal medicinal products containing Saccharomyces cerevisiae CBS 5926 were prepared in line with EMA/HMPC standard procedures and principles based on Article 10a of Directive 2001/83/EC (well-established use) and based on Article 16d(1), Article 16f and Article 16h of Directive 2001/83/EC (traditional use).

However, no absolute majority required for adoption of the monograph was achieved and the HMPC is of the opinion that a European Union herbal monograph on Saccharomyces cerevisiae CBS 5926 cannot be established due to different opinions on the classification of this yeast. On one hand, yeasts are considered 'Fungi' in a broad sense and as such could be considered falling under the herbal substance definition of Directive 2001/83/EC, however on the other hand living yeast could also be considered covered by the Live Biotherapeutic Products general monograph of the European Pharmacopoeia.

Therefore, this assessment report is published for information and transparency purposes only. This document is without prejudice to any classification of Saccharomyces cerevisiae either as a herbal substance or a live biotherapeutic substance, which is ultimately within the remit of individual EU/EEA Member States. The present HMPC assessment report shall in no way be regarded as a HMPC position on the classification of Saccharomyces cerevisiae as an herbal or biological active substance of medicinal product.

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

The HMPC has discussed whether living yeast cells (fungi) such as *Saccharomyces cerevisiae* (strain CBS 5962) may be considered herbal substances/preparations, based on Directive 2001/83/EC which defines herbal substances as "All mainly whole, fragmented or cut plants, plant parts, algae, fungi, lichen in an unprocessed, usually dried, form, but sometimes fresh. Certain exudates that have not been subjected to a specific treatment are also considered to be herbal substances. Herbal substances are precisely defined by the plant part used and the botanical name according to the binomial system (genus, species, variety and author)."

For the purposes of this assessment report, a specific yeast strain of *Saccharomyces cerevisiae* was considered as a herbal substance. However, the proposal for adoption of a monograph of herbal substance did not reach the required absolute majority in the HMPC in view of divergent views on the classification of *Saccharomyces cerevisia* CBS 5927 (see section 6. Overall conclusions (benefit-risk assessment)).

- Herbal substance(s)

Yeasts are eukaryotic single-cell microorganisms classified in the kingdom Fungi but they do not form a single taxonomic or phylogenetic group. As it is known from fungi yeasts can have asexual and sexual reproductive cycles. Mainly, the vegetative growth in yeasts is asexual reproduction by budding or fission.

Yeast species either require oxygen for aerobic cellular respiration (obligate aerobes) or are anaerobic, but also have aerobic methods of energy production (facultative anaerobes). They use different kinds

of carbohydrates to produce carbon dioxide and water or in the absence of oxygen a fermentation process is used to produce ethanol and carbon dioxide.
The use of different carbohydratesis a characteristic property to differentiate between various yeast species.
Yeasts grow best in a neutral or slightly acidic pH environment.

At present, a correct definition and classification for yeast species are still a matter of debate. The common classification is listed below:
According to the common scientific classification system, there are two separate phyla: Ascomycota and Basidiomycota. Within the phyla of the Ascomycota two different subphyla are known, the Saccharomycotina and the Taphrinomycotina. The only class in the subphylum Saccharomycotina is Saccharomycetes (Eriksson and Winka, 1997). The class of Saccharomycetes contains the order Saccharomycetales, a budding yeast. Twelve families are included in the order Saccharomycetales. One of these families is the so-called Saccharomycetaceae, which is presented in a wide variety of habitats. This family contains among others the species *Saccharomyces cerevisiae*.
Saccharomyces cerevisiae is one of the most important and useful yeasts. A strain of *Saccharomyces cerevisiae* was the first eukaryotic genome to be completely sequenced (Sankoff, 2009). Because of their useful physiological properties, many yeast species are considered of importance in food production. Strains of *Saccharomyces cerevisiae* have a long tradition of use in baking and brewing. It is assumed that *Saccharomyces cerevisiae* strains are isolated from fruits. This is also known for the *Saccharomyces cerevisiae* strain described in the current monograph (McFarland, 2010).

The correct taxonomy of the *Saccharomyces cerevisiae* strain has been discussed over years. The strain has received considerable discussion about its valid nomenclature (McFarland, 1996; McCullough *et al*., 1998). Before the actual era of yeast taxonomy by means of gene sequences and other molecular criteria, identifications were based on phenotypic tests and assimilation profiles.

In 1996, McFarland postulated that *Saccharomyces boulardii* has been shown to be a separate species of *Saccharomyces* on the basis of several taxonomic, metabolic, and molecular parameters. *Saccharomyces boulardii* is a wild *Saccharomyces* strain that does not produce ascospores or uses galactose as a carbon source (as do wild *Saccharomyces cerevisiae* strains). This strain is also given a separate designation by the American Type Culture Collection (ATCC 74012). *Saccharomyces boulardii* has different oxidative utilization and fermentation patterns that can distinguish it from *Saccharomyces cerevisiae* (McFarland, 1996).

Although the phenotype sometimes can be used to correctly identify species, in the following years molecular comparisons have shown that many earlier identifications based on phenotype have been incorrect (Kurtzman and Fell, 2006). Rapid detection and accurate identification of yeasts are now possible by use of a variety of molecular methods. Many phylogenetic relationships among the yeasts and other fungi have been clarified by analysis of gene sequence divergence (Kurtzman and Fell, 2006). Based on various molecular methods it is consistently shown that *Saccharomyces boulardii* is similar to *Saccharomyces cerevisae* and cannot be differentiated as a separate species (Vaughan-Martini and Martini, 1987; McCullough *et al*., 1998; Blaschek *et al*., 2013; Vaughan-Martini, 2003). Therefore, the yeast previously named *Saccharomyces boulardii* is considered as a special strain within the heterogeneous species *Saccharomyces cerevisiae* (Bassetti *et al.,* 1998; Piarroux *et al.,* 1999; Perapoch *et al*., 2000; Lherm *et al.,* 2002; Mitterdorfer, 2002a; Mitterdorfer, 2002b; Cassone *et al*., 2003; Riquelme *et al.,* 2003).

However, the term *Saccharomyces boulardii* has been used more frequently as medicinal term in the therapeutic usage than the taxonomically correct term *Saccharomyces cerevisiae* strain Boulard. Instead of labelling the strain, it is more common to use the name of the culture collection indicating the respective number of the deposit: "*Saccharomyces cerevisiae* CBS 5926", which is the primarily

used reference for this strain. The strain is also deposited in the American Type Culture Collection with the number ATCC 74012 and additionally in the Institute Pasteur de Paris as I-745.

Differences and similarities of *Saccharomyces cerevisiae* CBS 5926 and laboratory strains of *S. cerevisae* – without any characterization - are observed (Tab. 1) (Edwards-Ingram *et al*., 2007; Lukaszewicz, 2012). The majority of the studies shows that especially the ability for pseudohyphal swtiching, the survival at low acid pH and higher optimal growth temperature are features having a direct influence on the use in medicinal products.

Table 1: Differences and similarities of *Saccharomyces cerevisiae* CBS 5926 and laboratory strains of *Saccharomyces cerevisae* (taken from McFarland, 1996; Edwards-Ingram *et al*., 2004; Malgoire *et al*., 2005; Edwards-Ingram *et al*., 2007; Lukaszewicz, 2012)

***Saccharomyces cerevisiae* CBS 5926**	***Saccharomyces cerevisiae* (laboratory strain)**
higher optimal growth temperature (ca. 37°C)	lower optimal growth temperature (ca. 30°C)
higher resistance to low pH	lower resistance to low pH
does not use galactose	uses galactose
asporogenous in contrast to *Saccharomyces cerevisiae* but may produce fertile hybrids with *Saccharomyces cerevisiae* strains	sporogenous
lost all intact Ty1/2 elements	contains several Ty1/1 elements
microsattelite typing shows genotypic differences	
trisomic for chromosome IX	stable strains with various ploidy

Ty 1/1 is a specific transposable element for yeast

- Herbal preparation(s)

Saccharomyces cerevisiae CBS 5926 is being commercialized as a freeze-dried powder obtained from an aqueous suspension using a fermentative culture procedure referring to a Cell-Bank-System. For this reason, the manufacture is inspected according to GMP Annex 2 "Manufacture of Biological active substances and Medicinal Products for Human Use" and authorized by the Regulatory Authorities as a biological active substance. Usually, the freeze-dried yeast is a preparation containing lactose as excipient during the freeze-drying process. However, there are also some preparations, which are produced by fluid bed drying. These two possible manufacturing routes can be considered as comparable taking into account the specification characteristics of the yeast preparation, as described below.

As main characteristic of the yeast, the number of viable cells is determined in addition to the strain identity. The respective specification for the content of viable cells is set according to the yeast used in the clinical studies and is assayed by dilution series, smear, and count of the colonies grown. Regarding the specification of the yeast used in the clinical studies the acceptance criterion for the parameter "content of viable yeast cells" is set to "$\geqslant 2 \times 10^{10}$ CFU (cell forming units)/g dried (native) yeast". As it is common for microbiological results the limit of the result must be interpreted in such a way that a deviation of +/- ½ power of ten is still considered "within the specification".

The identity is proven based on microscopic and macroscopic tests, a visual test based on the form and color of the cell colonies grown on Sabouraud agar. A DNA-fingerprint is used to compare the yeast with reference to *Saccharomyces cerevisiae* CBS 5926. Furthermore, a biochemical identification test should be performed by means of the different carbohydrate assimilation profiles of yeast strains resulting in a specific code by metabolizing different substrates.

In the majority of the clinical studies included in this assessment report hard capsules containing the dried yeast powder are used as pharmaceutical dosage form. In the release specification, the acceptance criterion for the parameter assay the content of viable cells is defined as "$\geqslant 2 \times 10^{10}$ CFU/g dried yeast powder". During stability, at least "4×10^{9} CFU/g dried yeast powder" should be ensured. As it is mentioned before, the limit of the result must be interpreted so that a deviation of +/- ½ power of ten is still considered "within the specification". These acceptance criteria are set according to the specification of the finished product used in the clinical studies. On the market, there is also the dried yeast powder in sachets which is considered a comparable dosage form.

For an application of other strains than the above-mentioned *Saccharomyces cerevisiae* CBS 5926, on which this assessment report is based, the genetic identity should be documented. Referring also to the respective decision of the EFSA within a Scientific Opinion on the substantiation of health claims to Article 13(1) of Regulation EC No 1924/2006 for identification of a yeast species restriction fragment length polymorphism analysis (RFLP) or sequencing analysis of DNA taxonomic markers should be used. The strain identification must be supported by chromosome length polymorphism analysis by PFGE, RAPDs, microsatellite DNA polymorphism analysis or other internationally accepted genetic typing molecular techniques (EFSA, 2010; 2012a; 2012b). Furthermore, characteristics reported from literature in table 1 should also be considered concerning properties of different strains.

In literature different terms have been used for *Saccharomyces cerevisiae* CBS 5926, *Saccharomyces cerevisiae* as well as *Saccharomyces boulardii*. For the investigations mentioned cited in this assessment report the terms used by the authors are maintained.

- Herbal medicinal products

In the clinical studies assessed different medicinal products have been administered to the patients. According to the homepage of Biocodex (http://www.biocodex.com/en/therapeutic-areas-products) *Saccharomyces boulardii* CNCM I-745 (*Saccharomyces cerevisiae* CBS 5926) is the active principle of the several medicinal products. Hence, it is concluded that all these medicinal products are identical with regard to *Saccharomyces cerevisiae* strain and number of living cells. Recent package leaflets mention amounts of living cells as follows: e.g. $\geqslant 1.8 \times 10^{10}$ viable cells/g lyophilisate or 5×10^{9} viable cells/250 mg sachet (corresponding to 2×10^{10} viable cells/g).
For Floratil® contradictory information is available. On the Biocodex homepage, Floratil® is characterised as a natural yeast-based probiotic and thus differs to the medicinal products mentioned before. Based on the patent description and on package leaflets available Floratil does not differ from medicinal products containing *Saccharomyces boulardii*. A recent package leaflet mentions not less than 5×10^{9} cells *Saccharomyces boulardii*/g lyophilisate whereas the reference Corrêa *et al* (2011) mentions 4×10^{9} viable cells in one 200 mg capsule which correlates to 2×10^{10} viable cells/g.

1.2. Search and assessment methodology

This assessment report was based on the literature on *Saccharomyces boulardii/cerevisiae* available at the „Federal Institute for Drugs and Medicinal Devices" in Germany and the publications provided by the AESGP (Association of the European Self-Medication Industry), Biocodex and Pierre-Fabre in response to the EMA HMPC call for data on *Saccharomyces boulardii/cerevisiae* of 14.02.2014. Furthermore, a literature search in the DIMDI database was performed in July 2015 using the following

terms: *Saccharomyces boulardii/cerevisiae*, humans, clinical, preclinical, safety. A separate literature research was performed for the therapeutic use of *Saccharomyces boulardii/cerevisiae* in the indication irritable bowel syndrome in the database XMEDALL (terms used: irritable bowel syndrome, *Saccharomyces*).

To update the list of references, in March and April 2020 a second literature search was performed in Pubmed database for articles published in the last 5 years using the following terms: "*Saccharomyces boulardii/cerevisiae,* humans, diarrhea", and "*Saccharomyces boulardii/cerevisiae*, humans, irritable bowel syndrome" respectively and using the EBSCO Discovery Service (EDS) with the following terms: (*saccharomyces boulardii* OR *saccharomyces cerevisiae*) AND humans AND (clinical OR preclinical OR safety) AND (diarrhea OR diarrhoea) from July 2015 to April 2020. Literature was also included from the single PSUR assessment PSUSA/00009284/201702).

As diarrhoeal diseases still mean a major threat to global health, in the recent years the number of publications on *Saccharomyces boulardii/cerevisiae* has increased continuously (Lukaszewicz, 2012). Only the articles considered as relevant for the establishment of this assessment report on a well-established and traditional use of *Saccharomyces cerevisiae* CBS 5926 within the European Union were included in the reference list. In view of the large quantity of articles on *Saccharomyces boulardii/cerevisiae* published, it was not attempted here to give a complete overview on all studies and medical indications having been investigated with this yeast preparation.

2. Data on medicinal use

2.1. Information about products on the market

2.1.1. Information about products on the market in the EU/EEA Member States

Information obtained from the request on 17.4.2014 concerning drug preparations containing *Saccharomyces cerevisiae/boulardii* on the market in the European Union showed the following results:

No products have been authorized in Bulgaria, Croatia, Ireland, Poland, Serbia, United Kingdom, and Slovenia.

Table 2: Overview of data obtained from marketed medicinal products

Active substance	**Indication**	**Pharmaceutical form** **Strength (where relevant)** **Posology** **Duration of use**	**Regulatory Status** **(date, Member State)**

1) *Saccharomyces boulardii*	a) Prevention of diarrhoea caused by ingestion of broad spectrum antibiotics in individuals who are predisposed to diarrhoea by *C. difficile* b) Prevention of a relapse (recurrence) of diarrhoea caused by *C. difficile* c) For the treatment of acute diarrhoea in children under 12 years, in addition to oral hydration.	hard capsules 125 mg/ 250 mg (2×10^{10} viable cells/g) a and b) 4 x 125 mg/d resp. 2 x 250 mg/d for 4 weeks corresponding to 500 mg/day c) 2-4 x 125 mg/d resp. 1-2 x 250 mg/d for 4-7 days corresponding to 250-500 mg/day	WEU; 2012; Belgium
2) *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926)	see 1)	hard capsules, powder for oral suspension (sachet) 250 mg ($\geq 2.4 \times 10^{10}$ viable cells/g) adults: 2 x 1-2 doses (250 mg)/d children: 2 x 1 dose (250 mg)/d	WEU; 2004; Belgium
3) *Saccharomyces boulardii*		containing 50 mg	1968–2005; Belgium

4) *Saccharomyces boulardii* (dried) - Institute Pasteur: I-745 (*Saccharomyces cerevisiae* CBS 5926)	- adjuvant therapy of acute infectious diarrhoea - prevention and therapy of colitis and diarrhoea caused by antibiotics - in combination with vancomycin and metronidazol for therapy of colitis caused by *Clostridium difficile* -adjuvant therapy of irritable bowel syndrome	powder for oral suspensions in sachets/ hard gelatine capsules 250 mg (containing not less than 1 x 10^9 total viable cells count)/sachet or hard capsule adolescents and children over 4 years of age: 1-2 bags/hard capsules once to twice daily (250-1000 mg/day) children 3–4 years of age: 1 to 2 bags/hard capsules daily (250-500 mg/day)	Full MA; 2002; Czech Republic
5) *Saccharomyces boulardii* (lyophilized) - Institute Pasteur: I-745 - ATCC (American Type Culture Collection): ATCC 74012 - CBS (Centraalbureau voor Schimmelcultures): CBS 5926	- disorders of the intestine caused by disturbances of the intestinal microflora - prophylaxis and treatment of diarrhoea caused by use of antibiotics - in adjunct to vancomycin or metronidazole treatment to prevent the recurrence of *Clostridium difficile* induced diarrhoea	powder for oral suspension 250 mg ($\geq$18 x 10^9 cells/g; $\geq$2.5 x 10^9 cells/sachet) 1 or 2 sachets, given once or twice daily (250-1000 mg/day)	Full MA; 1997; Estonia

6) *Saccharomyces boulardii* (lyophilized) (details see 5)	- disorders of the intestine caused by disturbances of the intestinal microflora - prophylaxis and treatment of diarrhoea caused by use of antibiotics - in adjunct to vancomycin or metronidazole treatment to prevent the recurrence of *Clostridium difficile* induced diarrhoea	hard capsule 250 mg ($\geq$18 x 10^9 cells/g; $\geq$2.5 x 10^9 cells/capsule) 1 or 2 capsules, given once or twice daily (250-1000 mg/day)	Full MA; 2003; Estonia
7) *Saccharomyces boulardii*	- acute diarrhoea antibiotic-associated diarrhoea - travellers' diarrhoea recurrent *Clostridium difficile* infection	hard capsule/sachet for oral suspension 250 mg lyophilized powder ($\geq$31.8 x 10^9 CPU/g, >5x10^9 CPU/g in the finished product in the release specification, respectively)	Full MA, bibliographic (national) application; 1996; Finland
8) *Saccharomyces boulardii*	Symptomatic treatment of diarrhoea in addition to rehydration.	hard capsule 50 mg of *Saccharomyces boulardii* children over 6 years of age and adults: 100 mg twice daily	WEU; 1961; France
9) *Saccharomyces boulardii*	Symptomatic treatment of diarrhoea in addition to rehydration.	hard capsule 200 mg of *Saccharomyces boulardii*	WEU; 1970; France
10) *Saccharomyces boulardii* *(Saccharomyces cerevisiae* CBS 5926)	Symptomatic treatment of diarrhoea in addition to rehydration.	powder for oral solution in sachet (1 sachet = 100 mg of *Saccharomyces boulardii*) children over 2 years of age and adults: 1 sachet twice daily (= 200 mg/d)	WEU; 1997; France

11) *Saccharomyces boulardii*	- prophylaxis and treatment of antibiotic-associated adverse reactions of the intestine - complementary treatment in acute diarrheas of adults and children - preventive treatment of recurrence of *Clostridium difficile* disease in addition to vancomycin/ metronidazole - preventive treatment of broad spectrum antibiotic-associated diarrhea	capsule 250 mg adults: 2-4 capsules/day (500-1000 mg/day) children: 1-2 capsules (250-500 mg/day)	WEU; 2010; Greece (capsules containing 50 mg *Saccharomyces boulardii* were approved 1968)
12) *Saccharomyces boulardii* (lyophilised cells)	- acute diarrhoea of bacterial and viral aetiology in children and adults - treatment and prevention of antibiotic-related colitis - prevention of recurrence of *Clostridium difficile* diseases -irritable bowel syndrome - prevention of diarrhoea associated with tube-feeding	capsule 250 mg and powder for oral suspension 250 mg/sachet 250-500 mg 1-2 times per day (250-1000 mg/day)	Full MA; 1997; Latvia

13) *Saccharomyces boulardii* (freeze-dried)	adults: - adjuvant to antibiotics to prevent relapse of *Clostridium difficile* diarrhoea (CDD) - prophylaxis of antibiotic-associated diarrhoea (ADD) children: - prophylaxis of antibiotic-associated diarrhoea (ADD) in vulnerable patients Immunosuppressed patients have been excluded in the studies.	capsule, hard 250 mg adults: 2 capsules 2 times daily (1000 mg daily) children: ≥3 years of age: 1 capsule 2 times daily (500 mg daily) 2-3 years of age: 1 capsule daily (250 mg daily) Duration of use: Not more than 4 weeks. Treatment should be initiated 48-72 h from the start of the antibiotic treatment and continue until at least 3 days after completed antibiotic treatment.	Full MA; 1995; Sweden
14) *Saccharomyces cerevisiae* CBS 5926	a) symptomatic treatment of acute diarrhoea b) prevention and symptomatic treatment of traveller's diarrhoea c) diarrhoea associated with tube feeding d) adjuvant treatment of chronic acne	capsule à 250 mg powder 250 mg/sachet (>$1.8x10^{10}$ viable cells/g lyophilisate) children >2 years of age and adults: a) single dose: 250 mg 1-2 times daily daily dose: 250-500 mg b) single dose: 250 mg 1-2 times daily daily dose: 250-500 mg starting 5 d before departure c) 750 mg/ 1.5 l nutrient solution d) single dose: 250 mg 3 times daily daily dose: 750 mg	WEU; 1995; Germany

15) *Saccharomyces cerevisiae* CBS 5926	see 14)	hard capsule à 50 mg (>1.8 x 10^{10} viable cells/g lyophilisate) children >2 years of age and adults: a) single dose: 100–150 mg 3 times daily daily dose: 300-450 mg b) single dose: 100-150 mg 3 times daily daily dose: 300-450 mg starting 5d before departure c) 750 mg/ 1.5 l nutrient solution d) single dose: 250 mg 3 times daily daily dose: 750 mg	WEU; 1995; Germany
16) *Saccharomyces cerevisiae* CBS 5926	see 14)	125 mg/hard capsule (≥ 2.5 x 10^9 viable cells/cps fluid bed drying) posology see no. 13)	WEU; 2001; Germany
17) *Saccharomyces cerevisiae* CBS 5926	see 14)	250 mg/capsule (≥ 5.0 x 10^9 viable cells/cps fluid bed drying) posology see 13)	WEU; 2001; Germany
18) *Saccharomyces cerevisiae* CBS 5926	see 14)	hard capsule à 375 mg (≥7.5 x 10^9 viable cells/cps fluid bed drying) a) 375 mg/d b) 375 mg/d starting 5 d before departure c) 750 mg/1.5 l nutritive solution/d d) 750 mg/d	WEU; 2001; Germany

19) *Saccharomyces cerevisiae* CBS 5926	a) symptomatic treatment of acute diarrhoea b) prevention and symptomatic treatment of traveler's diarrhoea and diarrhoea associated with tube feeding	hard capsule à 250 mg (≥5.0 x 10^9 viable cells/capsule, fluid bed drying) a) single dose: 250 mg 1-2 times daily daily dose: 250-500 mg b) traveler's diarrhoea: single dose: 250 mg 1-2 times daily daily dose: 250-500 mg starting 5 d before departure tube feeding: 750 mg/ 1.5 l nutrient solution	WEU; 2001; Germany
20) *Saccharomyces cerevisiae* CBS 5926	see 19)	hard capsule à 250 mg (≥1 x 10^{10} viable cells/g lyophilisate) posology see 18)	WEU; 2005; Germany
21) *Saccharomyces cerevisiae* CBS 5926	- symptomatic treatment of acute diarrhoea - prevention and symptomatic treatment of traveler's diarrhoea	hard capsule à 250 mg (≥5.0 x 10^9 viable cells/capsule, fluid bed drying) 1 hard capsule once to twice daily (250-500 mg daily)	WEU; 2013; Germany
22) *Saccharomyces cerevisiae* CBS 5926	- diarrhoea: enteritis, colitis - prevention and treatment of traveler's/summer diarrhoea - enteral dysbiosis - diarrhoea associated with antibiotics and chemotherapeutics - acne	capsule à 50 mg daily dosage in babies, children and adults: acute diarrhoea: 3 x 100 mg; max. 3 x 200 mg chronic diarrhoea, prophylaxis, during antibiotic treatment: 3 x 50 mg acne: 3 x 100 mg for 14 days; thereafter 3 x 50 mg; for a treatment period of 3 months at least	WEU; 1975 to 2003; Germany

23) *Saccharomyces cerevisiae* CBS 5926	see 14) (without acne)	capsule à 250 mg posology see 14)	WEU; 1978 to 2003; Germany
24) *Saccharomyces boulardii*	antimycotic	ointment (44 mg/g)	1962-2008; Belgium
25) *Saccharomyces cerevisiae* CBS 5926	a) symptomatic treatment of acute diarrhoea b) symptomatic treatment of traveller's diarrhoea and diarrhoea associated with tube feeding c) adjuvant treatment of chronic acne	capsule à 250 mg (≥ 2.5 x 10^9 viable cells) children from 2 years of age and adults: a) 250-500 mg/d b) traveller's diarrhoea: 250-500 mg/d tube feeding diarrhoea: 500 mg /l nutritive solution c) 750 mg/d	Full MA; 2000; Austria

This overview is not exhaustive. It is provided for information only and reflects the situation at the time, when it was established.

Further data on the administration of *Saccharomyces boulardii/cerevisiae* were provided by the different countries:

Undesirable effects:

BE: very rare (< 1/10.000): fever in case of fungemia, mycosis, anaphylaxia, dyspnea, itching, exanthema, Quincke edema, epigastric pain and abdominal meteorism (these symptoms were observed in clinical studies and do not require to interrupt the course of treatment), thirst.

EE: Rare cases of digestive tract disturbances have been reported, not requiring that treatment to be discontinued.

EL: rare: flatulence; very rare: rash, cutaneous allergy, urticaria, pruritus, anaphylactic reaction, angiooedema, exanthema

FR: very rarely: allergic reactions (Quincke oedema), flushes, itches; rarely: urticarial

SE: uncommon (≥1/1,000 to <1/100): urticarial, allergic reactions, constipation, thirst; rare (≥1/10,000 to <1/1,000): angioedema, exanthema

Cases of sepsis caused by *Saccharomyces boulardii* have been reported for patients with seriously impaired general condition.

CZ: gastrointestinal disorders (stomach pain, bloating, constipation), allergic reactions (itching, redness, urticaria), systemic mycotic infection

DE: flatulence, allergic reactions (itching, urticarial, localized or generalized exanthema, Quincke edema, dyspnea, anaphylaxia), fungemia in patients with central venous catheter, life-threatening diseases, severe underlying disease, reduced immune defense

AT: flatulence, hypersensitivity reactions (itching, urticaria, localized or generalized exanthema, Quincke edema)

Use in children and adolescents:

EE: The contents of the sachet should be mixed with water or another liquid or food and the contents should be poured in a baby`s feeding-bottle. In young children under 6 years of age, it is recommended not to swallow capsules (risk of false passage) but to open them and tip the contents into a beverage or food.

EL: In order to be administered to children, the capsule should be opened and the content should be mixed with milk (feeding-bottle) or food.

LV: For children less than 6 years of age capsules should be opened and powder mixed with liquid.

DE: The content of the capsule may be mixed into food or beverages. The food may not be too hot (>50° C) or ice cold.

Due to missing data on posology, the medical product should not be used in children <2 years of age.

AT: Due to missing data, the medicinal product may be used in chchildren < 2 vears on doctor's orders only

Duration of treatment:

BE: adults: prevention of relapses of diarrhoea caused by *C. difficile*: 4 weeks; children: Treatment of diarrhoea in addition to rehydration therapy: 1 week
The treatment should not be interrupted too early, because diarrhoea can recur.

EL: If diarrhea persists after 2 days or if blood in faeces appears or if fever appears, treatment should be reconsidered and the necessity to introduce oral or parenteral rehydratation should be considered, orally or parenteral. After diarrhea is stopped, the treatment may be continued for some days.

CZ: If the symptoms persist for more than 2 days during the use of the medicinal product, the treatment method should be reconsidered.

FI: 1 – 4 weeks depending on the indiaction

DE and AT: No restriction of treatment duration: Treatment of diarrhoea should be continued for several days following a cessation of complaints. For the treatment of chronic acne administration for several weeks is recommended.

Contraindications:

BE: Hypersensitivity to one of the ingredients or other yeast, patient with central venous catheter.

CZ: Patients with central venous catheter.

EE: Hypersensitivity to *Saccharomyces boulardii*. Patient with central venous catheter.

EL: Known hypersensitivity to one of the components. Patients having a Central Venous Catheter-CVC-, allergy to yeast, especially *Saccharomyces boulardii*.

LV: Hypersensitivity to one of the ingredients. Patient with central venous catheter and with immune deficiency disorders.

FI: Hypersensitivity to the active substance

SE: Hypersensitivity to the active substance. Patients with central venous catheter.

DE: Hypersensitivity to *Saccharomyces boulardii*. Due to the risk of fungemia treatment is contraindicated in patients with life-threatening diseases, reduced immune defense, and in patients swith central venous catheter.

Children <2 years are excluded from self-medication and have to be treated only after consultation of a medical doctor.

AT: Hypersensitivity to ingredients of the medicinal product.

Special warnings/precautions:

BE: As diarrhoea causes serious loss of water and electrolytes, substitution of water and electrolytes is important.
If freezing liquids or food, or liquids or food, which can be heated above 50°C, are used to prepare a suspension for oral use, the acticity of the medicinal product can decrease.

Precautions in fever above 38°C, severe abdominal pain, blood in stool, vomiting associated with diarrhoea, diarrhoea for more than 3 days.
In case of microbiological stool examination *Saccharomyces boulardii* can cause false-positive results.
The medicinal product is contraindicated in patients with severe immune deficiency (e.g. HIV-infection, organ transplantation, leukaemia, progredient malignant tumour, radiotherapy, chemotherapy, large-dose long-term glucocorticoids treatment) should not use this medicinal product without consulting a doctor.

CZ: If the symptoms persist for more than 2 days during the use of the medicinal product, the treatment method should be reconsidered. Not to be mixed with hot (more than 50°C) or iced beverages or meal. Not to be combined with alcohol.

EE: Contains living cells. This drug should therefore not be mixed with very hot (over 50°C), iced or alcoholic drinks or food.

The treatment does not replace rehydration when this is necessary. The rehydration dose and its route of administration (oral) should be adapted to the severity of the diarrhoea and to the age and state of health of the patient.

Saccharomyces boulardii is a living organism associated with risks of sytemic fungus infection by gastro-intestinal translocation or hand-borne contamination. Rare cases of fungemia have been reported in hospitalized severely ill patients, most often because of gastrointestinal disease, with a central venous catheter.

EL: In children below 2 years of age, a medical advice is recommended due to risk of dehydration. The treatment does not replace rehydration when this is necessary. The rehydration dose and its route of administration (oral-IV) should be adapted to the severity of the diarrhoea and to the age and state of health of the patient.

Contains living cells. Therefore it must not be mixed into very hot (above 50°C), icy drinks or foods (e.g. drinks stored in the refrigerator, ice creams, very hot meals) and alcohol-containing drinks.

It is advisable not to open the capsules in the surroundings of patients with a central venous catheter-CVC-, to avoid any colonization, especially hand-borne of the catheter. There have been reports of patients with a central venous catheter-CVC-, even not treated with *Saccharomyces boulardii*, of very rare cases of fungemia (penetration of blood by yeast), most often resulting in pyrexia and blood

cultures positive for *Saccharomyces*. The outcome in all these cases has been satisfactory after administration of antifungal treatment and, when necessary, removal of the catheter.

It is not possible to draw safe safety data on the content of 250 mg for patients who are more prone to side effects or systemic fungal infections, such as immunosuppressed patients, patients with AIDS, patients in Intensive Care Units and patients receiving enteral feeding in the hospital environment.

SE: There have been reports in patients with a central venous catheter of very rare cases of fungemia (blood cultures positive for *Saccharomyces*). It is advisable not to open capsules in the surroundings of patients with a central venous catheter, to avoid any colonization, especially hand-borne, of the catheter.

DE: Risk of fungemia in patients with life-threatening diseases, reduced immune defense, and in patients with central venous catheter.

If diarrhoea lasts longer than 2 days, aggravates or is associated with blood or elevated body temperature, a doctor should be consulted.

Especially in children the most therapeutic measure is a substitution of water and electrolytes.

In case acne deteriorates or does not improve a doctor should be consulted.

In case of microbiological stool examination during or shortly after treatment with this medicinal product the laboratory should be informed on its administration, as false-positive results may be possible.

Due to missing data on posology, the medical product should not be used in children <2 years of age.

AT: Especially in children the most therapeutic measure is a substitution of water and electrolytes.
If diarrhoea lasts longer than 2 days or is associated with blood or elevated body temperature, a doctor should be consulted.
In case of microbiological stool examination during or shortly after treatment with this medicinal product the laboratory should be informed on its administration, as false-positive results may be possible.
Due to missing data, the medicinal product may be used in children < 2 vears on doctor's orders only
Because of unratable risk of fungemia patients with impaired immune status (e.g. HIV-infection, chemotherapy, or radiotherapy) should not use this medicinal product without consulting a doctor.

Drug interactions:

BE: The intake of antimycotics neutralizes the effect of the medicinal product.

CZ: Not to be used concomitantly with systemic oral and parenteral antimycotics.

EE: Because of its fungal nature, *Saccharomyces boulardii* must not be administered with systemic or oral antifungal drugs.

EL: Because of its fungal nature, it must not be administered with parenteral or oral antifungal drugs.

SE: Should not be used concomitantly with antimycotic drugs.

DE and AT: The co-administration of antimycotics may impair the treatment results of *Saccharomyces boulardii*.

The concomitant use of monoamine oxidase inhibitors may increase blood pressure.

Fertility, pregnancy and lactation:

BE: Due to the lack of data, the use in pregnancy and lactation is not recommended.

CZ: Safety during pregnancy and lactation has not been established. In absence of sufficient data, the use during pregnancy and lactation is not recommended.

EE: *Saccharomyces boulardii* does not absorb through the gastrointestinal tract. *Saccharomyces boulardii* does not transfer into breast milk. No data about fertility.

EL: There are not reliable animal teratogenesis data. Clinically, no malformed nor foetotoxic effect has been reported to date. However, monitoring of pregnancies exposed to this medicine is insufficient to rule out any risk. Therefore, although *Saccharomyces boulardii* is not absorbed, as a precautionary measure, it is preferable to weight benefit/risk before using this medicine during pregnancy.

Saccharomyces boulardii is not absorbed. In the absence of data, it is preferable to weight benefit/risk for the breastfed infant before using it during lactation.

SE: Pregnancy and breast-feeding: *Saccharomyces boulardii* is not absorbed.

DE: Due to the lack of data, *Saccharomyces boulardii* should not be used in pregnancy.

AT: The widespread use of yeast as food indicates no risk for pregnancy and lactation.

Overdose:

Not known.

AT: Flatulence can increase. A specific antidote is not known.

Effects on ability to drive or operate machinery or impairment of mental ability:

BE and CZ: The product does not influence the ability to drive or operate machinery.

SE: No effects have been observed.

DE: No limitations

AT and EL: Not known

Information on relevant combination medicinal products marketed in the EU/EEA

Combination products (ointment, suppositories) containing alcoholic extracts of *Saccharomyces cerevisiae* (unknown DER) and oil from shark liver for the symptomatic treatment of haemorrhoids have been reported by Czech Republic. These products are not regarded as relevant for the assessment of the medicinal use of *Saccharomyces cerevisiae* CBS 5926.

In France, several combination products containing *Saccharomyces cerevisiae* are on the market. They refer to various indications and differ with regard to their compounds and pharmaceutical form.

These products are not regarded as relevant for the assessment of the medicinal use of *Saccharomyces cerevisiae* CBS 5926.

Information on other products marketed in the EeU/EEA (where relevant)

Latvia:
Several food supplements containing *Saccharomyces cerevisiaeboulardii* are included in the database of Latvian Food Centre (mono and combination products). No further information is given.

2.1.2. Information on products on the market outside the EU/EEA

In the United States, *Saccharomyces boulardii* is regarded as a probiotic which as a dietary supplement does not require approval by the Food and Drug Administration. Due to its regulation as a dietary

supplement, it is intended for use by the general healthy population, not as a drug to prevent, treat, or mitigate disease (Venugopalan *et al*., 2010).

According to CFR – Code of Federal Regulations Title 21 (FDA), dried yeast (*Saccharomyces cerevisiae* and *Saccharomyces fragilis*) may be safely used in food provided the total folic acid content of the yeast does not exceed 0.04 mg /g yeast (approximately 0.008 mg of pteroyglutamic acid per gram yeast) (21CFR172.896).

2.2. Information on documented medicinal use and historical data from literature

During his visit to Indochina in 1920, the French biologist Henri Boulard discovered a closely related strain of *Saccharomyces cerevisiae* which was named *Saccharomyces boulardii*. Its discovery was based on the observation that during an outbreak of cholera some people who either had chewed on the skins of lychee and mangosteen fruits or drunken a special tea prepared from the lychee fruit were not affected by the disease. In 1947, Laboratories Biocodex bought the patent for this yeast strain and started the process of research and manufacturing (McFarland, 2010). Since then, this *Saccharomyces* strain (initially named "*boulardii*") has been used in a variety of indications which mainly refer to the gastrointestinal tract.

In 1988 Komission E (BAnz, 1988) published a first monograph on *Faex medicinalis*, which consists of fresh and dried cells *of Saccharomyces cerevisiae* MEYER and/or *Candida utilis* and contains vitamins, glucanes and mannans. As indications lack of appetite and adjuvant treatment of acne and furunculosis are mentioned. The dosage recommended is 6 g/d.

According to the monograph of the Kommission E which was published thereafter in 1994 (BAnz, 1994) and Blaschek *et al*. (2013), there is a well-established use (WEU) of *Saccharomyces boulardii* (*Saccharomyces cerevisiae* HANSEN CBS 5926) in the following indications:

- Symptomatic treatment of acute diarrhoea
- Prophylaxis and symptomatic treatment of travelers' diarrhoea
- Diarrhoea associated with tube feeding
- Adjuvant treatment of acne.

The corresponding posologies are listed in table 3.

Table 3: Overview of historical data

Country	**Herbal preparation**	**Documented Use / Traditional Use**	**Pharmaceutical form Strength (where relevant)** **Posology** **Duration of use**	**Reference**

Germany	*Saccharomyces cerevisiae* HANSEN CBS 5926 (1 g of the lyophilisate contains ≥ 1.8 x 10^{10} viable cells)	WEU: a) symptomatic treatment of acute diarrhoea b) prophylaxis and symptomatic treatment of travelers' diarrhoea c) diarrhoea associated with tube feeding d) adjuvant treatment of acne	adults and children >2 years of age: a) 250-500 mg/d b) 250-500 mg/d; 5 d before departure c) 500 mg/l nutrient solution d) 750 mg/d	monograph of Kommission E (15.4.1994) Blaschek *et al*. (2013)

2.3. Overall conclusions on medicinal use

According to the results of the market overview, preparations from *Saccharomyces cerevisiae* are used in many European countries with the first medicinal products having been marketed in 1968 (Belgium). The following preparations fulfil the requirements of 30 years of medicinal use in the EU: symptomatic treatment of acute diarrhoea, prevention and symptomatic treatment of travelers' diarrhoea and adjuvant treatment of chronic acne (for assessment of the clinical data see chapter 4).

Table 4: Overview of evidence on period of medicinal use

Herbal preparation Pharmaceutical form	Indication	Strength Posology	Period of medicinal use
Saccharomyces cerevisiae CBS 5926 (>1.8x10^{10} viable cells/g lyophilisate)	Treatment of acute diarrhoea	Adolescents, adults and elderly: single dose: 250 mg 2 times daily daily dose: 250-500 mg children (6 months to 11 years): single dose: 250 mg 1-2 times daily	since 1978
Saccharomyces cerevisiae CBS 5926 (>1.8x10^{10} viable cells/g lyophilisate)	Prevention of travelers' diarrhoea	Adolescents, adults and elderly: single dose: 250 mg 1-2 times daily daily dose: 250-500 mg starting 5 d before departure	since 1978

Saccharomyces cerevisiae CBS 5926 (>1.8x10^{10} viable cells/g lyophilisate)	Adjuvant treatment of acne	Adolescents and adults: single dose: 250 mg 3 times daily daily dose: 750 mg	monograph of Kommission E (15.4.1994) since 1995

3. Non-Clinical Data

The pharmacological properties of *Saccharomyces cerevisiae* have been investigated extensively both by *in-vitro* and *in-vivo* methods and the results have been published in numerous publications. *Saccharomyces cerevisiae* is considered as a probiotic according to the definition of the World Health Organisation (2002) i.e. "live microorganisms which, when administered in adequate amounts, confer a health benefit on the host." A comprehensive review on the various mechanisms of action of *Saccharomyces cerevisiae* is given by Kelesidis and Pothoulakis (2012), Im and Pothoulakis (2010), McFarland (2010) and Moslehi-Jenabian *et al* (2010).
Considering the multitude of studies with preclinical data on *Saccharomyces cerevisiae* a systematic review is not given here. This is rather the attempt to select exemplary studies relevant for the medicinal use.

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

3.1.1. Primary pharmacodynamics

As summarized by Kelesidis and Pothoulakis (2012) and McFarland (2010) the pharmacodynamic effects of *Saccharomyces boulardii* can be classified into three main domains of action:

- luminal action, including effects against several bacterial toxins, antimicrobial activity, modulation of intestinal flora, metabolic activity;
- trophic action on the intestinal mucosa, including enzymatic activity;
- regulation of the immune response by acting as immune stimulant, by reducing pro-inflammatory responses and promoting mucosal anti-inflammatory signaling effects.

While some of the effects were only investigated in humans and therefore are not further mentioned here, others were also seen in non-clinical tests.
For at least some of the effects it was claimed, that the substances responsible for are released by the cells of *Saccharomyces cerevisiae* during their intestinal catabolism rather than being secreted by viable cells (Kelesidis and Pothoulakis, 2012; McFarland, 2010):

Luminal action:

- antitoxin effects

In several studies inhibition of toxin receptor binding sites, stimulation of antibody production against *Clostridium difficile* toxin A and direct proteolysis of the pathogenic toxins/secretion of enzymatic proteins (production of a serine protease that cleaves *C. difficile* toxin A, production of a 63 kDa phosphatase that destroys the endotoxin of pathogenic *Escherichia coli* and production of a 120 kDa protein that reduces the effects of cholera toxin) was shown.

- antimicrobial activity

Saccharomyces boulardii is capable of directly or indirectly interfering with intestinal pathogens. Inhibition of growth of bacteria and parasites, reduction of gut translocation of pathogens,

neutralization of bacterial virulence factors and suppression of host cell adherence that interferes with bacterial colonization was reported.

- modulation of intestinal flora

Rapid re-establishment of normal microbiota could be shown in antibiotic-exposed mice could be shown after administration of *Saccharomyces boulardii*.
According to a review by Moré and Swidsinski (2015) *Saccharomyces boulardii* is able to create a favorable growth environment for the beneficial intestinal microbiota, while constituting extra protection to the host mucus layer and mucosa.

Trophic action:
Saccharomyces boulardii can reduce the number of infected cells and stimulates the growth and differentiation of intestinal cells in response to trophic factors, prevent apoptosis and synthesis of TNFα. It reduces mucositis, restores fluid transport pathways, stimulates protein and energy production and restores metabolic activities in colonic epithelial cells, secretes mitogenic factors that enhance cell restitution, enhances release of brush-border membrane enzymes, stimulates the production of glycoproteins in the brush border and the production of intestinal polyamines, restores normal levels of colonic short chain fatty acids (SCFAs), stabilizes gastrointestinal barrier function and strengthens enterocyte tight junctions and can reduce crypt hyperplasia and cell damage in colitis models.

The trophic effects of *Saccharomyces boulardii* are the focus of a review by Moré and Vandenplas (2018). They report that *Saccharomyces boulardii* CNCM I-745 synthesizes and secretes polyamines, which have a role in cell proliferation and differentiation. The administration of polyamines or *Saccharomyces boulardii* CNCM I-745 enhances the expression of intestinal digestive enzymes as well as nutrient uptake transporters. The signalling mechanisms leading to enzyme activation are not fully understood. However, polyamines have direct nucleic acid–binding capacity with regulatory impact. *Saccharomyces boulardii* CNCM I-745 induces signalling via the mitogen-activated protein kinase pathway. In addition, effects on the phosphatidylinositol-3 kinase (PI3K) pathway have been reported. As an additional direct effect, *Saccharomyces boulardii* CNCM I-745 secretes certain enzymes, which enhance nutrient acquisition for the yeast and the host. The increased availability of digestive enzymes seems to be one of the mechanisms by which *Saccharomyces boulardii* CNCM I-745 counteracts diarrhoea; however, also people with certain enzyme deficiencies may profit from its administration. More studies are needed to fully understand the mechanisms of trophic activation by the probiotic yeast.

Regulation of the immune response:

- acting as immune stimulant

It could be shown that *Saccharomyces boulardii* triggers activation of complement and migration of monocytes and granulocytes and enhances the number of Kupffer cells in germfree mice. Furthermore, experiments revealed that it enhances the mucosal immune response and secretory IgA intestinal levels, enhances systemic immune response and levels of serum IgG to *C. difficile* toxins A and B, contributes to earlier production of IFN-γ and IL-12, inhibits dendritic cell-induced activation of T cells, modifies migration of lymphocytes in a chronic inflammatory bowel disease model, modifies lymphocyte adherence to endothelial cells and improves cell rolling and adhesion.

- reducing pro-inflammatory responses and promoting mucosal anti-inflammatory signaling effects

In several studies *Saccharomyces boulardii* decreased expression of pro-inflammatory cytokines (IL-8, IL-6, IL-1β, TNF-α and IFN-γ), increased expression of the anti-inflammatory cytokine IL-10, interfered with NF-κB-mediated signal transduction pathways, in immune and colonic epithelial cells, blocked

activation of $ERK_{1/2}$ and MAP kinases, decreased nitric oxid and inhibited production of inducible nitric oxid synthase, modulated T-cell migratory behavior and increased trapping of T helper cells into mesenteric lymph nodes and stimulated production of anti-inflammatory molecules in human colonocytes such as peroxisome proliferator-activated receptor-gamma.

To be an effective probiotic agent, *Saccharomyces cerevisiae* must fulfill several conditions, such as survival of the passage to its target organ (most commonly the colon) or resistance to stomach acids and bile acids.
Although much of the oral dose seems to be destroyed (usually stool levels are 100-1000 times lower than the oral dose), surviving oral doses have been found to be effective (usually at levels over 10^8 organisms/g stool).

Table 5: properties of *Saccharomyces cerevisiae* which should be considered to determine the efficacy [taken from Kelesidis and Pothoulakis, 2012]

Properties of *Saccharomyces boulardii*	References
survives passage to its target organ (most commonly the colon): although much of the oral dose is destroyed (usually stool levels are 100–1000 times lower than the oral dose), surviving oral doses have been found to be effective (usually at levels over 10^8 organisms/gram stool)	Gorbach, 2000
survives at body temperature (37°C): unique advantage of being one of the few yeasts that do best at human body temperatures	Graff *et al*., 2008
in lyophilized form, *Saccharomyces boulardii* survives gastric acid and bile	Graff *et al*., 2008
as is the case with all yeasts, *Saccharomyces boulardii* is naturally resistant to antibiotics	Graff *et al*., 2008
Saccharomyces boulardii is resistant to proteolysis	Buts, 2009
Saccharomyces boulardii exists in the competitive milieu of the intestinal tract	Buts, 2009
Saccharomyces boulardii levels are higher in patients with disturbed intestinal microbiota (due to antibiotic exposure) compared to patients without antibiotic exposure	Klein *et al*., 1993
when given orally, achieves steady-state concentrations within three days and is cleared within 3–5 days after it is discontinued	Blehaut *et al*., 1989; Elmer *et al*., 1999b
some types of fiber (psyllium) increased *Saccharomyces boulardii* levels by 22%, while other types of fiber (pectin) showed no effect	Elmer *et al*., 1999a

Czerucka and Rampal (2019) described the diversity of *Saccharomyces boulardii* CNCM I-745 mechanisms of action against intestinal infections as follows:
"*Saccharomyces boulardii strain CNCM I-745 is a probiotic yeast that by virtue of being a eukaryote differs from other probiotic strains, which are of bacterial origin (prokaryote). The research shows a great diversity in its mode of action and types of targets: pathogens, pathogenic toxins, gut microbiota and intestinal epithelium. Two main mechanisms were demonstrated: the first one is a large capacity of the wall to fix bacteria and toxins, which facilitates their elimination during intestinal*

transit and the second one is the synthesis by this yeast of several active factors. These factors include high molecular weight proteins, some of which have antisecretory effects, others act as proteases that degrade toxins or their receptors. Factors of small size and protein or non-protein nature that exhibit anti-secretory or anti-inflammatory activities are also involved in its action. Finally, Saccharomyces boulardii CNCM I-745 acts on different components that maintain the intestinal barrier: Tight junctions that regulate permeability; reconstitution of the microbiota after antibiotic therapy; and, activation of innate immunity, which stimulates innate defenses of the host during infection. The optimization of the use of this probiotic in infections requires a better knowledge of the different mechanisms of action."

3.1.2. Secondary pharmacodynamics

Kim *et al*. (2004) described the purification and characterization of a novel antihypertensive angiotensin I-converting enzyme (ACE) inhibitory peptide from *Saccharomyces cerevisiae*. The purified inhibitor competitively inhibited ACE and showed a clear antihypertensive effect in spontaneously hypertensive rats at a dosage of 1 mg/kg body weight. These results were similar to that of the antihypertensive drug captopril.

Investigations of Karen *et al*. (2010) in rats suggest that *Saccharomyces boulardii* may reduce lung injury by reducing bacterial translocation, which results in reduced infection, inflammation, and generation of proinflammatory cytokines in an experimental model of acute necrotizing pancreatitis.

3.1.3. Safety pharmacology

Not available.

3.1.4. Pharmacodynamic interactions

Blackwell and Marley (1966) tested yeast extracts in rats, cats and guinea-pigs concerning interactions with monoamine oxidase inhibitors because attacks of hypertension and headache have occurred in patients taking monoamine oxidase inhibitors and Marmite (a food product). The explored yeast extracts were obtained locally except for some samples of Marmite (Salt Marmite) and Salt-free Marmite given by Bovril Ltd.
The product injected i.v. into untreated rat and cat produced a fall followed by a rise in blood pressure and contracted the nictitating membrane. These sympathomimetic effects were prolonged by previous treatment with an amine oxidase inhibitor.
The effects of yeast extract after intraduodenal injection were only obtained in the rat or cat after treatment with an amine oxidase inhibitor. The inhibitors used were of the amine, hydrazine and hydrazide variety.
Furthermore, some histamine-like effects of the yeast extracts both after i.v. injection and intraduodenal injection were observed. The histamine-like effects could be attributed to the absorption of a histamine-like substance from the yeast extract in the intestine which was facilitated by inhibition of monoamine oxidase.
In the guinea-pig the intravenous or intraduodenal injection of yeast extract increased resistance on inflation of the lungs.
The authors summarized that yeast extracts had sympathomimetic and histamine-like properties.

Assessor's comment:

Since the product tested consists not only yeast extract, but also high amounts of salt, vegetable extract and various vitamins, the results should be regarded as not appropriate for the assessment of the medicinal use of Saccharomyces cerevisiae.

Izquierdo-Pulido *et al*. (1995) examined the influence of *Saccharomyces cerevisiae* var. *uvarum* on histamine and tyramine formation during beer fermentation. They found that the yeast did not produce histamine or tyramine during fermentation. Yeast recycling did not influence biogenic amines formation.

3.1.5. Conclusions

In many review articles, the pharmacodynamic effects of *Saccharomyces cerevisiae* were classified into three main domains of action: a luminal and a trophic action as well as an influence on the immune response. Several mechanisms of actions are described within these 3 categories. Some of the knowledge was obtained using *in-vitro* data; therefore, the biological relevance of these results cannot be evaluated. Also *in-vivo* data obtaind in animals with different physiology concerning the digestive system might only be supportive for the human situation. Nevertheless, since some of the effects could also be seen in patients it seems that at least basically the non-clinical results support the usage of *Saccharomyces cerevisiae* in the conditions linked to diarrohea.

Concerning possible interactions with monoamine oxidase inhibitors, the available preclinical data do not refer to the specific *Saccharomyces* strain covered by this assessment report and monograph. Furthermore, these data are inconsistent with regard to histamine and tyramine findings.

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

In 1989, Blehaut *et al*. investigated the kinetics of *Saccharomyces boulardii* in humans, rats and mice (see chapter 4.1.2). They conducted a multiple dose study and a single dose study in rats and presented the results of continuous administration of *Saccharomyces boulardii* in mice (lyophilized *Saccharomyces cerevisiae* CBS 5926 with a total number of yeast cells (±SD) of $10^{10.98\pm0.02}$ per gram resp. $10^{10.55\pm0.08}$ viable yeast cells per gram).

Single dose in rat:
A total of 54 rats weighing 200±20 g on a yeast free regimen received 0.4 g/kg of *Saccharomyces boulardii* suspension as a single dose (lyophilized *Saccharomyces cerevisiae* CBS 5926 with a total number of yeast cells (±SD) of $10^{10.98\pm0.02}$ per gram resp. $10^{10.55\pm0.08}$ viable yeast cells per gram) (oesophageal tube). A group of 6 rats were sacrificed at each of the following times: control, 0 (immediately post-dosing), 0.08, 0.25, 2, 3, 4, 8, and 24 h.
Measurements made in three levels of the gastrointestinal tract (oesophagus and stomach, small intestine, and large intestine) show the progression of live and dead cells through the length of the gastrointestinal tract. Live cells appeared to move faster than dead cells.
The recovery of live cells decreased from 87% at 15 min to a value of 13% at 24 h. An apparent disappearance half-life of 9 h was calculated from the last four time points (3, 4, 8 and 24 h).
The time course of dead cells was much slower. Initially, there was a plateau with a slight increase in recovery to 108% at 2 h. This phenomenon was explained with the addition of dead cells arising from the rapid disappearance of live cells. The apparent disappearance half-life calculated from the last four points was 44 h. This value is only an estimate since measurements were available for less than one half-life.

Multidose study in rat:
Six male rats weighing 200±20 g received 0.8 g/kg of *Saccharomyces boulardii* suspension (lyophilized *Saccharomyces boulardii* with a total number of yeast cells (±SD) of $10^{10.88\pm0.04}$ per gram resp. $10^{10.28\pm0.04}$ viable yeast cells per gram) as a single daily dose (oesophageal tubes) from day 1 to 14 and were fed with a yeast-free regimen.
Twenty-four hours after the beginning of *Saccharomyces boulardii* treatment, faecal concentration of live cells reached a steady-state in all animals. Less than 1% of the administered live cells were recovered in the faeces. In the period following *Saccharomyces boulardii* treatment (days 15-18), the concentrations of live cells decreased by three orders of magnitude within 24 h. Consequently, a precise measure of half-life could not be obtained. However, the authors estimated that its value is shorter than 3 h. Dead cells reached a steady-state within 24 h, too.
The recovery of dead cells based on total dose of *Saccharomyces boulardii* ranged from 9.3% to 15.8% with an overall mean of 12.4±2.4%. The recovery based on the dose of dead cells ranged from 12.4% to 21.2% with an overall mean (±SD) of 16.6±3.2%. The half-life time is estimated on 36 h.

Continuous administration in mice:
Ten nude and ten control mice (haired, thymic) received *Saccharomyces boulardii* (lyophilized *Saccharomyces boulardii* with a total number of yeast cells (±SD) of $10^{10.98\pm0.02}$ per gram resp. $10^{10.55\pm0.08}$ viable yeast cells per gram) in a 5% (W/V) suspension as the source of drinking water. In addition, 10 nude and 10 control mice received only saline.
After 70 days of oral administration the caecum only contained detectable levels of yeast. No live cells could be detected in the mesenteric lymph nodes, in the liver, lungs, heart or kidney.

In-vitro tests:
Graff *et al*. (2008) investigated to what extent *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) is sensitive to gastrointestinal pH conditions. The survival of different concentrations of *Saccharomyces boulardii* applied as freeze dried yeast or as aqueous suspension was examined in conditions mimicking the stomach pH (pH 1.1; 0.1 N HCl) and intestinal pH (pH 6.8; phosphate buffer). The viability of both forms of *Saccharomyces boulardii* remained stable for 6 h in phosphate buffer, whereas under acidic conditions for both forms of *Saccharomyces boulardii* from 5 min a significant decrease of viability was observed. At the highest concentration of 200 g/l the initial pH value of 1.1 increased to 3.2 demonstrating a protective effect. This investigation shows that in order to improve oral availability of viable *Saccharomyces boulardii* it has to be protected from gastric destruction.

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

3.3.1. Single dose toxicity

The monograph of the German Kommission E (1994) states that a single oral administration of 3 g/kg b.w. produced no toxic reactions in mice and rats.
Acute toxicity tests were performed in rats (Sudha, 2011). *Saccharomyces boulardii* (corresponding to 5×10^9 cfu/g) administered at single oral doses of 6,500 mg per kg of b.w. produced no treatment-related changes in the test animals. The animals were observed over a period of 14 days.

3.3.2. Repeat dose toxicity

The administration of 330 mg/kg b.w. over a period of 6 weeks on 6 days per week to dogs and the oral administration of 100 mg/kg b.w./day to rats and rabbits showed no drug induced changes

(Kommission E, 1994). For sub-acute toxicity studies Sprague-Dawley rats were fed with oral doses of *Saccharomyces boulardii* (corresponding to 5 x 10^9 cfu/g) up to 1,300 mg/kg b.w. for 14 consecutive days (Sudha, 2011). During the observation period of 28 days *Saccharomyces boulardii* was well tolerated and there was no morbidity or any toxic clinical symptom displayed either in male or female rats.

3.3.3. Genotoxicity

According to the Kommission E (1994) monograph, no mutagenic effects of *Saccharomyces cerevisiae* HANSEN CBS 5926 have been detected in an AMES-test with *Salmonella typhimurium* TA 98, TA 100, TA 1535, TA 1537 and TA 1538 with and without S9 activation.

3.3.4. Carcinogenicity

No studies on cancerogenicity for the yeast *Saccharomyces boulardii* are available.

3.3.5. Reproductive and developmental toxicity

No studies are available either on reproduction toxicology and the fertility-influencing, or on embryo-foetal and peri-/postnatal toxicity effect for the yeast *Saccharomyces boulardii*.

3.3.6. Local tolerance

Not applicable.

3.3.7. Other special studies

Pathogenicity
Maejima *et al*. (1980) examined *Saccharomyces cerevisiae* for its pathogenicity and colonization in mice and cynomolgus monkeys as the models of the biological containment level. Adult mice given perorally 5.5 or 2.4 x 10^7 cells of *Saccharomyces cerevisiae* strain MC16 excreted them rapidly and no colonization of the cells in the abdominal organs, lymph nodes or gastrointestinal wall was demonstrated. No change in the faecal flora was observed. After peroral administration of 4.9 x 10^7 or 7.8 x 10^8 cells, cynomolgus monkeys showed a similar tendency of rapid excretion and lack of colonization. Cortisone acetate treatment had no significant effect. Intravenous administration of 3.9 x 10^7 yeast cells had no pathogenic effect and no viable yeast was detected in blood. The biological containment level of *Saccharomyces cerevisiae* was suggested not to be lower than that of *E. coli* K12 biosafety level 1, and the possibility of achieving the biosafety level 2 was suggested. (Biosafety level 1: suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and or minimal potential hazard to laboratory personnel and the environment; Biosafety level 2 is similar to Biosafety level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. These agents cause only mild disease to humans, or are difficult to contract via aerosol in a lab setting (Chosewood and Wilson, 2009).

To assess the pathogenic potential Clemons *et al*. (1994) analyzed 13 clinical isolates (e.g. lung, blood, peritoneal fluid), 10 nonclinical isolates (e.g. bread yeast, wine yeast), and 5 constructed strains of *Saccharomyces cerevisiae*. All were *Saccharomyces cerevisiae* by biochemical profiles, sporulation, or genetic evidence. To initiate the model of infection, 4-week-old male CD-1 mice from the virus antibody-free colony were inoculated intravenously with 2 x 10^7 viable cfu of *Saccharomyces cerevisiae*. At various times after infection, 4-10 mice were killed, and brains, spleens, livers, kidneys,

and lungs were removed aseptically and residual burden of *Saccharomyces cerevisiae* in each organ was determined. Prolonged persistence of *Saccharomyces cerevisiae* especially in the brains was found. Furthermore, some of the clinical isolates tested demonstrated a modest proliferation in the brain over the first 7 days of infection (5-fold); infection due to nonclinical isolates declined. By authors' definition of virulence, the results indicate that the comparative pathogenicity of *Saccharomyces cerevisiae* isolates is represented by a continuum rather than a clear virulent or avirulent result. The majority of the clinical isolates were better able to persist *in-vivo* than isolates from nonclinical sources. 12 of the 13 clinical isolates were assigned to groups considered as being virulent or intermediate in virulence. This is in contrast to the nonclinical isolates where only 4 of 10 were assigned to the intermediate virulence group and the rest were considered avirulent. However, even within the latter group there were degrees of resistance to eradication by host mechanism. The authors concluded that these data indicate that recovery of *Saccharomyces cerevisiae* in a clinical setting, especially from severely compromised patients, should not be ignored.

To further characterize *Saccharomyces cerevisiae* pathogenesis Byron *et al*. (1995) studied a virulent clinical isolate and an avirulent nonclinical isolate in C5-deficient mice. Complement deficiency (in particular, of the fifth component [C5]) has been implicated in increasing susceptibility of humans and mice to fungal infections with *Aspergillus* fumigatus, *Candida albicans*, and *Cryptococcus neoformans* as well as infections caused by gram-negative bacteria. The mice were infected intravenously with 10^7 CFU and temporal burdens of yeast cells in various organs were determined. After infection the virulent clinical isolate increased by 20-fold in the brain from day 0 to 3 and by 4-fold in the kidneys. The avirulent nonclinical isolate increased by 13-fold in the brain from day 0 to 3 and decreased by 16-fold in the kidneys. Both isolates declined in number in other organs (spleen, liver, lungs). In all studies, 90% of mice infected with 10^7 CFU of the virulent clinical isolate died between days 2 and 7, whereas no mice infected with equivalent numbers of the avirulent nonclinical isolate died. No mice died after infection with 10^6 CFU of the virulent clinical or the avirulent nonclinical isolate. The importance of C5 was confirmed by studies using C5 deficient mice and their congenic C5 sufficient counterparts. Again the C5- mice were most susceptible to infection with *Saccharomyces cerevisiae*, with 63% infected with the virulent clinical isolate dying by day 7; no $C5^+$ mice died. No mice infected with the avirulent nonclinical isolate died, and mean burdens in the brain at day 14 were sevenfold lower in C5+ mice than in C5- mice. The authors concluded that C5 is important in the early innate host response to infection with some isolates of *Saccharomyces cerevisiae* able to cause mortality. However, in comparing the relative degrees of virulence of the various isolates in C5-sufficient and C5-deficient mice, it is apparent that different isolates have developed differing genetic strategies for the manifestation of virulence, regardless of the status of host response. *Saccharomyces cerevisiae* has been added to the list of emerging pathogens to which an immunocompromised host is susceptible and must now be looked at in the same light as other more common fungal pathogens.

3.3.8. Conclusions

There are only very limited non-clinical toxicological data available.

As some clinical case reports describe, immunocompromised patients run the risk of a *Saccharomyces cerevisiae* sepsis and therefore the use of *Saccharomyces cerevisiae* is contraindicated in these patients. This contraindication is supported by the preclinical data concerning pathogenicity mentioned above.

3.4. Overall conclusions on non-clinical data

In many review articles, the pharmacodynamic effects of *Saccharomyces cerevisiae* were classified into three main domains of action: a luminal and a trophic action as well as an influence on the immune

response. Since many results reflect on *in-vitro* data or were obtaind in animals with different physiology concerning the digestive system, the biological relevance of these results cannot be evaluated. Nevertheless, since some of the effects could also be seen in patients it seems that at least basically the non-clinical results on pharmacodynamic support the usage of *Saccharomyces cerevisiae* in the conditions linked to diarrhea.

Immediately after introduction in the oesophagus live and dead cells are moved through the gastrointestinal tract simultaneously with an irreversible degradation. Disappearance from the stomach and oesophagus was followed by appearance in the small intestine. Ultimately, *Saccharomyces cerevisiae* appears in the bowel and faeces thereby were less than 1% of the administered live cells recovered in faeces. The recovery of dead cells is incomplete, since there is an irreversible loss in the gastrointestinal tract by digestion of the cell wall. The majority of dead cells (83% in rat) introduced in the gastrointestinal tract are destroyed in a 24 h period. Cells of irreversible loss in the gastrointestinal tract by digestion of the cell wall do not cross the gastrointestinal wall.

There are only very limited toxicological data available. Studies on reproductive and developmental toxicity are not available. In the case of intravenous application, the preclinical data suggest that virulence of *Saccharomyces boulardii* cannot be excluded particularly in immunocompromised patients.

As there is no information on reproductive and developmental toxicity, the use during pregnancy and lactation cannot be recommended.

4. Clinical Data

4.1. Clinical pharmacology

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

As already presented in 3.1.1 according to Kelesidis and Pothoulakis (2012) and McFarland (2010) the pharmacodynamic effects of *Saccharomyces boulardii* can be classified into three main domains of action: a luminal and a trophic action as well as an influence on the immune response. The following effects have been confirmed in humans:

Luminal action:
Within the gastrointestinal lumen both anti-toxin and antimicrobial effects have been described. Anti-toxin effects are directed against *C. difficile* toxin A and B (54 kDa protease), endotoxins of pathogenic *E. coli* (63 kDa protein phosphatase), and cholera toxins (120 kDa protein). Anti-toxin effects are due to different mechanisms: inhibition of pathogen receptor sites or direct proteolysis of pathogenic toxins via a secretion of enzymatic proteins.
Antimicrobial effects include a direct or indirect inhibition of growth of pathogens, preservation of the tight junctions between enterocytes, reduction of gut translocation of pathogens, and suppression of host cell adherence that interferes with bacterial colonization. Investigations both in mice (Barc *et al*., 2008) and patients with chronic idiopathic diarrhoea (Swidsinski *et al*., 2008) showed that after the discontinuation of antibiotic treatment the restoration of normal intestinal microbiota is achieved faster under the administration of *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926). Garcia Vilela *et al*. (2008) investigated the effect of *Saccharomyces boulardii* on intestinal permeability in 34 patients with Crohn's disease in remission. As compared to placebo, the administration of 200 mg lyophilized *Saccharomyces boulardii*-17 (about 4 x 10^8 cells; *Saccharomyces cerevisiae* CBS 5926) every 8 h over a period of 3 months in addition to baseline treatment improved intestinal barrier function as measured by lactulose/mannitol ratio.

Akil *et al*. (2006) evaluated how the oral intake of *Saccharomyces boulardii* affects the number of *E. coli* colonies in the colon. 24 healthy children (age 3-16 years) received 5 billion CFU of *Saccharomyces boulardii* once daily for 5 days. Following treatment with *Saccharomyces boulardii*, the mean number of *E. coli* colonies in g/ml stool significantly decreased from 384,625±445,744 to 6,283±20,283. During the same interval, the number of *Saccharomyces boulardii* colonies increased from 0 to 11,047±26,754 in g/ml stool.

Furthermore, within the gut lumen *Saccharomyces boulardii* shows metabolic activity by increasing short chain fatty acids (SCFA) which favor normal colonic function. Following the oral administration of *Saccharomyces boulardii* (1 g/d) on 6 consecutive days, Girard-Pipau *et al*. (2002) observed an increase in the SCFA acetic, propionic and butyric acid in patients with enteral nutrition.

Schneider *et al*. (2005) assessed the effects of *Saccharomyces boulardii* on fecal flora and SCFA in 10 patients on long-term TEN (total enteral nutrition). Treatment with *Saccharomyces boulardii* (500 mg b.i.d. lyophilized powder) significantly increased total fecal SCFA as compared to 15 healthy controls. At the end of treatment, fecal butyrate was significantly higher whereby fecal flora remained unchanged. According to the author, the preventive effects of *Saccharomyces boulardii* on TEN-induced diarrhoea may be due to the increase of faecal SCFA.

Swidsinski *et al.* (2016) investigated the impact of antibiotics and *Saccharomyces boulardii* on bacterial composition in human feces. Samples were collected from three groups of women (n=20 each) treated for bacterial vaginosis with ciprofloxacin + metronidazole. Group A received the combined antibiotic regimen, whereas the A/Sb group received concomitant *Saccharomyces boulardii*, and the A_Sb group received *Saccharomyces boulardii* prophylaxis following the 14-day antibiotic course. One 250 mg capsule *Saccharomyces boulardii* ($1.8x10^{10}$ CFUs/g lyophilisate) three times daily was administered. The decrease in the concentrations of the fermenting biomass and inconsistency of the bacterial diversity caused by antibiotics were effectively averted by *Saccharomyces boulardii* prophylaxis. A total of 88% of patients receiving *Saccharomyces boulardii* quickly restored their initial individual microbial profiles. The total microbial concentrations recovered completely in the two *Saccharomyces boulardii* groups within 3 months post antibiotic treatment. However, although they were different shortly after antibiotic treatment, both concomitant and subsequent *Saccharomyces boulardii*-treated groups were similar at the end of the observational period.

Kabbani *et al.* (2017) compared and contrasted the effects of *Saccharomyces boulardii* (SB), an antibiotic (Amoxicillin-Clavulanate, AC) and the combination on the intestinal microbiota of healthy humans in a single-center, open-label, randomized controlled trial. In addition, they examined the effects of these interventions on gastro-intestinal symptoms with a particular focus on AAD. 53 subjects were enrolled, and 49 were considered for statistical analyses. They were randomized to one of 4 study groups: SB for 14 days, AC for 7 days, SB plus AC, control (no treatment). Participants gave stool samples and completed gastro-intestinal symptom questionnaires. Microbiota changes in the stool specimen were analysed using 16s rRNA gene pyrosequencing (bTEFAP). Control subjects had a stable microbiota throughout the study period. Significant microbiota changes were noted in the AC alone group during treatment, which reverted toward baseline, but were not yet completely restored 2 weeks after antibiotic therapy. No significant shifts in bacterial genera were noted in the SB alone group. Adding SB to AC led to less pronounced microbiota shifts including less overgrowth of *Escherichia* and to reduction in antibiotic-associated diarrhoea scores.

Trophic action:

Buts *et al*. (1986) investigated the response of the small intestinal mucosa to *Saccharomyces boulardii* in 7 healthy adults. Following the administration of high doses of lyophilized *Saccharomyces boulardii* (250 mg four times per day; 250 mg with a biological activity of 9.4×10^9 viable cells) over a period of 2 weeks a peroral suction biopsy was performed. As compared to the initial biopsy, the histological

examination of the post-trial biopsy showed no morphological alterations with regard to villus height or crypt depth. After treatment, a statistically significant increase of the specific enzymatic activity of disaccharidases (sucrase, lactase, maltase) was observed, whereas mucosal protein content remained unchanged.

The effects of *Saccharomyces boulardii* on duodenal mucosa were also investigated by Jahn *et al*. (1996) by means of morphometry and determination of brush border enzyme activity. Twelve healthy volunteers received lyophilized *Saccharomyces boulardii* 5 capsules three times a day (t.i.d.) (one capsule containing 50 mg (10^9 viable cells) over a period of 21 days). A comparison of intra-individual histochemical results pre- and post-administration of *Saccharomyces boulardii* revealed a statistically significant increase of enzymatic acitivity of lactase, α-glucosidase, and alkaline phosphatase in the brush border of enterocytes. According to the authors, this effect as well as a tendency of an increased villous surface possibly may be caused by an accelerated maturation of enterocytes.

Influence on the immune response:
Both locally in the gut and systemically, *Saccharomyces boulardii* modulates the immune response either by its action as an immune stimulant or by reducing pro-inflammatory responses. Based on their studies in growing rats Buts *et al*. (1990) assume that glucan and mannans, which are complements of the yeast external capside, could play an immunogenic role.

Ozkan *et al*. (2007) described an enhancement of immune response for *Saccharomyces boulardii*. Following the oral administration of 250 mg *Saccharomyces boulardii* two times a day (b.i.d.) for 7 days to 16 patients (age 6 months to 10 years) with acute diarrhoea as compared to 11 children on placebo a significant increase in serum IgA and decrease in C-reactive protein was observed. The percentage of CD8 lymphocytes at the end of treatment was significantly higher, too. Secretory IgA is important for the maintenance of intestinal barrier function.

Machado Caetano *et al*. (1986) investigated the effects of *Saccharomyces boulardii* on specific and non-specific defense in 96 immunocompetent human volunteers. Following the oral administration of 4 x 250 mg/day for 7 days (250 mg corresponding to 3.5 x 10^9 yeast cells) a significant increase of erythrocytes, leucocytes, neutrophils, complement components, serum anti-complementary activity and leukocyte chemokinesis, especially when autologous serum and antigen have been added to the culture medium, and decrease of complement haemolytic activity were observed. The overall changes in serum proteins suggested changes of acute phase proteins typical of an inflammatory process. *Saccharomyces boulardii* had no mitogenic response of lymphocyte populations. These results demonstrate that *Saccharomyces boulardii* oral ingestion induces cellular and humoral changes in humans, which result from reticulo-endothelial system stimulation and activation of the complement system. *Saccharomyces boulardii* did not affect the specific immune response – as it lacks mitogenic effects of its own and did not modify lymphocyte populations nor their response to both PHA (phytohaemaglutinin) and Cowan I mitogens.

Stickl (1987) performed a double-blind study and investigated the effects of *Saccharomyces cerevisiae* Hansen (either 500 mg/d or 200 mg/d *Saccharomyces cerevisiae* Hansen versus placebo) on IgA in saliva. Within the 3 weeks of treatment, no effects on IgA in saliva were observed. While *Saccharomyces* had no influence on T4-lymphocytes, T8-lymphocytes slightly increased. As T8-lymphocytes are known to possess cytotoxic and suppressor properties the author concluded from the study results that *Saccharomyces cerevisiae* might have positive effects on the treatment of diseases such as atopia or autoimmune diseases.

Apart from the trophic effects on intestinal mucosa, Jahn *et al*. (1996) also investigated the effects of *Saccharomyces boulardii* on lymphocytes. Only minimal effects on the immune system of the

peripheral blood and the intestine were observed. The percentage of activated $CD25^+$ T-helper/inducer cells increased only slightly, while the concentration of IgA was not affected.

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

An overview on pharmacokinetics of *Saccharomyces boulardii* and other biotherapeutic agents is given by Martin *et al*. (1999). According to them in healthy humans *Saccharomyces boulardii* is not a natural colonizer of the gut and is not absorbed following oral administration.
Pharmacokinetics of *Saccharomyces boulardii* and *Saccharomyces cerevisiae* were investigated in healthy volunteers or patients with *Clostridium difficile* disease (CDD) or HIV-infections. Furthermore, the concomitant use of antibiotics and antifungals was studied.

In 1989, Blehaut *et al*. investigated the kinetics of *Saccharomyces boulardii* in humans. Eight healthy volunteers received 500 mg *Saccharomyces boulardii* each in the morning and in the evening for a period of 14 days. The capsules contained lyophilized *Saccharomyces boulardii* with a total number of yeast cells (±SD) of $10^{10.98\pm0.02}$ per gram resp. $10^{10.55\pm0.08}$ viable yeast cells per gram. The examination of stool samples showed that the concentration of *Saccharomyces boulardii* increased rapidly over the first 2 days of administration and achieved a steady-state on day 3. Less than 1% (mean 0.36±0.31%) of the dose of live cells administered was recovered in feces and a half-life of 6 h was reported. The rate of recovery from dead cells ranged from 17% to 58% with an overall mean of 31.5±12.1%. Within 7 days following the termination of administration yeast cells were not detected in feces anymore. There was no evidence that cells of *Saccharomyces boulardii* cross the gastrointestinal wall in this study.

Another pharmacokinetic trial was performed by Pecquet *et al*. (1991) in 6 healthy volunteers who received a daily dosage of 3 times 10^8 life-dehydrated *Saccharomyces cerevisiae* cells for 5 days. Counts of *Saccharomyces cerevisiae* faecal excretion gradually increased to a maximum of 10^5 CFU/g feces. Within 5 days after the end of treatment yeast cells disappeared from faeces.

Klein *et al*. (1993) studied the recovery and elimination of *Saccharomyces boulardii* in healthy human volunteers. For the investigation of *Saccharomyces boulardii* dose on recovery of stool daily doses of 0.2, 1.0 and 3.0 g *Saccharomyces boulardii* were administered sequentially to 8 healthy volunteers over a period of one week each. For each dose steady-state levels occurred by 72 h. With increasing *Saccharomyces boulardii* doses, the main steady-state concentration of *Saccharomyces boulardii* increased significantly, whereas the percentage of recovery was independent from dose. The effect of ampicillin on single-dose elimination kinetics was investigated in 10 volunteers. Following the single oral administration of 1 g *Saccharomyces boulardii* total 24 hour stool was collected for 7 days. After a wash-out period of 7 days, ampicillin was given for another 7 days at a dose of 250 mg four times a day. 24 hours after the first dose of ampicillin, the test persons received a single dose of 1.0 g *Saccharomyces boulardii*. For the following 7 days, total 24 h stool was collected. Ampicillin significantly increased ($p<0.01$) the area under the concentration versus time curve and maximum faecal concentration. In order to investigate the effect of ampicillin on *Saccharomyces boulardii* elimination at steady state in 6 volunteers the same procedure was used as for the study of single-dose elimination kinetics with the exception that 0.5 g *Saccharomyces boulardii* was taken b.i.d for 7 days. Mean faecal output of *Saccharomyces boulardii* was measured between 72 and 120 h, when steady-state levels were reached. With ampicillin steady-state recovery of the drug increased about twofold ($p<0.05$) and steady-state levels were about 2.4 times higher ($p<0.01$).

Elmer *et al*. (1999b) reported a rapid clearance of *Saccharomyces boulardii* in 97 patients with recurrent *Clostridium difficile* disease (CDD). *Saccharomyces boulardii* was given in addition to standard antibiotic treatment (either vancomycin 2 g/d or 500 mg/d or metronidazole 1 g/d) for 10

days. On day 7 - according to randomization - 47 patients received placebo, 50 patients oral treatment with 1 g of *Saccharomyces boulardii* (500 mg b.i.d; 1 g containing about 10^{10} viable organisms) for 4 weeks. In 48 patients, the clearance of *Saccharomyces boulardii* was investigated. In 94% of the patients, *Saccharomyces boulardii* was cleared by the third day after the discontinuation of treatment. As compared to studies in healthy volunteers having received the same dosage of *Saccharomyces boulardii* fecal concentrations were 1-2 log lower. Patients who were asymptomatic at the time of stool collection in this study had significantly higher concentrations than patients with disease symptoms. This clinical study also showed that patients with low faecal concentrations of *Saccharomyces boulardii* had a higher risk of recurrence of CDD.

Controversial results to Graff *et al*. (2008) (see non-clinical pharmacokinetics) were achieved by Scevola *et al*. (2003). They, too, evaluated acid tolerance *in-vitro* and fecal recovery *in-vivo* of a *Saccharomyces cerevisiae* strain (*Saccharomyces* strain unknown) after oral administration to 16 healthy volunteers. From pH 1.0 to pH 7.0 the release of *Saccharomyces cerevisiae* in buffer solutions increased. The selected yeast strain showed good tolerance to low pH, which mimics the gastric environment. After one month of treatment at a dose of 100 million cells per day, *Saccharomyces cerevisiae* grew from the feces of 6 (37.5%) of the 16 healthy, treated volunteers. These findings, however, are of limited validity, since the dose of *Saccharomyces* administered was low (4.7×10^7 cfu/ml) and the drug product also contained vitamins (B1, B2, B6, B8, B12 and folic acid).

4.2. Clinical efficacy

4.2.1. Dose response studies

Not available.

4.2.2. Clinical studies (case studies and clinical trials)

At the beginning of this section, it is emphasized that in the clinical studies assessed different medicinal products containing *Saccharomyces boulardii* have been administered to the patients. Unfortunately, only in a few studies information on the *Saccharomyces boulardii strain* and number of living cells was given. In these cases, the information was included in the assessment report. If no information is given, the authors did not include these data in their publication.

Treatment of acute diarrhoea

Höchter *et al.* (1990) assessed safety and efficacy of a *Saccharomyces boulardii* treatment for patients with acute diarrhoea (more than 3 watery stools during the last 24 h) in a randomized double-blind, placebo-controlled, multicenter clinical trial for the reduction of stool frequency. 107 ambulatory patients were randomized. The data of 15 patients were not included in the statistical evaluation because of violation of the inclusion and exclusion criteria (to one patient additionally one antibiotic was applied which could cause diarrhoea, 14 patients had less than 3 watery stools). A total of 92 patients were included in the statistical evaluation (*Saccharomyces boulardii*: n=43; placebo: n=49). Treatment lasted 7 days. Control visits were performed on day 1, 3, and 8. Frequency and consistency of stool were the main efficacy variables which were assessed by a score for consistency (1=formed, 2=soft, 3=liquid), multiplied by the number of stools per day. Efficacy and tolerability were evaluated on basis of a score ranging from very good to bad separately by patient and investigator. Under *Saccharomyces boulardii* the reduction of score derived from stool frequency and consistency was significantly higher than under placebo (-17.2 and –13.6, respectively; p=0.035) after 2 days of therapy. Concerning accompanying variables there were significant advantages of the *Saccharomyces boulardii* treatment in comparison to placebo: improvement of nausea (day 3: 78.4% and 51.3% respectively (p=0.014); day 8: 100% and 81.1%, respectively (p=0.022)) and positive judgement of

therapy on day 3 by patients (very good/good) 95.1% and 76.1%, respectively (p=0.013). Positive judgement of therapy on day 3 by investigator was 88.4% for *Saccharomyces boulardii* and 78.8% for placebo. On day 3 treatment with *Saccharomyces boulardii* resulted in 21%±40% liquid stools and with placebo in 22%±40%, at day 8 under treatment with *Saccharomyces boulardii* 3%±16% and under placebo 12%±33% (p=0.026).
Two adverse reactions (slight constipation and vomiting) were reported during the study drug treatment period by two patients receiving *Saccharomyces boulardii* and by two patients in the placebo group.

Mayr *et al.* (1996) treated 222 patients with acute diarrhoea with 500 mg *Saccharomyces boulardii* daily for 7 days (either 2 capsules A (n=110) or 2 capsules B (*Saccharomyces cerevisiae* CBS 5926) (n=112)) in a controlled, randomized multicentre double-blind study. Both products differed only in their excipients: lactose was contained only in B. According to the publication both products contain identical strains with, at least 10^{10} live micro-organisms/g. The average number of stools per day decreased from 7.0 (A) and/or 6.8 (B) on day 0 to 1.94 and 2.25 on days 3-6. The statistical analysis showed at least an equivalent efficacy of both products. In total 13 patients reported adverse events. The symptoms mentioned were dizziness, recurrence of diarrhoea, nausea, vomiting, abdominal pain, dyspnoea and weakness. In one patient myocardial infarction was suspected. In the publication the causal evaluation was missing. However, the authors concluded that the safety of treatment with *Saccharomyces* was good.

Lacarrière and Rieckhoff (1986) assessed *Saccharomyces cerevisiae* Hansen for its efficacy in acute diarrhoea with at least three loose stools per day in a post marketing uncontrolled study with 3026 patients from 12 European and African countries. The patients should be older than one year; diet and concomitant oral treatment e.g. for rehydration should be applied as stated by the investigator. *Saccharomyces cerevisiae* was taken alone as a single therapy. The time of the medication was limited to 4 days. The efficacy was assessed on basis of the duration of diarrhoea. A total of 92.3% out of 2911 cases analysed responded well to a monotherapy with *Saccharomyces cerevisiae* in acute diarrhoea. *Saccharomyces cerevisiae* was also effective in children aged between one and five years (469; 19 excluded) and the efficacy was as good as in the age group of ≥6 years (92% and 92.4%, respectively). Efficacy was also assessed in consideration of the geographic origin. In Europe 93% of patients ≤5 years and 92% of patients >5 years responded well, in Africa 86% and 94% respectively. In 77% of the patients diarrhoea stopped within the first 3 days or less of treatment.
The tolerability of the treatment was very good. A total of eight patients reported side effects such as abdominal pain or meteorism which did not lead to a discontinuation of treatment.

Cottrell *et al.* (2015) conducted a prospective, randomized, single (investigator)-blind, three-arm, parallel group, non-inferiority clinical trial in adults with acute diarrhoea at clinics in Mexico and India. The patients were international travellers, expatriates and local residents and aged 18 years or over. They had symptoms of diarrhoeal illness with onset during the prior 48 h, a minimum of three unformed stools in the 24 h before study entry with the most recent stool unformed, and with abdominal discomfort within the prior 4 h. 415 subjects (ITT) were randomly assigned to Loperamide–simeticone 2/125 mg capsule-shaped tablet (caplet) (n=139), Loperamide–simeticone 2/125 mg chewable tablets (n=139) or *Saccharomyces boulardii* 250 mg capsules (≥1.8 x 10^{10} viable cells/g) (n=137). Two dosage units of Loperamide were taken initially at the investigator site, followed subsequently by one dosage unit after each unformed stool, with a maximum of 4 dosage unit in a 24-h period, for up to 48 h. *Saccharomyces boulardii* was administered twice daily for 5 days. The primary endpoint was the number of unformed stools passed between 0 and 24 h following the initial dose of study medication (NUS 0–24). Both Loperamide-simeticone groups had a significantly lower mean NUS 0-24 than the *Saccharomyces boulardii* group (both p<0.001). Mean NUS 0–24 values were 3.3 in the loperamide–simeticone caplet group, 3.2 in the chewable tablet group and 4.3 in the Saccharomyces

boulardii group. The (upper) limit of the one-sided 97.5 % CI for the difference (caplets - tablets) was 0.48, within the predetermined non-inferiority margin. Median time to last unformed stool, to complete relief of diarrhoea and of abdominal discomfort were also significantly lower in the loperamide-groups compared to *Saccharomyces boulardii*. At 7-day follow-up most subjects reported passing stool at least once since the final study visit (loperamide–simeticone caplet 94.1%, loperamide–simeticone chewable tablet 94.8%, *Saccharomyces boulardii* 97.0%), did not experience continued or recurrent diarrhoea [loperamide–simeticone caplet 3.7% (p<0.03 vs. *S.boulardii*), loperamide–simeticone chewable tablet 3.7%, *Saccharomyces boulardii* 5.7%] and felt completely well [loperamide–simeticone caplet 96.3% (p<0.02 vs. *Saccharomyces boulardii*), loperamide–simeticone chewable tablet 96.3% (p<0.02 vs. *Saccharomyces boulardii*), *Saccharomyces boulardii* 88.6%].
A total of 17 (4.1%) subjects experienced at least one adverse event; three (2.2%) subjects in the loperamide–simeticone caplet group, seven (5.0%) subjects in the loperamide–simeticone chewable tablet group and seven (5.1%) subjects in the *Saccharomyces boulardii* capsule group. The most commonly reported adverse events were nausea, asthenia and anorexia [in three (0.7%) subjects each, overall]. Constipation was reported by two (1.4%) subjects in the loperamide–simeticone chewable tablet group, with no reports in the other two groups. No subject experienced a serious adverse event or was withdrawn from the study due to an adverse event.
This study was single blinded and performed in Mexico and India without a placebo-controlled group. Therefore, this study did not assess the efficacy of *Saccharomyces boulardii* compared to placebo.

Table 6: Clinical studies (controlled and uncontrolled) on the treatment of acute diarrhoea

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute diarrhoea Höchter *et al.,* 1990	placebo-controlled, double-blind, randomized, multicenter duration: 8 days	per capsule:50 mg *Saccharomyces cerevisiae* CBS 5926 (≥1.8 x 10^{10} viable cells/g), 6.5 mg lactose, 93.5 mg saccharose 3 x 4 capsules at day 1 and 2, 3 x 2 capsules from day 3 to day 7	92 patients out of 107 randomized patients 41 female, 51 male verum: 43 placebo: 49	acute adult diarrhoea with more than 3 loose stools during the last 24 hours before consultation of a doctor	primary endpoint: score derived from stool frequency and quality (number of stools x consistency 1=formed, 2=soft, 3=loose) at day 3 day 1: verum 22.7±12.5 placebo 20.3±9.7 day 3 verum 5.5±6.8	Mann-Whitney-U-test for the primary endpoint and separate evaluation of stool frequency and quality	Under *Saccharomyces boulardii* the reduction of score derived from stool frequency and consistency was significantly higher than under

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		oral use	age: 18-65 years, mean age: 38 years		placebo 6.7±8.7 P=0.035 subgroup of patients with diarrhoea ≤2 days at time of study inclusion day 1 verum 22.0±11.7 placebo 19.3±7.1 day 3 verum 4.5±4.3 placebo 6.6±9.2	chi-square-test for the other secondary endpoints	placebo (-17.2 and –13.6, respectively; p=0.035) after 2 days of therapy.

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					without significance calculation secondary endpoints day 3 and day 8: stool frequency, stool quality, nausea, abdominal pain, vomiting, temperature, global efficacy and global tolerability		
Treatment of acute diarrhoea	controlled, randomized multicente	2 capsules A(n=110) or 2 capsules B(n=112) according to 500 mg	222 patients ≥18 years (128 male, 94 female)	acute diarrhoea (not longer than 24 hours) with more than 3 loose stools during the last 24	primary endpoint: stool frequency on day 2; cumulated stool frequency day 3 to day 6	noninferiority: confidence-interval-inclusion-	missing placebo control group, therefore

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Mayr *et al.,* 1996	r double-blind	*Saccharomyces cerevisiae* CBS 5926 (at least 10^{10} live micro-organisms/g) duration: 7 days (?)	ITT-population 219, because of 3 patients with missing data at day 2 (108 A, 111 B) PP-population 198: 6 patients not conform with inclusion	hours before inclusion	secondary endpoints: stool consistency, nausea, vomiting , overall assessment by physician and patient statistical analysis showed at least an equivalent efficacy of both products additional explorative analysis showed a significant superiority of A concerning the cumulated stool frequency and	method, α=0.025 superiority (explorative): covariance analysis, one-sided U-test, one-sided exact Fisher-test	efficacy cannot be assessed objectively, equivalent result of both products

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			criteria, 12 with medicine intake too low, 3 with incomplete documentation		consistency day 3 to day 6		
Treatment of acute diarrhoea Cottrell *et al.,* 2015	prospective, randomised, single (investigator)-blind, three-arm, parallel group,	Loperamide–simeticone 2/125 mg capsule-shaped tablet (caplet) (n=139): Loperamide–simeticone 2/125	415 (ITT) international travellers, expatriates and local residents and aged 18 years or over with mean	onset of diarrhea during the prior 48 h, a minimum of three unformed stools in the 24 h before study entry with the most recent stool unformed, and with	primary endpoint: number of unformed stools passed between 0 and 24 h following the initial dose of study medication (NUS 0–24): Both Loperamide-	The test for non-inferiority (NUS 0–24) of loperamide – simeticone caplets	single blinded and performed without a placebo-controlled group

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
	non-inferiority clinical trial in Mexico and India duration 7 days	mg chewable tablets (n=139): 2 dosage units initially, followed subsequently by one dosage unit after each unformed stool, with a maximum of 4 dosage unit in a 24-h period, for up to 48 h. *Saccharomyces boulardii* 250 mg capsules (≥1.8 x 1010 viable cells/g)	age of 36.4 years (range 18-79), male 64% female 36% Origin Mexico 27%, USA/Canada 23%, India 20%, Europe 17%, Russia 7%, South	abdominal discomfort within the prior 4 h	simeticone groups had a significantly lower mean NUS 0-24 than the Saccharomyces boulardii group (both p<0.001). Some secondary endpoints: Median time to last unformed stool, to complete relief of diarrhoea and of abdominal discomfort were also significantly lower in the loperamide-groups compared to	versus tablets was one-sided with aequal to 0.025; all other tests were two-sided at a 0.05 level Chisquare or Fisher's exact test.	

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		(n=137): twice daily for 5 days	America 2%, other 3%		Saccharomyces boulardii.		
Treatment of acute diarrhoea Lacarrière and Rieckhoff, 1986	open post marketing uncontrolled study duration: 4 days treatment	capsules containing *Saccharomyces cerevisiae* CBS 5926: ≥1.8 x 10^{10} viable cells/g (the amount of S.*cerevisiae* /capsule is not given) children 1-5 years: 3 x 3 capsules	3026 subjects ≥1 year; 115 excluded resulted in 2911 patients >5 years and 450 ≤5 years	acute diarrhoea with more than 3 loose stools during the last 24 hours before consultation of a doctor	duration of diarrhoea cured: 1 to 2 formed stools during 24 hours not cured: diarrhoea lasts more than 4 days; need for another antidiarrhoeal after 2 days	total population: 92.3% cured patients ≤5 years: 92% cured patients >5 years:	limited relevance for efficacy because uncontrolled no differences in consideration of age

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		patients >5 years: 3 x 4 capsules				92.4% cured	and geographical origin good tolerability

Special kinds of diarrhoea and gastrointestinal disorders

Mansour-Ghanaei *et al.* (2003) enrolled 57 adult patients with acute amebiasis in a double blind, randomized clinical trial in Iran. Three patients were excluded because of non-compliance. The patients were randomized either to regimen 1 (metronidazole (750 mg t.i.d.) and iodoquinol (630 mg t.i.d.) for 10 days, n=27) or regimen 2 (capsules of lyophilized *Saccharomyces cerevisiae* CBS 5926, 250 mg t.i.d.) orally in addition to regimen 1, n=27). Patients were re-examined at 2 and 4 weeks after the treatment, and stool was examined at the end of week 4. Student's t-test, Chi-square and McNemar's tests were used for statistical analysis. In group 1, diarrhoea lasted 48.0±18.5 hours and in group 2, 12.0±3.7 hours ($p<0.0001$). In group 1, the durations of fever and abdominal pain were 24.0±8.8 and 24.0±7.3 and in group 2 they were 12.0±5.3 and 12.0±3.2 hours, respectively ($p<0.001$). Duration of headache was similar in both groups. At week 4, amebic cysts were detected in 5 cases (18.5%) of group 1 but in none of group 2 ($p<0.02$).

Besirbellioglu *et al.* (2006) evaluated the efficacy of *Saccharomyces boulardii* against *Giardia lamblia* infections in a double-blind, placebo-controlled study in Turkey. Group 1 (30 adult patients with giardiasis) included metronidazole 750 mg 3 times daily along with *Saccharomyces boulardii* capsules (250 mg b.i.d orally, *Saccharomyces cerevisiae* CBS 5926) while group 2 (35 patients) was treated with metronidazole 750 mg 3 times daily and placebo for 10 days. In group 1 17 patients were symptomatic (diarrhoea, abdominal pain etc.) and the other 13 patients were asymptomatic. In group 2 15 patients were symptomatic and 20 asymptomatic. Patients were re-examined at 2 and 4 weeks after treatment, and stool examinations were performed. Statistical analysis was performed by the Mann-Whitney U-test and Fisher's exact test. At week 2, *G. lamblia* cysts were detected in 6 cases (17.1%) of group 2 and none in group 1. The proportion of patients with clearance of microscopical findings after 2 weeks was 100% (group 1) and 82.8% (group 2) ($p=0.027$). At the end of the fourth week, presence of the cysts continued in the same 6 cases in group 2. These findings indicated that *Saccharomyces boulardii* may be effective in treating giardiasis when combined with metronidazole therapy.

Saint-Marc *et al.* (1995) conducted a double-blind, placebo controlled, parallel group trial in 35 patients with stage IV AIDS to evaluate the efficacy of *Saccharomyces boulardii* in AIDS-related diarrhoea unresponsive to standard therapy. Mean age was 34.9 years. Most patients were male. The cause of diarrhoea was identified in 54.3% of cases (cryptosporidiosis in 20%). Eighteen patients were assigned to *Saccharomyces boulardii* therapy (3 x 1000 mg/day) and 17 to placebo. Resolution of diarrhoea was recorded in 61% of *Saccharomyces boulardii* patients versus 12% of placebo patients after one week ($p<0.002$). Significant improvements were also noted in the *Saccharomyces boulardii* group regarding the daily diarrhoea score based on stool number, weight, and volume ($p<0.002$), abdominal pain, abdominal distension, asthenia, weight gain, and Karnofsky index. Tolerability was very good. These data show that over a one-week period *Saccharomyces boulardii* is an effective treatment for persistent AIDS-related diarrhoea.

Prevention of acute antibiotic-associated diarrhoea (AAD)

The definition of antibiotic-associated diarrhoea (AAD) is "otherwise unexplained diarrhoea that occurs in association with the administration of antibiotics." (Bartlett 2002) According to McFarland (2006) the primary outcome for AAD is diarrhoea (≥3 loose stools/d for at least 2 days or ≥5 loose stools/48 h) within 2 months of antibiotic exposure. Its incidence depends on the kind of antibiotic administered, patient's characteristics such as age and general health status, severity of disease requiring intensive care, in- or outpatient treatment. Rates of up to 60% have been reported during hospital outbreaks (McFarland 2006). Symptoms vary in intensity from mild to life-threatening and may occur soon after the initiation of antibiotic treatment or up to 2 weeks after the end of antibiotic therapy (Micklefield

2014). The antidiarrhoeal effects of *Saccharomyces boulardii* may be explained pharmacologically by its trophical (increase of short chain fatty acids in the colon) and antitoxin effects (Micklefield 2014).

In a review by McFarland (2010) on the use of *Saccharomyces boulardii* in adults 10 randomized controlled clinical studies (see table 7, no. 1-7, Cremoni *et al.*, 2002a, Duman *et al.*, 2005, Cindoruk *et al.*, 2007, see table 8) have been included. According to the meta-analysis of these studies, *Saccharomyces boulardii* was assessed as significantly protective of AAD.

This effect has also been described in another meta-analysis by McFarland (2006) who investigated the efficacy of probiotics for the prevention of AAD and treatment of CDD (*Clostridium difficile* disease) on the basis of published randomized, controlled clinical trials (no. 1; 3; 4; 5; Kotowska *et al*., 2005; Cremoni *et al.*, 2002a). From their meta-analysis on the effect of probiotic administration on AAD which included the clinical trials no. 3-5 Cremonini *et al*. (2002b) concluded that the results obtained suggest a strong benefit of probiotic administration on AAD, but that the evidence for beneficial effects is still not definitive. According to the authors, published studies are flawed by the lack of a placebo design and by peculiar population features.

D'Souza *et al*. (2002) included 9 randomized, double-blind, placebo-controlled clinical trials in their meta-analysis on probiotics in the prevention of AAD. In 4 of these trials (no. 1; 3; 4; 5) *Saccharomyces boulardii* was administered. The authors concluded that probiotics can be used to prevent AAD and that *Saccharomyces boulardii* has the potential to be used in this situation. The efficacy of probiotics in this indication still remains to be proven.

Another meta-analysis published by Szajewska and Mrukowicz (2005) evaluated the effectiveness of *Saccharomyces boulardii* in the prevention of AAD in children and adults. Five randomized clinical studies (no. 1; 3; 4; 5; Kotowska *et al.*, 2005, n=1076 patients) have been included in the review and *Saccharomyces boulardii* was assessed as moderately effective in preventing AAD in children and adults treated with antibiotics. According to them, for every 10 patients receiving daily *Saccharomyces boulardii* with antibiotics, one fewer will develop AAD.

In 2012 another systemic review and meta-analysis on probiotics for the prevention and treatment of AAD was published by Hempel *et al*. (2012). They assessed parallel randomized controlled trials with different probiotics for the prevention of AAD and found that the pooled evidence suggests that probiotics are associated with a reduction in AAD. Nevertheless, more research is needed to determine whether this association varies systematically by population, antibiotic characteristic, or probiotic preparation.

Xie *et al*. (2015) published a systematic review concerning probiotics for the prevention of antibiotic-associated diarrhoea in older patients. Six trials with a total of 3562 patients were included. Only one of them investigated *Saccharomyces cerevisiae (boulardii)* in 69 patients, however, a benefit from *Saccharomyces boulardii* in preventing AAD and CDD in this patient group was not identified (Lewis *et al*. (1998). In their review, the authors concluded that probiotics may not reduce the risk of AAD and CDD (*Clostridium difficile* diarrhoea) in older patients. More robust studies were demanded, which should include large samples sizes, multi-centre and double blind designs and should isolate and examine factors such as probiotic strains and the types of antibiotic.

Szajewski and Kolodziej (2015b) published an update of their 2005 meta-analysis. In addition to the previously identified five randomized clinical trials, 16 new trials were included. A total of 4780 participants (2441 in the experimental group and 2339 in the control group) were included. 12 studies were placebo-controlled; in the remaining studies, there was no intervention in the control group. 15 trials (Chu *et al.* 2012, table 7 except Ehrhardt *et al.* (2016) and table 8 except Lee et *al.* (2011)) were performed in adults, and six trials in children (see below). Only two trials (Kotowska *et al.* 2005 and Pozzoni *et al.* 2012) were at low risk of bias. The daily dose of *Saccharomyces boulardii* ranged from

50 mg to 1000 mg. In adults, compared with placebo or no treatment, *Saccharomyces boulardii* reduced the risk of diarrhoea from 17.4% to 8.2% (15 RCTs, n=3114, RR: 0.49, 95% CI: 0.38-0.63; NNT: 11, 95% CI: 9-15). Subgroup analysis based on age, showed that the administration of *Saccharomyces boulardii* did not reduce the risk of *C. difficile*-associated diarrhoea in adults. One major limitation is that the methodological quality of included trials varied. Definition of AAD and/or diarrhea differed. There were wide differences in the duration of follow-up, which varied from 2 weeks to 1 year after cessation of antibiotic treatment, or it was not specified. The optimal dose of probiotics, including *Saccharomyces boulardii*, and the duration of treatment have not been established. The only study with low bias in adults (Pozzoni *et a*l. 2012) showed that *Saccharomyces boulardii* was not effective in preventing the development of AAD. Data regarding therapy-related adverse effects were available from 16 of the included trials. In these trials, *Saccharomyces boulardii* was well tolerated. Adverse events rate was similar in experimental and control groups.
One important question remains according to the authors whether the use of *Saccharomyces boulardii* shall be considered in all subjects receiving antibiotics or only in select populations. This will require clinical judgement.

Apart from the studies listed in the table 7 another clinical study has been identified in literature (Table 7: no.9) which has not been included in the meta-analyses mentioned above.

Ehrhardt *et al.* (2016) (Table 7: no.9) performed a multicentre, randomized, double-masked, placebo-controlled phase III study in hospitalized patients who received systemic antibiotic treatment in 15 hospitals in Germany between July 2010 and October 2012. Participants received 250 mg capsules *Saccharomyces boulardii* (at least 1.8×10^{10} live cells/g lyophilisate) or a matching placebo orally twice per day within 24 hours of initiating antibiotic treatment, continued treatment for 7 days after antibiotic discontinuation, and were then observed for 6 weeks. 2.444 patients ≥ 18 years were screened. The trial was stopped early for futility after inclusion of 477 participants. 246 patients aged 60.1±16.5 years and 231 patients aged 56.5±17.8 were randomized to the *Saccharomyces boulardii* group and the placebo group, respectively. 21 AADs occurred in the *Saccharomyces boulardii* group and 19 AADs in the placebo group (p=0.87). Two cases of CDAD (Clostridium difficile-associated diarrhea) were observed in each group. None of the prespecified factors (center, age, sex, duration of antibiotic treatment, and readministration of antibiotics) were associated with risk of AAD. Nine serious adverse events were recorded in the *Saccharomyces boulardii* group, and 3 serious adverse events in the placebo group. None were related to study participation. The authors found no evidence for an effect of *Saccharomyces boulardii* in preventing AAD or CDAD.

Table 7: Clinical studies on the prevention of acute antibiotic-associated diarrhoea (AAD) with *Saccharomyces boulardii* in adults

Type (aim) and objective(s) of Study Reference	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen; Route of Administration Duration of** **Treatment**	**Number of Subjects (including age, sex, drop out)**	**Healthy Subjects or Diagnosis of Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI** **95%)** **Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Prevention of AAD Adam *et al*., 1977	placebo controlled, randomized, multicenter	*Saccharomyces cerevisiae* CBS 5926: 4 cps/d (i.e. 200 mg /d) duration: 7 days	verum: n=199 (96 male/103 female) placebo: n=189 (96 male/93 female) mean age: verum: 39.3 yrs placebo: 37.6 yrs	administration of oral antibiotic treatment (tetracycline or beta-lactam) for ≥5 days because of broncho-pulmonary or ENT infection age: >15 years	occurrence of diarrhoea or candida infection diarrhoea: verum: 4.5% placebo: 17.5% candidiasis: verum: 2.0% placebo:12.2%	diarrhoea/ candidiasis: chi-square test both tests were highly significant	older investigation, publication in French only, follow-up too short

Prevention of AAD Monteiro *et al*., 1981	placebo controlled, double-blind	*Saccharomyces cerevisiae* CBS 5926: 4 cps/d duration: 6 days	verum: n=121 placebo: n=119	administration of oral antibiotic treatment (tetracycline or beta-lactam)	occurrence of diarrhoea (>2 bowel motions/d) or candida infection diarrhoea verum: 15.7% placebo: 27.7% candidiasis: verum; 1.7% placebo: 10.7%	diarrhoea/ candidiasis: Student's t-test	older investigation, publication in Portuguese only
Prevention of AAD Surawicz *et al*., 1989a	placebo-controlled, double-blind, 2:1 randomization	*Saccharomyces boulardii*: 1000 mg/d within 48 h after start of antibiotic treatment Duration: 2 weeks after last antibiotic therapy	318 patients enrolled 138 patients not included into statistical evaluation n=180 patients for statistical evaluation verum: n=116 placebo: n=64 mean age: 47.8 yrs male: 68.9%	in-patients receiving new antibiotic treatment for ≥3 days	incidence of diarrhoea (≥3 loose or watery stools/d for at least 2 days): verum: 9.5% placebo: 21.8%	incidence of diarrhoea: Chi-square (statistically significant difference)	antibiotic therapy not specified

Prevention of AAD McFarland *et al*., 1995	placebo-controlled, double-blind, multi-center, 1:1 randomization USA	lyophilized *Saccharomyces boulardii* 1 g/d (3×10^{10} cfu) within 72 h of the start of antibiotic treatment; administration until 3 days after the discontinuation of antibiotic treatment follow-up for 7 weeks	n=193 patients verum: n=97 mean age: 40.7 yrs male: 63.9% placebo: n=96 mean age: 42.3 yrs male: 65.6%	in-patients receiving new prescriptions for at least one beta-lactam antibiotic for ≥ 48 h	incidence of diarrhoea (≥3 loose or watery stools/d for at least 2 days): ITT-population verum: 7.2% placebo: 14.6%	incidence of diarrhoea binominal exact test (statistically significant)	efficacy investigated in the prevention of beta-lactam associated diarrhoea only
Prevention of AAD Lewis *et al*., 1998	placebo controlled, randomized UK	*Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) 113 mg b.i.d. throughout antibiotic therapy	n=69 patients verum: n=33 mean age: 75 yrs placebo : n=36 mean age: 77 yrs	patient older than 65 yrs with antibiotic treatment within the preceding 24 h	incidence of diarrhoea (≥3 loose stools/d) verum: 21% placebo: 13.9%	2-tailed Mann-Whitney or chi-square test no statistically significant difference	daily dose of *Saccharomyces boulardii* low no follow-up after the end of antibiotic therapy only elderly patients included

Prevention of AAD Can *et al*., 2006	randomized, antibiotic + placebo vs antibiotic + *Saccharomyces boulardii*, Turkish in-patients	*Saccharomyces boulardii* 48 h after initiation of antibiotic therapy at a dosage of 500 mg/d 250 mg contains 5 x 10^9 cfu observation period: 4 weeks after the end of antibiotic treatment	n=151 Verum + antibiotic: n=73 placebo + antibiotic: n=78	in-patients (age: 25-50 yrs) with chemotherapy not requiring intensive care	incidence of diarrhoea verum: 1.4% placebo: 9.0%	Student's t-test: statistical significance	Age was found to be a risk factor of AAD. Age was significantly higher in the AAD than in the non-AAD group. No information given, if the treatment groups were comparable initially.
Prevention of AAD Bravo *et al*., 2008	randomized, double-blind, Chile	*Saccharomyces boulardii* 500 mg/d for 12 days ($\geq$ 1,2 x 10^{10} viable cells)	n= 86 age: 15-81 yrs verum: n=41 placebo: n=45	adult patients with acute infectious diseases receiving treatment with amoxicillin for 5-10 days	incidence of diarrhoea verum: 9.8% placebo:11.1 %	no statistically significant differences between both treatments	only abstract in English
Prevention of AAD Pozzoni *et al*., 2012	placebo-controlled, randomized, double-blind, single-center	*Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) 5 x 10^9 cfu b.i.d within 48 h after the start of antibiotic therapy treatment duration: 7 days	n=275 verum: n=141 mean age: 79.9 yrs placebo: n=134 mean age: 78.5 yrs	Hospitalized patients >50 yrs of age with antibiotic therapy for <48 h	incidence of diarrhoea (>3 passages of liquid stool/d for at least 2 days or $\geq$5 passages within 48 h) verum: 15.1% placebo: 13.3%	no statistically significant difference chi-square test, Fisher's exact test	only elderly hospitalized patients included Saccharomyces boulardii was not effective in preventing the development of AAD

Prevention of AAD Ehrhardt *et al.* 2016	placebo-controlled, randomized, double-blind, multicenter	250 mg capsules *Saccharomyces boulardii* (at least 1.8 x 10^{10} live cells/g lyophilisate) or a matching placebo orally b.i.d. within 24 hours of initiating antibiotic treatment, continued treatment for 7 days after antibiotic discontinuation follow-up: 6 weeks after the end of antibiotic treatment	n=477 ITT analysis n=292 (with complete observations) PP analysis verum: n=246 mean age: 60.1 ± 16.5 years placebo: n=231 mean age: 56.5 ± 17.8 years 2.444 patients were screened. The trial was stopped early for futility after inclusion of 477 participants.	Hospitalized patients ≥ 18 yrs of age with systemic antibiotic therapy	Primary endpoint (ITT): Risk of AAD (diarrhea (passage of 3 or more loose or liquid stools (mostly in larger amounts) within 24 hours for at least 2 days) with onset not before the 3rd day of antibiotic treatment) Verum=21 episodes of AAD Placebo: 19 episodes of AAD P=0.87, hazard ratio of AAD in the verum group compared with the placebo group: 1.02 (95% CI, 0.55-1.90; p=0.94). Secondary endpoints: Risk of CDAD: 2 cases in each group Mean duration of AAD: Verum=4.48 days	no statistically significant differences between both treatments primary endpoint: Prentice, Williams and Peterson (PWP) model analyses Secondary endpoints: Cox proportional hazar model Cochran-Mantel-Haenszel χ^2 test The findings were robust in the PP as well as several sensitivity analyses.	no evidence for an effect of *Saccharomyces boulardii* in preventing AAD or CDAD limitations e.g.: Based on a 15 % incidence of AAD 686 patients would be needed in each group for 80 % power to detect 5 % difference in the cumulative incidence of AAD between treatment group. Unplanned, masked interim analysis according to the Müller and Schäfer procedure due to slow recruitement and unexpectedly few events.

					Placebo=4.26 days P=0.79 Mean time to onset: Verum=18.4 days Placebo=18.9 days		The authors concluded that based on the lower observed incidence of AAD in the 2 groups, they still would not have found a difference between the groups, if they had reached the target sample size. 1967 of 2444 screened patients were ineligibile for participation, one third due to contraindications. 100 cases (verum) and 85 cases (placebo) with missing or incomplete data concerning stool frequencies, however ITT analysis used all available data

Prevention of acute antibiotic-associated diarrhoea (AAD) caused by triple therapy of *Helicobacter pylori* infection

Helicobacter pylori, which was first discovered by Marshall and Warren in 1984, is an important factor in the pathogenesis of gastroduodenal ulcers and gastritis. In addition, chronic infections with *H.pylori* are associated with an increased risk for gastric carcinoma and gastric MALT (Mucosa-associated lymphoid tissue) - lymphoma. (Fischbach *et al.*, 2009). According to the guideline on *Helicobacter pylori* and gastroduodenal ulcer disease (Fischbach *et al.*, 2009) in Germany the prevalence of *H. pylori* infections ranges between 5% (children) and 24% (adults) and is higher in immigrants (36-86%). Triple therapy (combination of protonpump inhibitor plus amoxicillin plus clarithromycin) is indicated for the eradication of *H. pylori* infections. In order to evaluate, if the administration of *Saccharomyces boulardii* is effective in preventing side-effects (e.g occurrence of diarrhoea) of triple therapy for *Helicobacter pylori* infection several clinical studies have been performed (Table 8).

Cremonini *et al*. (2002a) included 85 *H. pylori* positive, asymptomatic patients in their clinical study who were randomized in four groups (*Lactobacillus* GG, *Saccharomyces boulardii*, *Lactobacillus* spp/biphidobacteria, placebo) to receive probiotic or placebo treatment during and for 7 days after a 1-week triple therapy (20 mg rabeprazole b.i.d., clarithromycin 500 mg b.i.d., tinidazole 500 mg b.i.d). 21 patients were randomized to receive *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926, $5x10^9$ cfu/sachet, b.i.d.), 21 patients to placebo. For the assessment of side-effects a questionnaire developed by de Boer *et al*. (1996) was used. This questionnaire was proposed as a standard side-effect scoring instrument for exploring *H. pylori* treatment regimens. The questionnaire includes the following items: loss of appetite, nausea, vomiting, taste disturbance, dizziness, stomach pain, diarrhoea during treatment, diarrhoea after treatment, headache, rash, others). During the first week of treatment the incidence of diarrhoea was 5% in the *S.boulardii* group and 30% in the placebo group (p=0.018). As compared to the placebo group, during week 2 incidence of diarrhoea remained lower in the *Saccharomyces boulardii* group, however, with borderline significance. Taste disturbance, too, was observed significantly less frequently in the *Saccharomyces boulardii* group (5% vs. 40%, p=0.0027). No major adverse events were reported.

In a multicentre open clinical trial Duman *et al.* (2005) enrolled 389 patients with peptic ulcer disease or non-ulcer dyspepsia for *H. pylori* eradication therapy with clarithromycin (500 mg b.i.d.), amoxicillin (1000 mg b.i.d.), and omeprazole (20 mg b.i.d.) for 14 days. These patients were then randomized to *Saccharomyces boulardii* (500 mg b.i.d.) (n=204) or no treatment (n=185). The aim of the study was to assess the efficacy and safety of *Saccharomyces boulardii* in the prevention of antibiotic-associated diarrhoea due to *H. pylori* eradication. Diarrhoea was defined as a change in bowel habits with at least 3 semi-solid or watery bowel movements per day for at least two consecutive days. The patients were observed during the entire treatment period and the following 4 weeks. The incidence of diarrhoea during the overall study period was 6.9% in the treatment and 15.6% in the control group (p=0.007). In one patient treatment with *Saccharomyces boulardii* was discontinued because of a skin reaction. Although the validity of this study is limited due to its open design it indicates that *Saccharomyces boulardii* may be effective in preventing antibiotic associated diarrhoea in patients receiving triple treatment for eradication of *H. pylori* infection.

In a double-blind clinical study Cindoruk *et al.* (2007) randomized 124 patients with *H. pylori* infection receiving 2 weeks of triple therapy (clarithromycin 500 mg b.i.d, amoxicillin 1000 mg b.i.d, lansoprazole 30 mg b.i.d) to *Saccharomyces boulardii* (n=62) at a dosage of 500 mg b.i.d. (*Saccharomyces cerevisiae* CBS 5926) or placebo (n=62). Side-effects of treatment were recorded by the questionnaire according to de Boer *et al* (1996). This questionnaire was filled out by the patients during the 2 weeks of treatment and the following 2 weeks. During the overall study period nine patients (14.5%) in the treatment group and 19 (30.6%) patients of the control group experienced diarrhoea (p<0.05). Statistically significant differences between the groups were also detected with

regard to epigastric discomfort: 9 (14.5%) patients in the *Saccharomyces boulardii* versus 27 (43.5%) patients in the control group experienced epigastric discomfort ($p<0.01$). The incidence of diarrhoea during the treatment phase was 11.2% in the *Saccharomyces boulardii* group and 25.8% in the control group, during follow-up the respective incidences were 3.2% and 4.8%. The overall assessment of tolerability on a 5-point scale was significantly superior in the treatment group ($p<0.001$). No major side-effects were observed.

Song *et al* (2010) evaluated the additive effects of *Saccharomyces boulardii* and *S.boulardii*/mucoprotective agent to proton pump inhibitor (PPI)-based triple therapy alone. 991 patients with *H. pylori* infections were randomized to one of the 3 groups. The first group received triple therapy alone which consisted of 20 mg omeprazole, 1000 mg amoxicillin, and 500 mg clarithromycin, twice a day for 7 weeks (n=331). In addition to triple therapy in the second group one capsule with *Saccharomyces boulardii* ($3x10^{10}$ cfu/g, 250 mg *Saccharomyces cerevisiae* CBS 5926) was given three times a day for 4 weeks (n=330). The third group receiving also a mucoprotective agent is not considered here. During the study period patients had to note the side-effects into a diary. Diarrhoea occurred more frequently in the group with triple therapy alone (6%) than in the group with the addition of *Saccharomyces boulardii* (3.3%). The frequency of overall side-effects in the *Saccharomyces boulardii* group was statistically significantly lower in the *Saccharomyces boulardii* group (14.5% vs. 19%; $p<0.05$).

Another randomized clinical trial in order to investigate the efficacy and safety of adding the probiotic *Saccharomyces boulardii* to standard triple therapy for eradication of *H. pylori* was performed by Zojaji *et al.* (2013). 80 patients were randomized to treatment with amoxicillin (1000 mg, b.i.d), clarithromycin (500 mg, b.i.d.), omeprazole (20 mg, b.i.d), and *Saccharomyces boulardii* (250 mg, b.i.d., *Saccharomyces cerevisiae* CBS 5926) for 14 days. 80 patients received triple therapy alone. Patients were asked to report any side effects of therapy during the treatment period (end of first, second, third and fourth weeks of treatment) and were given a possible side effect list, such as epigastric pain, diarrhoea, taste disturbance, constipation, and stomatitis. The frequency of side effects as nausea, diarrhoea, abdominal discomfort and bloating in the *Saccharomyces boulardii* group A, were significantly lower than group B in first and second weeks ($p<0.05$), Remarkable side-effects of treatment with *Saccharomyces boulardii* were not noted.

Kyriakos *et al.* (2013) investigated, if *Saccharomyces boulardii* enhances the efficacy of classic triple therapy in eradicating *H. pylori*. 70 patients with peptic ulcer or functional dyspepsia according to Rome III criteria and *H. pylori* infection were treated with omeprazole 20 mg b.i.d., clarithromycin 500 mg b.i.d., and amoxicillin 1 g b.i.d. for 14 days. A total of 36 out of 70 (51%) patients were randomized to *Saccharomyces boulardii*(*Saccharomyces cerevisiae* CBS 5926) , two capsules t.i.d. for 14 days (group A) and 34 (49%) on no intervention (group B). Seven patients in group B (20.6%) and 1 patient in group A (2.8%) stopped treatment because of diarrhoea (95% CI 3.3% to 32.7%, P=0.026).

In another study by Chu *et al*. (2012) on the efficacy and safety of *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926 250 mg b.i.d. for 14 days) in combination with proton pump inhibitor (PPI)-based triple therapy (omeprazole 20 mg b.i.d., amoxicillin 1 g b.i.d, clarithromycin 0,5 g b.i.d. for 14 days) for *H. pylori* related peptic ulcer a significant difference in the rate of adverse effects were observed. 100 patients were randomly assigned to treatment with standard triple therapy alone (n=50) or standard triple therapy plus *Saccharomyces boulardii* (n=50). The most common side-effects were mild to moderate and self-limiting (e.g. nausea, vomiting, diarrhoea, melena, dizziness). The incidence of side-effects in the group receiving *Saccharomyces boulardii* was statistically significantly lower than in the control group (15.6% vs. 57.8%; $p<0.01$). As in this clinical study the incidence of the diarrhoea was not stated explicitly, the study was not included in Table 8.

The effect of *Saccharomyces boulardii* as an adjuvant to a 14-day triple therapy (clarithromycin 500 mg b.i.d., amoxicillin 1000 mg b.i.d, lansoprazole 30 mg b.i.d) for eradication of *H. pylori* has been investigated by Lee *et al* (2011) and published as an abstract. 223 patients infected by *H. pylori* were randomized to 14 days of triple therapy alone (n=116, control group) or triple therapy in combination with *Saccharomyces boulardii* (n=107, treatment group). Side-effects of treatment were assessed by means of a validated questionnaire for 2 weeks from the start of therapy. The incidence of diarrhoea was 29.9% in the treatment group and 43.1% in the control group. This difference was statistically significant (p=0.036). Two patients from the treatment goup and 5 patients from the control group stopped treatment because of adverse events. The validity of this clinical study is also limited due to its open design. The dosage of *Saccharomyces boulardii* is not mentioned in the abstract.

In 2010 a meta-analysis on randomized controlled trials was performed by Szajewska *et al*. in order to investigate the effects of *Saccharomyces boulardii* as supplementation to standard triple therapy on *H. pylori* eradication rates and therapy-associated side effects. Five clinical studies with 1307 patients in total were included: 4 studies were performed in adults (Cindoruk *et al.* 2007, Cremonini *et al.* 2002a, Duman *et al.* 2005, Song et al. (2010), one study included children (Hurduc *et al*. 2009; see: prevention of AAD in children). Szajewska *et al*. (2010) observed a statistically lower risk of therapy-related diarrhoea in patients receiving concomitant treatment with *Saccharomyces boulardii*. Treatment with *Saccharomyces boulardii* compared with placebo reduced the risk of AAD from 17.2% to 6.7% (RR 0.47, 95% CI 0.32–0.69, NNT 16, 95% CI 11–30). Due to the majority of patients being adults, the authors stated that their results may be applicable only to this population. The daily doses of *Saccharomyces boulardii* ranged from 500 mg to 1000 mg. The authors conclude that in patients with *H. pylori* infection, there is evidence to recommend the use of *Saccharomyces boulardii* along with standard triple therapy as an option for decreasing overall therapy-related side effects, particularly diarrhoea.

In 2015 Szajewska *et al.* updated their meta-analysis on supplementation of triple therapy with *Saccharomyces boulardii*, as since then more randomized clinical studies have been published in this indication: Zhao *et al.* (2014), Zojaji *et al.* (2013), Kyriakos *et al.* (2013), Chu *et al.* (2012), Gao *et al.* (2012), Lee *et al.* (2011). The studies by Chu *et al*. (2012) and Gao *et al.* (2012) were published in Chinese and have been translated in order to allow an assessment by the authors.

In this assessment report, these two studies are not included, since a translation is not available. In the meta-analysis of Szajewska *et al.* (2015) a total of 2000 patients were included, of them 330 children with an age range from 3-18 years. As compared to triple treatment alone, the concomitant administration of *Saccharomyces boulardii* reduced the risk of overall adverse effects, especially diarrrhoea (RR 0.51, 95% CI 0.42-0.62; high quality evidence). None of the trials reported on adverse effects other than those related to eradication therapy. As the majority of patients included in this meta-analysis were adults, more studies are needed in children.

Table 8: Clinical studies on the prevention of acute antibiotic-associated diarrhoea (AAD) with *Saccharomyces boulardii* in adults caused by triple therapy of *Helicobacter pylori* infection

Type (aim) and objective(s) of Study Reference	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen; Route of Administration Duration of** **Treatment**	**Number of Subjects (including age, sex, drop out)**	**Healthy Subjects or Diagnosis of Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI** **95%)** **Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Prevention of AAD due to triple therapy of H. pylori infection Cremonini *et al*., 2002a	placebo-controlled, triple-blind, randomized	*Saccharomyces cerevisiae* CBS 5926, 5 x 10^9 cfu b.i.d. during the antibiotic week and 1 week thereafter observation period: 4 weeks	n=97 verum: n=22 placebo: n=21 age: 18-61 yrs male: n=43 female: n=54	symptom-free, *H. pylori* positive patients receiving triple antibiotic treatment (rabeprazole/clarithromycin/tinidazole) for 7 days	incidence of diarrhoea during the first week verum: 5% placebo: 30%	chi-square test: statistical significance (p=0.018)	small study groups

Prevention of AAD due to triple therapy of H. pylori infection Duman *et al*., 2005	open, randomized, *Saccharomyces boulardii* vs. no treatment, multicenter	*Saccharomyces cerevisiae* CBS 59262 x 500 mg/d for 2 weeks follow-up: 4 weeks	n=389 verum: n= 204 control: n= 185 verum: male: 50% mean age: 45.7 yrs control: male: 47.5% mean age: 44.7 yrs	patients with peptic ulcer or non-ulcer dyspepsia receiving triple antibiotic therapy (clarithromycin/amoxicillin/ omeprazole) for *H. pylori* for 2 weeks	incidence of diarrhoea (≥3 semi-solid or watery stools/d on 2 consecutive days) verum: 6.9% control: 15.6%	chi-square test: statistical significance p=0.007	due to open study design only limited validity
Prevention of AAD due to trrriple therapy of H. pylori infection Cindoruk *et al*., 2007	placebo-controlled, randomized, double-blind	*Saccharomyces boulardii* 2 x 500 mg/d for 2 weeks follow-up: 6 weeks after the end of treatment	n=124 verum: n=62 male: 41.9% mean age: 45.8 yrs placebo: n=62 male: 29% mean age: 47.6 yrs	patients receiving 2 weeks of triple therapy (clarithromycin, amoxicillin, lansoprazole) for *H. pylori* eradication	prevention of side-effects related to eradication therapy; frequency of diarrhoea verum: 11.2% (treatment phase) 3.2% (follow-up) 14.5% (overall) placebo: 25.8% (treatment phase) 4.8% (follow-up) 30.6% (overall)	chi-square test: statistical significance	*Saccharomyces boulardii* improved treatment tolerability

Prevention of AAD due to triple therapy of H. pylori infection Song *et al*., 2010	randomized, controlled	*Saccharomyces cerevisiae* CBS 5926 3 x 10^9 cfu /250 mg ? 750 mg/d for 4 weeks follow-up: 5-8 weeks	group + *Saccharomyces boulardii*: n=330 mean age: 49.8 yrs control group: n=331 mean age: 49.8 yrs	patients with *H. pylori* infection receiving proton pump inhibitor based triple treatment (omeprazole/amoxicillin/clarithromycin) for 7 days	incidence of diarrhoea group + *Saccharomyces boulardii*: 3.3% control group: 6.0%	statistically significant difference (p<0.05)	no double-blind study design
Prevention of AAD due to triple therapy of H. pylori infection Zojaji *et al*., 2013	randomized, controlled,	*Saccharomyces boulardii* (250 mg b.i.d) for 14 days group A: triple therapy + *Saccharomyces boulardii* Group B: triple therapy alone observation period: 4 weeks	n=160 mean age: 47.1 yrs female: 58.7% n=80 in each group	adult patients (>15 yrs of age) with *H. pylori* infection receiving triple treatment with amoxicillin/clarithromycin/ omeprazole for 14 days	occurrence of diarrhoea group A: week 1/ 2/ 3/ 4 13% / 12.5% / 6.3% / 1.3% group B: 30% / 26.3% / 11.3% / 5%	non-parametric t-test, chi-square test statistically significant difference in favor of *Saccharomyces boulardii* in week 1 and 2	no double-blind study design

Prevention of AAD due to triple therapy of H. pylori infection Kyriakos *et al*., 2013	prospective, randomized, controlled, single-center	group A: triple therapy plus *Saccharomyces boulardii* for 14 days group B: triple therapy alone triple therapy: Clarithromycin/amoxicillin/omeprazole	n=70 group A: n=36 mean age: 47 yrs group B: n=34 mean age: 45 yrs	adult patients (18-75 yrs) with *H. pylori* related peptic ulcer disease or functional dyspepsia during a 3-yr period	withdrawals because of the occurrence of diarrhoea: group A: 2.8% group B: 20.6%	statistical significance p=0.026	no double-blind clinical trial
Prevention of AAD due to triple therapy of H. pylori infection Lee *et al*., 2011	randomized, controlled	control group: triple therapy for 14 days treatment group: triple therapy for 14 days + *Saccharomyces boulardii*	control group: n=116 treatment group: n=107	223 patients infected with *H. pylori*	side-effects of treatment, tolerability by means of a validated questionnaire for 2 weeks from the start of the study frequency of diarrhoea: treatment group: 29.9% control group: 43.1%	statistical significance p= 0.041	dosage of *Saccharomyces boulardii* not mentioned; open study design; abstract only

Prevention of recurrence of *Clostridium difficile* disease (CDD)

According to Bartlett (2002) about 10 to 20% of the cases of AAD are caused by infections with *Clostridium difficile*. Most of the cases of colitis due to antibiotic therapy, however, are associated with *C. difficile*. Antibiotics which are most commonly implicated with CDD are clindamycin, cephalosporins, and penicillins. Clinical symptoms of CDD may range from uncomplicated diarrhoea to pseudomembranous colitis (PMC). For the standard treatment of CDD the antibiotics vancomycin and metronidazole are used. Although this medication is effective in 80% of the patients with CDD, in 20% further episodes of diarrhoea or colitis occur within 3 to 28 days after the end of antibiotic treatment (Mc Farland *et al*., 1994).

Several clinical studies have been performed in this indication with *Saccharomyces boulardii* given as an adjunct to standard treatment. In 1989b Surawicz *et al*. published the results of an open trial which investigated the efficacy of *Saccharomyces boulardii* for the treatment of recurrences of *C. difficile* associated colitis in humans. Thirteen patients with recurring *C. difficile* cytotoxin-positive diarrhoea were treated with 10 days of vancomycin and a 30-day course of *Saccharomyces boulardii* (500 mg b.i.d). Eleven (85%) had no further recurrences: The authors concluded that *Saccharomyces boulardii* may have a role in treating recurrent *C. difficile* diarrhoea and colitis. Due to its open design, the study is not included in the following table.

According to McFarland (2010) two randomized, double-blind clinical studies have been performed with *Saccharomyces boulardii* for the treatment of CDD: Both trials included patients who suffered from diarrhoea and had a positive culture for *C. difficile*. While McFarland *et al.* (1994) observed a statistically significant reduction of relapses with *Saccharomyces boulardii* only in the subgroup of patients with recurrent disease (34.6% vs. 64.7%; p=0.04), Surawicz *et al*. (2000) reported that only in the subgroup of patients receiving treatment with high dose vancomycin and *Saccharomyces boulardii* relapses of CDD tended to decrease. Statistically significant differences were not demonstrated in this study.
Both clinical trials have also been assessed in a review by Tung *et al*. (2009). The authors conclude that *Saccharomyces boulardii* may be effective for secondary prevention of CDD in some specific patient populations with particular concurrent antibiotic treatment.
Dendukuri *et al*. (2005) included the randomized clinical studies listed here in their review on probiotic therapy for the prevention and treatment of *C. difficile*-associated diarrhoea.
They calculated the following risk difference (95% CI) (placebo-probiotic):

McFarland *et al.*, 1994:		20.5 (2.2 to 37.0)
	First time CDD:	4.9 (-17.6 to 26.4)
	Recurrent CDD:	30.0 (2.3 to 50.6)
Surawicz *et al*., 2000:		9.8 (-6.7 to 25.6)
	High dose vancomycin:	33.0 (-0.3 to 62.0)
	Low-dose vancomycin:	-6.4 (-16.2 to 28.1)
	Metronidazole:	-1.9 (-25.8 to 29.2).

Only McFarland *et al.*(1994) reported a significant beneficial effect overall. In a post hoc analysis, the authors found that this effect was almost entirely limited to the subgroup with recurrent CDD.

According to Dendukuri *et al*. (2005) the studies conducted to date provide insufficient evidence for the routine clinical use of probiotics to prevent or treat CDD in adults. Better designed and larger studies are needed.

In their Guidelines for Clostridium difficile infections (CDI) in adults Cohen *et al*. (2010) assess the use of probiotics for the primary prevention of CDD as follows:
"Administration of currently available probiotics is not recommended to prevent primary CDI, as there are limited data to support this approach and there is a potential risk of bloodstream infection."

Flatley *et al.* (2015) performed a retrospective chart review to evaluate the effects of the removal of *Saccharomyces boulardii* from an automatic antibiotic order set and hospital formulary on hospital onset *C. difficile* infections (CDI) rates. At an USA hospital, an increase in the incidence of CDI in 2006 had prompted the following changes: 1) more stringent cleaning procedures; 2) CDI positive patients were isolated and placed in contact precautions and 3) implementation of a protocol that linked an order to initiate *Saccharomyces boulardii* therapy (250 mg b.i.d.) to patients receiving antibiotics highly associated with causing CDI like intravenous formulation of clindamycin, cefepime, ceftazidime, ceftriaxone, cefuroxime, and fluoroquinolones. In 2009, the hospital re-evaluated its CDI prevention strategies and *Saccharomyces boulardii* was removed. In their retrospective analysis, Flatley et al. compared a control group admitted from November, 2008 through November, 2009 to the study group admitted from January, 2010, through January, 2011. The primary outcome was the incidence rates of hospital onset CDI (hCDI) in all hospitalized patients during the control and study group. There were 167,157 hospital patient days with 167 hCDI cases in the control group and 183,867 hospital patient days with 191 hCDI cases in the study group without statistically significant difference in the incidence of hCDI between the two groups. Receipt of linked antibiotics was similar in patients acquiring hCDI. The secondary outcome was the incidence of hCDI among patients who received a linked antibiotic. In total, 8708 patients in the control group and 8411 patients in the study group were treated with linked antibiotics. 109 (1.25%) and 127 (1.51%) patients acquired hCDI in the control and study group, respectively (p=0.698). The authors concluded that administering *Saccharomyces boulardii* with intravenous broad spectrum antibiotics as prophylaxis was not effective for preventing hospital onset CDI.

Table 9: Clinical studies on the prevention of recurrence of CDD

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** **e.g. Jadad score**	**Comments on clinical relevance of results**
Prevention of recurrent CDD McFarland *et al.,* 1994	placebo-controlled, double-blind, randomized, parallel-group	2 x 2 capsules with 250 mg *Saccharomyces boulardii*; according to 3 x 10^{10} cfu per day or placebo in addition to a standard antibiotic therapy duration of therapy: 4 weeks follow-up: 4 weeks	124 adults 57 *Saccharomyces boulardii* mean age: 56.8 years ± 20.4 67 placebo mean age: 59.2 ± 21.1 9 drop outs during treatment, 5	active CDD (initial CDD and/or recurrent CDD) ranging from uncomplicated diarrhoea to pseudomembranous colitis (PMC) who were receiving one of two oral standard treatments (vancomycin or metronidazole) at the time of enrollment	efficacy calculated from the formula ($[I_P - I_T]/I_P$) x 100, I_P=incidence of CDD recurrence in placebo group, I_T=incidence of CDD recurrence in *Saccharomyces boulardii* group relative risks (RRs) calculated from cumulative incidence ratios, and two-tailed 95% test-based confidence interval (CIs) that excluded 1 were defined as significant	ITT-basis, Student's t test, Mann-Whitney U test, chi-square test, Fisher's Exact test, Kaplan-Meier method, mantel Log-Rank Test Jadad Score 5	it is not clear, if the use of multivariate logistic regression model to control for confounding factors was predefined, otherwise the primary outcome was not significant

Type (aim) and objective(s) of Study Reference	Study Design and Type of Control Study duration (if available)	Test Product(s): herbal preparation, pharmaceutical form; Dosage Regimen; Route of Administration Duration of treatment	Number of Subjects (including age, sex, drop out)	Healthy Subjects or Diagnosis of Patients (inclusion criteria)	Outcomes (primary and secondary endpoints)	Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score e.g. Jadad score	Comments on clinical relevance of results
			drop outs during follow up 5 patients died (1 pneumonia under *Saccharomyces boulardii*, 4 under placebo) 1 patient refused participation because of a side effect (rash due to vancomycin)		unadjusted RRs of *Saccharomyces boulardii* compared to placebo: 0.47; 95% CI, 0.22 to 1.00 using a multivariate logistic regression model to control for confounding factors (predefined?) the RRs of *Saccharomyces boulardii* compared to placebo was significantly reduced: 0.43; 95% CI, 0.20 to 0.97 overall: recurrence rate: placebo: 44.8 %	(Dendukuri, 2005)	in addition efficacy could only be confirmed in the subgroup with recurrent CDD and not in the subgroup with initial CDD neither the dose nor the duration of antibiotic

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** **e.g. Jadad score**	**Comments on clinical relevance of results**
					Saccharomyces boulardii: 26.3% Initial CDD: Recurrence rate: placebo: 24.2% *Saccharomyces boulardii*:19.3% recurrent CDD: recurrence rate: placebo: 64.7% *Saccharomyces boulardii*: 34.6%		was controlled
Prevention of recurrent CDD	placebo-controlled, double-	2 x 2 capsules with 250 mg *Saccharomyces*	168 adults (32 patients on high-dose	active diarrhoea (change in bowel habits with ≥3	diarrhoea cessation was defined as a return to normal	Student *t* test, Mann-Whitney	patients were treated for

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** **e.g. Jadad score**	**Comments on clinical relevance of results**
Surawicz *et al*., 2000	blind, explorative study	*boulardii* or placebo in addition to a 10-day course of a standard antibiotic therapy from day 7 duration of therapy: 4 weeks	vancomycin, 83 patients on low-dose vancomycin, 53 patients on metronidazole)	loose or watery stools per day for ≥2 consecutive days, or >8 loose stools within 48 h) before standard antibiotic treatment, positive *C. difficile* assay, ≥1 recent, prior episode of CDD within 1 year	bowel frequency (<3 loose or watery stools per day) for at least 48 h neither the 10-day course of lower dose of vancomycin nor metronidazole given with either *Saccharomyces boulardii* or placebo was significantly effective 3 (16.7%) of the 18 patients receiving high-dose vancomycin and *Saccharomyces boulardii* had a	ranked sum test, chi-square test, Fisher's Exact test. two-tailed tests of significance were used for all tests at level of p≤0.05 Jadad Score 3 (Dendukuri, 2005)	CDD according to the preference of their private physicians and not per randomizationand were then referred to the authors of the study only one subgroup is presented without indicating if

Type (aim) and objective(s) of Study Reference	Study Design and Type of Control Study duration (if available)	Test Product(s): herbal preparation, pharmaceutical form; Dosage Regimen; Route of Administration Duration of treatment	Number of Subjects (including age, sex, drop out)	Healthy Subjects or Diagnosis of Patients (inclusion criteria)	Outcomes (primary and secondary endpoints)	Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score e.g. Jadad score	Comments on clinical relevance of results
					recurrence of CDD, compared with 7 (50%) of 14 patients receiving high-dose vancomycin and placebo (p=0.05, by the Fisher exact test)		this design was predefined in the study protocol

Prevention of diarrhoea associated with tube-feeding

The occurrence of diarrhoea during tube feeding is a common complication. Possible causes are e.g. alteration of bacterial flora, hypoalbuminemia, infusion rate, or concomitant medication.
A list of randomized controlled clinical studies which investigated the anti-diarrhoea efficacy of *Saccharomyces boulardii* in tube-fed patients has been published by McFarland (2010).

One of the causes of diarrhoea in tube-fed patients may be the consequences on colonic trophicity of a deficiency in luminal short-chain fatty acids. Schneider *et al*. (2005) investigated the effects of *Saccharomyces boulardii* (0.5 g b.i.d for 6 days) on fecal flora and short-chain fatty acids (SCFA) in 10 patients on long-term total enteral nutrition. As compared to 15 healthy volunteers treatment with *Saccharomyces boulardii* increased total fecal SCFA levels – especially butyrate - which remained high even 9 days after the discontinuation of treatment. According to the authors, *Saccharomyces boulardii* induced increase of fecal SCFA concentrations may explain its preventive effects on diarrhoea induced by total enteral nutrition (see also 4.1.1).

Clinical experience with the use of *Saccharomyces boulardii* in prevention of nutrition-related diarrhoea is very limited. Only a low number of patients has been included in clinical studies. The dosages applied ranged from 1 g/d (Bleichner *et al*., 1997) to 2 g/day (Tempé *et al*., 1983, Schlotterer *et al*., 1987).

Table 10: Randomized controlled clinical studies for the prevention of diarrhoea in tube-fed patients

Type (aim) and objective(s) of Study Reference	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen; Route of Administration Duration of** **treatment**	**Number of Subjects (including age, sex, drop out)**	**Healthy Subjects or Diagnosis of Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI** **95%)** **Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Prevention of diarrhoea in tube-fed patients Tempé *et al*., 1983	randomized, double-blind	*Saccharomyces cerevisiae* CBS 5926 dosage: 1 x 10^{10}/d treatment duration: 11-21 d	n=40 placebo: n=20 mean age: 65.5 yrs *Saccharomyces boulardii*: n=20 mean age: 67.8 yrs	intensive care patients on enteral feeding for at least 10 d	days of diarrhoea: *Saccharomyces boulardii:* 34/389 d (8.7%) placebo: 63/373 d (16.9%)	p<0.001	small study population
Prevention of diarrhoea in tube-fed patients Schlotterer *et al*., 1987	randomized, double-blind	*Saccharomyces cerevisiae* CBS 59262 g/d vs. placebo	n=18 *Saccharomyces boulardii*: n=9 mean age: 32.9 yrs placebo: n=9 mean age: 43.4 yrs	patients 18 – 70 yrs with burns (18-70% of body surface) with enteral nutrition for ≥8-28 d	days of diarrhoea *Saccharomyces boulardii*: 3/204 d (1.5%) placebo: 19/208 d (9.1%)	p<0.001	small and special study population, higher posology than usually recommended for medicinal products in the European Union

Prevention of diarrhoea in tube-fed patients Bleichner *et al*., 1997	randomized, double-blind, placebo-controlled, multicenter	*Saccharomyces boulardii* 4 x 500 mg/d (no further information) vs. placebo	n=128 *Saccharomyces boulardii*: n=64 mean age: 61.6 yrs male: 45% placebo: n=64 mean age: 64.9 yrs male: 46%	critically ill patients with need for enteral nutrition for >6 d	% of days with diarrhoea per feeding days: 1) diarrhoea was defined according to a diarrhoea score based on volume and consistency of each stool: *Saccharomyces boulardii*: 14.2% placebo: 18.9% 2) diarrhoea was defined as three or more nonformed stools per day *Saccharomyces boulardii*: 7.7% placebo: 12.7%	p=0.0069 p<0.01	higher posology than usually recommended for medicinal products in the European Union

Irritable bowel syndrome

Irritabel bowel syndrome (IBS) is a functional gastrointestinal disorder in which abdominal discomfort or pain is associated with changes in bowel habits, stool consistency and other features of disordered defectation. IBS is considered to be one of the most frequent clinical problems in gastroenterology with an estimated prevalence in the Western world of up to 20% (CHMP 2014). In 2014 a guideline has been published by the CHMP on the evaluation of medicinal products for the treatment of IBS (CHMP 2014). Currently, the Rome III criteria, which replaced the Rome II criteria, are regarded to be the standard diagnostic criteria:
"Recurrent abdominal pain or discomfort at least 3 days per month in the last 3 months (with symptoms being present for the last three months and onset at least 6 months prior to diagnosis) associated with 2 or more of the following

- Improvement with defaecation
- Onset associated with a change in frequency of stool
- Onset associated with a change in form (appearance of stool)

Sub-typing of IBS patients is performed by the predominant stool pattern present in a patient. Short-term treatment intermittent use of compounds should be evaluated in repeated treatment courses shorter than 8 weeks. Long-term continuous treatment should be evaluated in studies with a duration of at least 6 month (CHMP 2014).

In 1983 Maupas *et al*. investigated, if *Saccharomyces boulardii* is effective in the treatment of IBS in a randomized, double-blind clinical study. 34 patients (*Saccharomyces boulardii*: n=16; Placebo: n=18) were treated with *Saccharomyces boulardii* (3 x 1 capsule/day) or placebo over a period of one months. The statistical evaluation showed that *Saccharomyces boulardii* was statistically significantly superior to placebo with respect to the opinion of the physician and patient, number and consistency of stools. Regarding the clinical symptoms pain, distension, dyspepsia, however, no statistically significant differences were observed between the groups. As this clinical study has already been performed in 1983 the current guidelines by the CHMP on the evaluation of medicinal products for the treatment of IBS have not been considered.

The clinical study performed by Maupas *et al*. (1983) was the only study performed with *Saccharomyces boulardii*, which McFarland and Dublin (2008) included in her meta-analysis of probiotics for the treatment of IBS. She concluded "*in summary, the present meta-analysis suggests that probiotics offer promise for the treatment of IBS. Results should be interpreted cautiously given the methodological limitations of published studies. Future studies are needed, in particular larger studies of longer duration with greater methodological rigor. In addition, more data are needed regarding which specific strains and doses are most likely to be effective. The use of probiotics for IBS warrants further study, particularly given the chronic nature of this condition, its major impact on patients' quality of life, and the dearth of other effective treatments.*"

Since then, more clinical studies investigating the effects of *Saccharomyces boulardii* in the treatment of IBS have been reported. Choi *et al*. (2011) evaluated the effects of *Saccharomyces boulardii* on quality of life and symptoms in 90 patients with diarrhoea-predominant or mixed-type IBS in a randomized, double-blind, placebo-controlled multicenter trial. Diagnosis of IBS was based on Rome II Criteria and in both groups about 72% of the patients suffered from diarrhoea-predominant IBS. Although the results of this study indicated a statistically significant higher improvement of quality of life in the patients treated with *Saccharomyces boulardii* (15.4% vs. 7.0%; $p<0.05$), IBS symptoms were reduced to a similar extent in both groups. Bowel frequency and stool consistency did not change in either group. The drop-out rate was high in both groups.
The studies performed by Maupas *et al*. (1983) and Choi *et al*. (2011) were included by Korpela and Niittynen (2012) in a review on the effects of probiotics on the gastrointestinal symptoms of IBS. The

authors concluded that general recommendations on the use of probiotics in IBS cannot be given at that time. Further clinical trials and data in the mechanism of action are required.

In 2011 the results of another randomized, double-blind, placebo-controlled clinical study with *Saccharomyces boulardii* in 70 patients with diarrhoea predominant IBS were published by Kabir *et al*. No significant difference between the two groups was found in any of the parameters evaluated (number of stools, consistency of stools, abdominal pain, abdominal distension, personal life and professional life) on any of the observation days (0, 30 and 60). *Saccharomyces boulardii* treatment for 30 days in diarrhoea predominant IBS patients did not result in any improvement.

Bafutto *et al*. (2013) evaluated the effects of mesalazine and *S.boulardii* given alone or in combination for the treatment of patients with diarrhoea-predominant IBS. A pilot study was performed in 53 patients, 12 patients received *Saccharomyces boulardii* alone. The results of the study showed a statistically significant improvement of symptom scores in all treatment groups. The improvement of the symptom score was greater with mesalazine alone or in combination with *Saccharomyces boulardii* as compared with *Saccharomyces boulardii* alone. Due to the small number of patients, the short study duration (30 days) and the pilot character of the study with lack of randomization and blinding the results obtained in this study do not support a WEU of *Saccharomyces boulardii* for the indication IBS.

Akhondi-Meybodi *et al.* (2014), too, studied the effect of *Saccharomyces boulardii* on the treatment of IBS in a randomized double-blind clinical in 60 patients. IBS was diagnosed by a gastroenterologist based on clinical symptoms, laboratory tests, and medical examinations such as colonoscopy, colon biopsy, sonography and stool culture. 30 patients were treated with *Saccharomyces boulardii* (1 capsule/d, *Saccharomyces cerevisiae* CBS 5926, at least 1x 10^{10} living cells/g), 30 patients received placebo for an interval of 3 weeks. The aim of the study was to investigate the effect of *Saccharomyces boulardii* on improving symptoms of IBS (abdominal pain, flatulence, urgent defecation, diarrhoea, obstipation, gurgling, eructation, gas release from the anus). Severity of symptoms was assessed by the patients by means of a Likert scale at the beginning and end of treatment. At the end of treatment statistically significant differences between the groups favouring *Saccharomyces boulardii* were found for pain severity (p=0.008), diarrhoea (p=0.001), and gurgling (p=0.317). The validity of this study, however, is limited due to the small number of patients included and the short treatment period. Furthermore, there are doubts with regard to the blinding procedure, since according to the authors placebo capsules were only "similar" to verum capsules "except that they did not contain *Saccharomyces* powder and had no coating". Another important aspect is that apart from abdominal pain symptoms of IBS which are essential for making the diagnosis IBS (e.g. improvement with defecation, frequency and consistency of stool) have not been included as primary endpoints in the statistical evaluation.

Pineton de Chambrun *et al*. (2015) performed a randomized clinical trial of *Saccharomyces cerevisiae* (n=86) versus placebo (n=93) in 179 patients with irritable bowel syndrome (Rome III criteria). The patients in the treatment group received *Saccharomyces cerevisiae* (CNCM I-3856) 500 mg once daily over a period of 8 weeks and as result a reduction of abdominal pain/discomfort scores without altering stool frequency and consistency was observed. Since genetic identity, however, was not proven for the strain of *Saccharomyces cerevisiae* administered in this clinical study the results cannot be transferred to the *Saccharomyces cerevisiae* CBS 5926 assessed here. Therefore, the clinical study was not included in the table below.

Table 11: Clinical studies in the indication irritable bowel syndrome (IBS)

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of IBS Maupas *et al*., 1983	double-blind, Randomized, three centers	3 x 1 cps/day (*Saccharomyces cerevisiae CBS 5926*) vs. placebo duration: 1 month	verum: n=16 placebo: n=18 sex: 20 male, 14 female mean age: 42 yrs	patients >18 yrs of age with IBS (abdominal pain, distension, episodes of diarrhoea)	statistically significant differences ($p<0.05$) with regard to physician's and patient's subjective opinion decrease in number of stools improvement in stool consistency	Student's t-test	Small number of patients, no AE mentioned

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					no statistically significant differences with regard to the symptoms pain, distension, dyspepsia		
Treatment of IBS Choi *et al*., 2011	double-blind, randomized, placebo-controlled, multicenter	*Saccharomyces cerevisiae* CBS 59262 x 10^{11} live cells 2 x 2 capsules/d for 4 weeks vs. placebo	*Saccharomyces boulardii*: n=45 mean age: 40.2 yrs male: 51.4% placebo:	patients (20-65 yrs of age) with diarrhoea-predominant IBS or mixed type IBS (Rome II criteria)	primary efficacy variable: IBS-QOL: statistically significant better improvement with *Saccharomyces boulardii* (15.4% vs. 7.0%) ($p<0.05$)	Student's t-test	IBS symptoms were secondary efficacy variables only One AE in the placebo group:

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			n=45 mean age: 40.6% male: 48.7% drop-outs: *Saccharomyces boulardii*: n=11 placebo: n=12		secondary efficacy variable: scores for IBS symptoms (abdominal pain, discomfort, hard/lumpy stool, loose/watery/stool, straining, urgency, sense of incomplete evacuation, mucus in stool, bloating, passage of gas): total scores were reduced in both		worsening of abdominal pain and flatulence High-rate of drop-outs

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					groups to a similar extent secondary variable: frequency and consistency of stool (Bristol stool scale): no significant change in either group		
Treatment of IBS Kabir *et al*., 2011	double-blind, randomized, placebo-controlled	*Saccharomyces boulardii* 250 mg sachet twice a day vs. placebo	*Saccharomyces boulardii*: n=35	patients with diarrhoea predominant IBS (Rome II criteria) age: 18-50 yrs	single scores for: number of stools, consistency of stools, abdominal pain, abdominal distension,	Student's t-test	Study from Bangladesh.

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		duration: 30 days	mean age: 32.4 yrs male: n=32 placebo: n=35 mean age: 29 yrs male: n= 32		personal life, professional life no difference between the groups in any score at days 0, 30 and 60		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of IBS Bafutto *et al*., 2013	pilot study	*Mesalazine alone* (MG): 800 mg t.i.d. *Mesalazine/S.boulardii* (MSbG): 800 mg t.i.d./200 mg t.i.d. *Saccharomyces boulardii alone* (SbG:): 200 mg t.i.d. duration: 30 days	*MG:* n=20 mean age: 46 yrs *MSbG*: n=21 mean age: 50 yrs *SbG:* n=12	53 patients with diarrhoea-predominant IBS (>18 yrs of age) (Rome III criteria)	symptom evaluation at baseline and after treatment (4-point likert scale): Stool frequency, form and consistency (Bristol Scale), abdominal pain and distension statistically significant improvement of symptom score (pre/post treatment comparison):	difference between baseline and end of treatment: Paired t-test Kruskal-Wallis test	short observation period small number of patients pilot study not randomized or blinded

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			mean age: 43 yrs		*MG*: 10.7 (pre) vs. 5.5. (post) ($p<0.0001$) MSbG: 10.67 (pre) vs. 5.0 (post) ($p<0.0001$) SbG: 9.75 (pre) vs. 7.08 (post) ($p<0.003$)		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					significant differences ($p<0.03$) were seen when comparing MG, MSbG and SbG		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of IBS Akhondi-Meybodi, 2014	double-blind, randomized, placebo-controlled	*Saccharomyces cerevisiae* CBS 5926(at least 1 x 10^{10} living cells/g) 1 capsule/d vs. placebo duration: 3 weeks	*S.boulardii* n=30 male: 43.3% mean age: 37.3 yrs Placebo: n=30 male: 40% mean age: 44.2 yrs	60 patients with IBS as diagnosed by a gastroenterologist	severity score of symptoms (Likert scale) on day 0 and after 3 weeks: abdominal pain, flatulence, urgent defecation, diarrhoea, obstipation, gurgling, eructation, gas release from anus aim of the study: effect of *Saccharomyces boulardii* on	t-test, paired samples t-test	small number of patients short treatment duration doubtful blinding

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					improvement of symptoms statistically significant difference between the groups regarding: pain severity (p=0.008) diarrhoea (p=0.001=) gurgling (p=0.317)		

Prevention of travellers' diarrhoea

In 2007 McFarland published a meta-analysis of randomized clinical studies with various probiotics for the prevention of traveller's diarrhoea. From 12 studies identified in literature, two were performed with *Saccharomyces boulardii*:

Kollaritsch *et al*. (1989) investigated the value of four different non-antibiotic preparations for prophylaxis or treatment of travellers' diarrhoea. All studies were placebo-controlled double blind field trials with Austrian tourists (n=2271) visiting countries with warm climates. Apart from *Saccharomyces* the following preparations were administered: *Lactobacillus acidophilus*, an oral vaccine consisting of heat-inactivated Enterobacteriaceae, a combination product containing carbo medicinalis, Bolus alba, pectin, lactose, whey powder. With regard to *Saccharomyces* healthy Austrian travellers were given either placebo (n=406; mean age: 43.2 years) or a daily dose of 125 mg b.i.d (n=426; mean age: 42.4 years; group I) or 250 mg b.i.d (n=399; mean age 41.5 years; group II) of *Saccharomyces cerevisiae* CBS 5926 containing 5 x 10^9 or 10^{10} revivable cells. Intake started 5 days prior to departure and continued during the whole stay abroad. In addition, the travellers received a questionnaire to record personal data, information of the holidays' conditions and information of diarrhoea.
Episodes of diarrhoea occurred in 42.6% of the placebo group and statistically significantly less frequent in the *Saccharomyces* group: 33.6% (group I; p<0.007) and 31.8% (group II; p<0.002). The incidence of diarrhoea as compared to placebo was reduced by 21.2% in group I and 25.4% in group II. Dose dependency was suggested but not significant in the study. Side-effects were not observed. 9 participants reported improvement of acne.
The clinical course in not preventable cases of diarrhoea was not influenced by this prophylaxis. Regional evaluation of efficacy, however, exhibited evident and statistically significant differences in protective capacity. The reduction of risk amounted to 58, 59 and 40% respectively in northern Africa, western Africa and various tropical islands and thus was more pronounced than the overall risk reduction rate. Due to the differing protection rates according to the destination of travelers, according to the authors, a selective efficacy of *Saccharomyces cerevisiae* has to be taken into account. Only the application of *Saccharomyces cerevisiae* (strain Hansen CBS 5926) decreased the incidence of diarrhoea significantly.

Table 12: Incidence of traveller's diarrhea according to region (from Kollaritsch et al. 1989)

Region	**Group**	**Frequency of diarrhoea**	**Reduction compared to placebo**	**Significant relating to overall reduction in group II**
North Africa (n=208)	placebo (n= 65) group I (n=73) group II (n=70)	50.7% 30.1% 21.4%	- 41% (p<0.01) 58% (p<0.01)	 P<0.0025
West Africa (n=51)	placebo (n= 19) group I (n=18)	52.6% 33.3%	- 37%	

	group II (n=14)	21.4%	59%	P<0.05
Middle East (islands) (n=123)	placebo (n= 45) group I (n=45) group II (n=33)	40.0% 28.9% 24.2%	- 28% (p<0.1) 40% (p<0.05)	P<0.05
East Africa (n=251)	placebo (n= 70) group I (n=98) group II (n=83)	48.6% 35.7% 36.1%	- 27% (p<0.05) 26% (p<0.1)	not significant
South America (n=97)	placebo (n= 38) group I (n=24) group II (n=35)	50.0% 33.3% 37.1%	- 33% 26%	not significant
World tour	placebo (n= 12) group I (n=12) group II (n=10)	50.0% 25.0% 40.0%	- 50% (p<0.1) 20%	not significant
Middle East (n=85)	placebo (n= 21) group I (n=32) group II (n=32)	66.6%* 68.6% 65.6%	- 0 0	not significant
Far East (n=228)	placebo (n= 86) group I (n=72) group II (n=70)	31.4%** 25.9% 30.0%	- 20% 5%	not significant
Central America (n=76)	placebo (n= 32) group I (n=18)	31.3% 38.9% 30.7%	- 0 0	not significant

	group II (n=26)			

* risk of diarrhoea significant higher (p<0.05) than the overall frequency in the placebo group;
** risk of diarrhoea significant smaller (p<0.05) than the overall frequency in the placebo group

The results of another placebo controlled double-blind clinical study were published by Kollaritsch *et al*. in 1993. 3000 Austrian travelers were randomly assigned to prophylactic treatment with *Saccharomyces boulardii* at daily dosages of 250 mg or 1000 mg (*Saccharomyces cerevisiae* CBS 5926) or to placebo. Prophylactic administration started 5 days before departure and was continued for the entire stay abroad. The study participants were asked to fill in questionnaires and the data of 1016 were available for the evaluation of efficacy (placebo: n=361; mean age 45.3 years; *Saccharomyces boulardii* 250 mg: n=352, mean age 43.9 years; *Saccharomyces boulardii* 1000 mg: n=303, mean age 47.7 years). The incidence of diarrhoea was 39.1% in the placebo group and statistically significantly lower during the prophylactic administration of *Saccharomyces boulardii*: 34.4% in the low dose and 28.7% in the high dose group. The success of the prophylactic administration of *Saccharomyces boulardii* (especially for the dosage 1000 mg), however, was shown to depend directly on the rigorous use of the preparation. In this study, too, a varying regional and dose-dependent effect was noted for *Saccharomyces boulardii* which was particularly marked in North Africa and the Near-East.

According to McFarland (2007) *Saccharomyces boulardii* showed significant efficacy for the prevention of traveler's diarrhoea.
In his publication on the therapy for and prevention of traveler's diarrhoea DuPont (2007) also mentions *Saccharomyces boulardii* - together with Lactobacillus GG - as one of the leading candidates for the prophylaxis in travel medicine. Although safe for use in immunocompetent subjects, the probiotic preparations have provided minimal protection against the development of traveler's diarrhoea.

In 2006 the Infectious Diseases Society of America (Hill *et al*., 2006) published guidelines for travel medicine. Regarding the prevention and management of traveller's diarrhoea probiotics are not recommended for use, as they did not demonstrate sufficient efficacy. This assessment was based on investigations with Lactobacillus GG and the clinical trial with *Saccharomyces boulardii* performed by Kollaritsch *et al*. (1993).

Acne

According to the monograph of the German Kommission E, *Saccharomyces boulardii* is also indicated as an adjuvant in the treatment of chronic acne at a daily dosage of 750 mg. The effects of *Saccharomyces boulardii* in acne are ascribed to its antimicrobial mechanism of action (Reuter *et al*., 2010).
Bedi and Shonefelt (2002) mention *Saccharomyces boulardii* for the use in acne in their review on herbal therapy in dermatology.
In an evidence-based review on botanicals in dermatology, Reuter *et al*. (2010) conclude that *Saccharomyces* may have the potential to become standard treatment in acne. They searched published literature for the use of botanicals in dermatological indications with the focus on controlled clinical studies. The level of evidence as suggested by the UK National Health Service was assessed as D (i.e. expert opinions without explicit critical appraisal or based on physiology, bench research, or first principles).

The following clinical studies have been performed in acne:

In 1973, Mandrella reported the treatment results of 166 patients (121 female, 45 male) with acne. The majority of patients (56%) suffered from juvenile acne. Majority of the patients (67%) were ⩽ 25 years old. The initial dose of *Saccharomyces boulardii* was 3 x 100 mg/d which was reduced to 3 x 50 mg/d after 2 weeks (*Saccharomyces cerevisiae* CBS 5926). The treatment period was longer than 6 months in about 80% of the patients. In almost 90% of the patients very good/good results were achieved. There was no case of treatment failure.

Kujath and Sipp (1978) treated 41 soldiers (age 18-25 years) with *Saccharomyces cerevisiae* CBS 5926 for acne vulgaris. During the first 3 days of treatment, the dosage was 3 x 100 mg/d, which was gradually reduced over a period of 2 months to 50 mg/d. After 2-8 months of treatment, results in 80% of the patients were assessed as good / very good. In 7% of the cases, treatment was rated as ineffective. Side-effects were not observed.

In a randomized, controlled double-blind study involving 139 patients with various forms of acne, the effectiveness and tolerance of *Saccharomyces cerevisiae* Hansen CBS 5926 (3 x 250 mg/d) was studied in comparison with placebo over a maximum period of five months (Weber *et al*., 1989). The results of therapy were assessed by the physician as very good/good in 74.3% of the patients receiving the preparation, as compared with 21.7% in the placebo group. In more than 80% of the former patients, the condition was assessed as healed/considerably improved, while the corresponding rate for the placebo group was only 26%. As the severity of acne and therapeutic efficacy were assessed mainly by subjective scores and statistical methods are only roughly described in the original publication, the study does not confirm the therapeutic efficacy of *Saccharomyces boulardii* in acne.

In another double-blind randomized clinical study by Stüttgen (1991). Ninety four patients with acne were treated with daily doses of 3 x 100 mg or 3 x 250 mg *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926). For the assessment of therapeutic efficacy the number of papules within a defined areal (3 x 5 cm), oiliness of the hair, and frequency of hair washing were determined. After 5 months of treatment, the number of papules had decreased by 56.1% (300 mg/d) and 56.5% (750 mg/d). Therapeutic success was rated by the doctors as very good/good in 32.6% resp. 29.1% of the patients. In 37% (300 mg/d) resp. 37.5% (750 mg/d) of the patients therapeutic efficacy was assessed as unsatisfactory. Statistically significant differences between the groups were not detected. As in this clinical study the rate of patients with unsatisfactory treatment effects was greater than the rate of patients with good treatment results, this study does not confirm a therapeutic efficacy of *Saccharomyces boulardii* in acne.

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

Acute unspecific diarrhoea in children

Controlled studies

Chapoy (1985, French publication) assessed lyophilised *Saccharomyces boulardii (Saccharomyces cerevisiae* Hansen CBS 5926 – Chapoy 1986, German publication) (2 x 250 mg/day) in combination with standard oral rehydration therapy for the treatment of acute diarrhoea in 38 infants and toddlers with acute diarrhoea based on an acute gastroenteritis in the age between two weeks and 30 months in a controlled clinical trial in France. After admission to hospital they were applied a special oral rehydration therapy. Normal nutrition was reintroduced in different episodes within about 72 h. Each second child was allocated to the additional treatment of *Saccharomyces cerevisiae* for five days. Control visits were performed on day 1 and 4. A significant reduction in the number of stools ($p<0.01$), weight and consistency of stools ($p<0.05$), and transit time ($p<0.05$) could be confirmed by statistical evaluation of all 19 children in the *Saccharomyces cerevisiae* group. No adverse effect was reported.

Table 13: Results of Chapoy (1986)

Mean ± SD	*Saccharomyces boulardii* group (n=19)	control group (n=19)	significance
stool frequency			
day 1	4.9±0.5	4.0±0.3	
day 4	2.1±0.2	3.4±0.4	
improvement	-2.8±0.5	-0.6±0.6	p<0.01
stool weight (g)			
day 1	283±42	192±25	
day 4	144±28	180±37	
improvement	140±43	-12±42	p<0.05
transit time via carmin red method (h)			
day 1	7.2±1.0	10.4±1.6	
day 4	16±1.6	12.3±1.6	
improvement	8.8±1.6	3.0±1.9	p<0.05
stool consistency after 4 days			
liquid	1	4	
soft	3	8	p<0.05
normal	15	7	

Cetina-Sauri and Sierra Basto (1989, 1991) evaluated the antidiarrhoeal effectiveness in acute diarrhoea and tolerance of *Saccharomyces boulardii* in a randomized double-blind placebo-controlled study, which included 130 Mexican infants from 3 months to 3 years of age. One group received 200 mg *Saccharomyces boulardii* every 8 h for 4 days, the other placebo. All children were treated with a rehydration therapy additionally. Efficacy was defined less than 4 bowel movements per day and no liquid stools. The frequency of stools decreased and the consistency of the faeces improved at 24 h of treatment in the *Saccharomyces boulardii* group as compared with the placebo group. The difference between the two groups became statistically significant at 48 and 96 hours, when clinical efficacy was assessed. Clinical cure rates were higher in the active treatment group than in the control group. The authors concluded that *Saccharomyces boulardii* may be used as an adjunct to oral rehydration for the treatment of acute diarrhoea in infants and very young children.

Table 14: Stool frequency: results of Centina-Sauri and Sierra Basto (1989, 1991)

Treatment day	***Saccharomyces boulardii* group (n=65)**	**Placebo group (n=65)**	**Significance**

start of treatment	7.50±2.15	6.66±2.26	
day 1	5.15±1.93	5.47±2.40	
day 2	3.76±2.31	4.38±2.73	p<0.05
day 3	2.53±1.78	3.63±2.53	
day 4	2.00±1.94	3.29±2.19	p<0.05

Urganci *et al.* 2001 evaluate the efficacy and tolerability of *Saccharomyces boulardii* (*Saccharomyces cerevisiae* Hansen CBS 5926) in a double-blind placebo-controlled study in 100 infants and small children (2–29 months; mean 10.8 months ± 0.9 months) with acute diarrhoea. 50 children got 250 mg *Saccharomyces boulardii* and rehydration solution and 50 children got placebo and rehydration solution. After 48 h and 96 h children treated with *Saccharomyces boulardii* scored significantly better than controls.

Table 15: Number of stools: results of Urganci *et al.* 2001

Treatment day	***Saccharomyces boulardii* group (n=50)**	**Placebo group (n=50)**	**Significance**
	Mean±SD	Mean±SD	
start of treatment	7.78±1.86	7.32±1.92	p>0.05
day 2	3.78±0.71	4.24±0.99	p<0.01
day 4	2.70±0.67	3.13±0.93	P<0.05

Kurugöl and Koturoglu (2005) evaluated the effect of *Saccharomyces boulardii* in Turkish children from 3 months to 7 years of age with acute diarrhoea. 232 children were enrolled, but 32 children were excluded. 23 children were prescribed antibiotics during the study period and 9 children were non-compliant to the protocol. Two hundred children received *Saccharomyces boulardii* in a granulated form in a daily dose of 250 mg or placebo for 5 days.

Table 16: Results of Kurugöl and Koturoglu (2005)

	***Saccharomyces boulardii* group (n=100)**	**Placebo group (n=100)**	**Significance**
duration of diarrhoea (d)	4.7±2.5*	5.5±3.2	p=0.03
duration of watery diarrhoea (d)	2.8±1.1	3.8±1.4	p<0.001
duration of vomiting (d)	1.2±1.0	1.3±1.0	p=0.61
duration of temperature >37.5°C (d)	1.0±0.8	1.1±0.9	p=0.28
length of hospital stay	2.9±1.2	3.9±1.5	p<0.001

*Values are mean ± SD

In addition the medians of the average stool frequency after the second day of the treatment were significantly lower in the *Saccharomyces boulardii* group than in the placebo group (p=0.003). Four children from the placebo group versus only one child from the *Saccharomyces boulardii* group had persisting diarrhoea. One child in the *Saccharomyces boulardii* group had meteorism.

Billoo *et al.* (2006) assessed the efficacy and safety of *Saccharomyces boulardii* in acute watery diarrhoea and its role in reducing the frequency of episodes of diarrhoea in subsequent two months. Pakistani children from 2 months to 12 years of age with acute diarrhoea were randomised in *Saccharomyces boulardii* group (treated with oral rehydration salt (ORS), nutritional support and *Saccharomyces boulardii*, 250 mg b.i.d) and in control group (treated with ORS and nutritional support only). Active treatment phase was 5 d and each child was followed for two months afterwards. Frequency and consistency of stools as well as safety of drug were assessed on every visit. A comparison of the two groups was made in terms of number of diarrhoeal episodes in subsequent two months. There were fifty patients in each group. Baseline characteristics such as mean age and the average frequency of stools were comparable in *Saccharomyces boulardii* and control group at the time of inclusion in the trial.

Table 17: Results of Billoo *et al.* (2006)

	***Saccharomyces boulardii* group (n=?)**	**Control group (n=?)**	**Significance (t-test)**
stool frequency day 0 (mean)	9.5	8.8	p=0.37
stool frequency day 3 (mean)	2.8	4.4	p=0.01
stool frequency day 6 (mean)	1.6	3.3	p=0.001
duration of diarrhoea (d)	3.6	4.8	p=0.001

Mean numbers of episodes of diarrhoea by the end of two months, were 0.56 in control group compared to 0.32 in *Saccharomyces boulardii* group (p=0.04). The drug was well accepted and tolerated. There were no reports of the side effects during treatment period. In conclusion, *Saccharomyces boulardii* significantly reduces the frequency and duration of acute diarrhoea. The consistency of stool also improves. The drug was welltolerated.

Canani *et al.* (2007) compared the efficacy of five probiotic preparations recommended to parents in the treatment of acute diarrhoea in Italian children. The study was a prospective single blind randomized controlled trial. 571 children aged between 3-36 months with acute diarrhoea (3 or more loose or liquid stools a day) were randomized to oral rehydration alone (n=91; 2 drop-outs; control group), or to oral rehydration in combination with a specific probiotic: *Lactobacillus rhamnosus* strain GG (n=98; drop out 1); *Saccharomyces boulardii* (n=87; drop out 2); *Bacillus clausii* (n=100; drop out 1); mix of *L delbrueckii* var *bulgaricus*, *Streptococcus thermophiles, L acidophilus*, and *Bifidobacterium bifidum* (n=94; drop out 1); or *Enterococcus faecium* SF68 (n=88; drop out 1). The primary outcome measures were the total duration of diarrhoea and the number of stools a day and their consistency. In contrast to *Lactobacillus rhamnosus* and the combination preparation *Saccharomyces boulardii* had no effect on duration of diarrhoea, stool outputs and

consistency as compared to rehydration therapy alone. None of the preparations had a significant effect on the secondary outcomes fever, vomiting and hospital admissions. No adverse events were recorded.

Ozkan *et al.* (2007) assessed the efficacy of *Saccharomyces boulardii* and its immune response in 27 Turkish children aged between 6 months and 10 years with acute diarrhoea. The patients were randomized in two groups. 16 children received 250 mg *Saccharomyces boulardii* dissolved in 5 ml water twice daily for 7 days, and 11 children received placebo. The decrease of stool frequency was significantly greater in the *Saccharomyces boulardii* group on days 3 and 4 as compared to the placebo group.

Table 18: Results of Ozkan *et al.* (2007)

	***Saccharomyces boulardii* group (n=?)**		**placebo group (n=?)**		**p value between the two groups**
	number of stools	**p value compared with baseline**	**number of stools**	**p value compared with baseline**	
baseline	6.23±0.53		5.92±0.44		ns
day 1	4.50±0.36	<0.001	5.36±0.38	<0.001	ns
day 2	3.06±0.33	<0.001	4.27±0.38	<0.001	ns
day 3	1.68±0.23	<0.001	3.36±0.38	<0.001	<0.05
day 4	0.43±0.22	<0.001	1.81±0.42	<0.001	<0.05

values are mean ± SD; ns = not significant

Clinical symptoms such as fever and dehydration were resolved on the second day, with no significant difference between the two groups. The *Saccharomyces boulardii* group demonstrated a significant decrease in serum immunoglobulin A and C-reactive protein levels on day 7. The percentage of CD8 lymphocytes on day 7 was significantly higher. No adverse reaction related to *Saccharomyces boulardii* therapy was observed during study.

Villarruel *et al.* (2007) evaluated the efficacy of *Saccharomyces boulardii* as an adjuvant to oral rehydration solution (ORS) in shortening the duration of acute, mild to moderate diarrhoea in 100 children aged between 3 and 24 months in ambulatory care in a randomized, double-blind, placebo-controlled study. 12 patients were excluded due to lack of compliance with protocol medication. Acute diarrhoea was defined as the presence of ≥3 liquid or loose stools in the preceding 24 h but for less than 7 days. 16 breastfed patients were included (5 in the *Saccharomyces boulardii* group and 11 in the placebo group). Children under 1 year received 250 mg *Saccharomyces boulardii* (no further specification), and those over 1 year 2 x 250 mg per day or placebo for 6 days. 72 patients had a follow-up for one month (35 in the *Saccharomyces boulardii* group and 37 in the placebo group).

Table 19: Results of Villarruel *et al.* (2007)

	***Saccharomyces boulardii* group**	**Placebo group**	**Significance**
number of stools on day 4	2.5±1.4	3.5±1.8	p<0.001

mean duration of diarrhoea	4.7 days (range 2-10 days)	6.16 days (range 2-13 days)	p<0.05
risk of having diarrhoea lasting more than 7 days/number of patients	3/44	12/44	RR 0.25; 95% CI 0.1-0.8

A statistically significant difference was observed in the number of stools on the 4th and 7th day favouring the subgroup that received early treatment (within the first 48 h of the onset of diarrhoea). However, no detailed information of stool frequency on day 7 is reported in the publication. No information of adverse events is given.

Vandenplas *et al.* (2007) evaluated the efficacy of *Saccharomyces boulardii* as an adjuvant to oral rehydration solution (ORS) in acute infectious gastroenteritis (GE) in children. 202 children less than 3 years old in 4 centres (3 in India, 1 in Indonesia) were included in the double-blind randomized placebo-controlled trial. All infants were treated according to WHO-recommendations (ORS for 6-7 h, rapid re-alimentation and ORS for every liquid stool) with 500 mg/day *Saccharomyces boulardii* or placebo as add-on treatment for 5 days. 188 (93%) children (93 *Saccharomyces boulardii*; 95 placebo) (age range 3-33 months) with diarrhoea (duration before inclusion 1-5 days) completed the study (drop-outs equal in both groups). Duration of diarrhoea was 66.57±52.52 h in the control group versus 53.65±38.74 h (difference ~13 h or 20% of duration) in the verum group (p=0.05). 86% of the children in *Saccharomyces boulardii* group versus 74% in placebo group were cured on day 3 (p=0.04). The number of cured patients on day 5 did not differ (97% vs 90%, p=0.133). The groups did not differ in quality of stools, vomiting or use of other medication. Sideeffects were not reported.

Htwe *et al.* (2008) evaluated the efficacy of *Saccharomyces boulardii* in acute diarrhoea in 100 hospitalized children in Myanmar (age range = 3 months to 10 years; 89 children were aged between 3 and 24 months, 11 children were older than 2 years). 50 children were treated with *Saccharomyces boulardii* (2 x 250 mg, no further specification) for five days in addition to oral rehydration solution (ORS) and 50 were given ORS alone (control group). The mean duration of diarrhoea was 3.08 days in the *Saccharomyces boulardii* group and 4.68 days (p<0.05) in the control group. Stools had a normal consistency on day 3 in 38 (76%) of 50 patients in the *Saccharomyces boulardii* group as compared with only 12 (24%) of 50 in the control group (p=0.019). No severe side effects were observed.

Table 20: stool frequency in the study population: results of Htwe *et al.* (2008)

	***Saccharomyces boulardii* (number of patients)**		**Control (number of patients)**		
Day	<3 stools/day	≥3 stools/day	<3 stools/day	≥3 stools/day	Chi-square test
1	0	50	0	50	ns
2	27	23	15	35	0.019
3	39	11	28	22	0.019
4	48	2	39	11	ns

5	50	0	48	2	ns
6	50	0	50	0	ns
7	50	0	50	0	ns

ns = not significant

Shen *et al.* (2008) evaluated the efficacy and safety of *Saccharomyces boulardii* for treatment of acute diarrhoea in 137 Chinese children aged from 1 month to 8 years in a multicenter, randomized and controlled trial. 75 children received *Saccharomyces boulardii* and 62 children dioctahedral smectite as control. The daily dose is not mentioned in the abstract. Frequency and consistency of stools as well as safety were assessed 48 h and 72 h after treatment. The improvement rate of diarrhoea after 48 h was 84% in the *Saccharomyces boulardii* group and 69.35% in control group (p=0.041), the improvement rates were 60% and 53.22%, respectively, after 72 h (p=0.425). The duration of diarrhoea was 3.12 days in *Saccharomyces boulardii* group whereas it was 3.58 days in the control group (p=0.080). The risk of diarrhoea lasting <7 days was more than 85% in two groups (p=0.347). No side effects were observed.

Ji *et al.* (2009) included in a randomized placebo-controlled trial 92 hospitalized Chinese children suffering from acute diarrhoea, aged from 2 months to 7 years. 46 children as control group received conventional treatment (ORS, oral montmorillonite, antibiotics if necessary), 46 children received *Saccharomyces boulardii* plus conventional therapy. The daily dose is not mentioned in the abstract. The mean diarrhoea duration was 6.54±1.74 days in the control group and 5.720±1.67 days in the *Saccharomyces boulardii* group (t=2.30, p<0.05). On the 4th day, the patients in the *Saccharomyces boulardii* group passed 3.13±0.95 stools/day versus 3.74±0.91 stools/day in the control group (t=3.14, p<0.01). On the 7th day, the patients in the *Saccharomyces boulardii* group passed 1.74±0.93 stools/day versus 2.24±0.95 stools/day in controlled group (t=2.55, p<0.05). A statistically significant difference was observed in the number of stools on the 4th and 7th day favouring the subgroup that received early treatment (within the first 48 h of the onset of diarrhoea) (t=3.90, 3.71, p<0.01).

Grandy *et al.* (2010) compared the effect of two probiotic products and placebo in addition to a rehydration therapy in the treatment of Bolivian children less than 2 years of age hospitalized for acute rotavirus diarrhoea in a randomized double-blind controlled clinical trial. Sample size was 20 per group and the outcomes were duration of diarrhoea, fever, vomiting and hospitalization. 64 cases finished the protocol. 20 patients received placebo (GC group), 21 patients *Saccharomyces boulardii* (4 x 10^{10} lyophilized cells b.i.d; GB group) and 23 patients a combination of *Lactobacillus acidophilus*, *Lactobacillus rhamnosus*, *Bifidobacterium longum* and *Saccharomyces boulardii* (1.375 x 10^{7} lyophilized cells/dose), GARLB group. Median duration of diarrhoea in the GB group (58 h) was shorter than in the GC group (84.5 h, p=0.04), also the duration of fever (p=0.041).

In a randomized prospective open-label study, Eren et al. (2010) compared the clinical efficacy and cost/effectiveness of *Saccharomyces boulardii* with yogurt fluid (YF) in acute non-bloody diarrhoea in Turkish children. Group A (n=28) received 250 mg lyophilized *Saccharomyces boulardii* twice a day in children ≥2 years and 125 mg twice a day in children <2 years of age. Group B (n=27) received YF (a fluid extracted from yoghurt made by a ferment containing *Lactobacillus bulgaricus* and *S. thermophilus*. The statistical analysis showed no significant difference between group A and B concerning duration of diarrhoea and duration of hospitalization in the PP and ITT analyses.

Table 21: Results of Eren *et al.* (2010)

	Group A	**Group B**	**Significance**

duration of diarrhoea in the ITT analysis (day)	4.45±2.46	5.38±3.14	p>0.05
duration of diarrhoea in the PP analysis (day)	4.54±2.36	4.81±1.79	p>0.05

At day 3 the PP analysis showed a resolution of diarrhoea in 13 patients (46.4%) in group A and in 6 patients (22. %) in group B (p=0.059). However the ITT analysis showed a statistically significant result for group A compared with group B (p=0.033). At day 5 there was no significant difference in both analysis concerning resolution of diarrhoea.

Le Luyer *et al.* (2010) compared in a double-blind, randomized, controlled, multicenter study the efficacy of a specific adapted formula (lactose-free, high-mineral, low-osmolarity formula, containing rice and pectin fortified with *Saccharomyces boulardii*) in the management of 70 infants (PPP; 77 ITTP) aged between 1 and 9 months suffering from acute diarrhoea with a standard formula used to feed healthy infants from birth. After a short phase of rehydration (oral or intravenous) 36 infants received the standard (control) formula, 34 infants received the modified formula. The duration of the diarrhoea was defined as the time needed until the occurrence of the first normal stool after the last liquid stool. The duration of diarrhoea from the time of inclusion was significantly reduced in the treated group (35.4±3.7 h) versus the control group (67.1±5 h); p<0.001). There were 15 infants with rotavirus in the treated group and 13 in the control group, however the duration of diarrhoea did not depend on the presence or absence of rotavirus but only on the treatment. The average daily weight gain was significantly higher in the treated group compared with the control group (74.2±26.4 g versus 23.7±6.7 g; p<0.05). On average 156 mg/d *Saccharomyces boulardii* were consumed in the treatment group. Efficacy of *Saccharomyces boulardii* cannot be assessed objectively because it was administered in combination with a specific adapted formula. No adverse event was reported. This trial was subsidized by the "Laboratoire United Pharmaceutical, 55, avenue Hoche, 75008 Paris, France".

Corrêa *et al.* (2011) investigated in a randomized double-blind, placebo-controlled study the efficacy of *Saccharomyces boulardii* in 186 Brazilian children (6 to 48 months old) with acute diarrhoea. The children received twice per day 200 mg *Saccharomyces boulardii* or placebo for 5 days. Among the 176 children who completed the trial, those treated with *Saccharomyces boulardii* (n=90) showed a reduction in diarrhoea duration (p <0.05) when compared with the placebo group (n=86).

Table 22: Results of Corrêa *et al.* (2011) I

		patients with diarrhoea 3 d after beginning of intervention				
analysis	groups (no. patients)	Yes (%)	No (%)	p	relative risk	95% confidence interval
intention to treat	*Saccharomyces boulardii* (95)	29 (30.5)	66 (69.5)	0.001	0.54	0.38-0.66
	placebo (91)	51 (56.0)	40 (44.0)			

per protocol	*Saccharomyces boulardii* (90)	29 (32.3)	61 (67.8)	0.0006	0.54	0.38-0.77
	placebo (86)	51 (59.2)	35 (40.8)			

The presence of rotavirus was detected in faecal samples from 162 patients. An exploratory analysis showed that the beneficial effect of *Saccharomyces boulardii* was observed essentially for patients presenting with rotaviral diarrhoea.

Table 23: Results of Corrêa *et al.* (2011) II

		Patients with diarrhoea 3 d after beginning of intervention					
groups (no. patients)		Yes (%)	No (%)	p	relative risk	95% confidence interval	P, ratio RR, 95% CI
rotavirus positive (93)	*Saccharomyces boulardii* (48)	14 (29.2)	34 (70.8)	0.0014	0.45	0.28-0.74	0.15 0.60
	placebo (45)	29 (64.4)	16 (35.6)				
rotavirus negative (69)	*Saccharomyces boulardii* (34)	14 (41.2)	20 (58.8)	0.395	0.76	0.46-1.26	0.30-1.20
	placebo (35)	19 (54.3)	16 (45.7)				

Dalgic *et al*. (2011) evaluated the effectiveness of zinc, *Saccharomyces boulardii* (no further specification), and lactose-free formula and their different combinations in the treatment of rotavirus diarrhoea in children from 1 to 28 months in Turkey. Time interval between onset of diarrhoea and hospitalization had to be less than 96 h. 480 children were enrolled in the prospective, single-blind and controlled trial and randomized to 8 groups each with 60 patients. Group 1 received 250 mg *Saccharomyces boulardii* once daily, group 2 received zinc, group 3 (received lactose-free formula, group 4 received *Saccharomyces boulardii* plus zinc, group 5 received *Saccharomyces boulardii* plus lactose-free formula, group 6 received zinc plus lactose-free formula, group 7 received *Saccharomyces boulardii* plus zinc plus lactose-free formula, control group 8 received only oral and/or parenteral rehydration solutions. No statistically significant differences were found in the time to resolution of fever after intervention between the treatment groups and the control group. The time to resolution of vomiting was significantly lower in group 4 compared with groups 1 and 5. The duration of diarrhoea was significantly reduced in groups 2 and 4 compared to control. A statistically significant difference in the duration of hospitalization was observed for the groups 2 and 4 in comparison to the control group. No adverse effects were observed.

Riaz *et al*. (2012) analysed the efficacy and safety of *Saccharomyces boulardii* in acute childhood diarrhoea in a double blind randomised controlled trial. 108 Indian children aged between 3 mo and 5 y were included in the study, 54 in the *Saccharomyces boulardii* and 54 children in the placebo group. Six cases in the *Saccharomyces boulardii* group and 3 cases in the placebo group left the study. There were 5 and 4 treatment failures in the *Saccharomyces boulardii* and placebo group, respectively. The children received either 250 mg *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) twice daily or placebo for 5 d or till the recovery whichever was earlier. Mean post intervention duration of diarrhoea was significantly (95% CI=-28.13 to -5.43) shorter in the *Saccharomyces boulardii* group (52.08±24.57 h) as compared to placebo group (64.04±30.43 h). The time of appearance of first semi formed stool in the *Saccharomyces boulardii* group (39.48±23.09 h) was significantly (95% CI=-25.4 to -3.87) shorter than the placebo group (54.13±28.21 h). Other parameters like total amount of oral rehydration solution, weight gain, and number of stools do not differ significantly. No adverse effects were observed.

Erdoğan *et al.* 2012 compared the clinical effectiveness of *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) and *Bifidobacterium lactis* in children aged between 5 months – 5 years who had been diagnosed with rotavirus gastroenteritis. The first group (25 children) received oral rehydration therapy and rapid refeeding with a normal diet with 282.5 mg/d *Saccharomyces boulardii*. The second group (25 children) received oral rehydration therapy and rapid refeeding with a normal diet with 30 mg/d *Bifidobacterium lactis*. The third group received only rehydration therapy and rapid refeeding with a normal diet. The mean duration time of diarrhoea was 6.6 ± 1.7 days for the first group, 4.1 ± 1.3 days for the second group, and 7,0 ± 1.6 days for the third group. The mean duration time for diarrhoea in the second group was significantly shorter than in the first and third groups ($p<0.001$). Vomiting rates had no significant difference in all groups.

Khan *et al.* 2012 performed a randomized controlled trial in 420 Pakistani children with acute watery diarrhoea, aged 2 months to 5 years. Group 1 (210 children) was given *Saccharomyces boulardii* (250 mg BD) and rehydration therapy, group 2 (210 children) rehydration therapy only for 5 days. Statistically significant differences in terms of stool consistency and frequency were noted in group 1 from 2nd day of treatment onwards. Group 1 also showed reduction in mean duration of diarrhoea by 1.1 das compared with group 2.

Burande 2013 performed a prospective, parallel, single-blind, randomized, controlled clinical trial in children with acute diarrhoea. 72 Indian children were randomized either to group I given 250 mg *Saccharomyces boulardii* twice a day, rehydration therapy and zinc or to group II given rehydration therapy and zinc, only. One patient in each group were lost to follow up. Average time for recovery from loose motions was 3.4 days ± 1.4 days in group I, and 5.5 days ± 2.1 days in group II (statistical significance).

Shaikh *et al*. (2015) conducted a randomized comparative study in 100 patients with acute diarrhoea (age 3 months to 5 years) in Pakistan. Cases were given low osmolar ORS, Zinc & *S.boulardii* 250 mg twice daily for three days and controls were given low osmolar ORS and Zinc. P-value ≤0.05 was considered as significant. The results showed that 51 patients were male and 49 patients were female. The overall mean age of study subjects was 26.73±12.65 months. 96 children were hospitalized. There were 50 children in each of the two groups. The results were evaluated according to 1st, 2nd, and 3rd day post intervention. Among 50 patients of the case group, 18 patients were compliant. 49 patients had decrease in duration. The same results were observed in the decrease in frequency. Consistency was improved in 39 patients. The duration of hospitalization was reduced in 36 patients. It was observed that improvement in the duration, frequency, and consistency was mostly observed on 2nd day of post intervention. There was no

significant difference between the two groups regarding the mean number of stools after 24 hours of beginning of treatment. However, the results showed statistically significant gradual reduction in favor of probiotic group from 48 hours onwards. The mean number of stools remained comparable between probiotic group and control group on day 0 and day 1. However, in probiotic group the mean number of stools was lower on day 2, day 3 and day 4 compared to control group. The information concerning study design and method is incomplete.

Das *et al*. (2016) studied the efficacy and safety of *Saccharomyces boulardii* (SB) in acute childhood rotavirus diarrhoea in India. In a double-blind, randomized controlled trial 60 children (3 months to 5 years) with WHO-defined acute watery diarrhoea (≥3 unformed or loose stools in the last 24 h) and stool rotaviruspositive were randomized into intervention (n = 30) and control (n = 30) groups. The intervention group received SB in lyophilized powered form (2x250 mg/day) for 5 days. The children remained in hospital till improvement in their clinical condition, and after discharge were followed till 7 days. The primary endpoint "the median duration (hours) of diarrhoea" (time (in hours) from the first to the last abnormal (loose or liquid) stools preceding normal stool return) was significantly shorter in the intervention group (60 vs. 89; 95% CI: -41.2 to -16.8). A significantly shorter duration of hospitalization (74 vs. 91; 95% CI: -33.46 to- 0.54) was also seen in the intervention group, but no significant difference was seen for fever and vomiting. There was also no difference between the two groups in the proportion of children requiring parenteral rehydration and persistence of diarrhoea lasting beyond day 7. There was no report of any adverse event.

Table 24: Results of Das *et al.* (2016)

Outcome	***Saccharomyces boulardii* group (n=30)**	**Placebo group n=28 (completed follow-up)**	**Difference (95% CI)**
Median (IQR) hours of diarrhoea[a]	60 (51-67)	89 (68-95)	-29 (-41.2 to -16.8)
Median (IQR) hours of hospitalization[a]	74 (64-90)	91 (76-105)	-17 (-33.46 to -0.54)
Median (IQR) duration of fever (hours)[a]	56 (48-67)	67 (55-81)	-11 (-23.04 to 1.04)
Median (IQR) duration of vomiting (hours)[a]	48 (39-56)	55 (43-61)	-7 (-16.41 to 2.41)
Proportion of children requiring parenteral rehydration[b]	2 (6.7)	5 (16.7)	0.36 (0.06 to 2.01)
Proportion of children having diarrhea lasting beyond day 7[b]	1 (3.3)	4 (14.3)	0.21 (0.02 to 1.98)

The difference between the two medians and calculation of 95% CI (confidence interval) has been done by the method proposed by Bonett and Price.
bData expressed in odds ratio (OR) and 95% CI.

In a randomized controlled trial in Pakistan Asmat *et al.* (2018) compared the efficacy of *Saccharomyces boulardii* and lactic acid producing probiotics in addition to usual treatment regimen to cure diarrhoea among children. Children at the age from 6 months to 5 years suffering from

diarrhoea (<14 days) were randomly assigned to oral treatment with *Saccharomyces boulardii* or lactic acid producing bacterial probiotics for 5 days. The dose for probiotics was administered orally twice a day in 20 ml of water, dosage for less than one year of age was 150 mg and 250 mg for children older than two years, divided into two doses for each of *Saccharomyces boulardii* and lactic acid producing probiotics groups. Efficacy of both probiotics was assessed as per operational definition (frequency and consistency of stools/day). In addition to probiotics, all patients were treated with intravenous antibiotics (ceftrioxne) and oral rehydration therapy as per hospital protocol. Chisquare test was applied to compare the efficacy of both groups. Data were stratified for age, gender, gestational, weight, duration of diarrhea and socioeconomic status to deal with effect of modifiers. Post-stratification Chisquare test was applied. P-value of <0.05 was considered as significant.Two hundred patients were randomly selected for trials; out of which, 100 were treated with *Saccharomyces boulardii* while the other 100 were supplemented with lactic acid concomitantly along with conventional diarrhea treatment. Results indicated that *Saccharomyces boulardii* treatment group has significantly higher efficacy rate (45%) compared to lactic acid producing probiotics (26%). According to the authors this study showed that *Saccharomyces boulardii* has a better efficacy compared to lactic acid and may be adopted as a probiotic of choice.

Table 25: clinical studies in children with acute unspecific diarrhoea

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of Administration** **Duration of treatment**	**Number of Subjects (including age, sex, drop out)**	**Healthy Subjects or Diagnosis of Patients (inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Chapoy, 1985 Chapoy, 1986	controlled	Chapoy 1985: *Saccharomyces cerevisiae* CBS 5926 (no information on number of viable cells) Chapoy 1986: *Saccharomyces cerevisiae* CBS 5926 ($\geq 1.8 \times 10^{10}$ viable cells/g) dosage:	38 French children (2 weeks to 30 months) *Saccharomyces boulardii: n*= 19 control: n= 19	gastroenteritis with acute diarrhoea	results of Day 1 and Day 4 were compared and calculated as mean ± SD. Frequency (days), consistency (watery, soft, normal), weight (g), transit time (h, assessed by the carmin red method) of stools significant reduction in the number of stools ($p<0.01$),	Student's t test, chi-square test	small European study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		2 x 250 mg and rehydration treatment or only rehydration treatment duration: 5 days			weight and consistency of stools ($p<0.05$), and transit time ($p<0.05$) could be confirmed by statistical evaluation of all 19 children in the *Saccharomyces cerevisiae* group		
Treatment of acute unspecific diarrhoea Cetina-Sauri and Sierra	multicenter, randomized, placebo-controlled, double-blind	200 mg *Saccharomyces cerevisiae CBS 5926* (no information on strain and number of viable cells are	130 Mexican children; (*Saccharomyces boulardii* n=65; placebo n=65)	acute diarrhoea	frequency (comparison between the two treatment groups=efficacy) and consistency (effective, ineffective) of stools after 4 days;	Student's t test, chi-square test	Non-European study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Basto, 1989 and 1991		given in the reference, according to Corrêa *et al.*(2011), who used the same product: 200 mg contain 4 x 10^9 viable cells) every 8 h or placebo and rehydration therapy duration: 4 days	age: 3 mo-3 yrs		clinical cure rate (efficacy was defined as return to 4 bowel movements or deterioration of symptoms) frequency of stools decreased and the difference between the two groups became statistically significant at 48 and 96 h ($p<0.05$)		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					clinical cure rate after 48 h was higher in the *Saccharomyces boulardii* group (48 h: ≈37%; 96 h: ≈87%) than in the placebo group (48 h: ≈7%; 96 h: ≈43%) judgement of effectivity showed significant results too ($p<0.01$)		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Urganci *et al.,* 2001	placebo-controlled, double-blind	*Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926: ≥1.8 x 10^{10} viable cells/g) 250 mg/d *Saccharomyces boulardii* or placebo together with rehydration therapy	100 Turkish children *S.cerevisiae* n=50 (24 boys, 26 girls) mean age=11.5±7.1 mo placebo n=50 (22 boys, 28 girls)	acute, non-bacterial diarrhoea lasting more than 48 hours	significant differences were found regarding numer of stools after 48 h ($p<0.01$) and 96 h ($p<0.05$) and percentage of cases cured after 48 h ($p<0.01$) and 96 h ($p<0.05$) in *Saccharomyces cerevisiae* treated children.	Student's t test for two independent samples, chi-square test	randomization not mentioned Non-European (Turkish) study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			mean age=10.1±5.1 mo age: 2–29 mo				
Treatment of acute unspecific diarrhoea Kurugöl and Koturoglu, 2005	randomized, placebo-controlled, double-blind	250 mg/d *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926, no information on number of viable cells) or placebo	200 Turkish children out of 232 enrolled children age: 3 mo - 7 yrs	acute diarrhoea (liquid, mucous of bloody stools passed at least twice as frequently than usual for a minimum of 24 h before admission	time from the start of the treatment until the appearance of the first normal stool: significant reduction in the *Saccharomyces boulardii* group compared to the placebo group	Student's t test, Chi-square test	Non-European (Turkish) study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		together with rehydration therapy duration: 5 days		but not for longer than 7 d	(4.7±2.5 vs 5.5±3.2, p=0.03) medians of the average stool frequency after the second day of the treatment were significantly lower in the *Saccharomyces boulardii* group than in the placebo group (p=0.003)		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control** **Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%)** **Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					WMD (fixed) 95% CI: -1.00 day (-1.35, -0.65), Vandenplas *et al.*, 2008		
Treatment of acute unspecific diarrhoea Billoo *et al.*, 2006	randomized, controlled	*Saccharomyces cerevisiae* CBS 5926, about 2×10^{10} viable cells/g (250 mg b.i.d) with rehydration therapy or rehydration therapy only	100 Pakistani children (2 mo - 12 yrs) (50/50)	acute watery diarrhoea of mild to moderate severity	by day 3 frequency of stools reduced to 2.8 and 4.4 stools per day, respectively (p=0.01) and by day 6 it reduced to 1.6 (*Saccharomyces boulardii* group) and 3.3 (control group), p=0.001	Student's t test	Non-European study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		duration: active treatment 5 days, follow-up for 2 months			duration of diarrhoea was 3.6 days in *Saccharomyces boulardii* group whereas it was 4.8 days in control group (p=0.001) WMD (fixed) 95% CI: -1.26 day (-1.73, -0.79), Vandenplas *et al.,* 2008		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Canani *et al.*, 2007	prospective single blind randomized controlled	*Saccharomyces cerevisiae* CBS 5926lt: 5 x 10^9 live micro-organisms/dose twice daily group 1: rehydration solution alone group 2: *Lactobacillus rhamnosus* strain GG	571 Italian children aged between 3-36 mo randomized 558 children received study intervention: group 1: n=91; drop out 2,	acute diarrhoea (3 or more outputs of loose or liquid stools a day)	*Saccharomyces boulardii* had no effect on duration of diarrhoea, stool outputs and consistency compared to rehydration therapy alone in contrast *to Lactobacillus rhamnosus* and the combination preparation none of the preparations had a	chi-square test, Mann-Whitney U test	single blind, European study population negative outcome for *Saccharomyces boulardii*

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		group 3: *Saccharomyces boulardii* group 4: *Bacillus clausii* group 5: mix of *L. delbrueckii* var *bulgaricus*, *Streptococcus thermophiles*, L. *acidophilus* and *Bifidobacterium bifidum*	group 2: n=98; drop out 1, group 3: n=87; drop out 2, group 4: n=100; drop out 1, group 5: n=94; drop out 1,		significant effect on the secondary outcomes fever, vomiting and hospital admissions. WMD (fixed) 95% CI: -0.11day (-0.48, 0.26), Vandenplas *et al*., 2008		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		group 6: *Enterococcus faecium* SF68 duration: 5 days	group 6:n=88; drop out 1				

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Ozkan *et al.*, 2007	randomized, double-blind, placebo-controlled	*Saccharomyces cerevisiae* CBS 5926, no information on number of viable cells (250 mg b.i.d) or placebo in addition to oral rehydration therapy duration: 7 days	27 Turkish children (6 mo - 10 yrs) verum=16 (9 male/7 female) mean age=23.4±6.6 mo placebo=11 (6 male/5 female) mean	acute diarrhoea	both groups experienced reduced daily stool frequency, the decrease being significantly greater in group 1 on days 3 and 4 compared with group 2 group 1 demonstrated significant increases in serum immunoglobulin A and decreases in C-	chi-square test, Fisher's exact test, Mann Whitney U-test, Wilcoxon's signed rank test	small Non-European study population (Turkish), significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			age=17.6±4.6 mo		reactive protein levels on day 7 percentage of CD8 lymphocytes on day 7 was significantly higher in group 1 than group 2		
Treatment of acute unspecific diarrhoea	randomized, double-blind, placebo-controlled	children under 1 year: 1 x 250 mg *Saccharomyces boulardii* (no further specification)	100 children (Argentine/ Belgian?) (3 - 24 mo) verum=44	acute mild to moderate diarrhoea	primary endpoints: number of stools on day 4 and 7 were significantly smaller in the *Saccharomyces boulardii* group than in the placebo group	Student's t test ANOVA	European or Non-european study population,

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Villarruel *et al.*, 2007		children over 1 year: 2 x 250 mg per day or placebo in addition to oral rehydration solution (ORS) duration of therapy: 6 days 72 patients had a follow-up for one	placebo=44 12 drop outs		Patients were also less likely to have diarrhoea on the 7th day or diarrhoea lasting more than 7 days in the *Saccharomyces boulardii* group compared to the placebo group WMD (fixed) 95% CI: -1.46 day (-2.68, -		significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		month (verum 35; placebo 37)			0.25), Vandenplas *et al.*, 2008		
Treatment of acute unspecific diarrhoea Vandenplas *et al.*, 2007	randomized, double-blind, placebo-controlled	500 mg/day *Saccharomyces boulardii* (no further specification) or placebo in addition to ORS for 5 days	202 Indian or Indonesian children <3 yrs old included 188 children (93 verum, 95 placebo) (age range 3-33 mo) completed	infectious gastroenteritis, diarrhoea, duration before inclusion 1-5 days	duration of diarrhoea was 66.57±52.52 h in the control group versus 53.65±38.74 h (difference ~13 h or 20% of duration) in the verum group (p=0.05) 86% in *Saccharomyces boulardii* group	Not mentioned	only one side publication available, Non-European study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					versus 74% in placebo group were cured on day 3 (p=0.04) number of cured patients on day 5 did not differ (97% vs 90%, p=0.133)		
Treatment of acute unspecific diarrhoea	open, controlled	*Saccharomyces boulardii* (250 mg b.i.d, no further specification) with rehydration therapy	100 children in Myanmar (3 mo - 10 yrs)	acute watery diarrhoea with a duration of less than 7 days	mean duration of diarrhea was 3.08 days in the *Saccharomyces boulardii* group and	chi-square test	Non-European study population,

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Htwe *et al.*, 2008		or rehydration therapy only duration: 5 days	verum=50 control=50	diarrhoea was defined as passing three or more loose stools per day (loose stool is a stool that takes the shape of the container)	4.68 days ($p<0.05$) in the control group stools had a normal consistency on day 3 in 38 (76%) of 50 patients in the *Saccharomyces boulardii* group compared with 12 (24%) of 50 in the control group ($p=0.019$)		significant results

Type (aim) and objective(s) of Study Reference	Study Design and Type of Control Study duration (if available)	Test Product(s): herbal preparation, pharmaceutical form; Dosage Regimen; Route of Administration Duration of treatment	Number of Subjects (including age, sex, drop out)	Healthy Subjects or Diagnosis of Patients (inclusion criteria)	Outcomes (primary and secondary endpoints)	Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score e.g. Jadad score	Comments on clinical relevance of results
					on day 2, 27 (54%) of 50 had less than three stools per day in the *Saccharomyces boulardii* group compared with 15 (30%) of 50 in the control group (p=0.019) WMD (fixed) 95% CI: -1.60 day (-2.03, -1.17), Vandenplas *et al.*, 2008		

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Shen *et al.*, 2008 (abstract only)	multi-center, randomized, controlled	*Saccharomyces boulardii* (n=75) or dioctahedral smectite as control (n=62)	137 Chinese children (1 mo - 8 yrs)	acute diarrhoea	improvement rate of diarrhoea after 48 h: 84% in the *Saccharomyces boulardii* group and 69.35% in control group (p=0.041) improvement rate after 72 h: 60% and 53.22%, respectively (p=0.425) duration of diarrhoea: 3.12 days in	Not mentioned	abstract only, Non-european study population, active-controlled, dosage not given, superiority of *Saccharomyces boulardii* in

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					Saccharomyces boulardii group and 3.58 days in the control group (p=0.080) risk of diarrhoea lasting <7 days was more than 85% in two groups (p=0.347)		improvement of diarrhoea after 48 h, otherwise comparable results
Treatment of acute unspecific diarrhoea	randomized, controlled	conventional treatment (ORS, oral montmorillonite,	92 Chinese children (2 mo - 7 yrs)	acute diarrhoea	4th day: *Saccharomyces boulardii group* 3.13±0.95 stools/day	Not exactly mentioned:	abstract only, Non-european

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Ji *et al.*, 2009 (abstract only)		antibiotics if necessary), or *Saccharomyces boulardii* (no further specification) plus conventional therapy duration: ?	verum=46 control=46		versus 3.74±0.91 stools/day in control group (t=3.14, p<0.01) 7th day: *Saccharomyces boulardii* group 1.74±0.93 stools/day versus 2.24±0.95 stools/day in control group (t=2.55, p<0.05)	Student's t test chi-square test	study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Grandy *et al.*, 2010	randomized, double-blind, placebo-controlled	in addition to a rehydration therapy: 21 patients (GB group): *Saccharomyces boulardii* (4 x 10^{10} lyophilized cells b.i.d) 23 patients (GARLB group): a combination of *L. acidophilus*, *L. rhamnosus*, *Bifidobacterium longum* and	76 Bolivian patients 12 patients were excluded from analysis (other etiologic agents, urinary infection, pneumonia, edema, kwashiorkor, severe vomiting)	acute rotavirus diarrhoea: presence of at least 3 bowel movements more than the normal number for the child and/or presence of watery stools per day, plus latex test positive for rotavirus	median duration of diarrhoea in the GB group (58 h; IRQ 41) was shorter than in the GC group (84.5 h (IRQ 94), p=0.04), also the duration of fever (p=0.041) median duration of diarrhoea in the GARLB group (60 h, IRQ 40) was shorter than in the GC group (84.5 h) without	Kruskall-Wallis testing, Mann-Whitney U test, chi square test	Non-european study population, small study groups, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		Saccharomyces boulardii (1.375×10^7 lyophilized cells/dose) b.i.d 20 patients (GC group) placebo duration: 5 days	64 analyzed patients (1 - 23 mo) 36 male, 28 female		statistical significance (p=0.06) IRQ = interquartile range		

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Treatment of acute unspecific diarrhoea Eren *et al.*, 2010	randomized, controlled, open	*Saccharomyces cerevisiae* CBS 5926 (no information on number of cells) group A (n=28): 250 mg *Saccharomyces boulardii* twice a day in children ≥2 years and 125 mg twice a day in children <2 years of age	67 Turkish children (5 mo - 16 yrs) 12 drop outs (extraintestinal infections, violation of the fermented milk restriction, secondary hemophagocytic syndrome,	acute diarrhoea (presence of 3 or more liquid or loose stools per day lasting for less than 14 days)	no significant difference between group A and B concerning duration of diarrhoea and duration of hospitalization in the PP and ITT analyses at day 3: PP analysis showed a resolution of diarrhoea in 13 patients (46.4%) in group A and in 6	Independent t test, chi square test, Mann-Whitney U tests, and Wilcoxon sign test	small Non-european study population (Turkish), open design, active-controlled, ITT analysis showed statistically significant result in resolution of iarrhoea for *Saccharomy*

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		group B (n=27) yogurt fluid (YF: a fluid extracted from yoghurt made by a ferment containing *Lactobacillus bulgaricus* and *S. thermophiles*) duration: until diarrhoea resolved	celiac disease) 55 children (36 boys, 19 girls; mean age=21±28. 2 mo)		patients (22.2%) in group B (p=0.059) ITT analysis showed statistically significant result for group A compared with group B (p=0.033) at day 5: no significant difference in both analysis concerning resolution of diarrhoea		*ces boulardii*. otherwise comparable results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
Treatment of acute unspecific diarrhoea Le Luyer *et al.*, 2010	randomized, controlled, double-blind, multicentric	lactose-free, high-mineral, low-osmolarity formula, containing rice and pectin fortified with *Saccharomyces boulardii* versus standard formula after a short phase of rehydration therapy oral or intravenous in average: 156 mg/d	77 (ITT) – 38 treatment group – 39 control group 70 (PP) – 36 treatment group – 34 control group age: 1-9 mo	acute diarrhoea (presence of 3 or more liquid or loose stools during the last 24 h)	duration of diarrhoea from time of inclusion was significantly reduced in the treated group (35.4±3.7 h) versus the control group (67.1±5 h) average daily weight gain was significantly higher in the treated group compared with the control group	mean value with standard deviation, variance analysis, "somme des carrés", Kruskal-Wallis-test	efficacy of *Saccharomyces boulardii* cannot be assessed because it was administered in combination with a specific adapted formula good tolerability

<table>
<tr><th>Type (aim) and objective(s) of Study

Reference</th><th>Study Design and Type of Control Study duration (if available)</th><th>Test Product(s): herbal preparation, pharmaceutical form;

Dosage

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Duration of treatment</th><th>Number of

Subjects (including age, sex, drop out)</th><th>Healthy

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e.g. Jadad score</th><th>Comments on clinical relevance of results</th></tr>
<tr><td></td><td></td><td>Saccharomyces boulardii

duration of treatment: 6 days (?)</td><td></td><td></td><td>(74.2±26.4 g versus 23.7±6.7 g; p<0.05)
<table><tr><td>No of stools</td><td>treat-ment</td><td>con-trol</td></tr><tr><td>begin</td><td colspan="2">6.6±0.4</td></tr><tr><td>Day 4</td><td>.9±.4</td><td>2.8±.7</td></tr><tr><td>Day 5</td><td>1.1±.5</td><td>2.3±.7</td></tr><tr><td>Day 6</td><td>.2±.2</td><td>1.3±.7</td></tr></table>p<0.005</td><td></td><td></td></tr>
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Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					no adverse event		
Treatment of acute unspecific diarrhoea Corrêa *et al.*, 2011	randomized, placebo-controlled, double-blind	*Saccharomyces cerevisiae* CBS 5926: 1 capsule contains 200 mg lyophilized *Saccharomyces boulardii* (4 x 10^9 viable cells) 200 mg *Saccharomyces*	186 Brazilian children (6-48 mo): verum=95 placebo=91 10 drop outs: verum=3 and placebo=2 due to need	acute diarrhoea within 72 h before hospitalization diarrhoea was defined as a change in bowel habits with a diminution of stool consistency and 3 or more evacuation per day	therapy with *Saccharomyces boulardii* showed a reduction in diarrhoea duration ($p<0.05$) when compared with the placebo group exploratory analysis showed that the beneficial effect of *Saccharomyces*	Yates continuity-corrected chi square test	Non-european study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		boulardii twice a day or placebo duration: 5 days	for antibiotic treatment verum=2 and placebo=3 because of withdrawal during the trial		*boulardii* was observed essentially for patients presenting with rotaviral diarrhoea		
Treatment of acute unspecific diarrhoea	randomized single-blind, controlled	1) 250 mg *Saccharomyces boulardii* once daily	480 Turkish children (1-28 mo)	episode of ≥3 watery or looser-than normal stools	duration of diarrhoea (from start of treatment until the first normal stool) was significantly	chi-square test, ANOVA	Non-European study

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Dalgic *et al.*, 2011		alone (no further specification) 2) zinc alone 3) lactose-free formula alone 4) *Saccharomyces boulardii* + zinc 5) *Saccharomyces boulardii* + lactose-free formula 6) zinc + lactose-free formula	60 patients in each group 288 boys, 192 girls mean age: 13.71±6.21 mo 470 partially breast-fed, 10 received cow's milk-	within 24 h and/or forceful vomiting time interval between onset of diarrhoea and hospitalization <96 h, stool positive for rotavirus antigen mild-moderate dehydration	reduced in groups 2 and 4 compared to control ($p<0.05$) time to resolution of vomiting was significantly lower in group 4 compared with groups 1 and 5 statistically significant difference in the duration of hospitalization was observed for the		population (Turkish), significant results for *Saccharomyces boulardii* + zinc

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		7) *Saccharomyces boulardii* + zinc + lactose-free formula 8) only oral and/or parenteral rehydration duration: minimum 5 days	based formula only		groups 2 and 4 in comparison to the control group no statistically significant differences were found in the time to resolution of fever after intervention between the treatment groups and the control group no adverse event		

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Treatment of acute unspecific diarrhoea Riaz *et al.*, 2012	randomized, placebo-controlled, double-blind	*Saccharomyces cerevisiae* CBS 5926 (no information on number of cells): 250 mg twice daily or placebo duration: 5 days or till recovery whichever was earlier	108 Indian children (3 mo – 5 yrs), 54 in each group 6 children in the *Saccharomyces boulardii* group and 3 in the placebo group left the study	acute onset diarrhoea (less than 48 h)	duration of post intervention diarrhoea (defined time from enrolment to recovery, recovery and discharge criterion was defined as passage of 3 consecutive semi formed stools or no stools for 12 h) significantly (95% CI=-28.13 to -5.43) shorter in the *Saccharomyces boulardii* group (52.08±24.57 h) as	Student's t test. Mann-Whitney *U* test, chi-square test, Fisher's exact test	Non-Europeaun study population, significant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
			treatment failures: 5 in the *Saccharomyces boulardii* group, 4 in placebo group		compared to placebo group (64.04±30.43 h)		
Treatment of acute unssspecccificdiarrhoea	prospective randomized controlled	group I (25): 282.5 mg/d *Saccharomyces cerevisiae* CBS 5926 (no information on numer of cells) +	75 Turkish children between (5 mo - 5 yrs) group I: 21.6 ± 11.5 mo,	rotavirus gastroenteritis with 3 or more times of watery diarrhoea per day in the last 48 h	mean duration time of diarrhoea group I: 6.6 ± 1.7 days,	SPSS 19 programme for windows, chi-square test,	Non-European study population (Turkish),

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Erdoğan *et al*. 2012		rehydration therapy, group II (25): 30 mg *Bifidobacterium lactis* + rehydration therapy group III (25): rehydration therapy only	11 boys, 14 girls group II: 22.1 ± 14 mo, 12 boys, 13 girls group III: 19.1 ± 13.3 mo, 14 boys, 11 girls		group II: 4.1 ± 1.3 days, group III: 7.0 ± 1.6 days, significance in group II compared with group I and group III vomiting rates had no significant difference in all groups	repeated ANOVA test	small study groups, treatment with *Saccharomyces boulardii* also shorten the duration of diarrhoea, significance ?
Treatment of acute	randomized controlled	group I (210): 250 mg BD *Saccharomyces*	420 Pakistani children (2 mo - 5 yrs)	acute watery diarrhoea	stool consistency and frequency: the improvement was	Student's t test, chi-	Non-European

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unspecific diarrhoea Khan *et al.* 2012		*boulardii* + rehydration therapy group II (210): rehydration therapy alone duration of treatmen: 5 days	group I: 107 children 1-12 mo; 85 children 1-3 yrs, 18 children 3-5 yrs; 107 boys, 103 girls. group II: 107 children 1-12 mo; 79 children 1-3 yrs; 24 children 3-5 yrs; 109		significantly rapid in group I compared with group II. mean duration of diarrhoea in group I was 3.43 days, compared with 4.5 days in group II ($p<0.05$)	square test,	study population, significant results

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			boys, 101 girls				
Treatment of acute unspecific diarrhoea Burande, 2013	prospective, parallel, single-blind, randomized, controlled	group I: 250 mg *Saccharomyces boulardii* twice daily (lyophilized powder in a sachet weighing 282 mg equivalent to 250 mg of yeast) + rehydration therapy group II: rehydration therapy only	100 Indian patients screened, 72 patients randomized (group I=36; group II=36), 1 drop out per group (lost to follow up) group I:	acute diarrhoea (≥ unformed stool in last 24 h with duration of <48 h)	average time for recovery from loose motions: group I: 3.4±1.4 days group II: 5.5±2.1 days (*Z* value = 4.9) statistical significance	Not exactly mentioned, Student *t*-test	Non-European study population, single-blind, signifcant results

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		in addition zinc 10 mg/d for children<6 months, and 20 mg/d for children >6 months for 14 days duration of treatment with *Saccharomyces boulardii* 5 days	mean age: 11.46±8.64 mo, 17 boys, 18 girls; group II: mean age 13.55±12.84 mo, 14 boys, 21 girls		vomiting (11 patients in group I, and 8 patients in group II): average time of recovery in group I 2.5±1.2 days, and in group II 3.3±1.2 days ($p<0.01$, two-tailed unpaired Student *t*-test)		
Treatment of acute unspecific diarrhoea	Randomized controlled study in Pakistan, no	Cases: low osmolar ORS, Zinc & *Saccharomyces boulardii* 250 mg	100 children (3 months – 5 years, mean age	acute diarrhoea	Primary outcome variables: duration, frequency, consistency of stools	SPSS-version 17(statistical package	Non-European study population,

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Shaikh, 2015	further information is given Duration 3 days?	twice daily for three days Controls: low osmolar ORS and Zinc	26.73±12.65 months) 51 male and 49 female patients 96 children were hospitalized. 50 children in each of the two groups.		and duration of hospitalization according to 1st, 2nd, and 3rd day post intervention: case group: 49 patients had decrease in duration and frequency. Consistency was improved in 39 patients. The duration of hospitalization was reduced in 36 patients.	for social sciences). Chi-square test. P-value ≤ 0.05 was considered as significant.	The information concerning study design and method is incomplete. statistical significance in favor of probiotic group from 48 hours onwards.

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
					Improvement in the duration, frequency, and consistency was mostly observed on 2nd day of post intervention. No significant difference between the two groups for the mean number of stools after 24 hours of beginning of treatment. Statistically significant gradual reduction in favor of		

Type (aim) and objective(s) of Study Reference	Study Design and Type of Control Study duration (if available)	Test Product(s): herbal preparation, pharmaceutical form; Dosage Regimen; Route of Administration Duration of treatment	Number of Subjects (including age, sex, drop out)	Healthy Subjects or Diagnosis of Patients (inclusion criteria)	Outcomes (primary and secondary endpoints)	Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score e.g. Jadad score	Comments on clinical relevance of results
					probiotic group from 48 hours onwards. The mean number of stools remained comparable between probiotic group and control group on day 0 and day 1. In probiotic group the mean number of stools was lower on day 2, day 3 and day 4 compared to control group 2.		

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Treatment of acute unspecific diarrhoea Das, 2016	double-blind, randomized controlled one center in India duration = 7 days	SB group: 250 mg *Saccharomyces boulardii* in lyophilized powdered form b.i.d. for 5 days Control group: placebo	60 children (3 months to 5 years) enrolled, each group 30	acute rotavirus diarrhea of <48 h duration, (diarrhea: ≥3 unformed or loose stools in the last 24 h)	Primary endpoint: Duration (in h) of acute diarrhea: median duration (hours) of diarrhoea was significantly shorter in the SB group (60 vs. 89; 95% CI: -41.2 to-16.8). Secondary endpoints: Significantly shorter duration of hospitalization, no significant difference	SPSS software (version 20.0 Chicago, IL, USA). Chi-Square test, Mann-Whitney U test, Bonett and Price. Intention to treat analysis was used	Significant results. Indian, partially hospitalized children, small groups

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					in fever, vomiting, require of parenteral rehydration and persistence of diarrhoea lasting beyond day 7.	for the primary outcome. P-value <0.05 was taken as significant.	
Treatment of acute unspecific diarrhoea Asmat *et al*., 2018	Randomized controlled	*Saccharomyces boulardii* resp. Lactic acid producing bacterial probiotics: < 1 yr: 150 mg	*Saccharomyces boulardii*: N= 100 6 mo- 3 yrs: 61%	Children of both genders from 6 months to 5 years of age, suffering from	Efficacy rate of both probiotics was assessed as per operational definition	Comparison of efficacy: chisquare	Non-Europeaun study population (Pakistan) Unblinded

Type (aim) and objective(s) of Study **Reference**	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen;** **Route of** **Administration** **Duration of treatment**	**Number of** **Subjects (including age, sex, drop out)**	**Healthy** **Subjects or** **Diagnosis of** **Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI 95%) Quality score** e.g. Jadad score	**Comments on clinical relevance of results**
		>2 yrs: 250 mg Each divided in 2 daily doses All patients: i.v. antibiotics (ceftrioxne) + oral rehydration treatment duration: 5 days	4-5 yrs: 39% Male: 48% Female: 52% Lactic acid: N= 100 6 mo- 3 yrs: 62% 4-5 yrs: 39% Male: 57% Female: 43%	diarrhoea (less than 14 days) (diarrhoea: ≥3 unformed or loose stools in the last 24 h)	(frequency and consistency of stools/day) *Saccharomyces boulardii*: 45% Lactic acid producing bacterial probiotics: 26% P= 0.004	Post-stratification chi-square (age, gender, gestational weight, duration of diarhoea, socioeconomic status)	Incomplete methodology, Dosage inconsistent, concomitant administration of antibiotics in both groups *Saccharomyces boulardii* not further specified

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							significant results

Meta-analyses

Szajewska *et al.* (2007) performed a meta-analysis in order to assess the effectiveness of *Saccharomyces boulardii* in treating acute infectious diarrhoea in children. Five randomized-controlled trials (310 participants in the verum group and 309 in the control group) met the inclusion criteria: Billoo *et al.* (2006), Cetina-Sauri and Sierra Basto (1994), Hafeez *et al.* (2002), Kurugöl and Koturoglu (2005) and Villarruel *et al.* (2007; not published up to the time of meta-analysis). Combined data from four randomized-controlled trials showed that *Saccharomyces boulardii* significantly reduced the duration of diarrhoea compared with control (Weighted mean difference (WMD) -1.1 day, 95 % CI: -1.3 t0 -0.83). Cetina-Sauri and Sierra Basto (1994) did not report the duration of intervention. Four studies (not Kurugöl *et al.* (2005)) provided information of stool frequency at various time intervals. The meta-analysis showed a reduction in the frequency of diarrhoea for those treated with *Saccharomyces boulardii* compared with the control at all-time intervals studied, except on day 1. The authors concluded that there exists a moderate clinical benefit of *Saccharomyces boulardii* therapy in otherwise healthy infants and children with acute gastroenteritis, mainly a shorter duration of diarrhoea. However, it should be noted that studies with methodological limitations are included in this meta-analysis and that the included studies were carried out mainly in non-European countries.

Vandenplas *et al.* (2008) updated the above mentioned meta-analysis including Canani *et al.* (2007) and Htwe *et al.* (2008). Based on the pooled results of six RCTs involving 756 children (treatment group: n=377 – control group: n=379), *Saccharomyces boulardii* as compared to placebo or no intervention, reduced the duration of diarrhoea by 0.92 day (22 h; weighted mean difference (WMD) -0.92, CI -1.11 to -0.74).

Szajewska and Skȯrka (2009) updated their meta-analysis 2007 including Canani *et al.* (2007), Htwe *et al.* (2008), and Vandenplas *et al.* (2007). The meta-analysis of seven RCTs (treatment group: n=470 – control group: n=474) showed a reduction in the duration of the diarrhoea (WMD -1.08 day, 95% CI -1.64 to -0.53, random effects model) in those treated with *Saccharomyces boulardii* as compared with placebo.

Pan *et al.* (2012) made a systemic review about clinical random control trials (RCTs) focused on *Saccharomyces boulardii* in treating acute childhood diarrhoea. The following selection criteria were used to identify published studies for inclusion in the meta-analysis: study design – RCTs; population - children with acute diarrhoea; intervention - *Saccharomyces boulardii* versus placebo or no additional intervention; outcome variable – duration of diarrhoea, stool frequency and adverse effects. Eight articles (Billoo *et al.* (2006), Canani *et al.* (2007), Hafeez *et al.* (2002), Htwe *et al.* (2008), Ji *et al.* (2009), Kurugöl and Koturoglu (2005), Shen *et al.* (2008) , Villarruel *et al.* (2007)) were included: 978 children with acute diarrhoea (*Saccharomyces boulardii* group: 487; control group: 491). According to the Jadad score, Billoo *et al.* (2006), Canani *et al.* (2007), Hafeez *et al.* (2002), Htwe *et al.* (2008), Kurugöl and Koturoglu (2005), and Villarruel *et al.* (2007) were regarded as high quality literature (Jadad score 4), and Ji *et al.* (2009) and Shen *et al.* (2008) as low quality literature (Jadad score 2). A meta-analysis of the 8 studies showed a reduction in the duration of the diarrhoea (MD: -0.92 day, 95 % CI: -1.32 to -0.52) for those treated with *Saccharomyces boulardii* as compared with placebo. The included studies were heterogeneous. Five studies (Billoo *et al.* (2006), Hafeez *et al.* (2002), Ji *et al.* (2009), Shen *et al.* (2008), Villarruel *et al.* (2007)) provided information of stool frequency at various time intervals. The analyses showed a reduction in stool frequency of stools for those treated with *S. boulardi* as compared with the control on day 3, day 4 and day 7. Adverse effects associated with *Saccharomyces boulardii* were not reported. The authors concluded that the therapeutic effects of *Saccharomyces boulardii* was demonstrated in children with acute diarrhoea, but the clinical trials included were of small samples with methodological limitations.

Dinleyici *et al*. (2012) performed a meta-analysis which studied 11 RCTs with a total of 1306 children (651 in the *Saccharomyces boulardii* group and 655 as controls). According to the authors all of these studies have been performed with the same *Saccharomyces boulardii* strain by the same company (*Saccharomyces cerevisiae* CBS 5926). The included studies are Billoo *et al.* (2006), Canani *et al.* (2007), Dalgic *et al.* (2011), Eren *et al.* (2010), Grandy *et al.* (2010), Hafeez *et al.* (2002), Htwe *et al.* (2008), Kurugöl and Koturoglu (2005), Riaz *et al.* (2012), Vandenplas *et al.* (2007), and Villaruel *et al.* (2007). *Saccharomyces boulardii* significantly reduced the duration of acute infectious diarrhoea as compared with controls. The pooled weighted mean difference (WMD) was -0.99 days (approximately 24 h, 95% CI: -1.40 to -0.58). Based on the results of nine RCTs involving 1128 children (Cetina-Sauri and Basto (1994), Corrêa *et al.* (2011), Eren *et al.* (2010), Hafeez *et al.* (2002), Htwe *et al.* (2008), Kurugöl and Koturoglu (2005), Riaz *et al.* (2012), Vandenplas *et al.* (2007), Villaruel *et al.* (2007)) *Saccharomyces boulardii* could significantly reduce the risk of dirrhoea on the third day of illness (RR: 0.52; 95% CI: 0.42-0.65).The authors stated that all included trials had a number of methodological limitations e.g. small sample size, different definition of diarrhoea. However, more than 80% of these studies have a follow-up and intention-to treat analysis. They concluded, nevertheless, that this analysis gives a strong evidence that *Saccharomyces boulardii* has a clinically significant benefit in the treatment of acute infections diarrhoea in infants and children. This benefit has been replicated worldwide and shown in developed and developing countries. Furthermore, treatment with *Saccharomyces boulardii* is safe in children with acute diarrhoea.

This statement is further supported by another systematic review and network meta-analysis by Florez *et al.* (2018). The aim was to determine the comparative effectiveness and safety of the pharmacological and nutritional interventions for reducing the duration of acute diarrhoea and gastroenteritis in children. For this purpose, a total of 174 studies (32,430 children) proved eligible. Studies were conducted in 42 countries of which most were low-and middle-income countries (LMIC). Most interventions analyzed (except vitamin A, micronutrients (MN), prebiotics, and kaolin-pectin) showed evidence of superiority to placebo in reducing diarrhoea. With moderate-to high-quality of evidence, *Saccharomyces boulardii* +Zinc and smectite + zinc demonstrated the best combination of evidence quality and magnitude of effect while symbiotics, loperamide and zinc proved being the best single interventions, and loperamide was the most unsafe. According to the authors the effect of zinc, *Saccharomyces boulardii* + zinc and smectite + zinc might only be applied to children in LMIC. Results suggest no further role for studies comparing interventions against no treatment or placebo, or studies testing loperamide, MN, kaolin-pectin, vitamin A, prebiotics and diluted milk.

Padayachee *et al.* (2019) have published a systematic review to assess the efficacy and safety of *Saccharomyces boulardii* in the treatment of acute gastroenteritis (AGE) in the paediatric population. Ten of 190 articles were selected for final inclusion: Billoo 2006, Burande 2013, Correa 2011, Dalgic 2011, Erdogan 2012, Eren 2010, Htwe 2008, Kurugöl 2005, Ozkan 2007, Riaz 2012 (see table 25). Overall, the results indicate that *Saccharomyces boulardii* shortened the duration of AGE caused by rotavirus (in days), when compared with the control/placebo group, with the included studies displaying little/no heterogeneity. In addition, no adverse effects were associated with the use of this yeast probiotic in treating AGE in otherwise healthy children. Therefore, the results of the current systematic review indicate that there is a potential benefit associated with the use of *Saccharomyces boulardii* to treat AGE in the paediatric patient. However, owing to factors such as small sample sizes, unclear and inconsistent quality of methodology, and reporting bias owing to source of funding and support, a definitive conclusion and recommendation for the use of a specific probiotic like *Saccharomyces boulardii* to be used as treatment or treatment adjunct for AGE in the paediatric hospitalised patient cannot yet be made. In order to offer specific treatment guidelines, future research initiatives investigating the subject of the benefits/harm associated with the use of *Saccharomyces boulardii* must therefore endeavour to consist of larger RCTs which: minimise heterogeneity associated

with study participants enrolled, clearly predefine aetiologies, e.g. GE or AGE, minimise methodological variability (e.g. blinding), standardise the presentation in which the intervention is offered, and conduct single-strain probiotic investigations. In addition, secondary outcomes like length of hospital stay and costeffectiveness can also be investigated.

Uncontrolled studies

Amorissani Folquet *et al.* (2011) conducted an open, observational study in the 3 West African countries Togo, Benin and Côte d'Ivoire. 331 children aged between 1 month and 15 years old (mean age 25.6 months), presenting with acute diarrhoea (at least 3 loose or liquid stools in a 24-hour period, which started less than 4 days before enrolment) were included. In addition to a rehydration therapy *Saccharomyces boulardii* was administered for 5 days at a dose of 250 mg (no further specification given) in the morning and evening. The patients were predominantly male. 292 patients could be analysed. The mean duration of the diarrhoea after enrolment was 2.64 days (SD: 1.09). In all but 5 patients, duration of diarrhoea was less than 5 days. The mean daily number of stools on day 2 was 2.43. *Saccharomyces boulardii* was well tolerated (excellent or good) in 93% cases. One child developed mild transient constipation which did not require discontinuation of the treatment.

Prevention of AAD in children

The efficacy of *Saccharomyces boulardii* in the prevention of AAD has also been investigated in children. Turck *et al*. (2003) investigated the incidence and risk factors of oral antibiotic-associated diarrhoea in an outpatient pediatric population. 650 children with an age range from 1 month to 15.4 years were included and in 11% of them AAD occurred. An even higher rate was observed in children <2 years (18%) and the administration of amoxicillin/clavulanate had a higher risk of AAD (23%) as compared to other antibiotics.

Benhamou *et al*. (1999) investigated the efficacy of *Saccharomyces boulardii* in the prevention of antibiotic-induced diarrhoea in children (age 1 to 5 years) (n=327) receiving antibiotic treatment during 8 days because of infections of the respiratory tract. The study was controlled and double-blind. As comparative agent Diosmectite (n=289) was chosen.

Erdeve *et al.* (2004) investigated the efficacy of *Saccharomyces boulardii* in 653 children (age: 1-15 years) in a randomized, controlled clinical study. Patients were treated with the antibiotics sulbactam-ampicillin (SAM) or azithromycin (AZT) as monotherapy or in combination with *Saccharomyces boulardii* (250 mg/d). 466 patients in total completed the study and were included into the statistical evaluation: SAM monotherapy (n=117), SAM/*Saccharomyces boulardii* (n=117), AZT monotherapy (n=105), AZT/*Saccharomyces boulardii* (n=127). AAD occurred in 18.9% of the patients receiving antibiotic monotherapy and in 5.7% of the patients with the combination of antibiotic and probiotic. ($p<0.05$; chi-square). In patients receiving AZT the use of *Saccharomyces boulardii* showed no statistically significant effect on the development of diarrhoea, whereas in the SAM group a statistically significant effect was observed. While 25.6% of the children on antibiotic monotherapy developed diarrhoea, only 5.7% of the children receiving combined treatment experienced diarrhoea ($p<0.05$; chi-square). The rate of SAM-associated diarrhoea was highest in the age group 1-5 years.

In 2005 Kotowska *et al*. performed a double-blind, randomized placebo-controlled clinical trial in order to investigate, if *Saccharomyces boulardii* prevents AAD in children. A total of 269 children (age 6 months to 14 years) with otitis media and/or respiratory tract infections were included in the trial and received standard antibiotic treatment plus 250 mg of *Saccharomyces boulardii* (n=132) or a placebo (n=137) orally twice daily for the duration of antibiotic treatment. Analyses were based on allocated treatment and included data from 246 children. Patients receiving *Saccharomyces boulardii* had a lower prevalence of diarrhoea (definition: ‡3 loose or watery stools/day for ⩾48 h occurring during or up to

2 weeks after the antibiotic therapy) than those receiving placebo (8% vs. 23%, relative risk: 0.3, 95% confidence interval: 0.2–0.7). *Saccharomyces boulardii* also reduced the risk of AAD (definition: diarrhoea caused by *Clostridium difficile* or otherwise unexplained diarrhoea) compared with placebo [3.4% vs. 17.3%, relative risk: 0.2; 95% confidence interval: 0.07–0.5]. No adverse events were observed. As AAD may occur up to 2 months after the end of antibiotic treatment, the follow-up interval in this study was short and some cases of AAD may have been missed.

Shan *et al*. (2013) investigated the efficacy of *Saccharomyces boulardii* in the treatment and prevention of AAD. A total of 333 hospitalised children with acute lower respiratory tract infection (age 6 months to 14 years) were enrolled in a 2-phase open randomized controlled trial. During the 1st phase, all children received intravenous antibiotics. They were randomly allocated to antibiotic treatment alone (B: n=166) or combination treatment with antibiotic and *Saccharomyces boulardii (*500 mg/day, A: n=167) and followed for 2 weeks. Diarrhoea was defined as ⩾3 loose/watery stools/day during at least 2 days, occurring during treatment and/or up to 2 weeks after antibiotic therapy had stopped. AAD was considered when diarrhoea was caused by *Clostridium difficile* or when stool cultures remained negative. In the 2nd phase of the study patients from group B who were treated with the antibiotic alone and developed diarrhoea were randomly allocated to two sub-groups: one group B1 (*Saccharomyces boulardii* + oral rehydration solution (ORS)) and group B2 (ORS alone). Data from 283 patients were available for analysis. Diarrhoea prevalence was lower in group A than in group B (7.9% vs. 29.2%; relative risk (RR): 0.27, 95% confidence interval (CI): 0.1-0.5). *Saccharomyces boulardii* reduced the risk of AAD (4.3% vs. 19.4%; RR: 0.22; 95% CI: 0.1-0.5). When group B patients developed diarrhoea (n=42), *Saccharomyces boulardii* treatment during 5 days (group B1) resulted in lower stool frequency ($P<0.05$) and higher recovery rate (91.3% in group B1 vs. 21.1% in B2; $P<0.001$). The mean duration of diarrhoea in group B1 was shorter (2.31±0.95 vs. 8.97±1.07 days; $P<0.001$). No adverse effects related to *Saccharomyces boulardii* were observed.

Casem (2013) performed a randomized clinical trial between June and October 2012 in the Philippines. 140 patients aged 6 months to 18 years with paediatric community acquired pneumonia (PCAP) and on IV or oral antibiotics within 24 hours of enrolment received either the standard antibiotic treatment alone (control n=71, aged 3.54±3.42 years) or antibiotic treatment plus 250 mg *Saccharomyces boulardii* (treatment group n=69, aged 4.19±3.09) twice a day for the entire duration of treatment. Analyses were based on treatment. 16 patients from the control group and 11 patients from the treatment group presented diarrhoea without reaching statistical relevance (p=0.391). The treatment group had a shorter duration of diarrhoea than the control group (p=0.032). *Saccharomyces boulardii* was generally well tolerated and there was no documented or reported adverse event.

A randomized, open, parallel study was conducted in an Indian tertiary care hospital by Jindal *et al*. (2017). 600 children in the age group of 6 months –12 years, receiving beta lactam antibiotics for various ailments like otitis media, tonsillitis, urinary tract infections etc. were included in the study. All these children did not have diarrhoea. 300 out of 600 children were also given *Saccharomyces boulardii* sachets b.i.d. (4-6 billions CFUs daily) for 7 days along with beta lactam antibiotic. 72 [24%] out of 300 patients in the control group developed diarrhoea whereas only 16 [5.3%] out of 300 in the treatment group developed diarrhoea. The results were stastistically significant [$p < 0.001$] as calculated by Chi-Square test.

In a meta-analysis, Johnston *et al*. (2007) investigated the efficacy and safety of probiotics for the prevention of AAD. Clinical studies included in their meta-analysis were those performed by Benhamou *et al.* (1999), Erdeve *et al*. (2004), and Kotowska *et al*. (2005) (see Table below). They concluded that the current data are promising but inconclusive, and that there is insufficient evidence to recommend the use of *Saccharomyces boulardii* for co-administration at this time. According to the meta-analysis of Szajewska *et al*. (2006) on the use of probiotics in the prevention of AAD in children only the clinical

study conducted by Kotowska *et al.* (2005) was included. According to her probiotics reduce the risk of AAD in children. For every 7 patients that would develop diarrhoea during antibiotic treatment one fewer will develop AAD if also receiving probiotics.

Johnston *et al.* (2011) again reviewed the data concerning probiotics for the prevention of pediatric antibiotic-associated diarrhoea. The same clinical studies with regard to *Saccharomyces boulardii* were included in this review (Benhamou et al. (1999), Erdeve et al. (2004), and Kotowska et al.(2005) (see Table below)).
The authors now draw the following conclusion: "Despite heterogeneity in probiotic strain, dose, and duration, as well as in study quality, the overall evidence suggests a protective effect of probiotics in preventing AAD. Using 11 criteria to evaluate the credibility of the subgroup analysis on probiotic dose, the results indicate that the subgroup effect based on dose (⩾5 billion CFU/day) was credible. Based on high-dose probiotics, the number needed to treat (NNT) to prevent one case of diarrhoea is seven (NNT 7; 95% CI 6 to 10). However, a GRADE analysis indicated that the overall quality of the evidence for the primary endpoint (incidence of diarrhoea) was low due to issues with risk of bias (due to high loss to follow-up) and imprecision (sparse data, 225 events). The benefit for high dose probiotics (*Lactobacillus rhamnosus* or *Saccharomyces boulardii*) needs to be confirmed by a large well-designed randomized trial. More refined trials are also needed that test strain specific probiotics and evaluate the efficacy (e.g. incidence and duration of diarrhoea) and safety of probiotics with limited losses to follow up. It is premature to draw conclusions about the efficacy and safety of other probiotic agents for pediatric AAD. Future trials would benefit from a standard and valid outcomes to measure AAD."

Szajewski and Kolodziej (2015b) published an update of their 2005 meta-analysis. Concerning children, they evaluated Kotowska *et al*. (2005), and in addition Shan et al. (2013), Erdeve *et al*. (2004), Casem (2013), Bin *et al*. (2015), and Zhao *et al*. (2014). In children compared with placebo or no treatment, *Saccharomyces boulardii* reduced the risk of diarrhoea from 20.9% to 8.8% (RR: 0.43, 95% CI: 0.30-0.60, NNT: 9, 95 % CI: 7-12). Without Bin *et al*. (2015), and Zhao *et al*. (2014), who studied antibiotics as part of eradication therapy (s. below), RR was 0.36 (95% CI: 0.21-0.61). Subgroup analysis based on age, showed that the administration of *Saccharomyces boulardii* reduced the risk of *C. difficile*-associated diarrhoea in children (2 RCTs, n=579, RR: 0.25, 95% CI: 0.08-0.73). However, the wide confidence interval calls for caution. One major limitation is that the methodological quality of included trials varied. Only Kotowska *et al*. (2005) was at low risk of bias. Definition of AAD and/or diarrhoea differed. The optimal dose of probiotics, including *Saccharomyces boulardii*, and the duration of treatment have not been established. One important question remains according to the authors: whether the use of *Saccharomyces boulardii* shall be considered in all subjects receiving antibiotics or only in select populations. This will require clinical judgement.

A current Cochrane review (Guo *et al.* 2019) concluded that overall evidence suggests a moderate protective effect of probiotics for preventing AAD (NNTB 9, 95% CI 7 to 13). Subgroup analysis indicated that high dose (⩾5 billion CFUs per day) is more effective than low probiotic dose (<5 billion CFUs per day) with moderate certainty evidence). Besides Benhamou *et al.* (1999), Erdeve *et al*. (2004), and Kotowska *et al.(*2005), the following studies with *Saccharomyces boulardii* were included: Shan *et al*. 2013, and Jindal *et a*l. (2017); Zhao et *al. 2014* and Bin *et al.* 2015 (both Helicobacter pylorii infection).

However, the authors concluded, the benefit of high dose probiotics (e.g. *Lactobacilus rhamnosus* or *Saccharomyces boulardii*) needs to be confirmed by a large well-designed multi-centered randomized trial. Only the study of Kotowska *et al.* (2015) was categorized as low risk of bias, the other studies were categorized as high risk of bias. Adverse event rates were low and no serious adverse event were attributed to probiotics.

Table 26: Clinical studies on the prevention of acute antibiotic-associated diarrhoea (AAD) with *Saccharomyces boulardii* in children

Type (aim) and objective(s) of Study Reference	**Study Design and Type of Control Study duration (if available)**	**Test Product(s): herbal preparation, pharmaceutical form;** **Dosage** **Regimen; Route of Administration Duration of** **treatment**	**Number of Subjects (including age, sex, drop out)**	**Healthy Subjects or Diagnosis of Patients** **(inclusion criteria)**	**Outcomes (primary and secondary endpoints)**	**Statistical analysis (e.g. ITT yes/no, CI** **95%)** **Quality score** **e.g. Jadad** **score**	**Comments on clinical relevance of results**
Prevention of AAD Benhamou *et al*., 1999	randomized, controlled, double-blind	*Saccharomyces boulardii* 226 mg/d (4.5 billion cfu/d) vs. Diosmectite 6 g/day (1-2 yrs), 9 g/day (>2 yrs)	included: n=779 completed: *Saccharomyces boulardii* n= 327 Diosmectite n=289	children (1-5 yrs) with respiratory tract infection receiving antibiotics during 8 days	occurrence of diarrhoea (>3 liquid stools/d): *Saccharomyces boulardii:* 7.6% Diosmectite: 5.5%		only abstract in English length of observation period not reported Probably too short in order to allow a reliable statement with regard to a prevention of AAD high rate of withdrawals

Prevention of AAD Erdeve *et al*., 2004, 2005	randomized no details given	1. sulbactam-ampicillin + *Saccharomyces boulardii* (n=117) 2. sulbactam-ampicillin (n=117) 3. azithromycin + *Saccharomyces boulardii* (n=127) 4. azithromycin (n=105)	n=653 patients age: 1-15 yrs 466 completed	children with antibiotic treatment (ampicillin-sublactam or azithromycin)	incidence of diarrhoea: >3 watery stools/d sublactam/ampicillin: 5.7% 25.6% p<0.05	chi-square test significant difference only for the sulbactam/ ampicillin group	high rate of discontinuations: 28.7% duration of follow-up unknown
Prevention of AAD Kotowska *et al*., 2005	double-blind, randomized, placebo-controlled	antibiotic therapy with 250 mg *Saccharomyces boulardii* b.i.d. vs. antibiotic therapy with placebo	*Saccharomyces boulardii* group n=132 mean age: 58.8 mo placebo: n=137 mean age: 55.8 mo	children (6 mo – 14 yrs) with acute otitis media and short-term antibiotic treatment within 24 h of enrolment, out- and in-patients	frequency of diarrhoea (≥3 watery/loose stools per day for ≥48 h occurring during or 2 weeks after antibiotic therapy) and AAD (*C. difficile*, unexplained) *Saccharomyces*	chi-square test, Fisher exact test significant reduction of diarrhoea caused by amoxicillin/ clavunate and cefuroxime i.v.	only 2 weeks of follow-up discontinuations: *Saccharomyces boulardii* n=13 placebo n=10

Prevention of AAD Shan *et al*., 2013	open, randomized, controlled follow-up: 2 weeks after the end of antibiotic therapy	group A: 2 x 250 mg *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926) during antibiotic therapy group B:	group A: n=167 mean age: 49.8 mo group B: n=166 mean age: 48.7 mo	hospitalised children (6 mo – 14 yrs) with acute infections of lower respiratory tract requiring iv antibiotic therapy	incidence of diarrhoea (≥3 watery/loose stools per day for ≥48 h occurring during or 2 weeks after antibiotic therapy) and AAD (C. *difficile*, unexplained) group A: 7.9% (diarrhoea) 4.3% (AAD)	chi-square test significant reduction of diarrhoea caused by amoxicillin/ clavunate and i.v. cefuroxime	open design follow–up of 2 weeks only wide-age range mainly younger children affected by diarrhoea
Prevention of AAD Casem 2013	Randomized, controlled	Treatment group: 250 mg *Saccharomyces boulardii* b.i.d. in addition to antibiotics Control group: antibiotics only	Treatment group: N=69 Drop out: 3 Control group N=71 Drop out 3	patients aged 6 months to 18 years with paediatric community acquired pneumonia (PCAP) and on IV or oral antibiotics within 24 hours of enrolement	Primary outcome: 16 patients from the control group and 11 patients from the treatment group presented diarrhea (3 or more loose or watery stools (MBSFS 4 or 5) per day, which lasts for a minimum of 48 hours) without reaching statistical relevance (p=0.391).	Chi-Square test or Fisher exact test was used to compare differences between groups. All statistical tests were two-tailed and were performed at the 5 % level of significance. A P value of <0.05 % was considered significant.	Open, not placebo controlled design follow–up of 2 weeks only wide-age range performed in the Philippines

					Secondary outcome: The treatment group had a shorter duration of diarrhoea than the control group (p=0.032). MBSFS=modified Bristol stool form scale		
Prevention of AAD Jindal 2017	Randomized, open, parallel study in Indian out-patients	Treatment group: 250 mg *Saccharomyces boulardii* b.i.d. (4-6 billions CFUs daily) for 7 days and antibiotics (Co-amoxyclav, Cefpodoxime, Cefdinir ,Cefixime and Cephalaxin) Control group: antibiotics only	Treatment group: N=300 Control group: N=300	Children aged 6 months to 12 years with urinary tract infection, otitis media, tonsillitis	Treatment group: 16 [5.3%] diarrhoea. Subgroups (each 60 patients): Amoxyclav = 2 Cefpodoxime = 4 Cephalaxin = 2 Cefixime = 8 Cefdinir = 0 Control group: 72 [24%] diarrhoea Subgroups (each 60 patients): Amoxyclav = 26 Cefpodoxime = 12 Cephalaxin = 14	SPPS software version 17.01 using Chi-Square test	Open, not placebo controlled design Performed in India No definition of diarrhoea is given Follow-up is not defined Beta-lactam antibiotics

					Cefixime = 16 Cefdinir = 4 p < 0.001		

Prevention of acute antibiotic-associated diarrhoea (AAD) caused by triple therapy of *Helicobacter pylori* infection in children

Clinical studies have been performed in children with *Saccharomyces boulardii* in addition to standard triple therapy for eradication of *H. pylori*.

The randomized, open clinical study by Hurduc *et al*. (2009) investigated the efficacy of *Saccharomyces boulardii* on the eradication rate of *H. pylori*. *H. pylori* infection was identified in 90 of 145 children (62%) and it correlated positively with age ($p < 0.002$) and inversely with socioeconomic status ($p < 0.005$). These 90 children (range 3 – 18 years) received standard triple treatment (omeprazole/ esomeprazole, amoxicillin, clarithromycin) for 7-10 day. The intervention group (48 patients) received additional therapy with *Saccharomyces boulardii* 250 mg b.i.d. Apart from efficacy the rate of side-effects was investigated and in the *Saccharomyces boulardii* group a significant reduction of the incidence of adverse reactions was observed (30.9% vs. 8.3%). The incidence of AAD resp. diarrhoea is not stated.

In an open randomized clinical study Bin *et al.* (2015) randomized 194 *H. pylori* positive children (age: 22 months to 16 years) to triple therapy (omeprazole, clarithromycin, amoxicillin or in case of penicillin allergy omeprazole, clarithromycin, metronidazole) alone (n=92) or triple therapy plus *Saccharomyces boulardii* (n=102). *Saccharomyces boulardii* was administered at a dosage of 2 x 250 mg/day (*Saccharomyces cerevisiae* CBS 5926). The incidence of diarrhoea was the primary outcome of the study. Diarrhoea was defined as an increase in the frequency of bowel movements (>3/day) or decrease in stool consistency (Bristol stool scale 5 or 6). In case of diarrhoea montmorillonite powder, a natural clay (3 g three times daily) was administered orally to patients in either group without interrupting treatment with *Saccharomyces boulardii*. In the group receiving concomitant treatment with *Saccharomyces boulardii* 12 children (11.76%) experienced diarrhoea, in the control group with triple therapy alone 28.26% of the children (n=26). This difference was statistically significant ($p<0.05$).

In their meta-analyses on *Saccharomyces boulardii* as concomitant therapy during eradication therapy for *H. pylori* Szajewska *et al.* (2010 and 2015a) the number of children included in clinical studies were assessed as not sufficient. Since then, only one clinical study (Bin *et al.* 2015) has been published. As this study, however, has been conducted in China it remains open, if the data presented also apply for European children.

Feng *et al.* (2017) aimed to identify the best probiotic supplementation in triple therapy for pediatric population with *Helicobacter pylori* infection in a systematic review and network meta-analysis. Children without symptoms and with gastrointestinal symptoms have been reported to have a seroprevalence rate of 15.7 and 40%, respectively. Twenty-nine trials (3122 participants) involving 17 probiotics regimens were identified among them five trials with *Saccharomyces boulardii* (Zhang 2013 (publication not available), Zhang 2012 (publication not available), Zhao *et al*. 2014 [in Chinese, only abstract in English], Zhang 2015 (seems to be Bin *et al.* 2015), Hurduc *et al.* (2015). Compared with placebo, probiotic-supplemented triple therapy significantly increased *H. pylori* eradication rates and reduced the incidence of total side effects. However, there were several limitations to the meta-analysis, e.g. diversity of antibiotics in triple therapy, confirmation of *H. pylori* eradication, different administration time of probiotics, and different design of trials.

***Clostridium difficile*-associated disease (CDD)**

Over a period of 10 consecutive months Buts *et al.* (1993) studied 19 eligible children (7 boys, 12 girls; median age 8 months; 2 months to 11 years) who presented with enteral symptoms lasting for >15 days and who had solely *C. difficile* in stools with positive cytotoxin B assay. *Saccharomyces boulardii* (ATCC 74012, *Saccharomyces cerevisiae* CBS 5926) was given orally in a lyophilized form over 15 days

(250 mg 2 x per day for infants <1year, 3 x per day for children 1-4 years of age, and 4 x per day for those >4 years of age). Within 1 week of treatment, enteral symptoms and physical findings resolved in 18 patients (95%) with marked decreases ($p<0.001$) in the number of stools, frequency of colic episodes, and total duration of colics per day. Clearing of toxin B was observed within 15 days of therapy in 16 cases (85%), whereas eradication of *C. difficile* from stools was complete after 1 month in 14 (73%). A clinical and bacteriological relapse occurred in two patients (11%), which resolved rapidly with a second 15-day course of *Saccharomyces boulardii*.

Other kinds of diarrhoea in children

Gaon *et al.* (2003) evaluated the effect of *Lactobacillus* and *Saccharomyces* on persistent diarrhoea (more than 3 stools per day for the last consecutive 14 days or more) in Argentine children in a randomized, double blind, placebo controlled study. Eighty-nine children, aged 6-24 months were randomly distributed to receive pasteurized cow milk containing *Lactobacillus* (10^{10}-10^{12} CFU/g, N=30), or lyophilized *Saccharomyces boulardii* (reconstituted in sterile distilled water at a concentration of 0.1 g/ml (1 g powder per ml contained 10^{10} CFU of *Saccharomyces boulardii*), n=30) or pasteurized cow milk as placebo (n=29); on each diet 175 g was given twice a day for a 5 day period. Patients with mild or moderate dehydration were rehydrated *ad libitum* orally or by gastric tube for 4 to 6 h before starting the study. Number of stools, duration of illness and frequency of vomiting were considered. Enteric pathogens were isolated from stools in 40% of the patients, 27% had rotavirus. *Lactobacillus* and *Saccharomyces* significantly reduced the number of stools on day 5 ($p<0.001$) and diarrhoeal duration ($p<0.005$). Similarly both significantly ($p<0.002$) reduced vomiting as compared with placebo. There was no difference between treatments depending on rotavirus status. In conclusion, *Lactobacillus* and *Saccharomyces* were both effective in the management of persistent diarrhoea in children.

Savaş-Erdeve *et al.* (2009) assessed the efficacy and safety of adding *Saccharomyces boulardii* to antibiotic treatment for amebiasis-associated acute diarrhoea in Turkish children aged from 1 to 15 years. 45 children in group I received metronidazole orally for 10 days, while 45 children in group II additionally received 250 mg lyophilized *Saccharomyces boulardii* (, 5 x 10^6 living microorganisms, *Saccharomyces cerevisiae* CBS 5926). The median duration of acute diarrhoea was 5 (1-10) days in group I and 4.5 (1-10) in group II ($p=0.965$). The median number on stools on follow-up and duration of bloody diarrhoea, fever, abdominal pain and vomiting were similar in the two groups. *Saccharomyces boulardii* was well tolerated withot any side effects. Addition of *Saccharomyces boulardii* does not seem to be more effective than metronidazole alone.

Dinleyici *et al.* (2009) evaluated the clinical efficacy of *Saccharomyces boulardii* in addition to metronidazole as compared to metronidazole alone in 50 Turkish children with acute bloody diarrhoea caused by amebiasis in a prospective, randomized, open label study. Group A and B (each n=25) were treated with metronidazole (30 mg/kg twice daily), but *Saccharomyces boulardii* (250 mg twice daily, *Saccharomyces cerevisiae* CBS 5926) during the 7 days was added to group B patients. Group A was composed of 25 children (12 girls, 13 boys, mean age 11.7±2.1 years), group B of 25 children, too (14 girls, 11 boys, mean age 10.9±2.2 years). Duration of bloody diarrhoea was significantly longer in group A (72.0±28.5 vs. 42.2±17.4 h, $p<0.001$). On day 5, amebic cysts had disappeared in all children in group B, whereas in group A, amebic cysts were still present in 6 children ($p<0.05$). On day 10, all children were cured and cysts had disappeared in all. The addition of *Saccharomyces boulardii* to metronidazole in amebiasis significantly decreased duration of (bloody) diarrhoea and enhanced clearance of cysts.

Dinleyici *et al.* (2011) compared the natural evolution of diarrhoea (no treatment – group C) to the efficacy of *Saccharomyces boulardii* (250 mg b.i.d, *Saccharomyces cerevisiae* CBS 5926 – group B) or metronidazole (group A) (30 mg/kg b.i.d) in Turkish children with gastrointestinal symptoms

(abdominal pain, diarrhoea, nausea-vomiting, flatulence) for more than two weeks and positive stool examination for *Blastocystis hominis* in a randomized single-blinded clinical trial. Group A was composed of 15 children (7 girls, 8 boys, mean age 94.6±37.4 months), group B of 18 children (7 girls, 11 boys, mean age 99.1±43.8 months) and group C of 15 children (8 girls, 7 boys, mean age 90.2±46.7). The primary end points were clinical evaluation and result of microscopic stool examination at day 15. Clinical cure was observed in 77.7% in group B; in 66.6% in group A; and in 40% in control group C (p<0.031, between *Saccharomyces boulardii* and control). Disappearance of the cysts from the stools on day 15 was 80% in group A, 72.2% in group B, and26.6% in group C (p=0.011 between group A and C; p=0.013 between group B and C). At the end of the first month after inclusion, clinical cure rate was 94.4% in group B and 73.3% in group A (p=0.11): Parasitological cure rate for *B. hominis* was very comparable between both groups (94.4% vs. 93.3%, p=0.43).

Overall Review / Meta-analysis

Feizizadeh *et al*., 2014 showed in their review and meta-analysis that *Saccharomyces boulardii* is safe and has clear beneficial effects in children with acute diarrhoea when administered in addition to rehydration therapy. However, the authors stated that additional studies using head-to-head comparisons are needed to define the best dosage of *Saccharomyces boulardii* for diarrhoea with different causes. The authors identified 22 articles, which met the inclusion criteria, although these studies varied in the definition and cause of diarrhoea, the termination of diarrhoea, inclusion and exclusion criteria, their methodological quality and the reported outcomes. 2440 patients between 1 month to 15 years of age were included (1225 interventions, 1215 controls). For most of the studies, the daily dosage of *Saccharomyces boulardii* was 250-750 mg (10^9 to 10^{10} cfu). Duration of intervention was 5 to 10 days. The pooled data of 17 studies, which reported duration of diarrhoea (Cetina-Sauri and Sierra Basto, 1994; Urganci *et al.*, 2001; Hafeez *et al.* , 2002; Kurugöl and Koturoglu, 2005; Billoo *et al.*, 2006; Canani *et al.*, 2007; Vandenplas *et al.*, 2007; Villarruel *et al.*, 2007; Htwe *et al.*, 2008; Savas-Erdeve *et al.*, 2009; Dinleycici *et al.*, 2009; Grandy *et al.*, 2010; Dalgic *et al.*, 2011; Erdoğan *et al.* , 2012; Khan *et al.*, 2012; Riaz *et al.*, 2012; Burande, 2013), showed that *Saccharomyces boulardii* significantly reduced the duration of diarrhoea (mean difference [MD], -19.7 h, 95% confidence interval [CD], -26.05 to -13.34). Subgroup analysis according to cause of diarrhoea showed the duration of diarrhoea was reduced in all 3 subgroups, including rotavirus, *Entamoeba histolytica*, and nonspecific cause. Subgroup analysis based on hospitalisation indicated that using *Saccharomyces boulardii* reduced duration of mild diarrhoea more than severe diarrhoea.

The European Society for Pediatric Gastroenterology, Hepatology, and Nutrition and the European Society for Pediatric Infectious Diseases updated 2014 the evidence-based guidelines for the management of acute gastroenteritis (AGE) in children in Europe (Guarino *et al*., 2014). Concerning treatment oral rehydration therapy should be used as first-line therapy for the management of children with AGE (strong recommendation, moderate-quality evidence). With regard to probiotics, these guidelines endorse the document developed by the ESPGHAN Working Group on Probiotics and Prebiotics (Szajewska *et al*., 2014), which provided recommendations for the use of probiotics for the treatment of AGE in infants and children. Probiotics reduced the duration of approximately 1 day. However, probiotic effects are strain-specific, so the safety and clinical effects of one probiotic microorganism should not be extrapolated to other probiotic microorganisms. The use of the following probiotics may be considered in the management of children with AGE in addition to rehydration therapy: *L. rhamnosus* GG (low-quality evidence, string recommendation), *Saccharomyces boulardii* (low-quality evidence, strong recommendation), based on a consistent amount of evidence in various settings.

Irritable bowel syndrome in children (IBS)

Chouraqui *et al*. (1995, abstract) investigated the effectiveness of *Saccharomyces boulardii* at a dosage of 25 mg/kg b.w./d (*Saccharomyces cerevisiae* CBS 5926) versus placebo treatment in infants with irritable bowel syndrome who did not improve after one month of dietary modifications. Disease severity was rated by means of a clinical score including gastrointestinal symptoms and signs. During the first month of treatment, the infants were randomly assigned to *Saccharomyces boulardii* or placebo. For the following 2 months, all patients received *Saccharomyces boulardii*. 41 children (mean age: 22.7 months) were elegible, of whom 20 responded to dietary modifications. 18 of the remaining 21 patients were included in the study (*Saccharomyces boulardii*: n=8; placebo: n=10). After the first month of treatment the clinical score improved by 59% with *Saccharomyces boulardii* and 24% with placebo ($p<0.01$). Two months later, when both groups received *Saccharomyces boulardii*, no difference was observed. According to the authors, this trial demonstrates the effectiveness of oral *Saccharomyces boulardii* in the treatment of infants with IBS who do not respond to dietary modifications alone. *Saccharomyces boulardii* seems to be effective after one month of administration. Due to the low number of patients included and missing details of the study (e.g. diagnostic criteria, statistical evaluation, clinical score) it cannot be accepted as evidence that Saccharomyces boulardii is effective in the treatment of IBS in children.

4.4. Overall conclusions on clinical pharmacology and efficacy

Pharmacology

Based on bibliographic data a protective mechanism of *Saccharomyces cerevisiae* CBS 5926 against pathogens causing diarrhoea is supposed. Experimental data suggest different mechanisms (luminal and trophic actions as well as mucosal-antiinflammatory signaling effects). It is assumed that the effects are tied to the viability of the yeast cells.

In a review Terciolo *et al*. (2019) summarized possible the impact of *Saccharomyces boulardii* on various gastrointestinal and systemic diseases associated with intestinal epithelial barrier defects. Through anti-inflammatory, anti-secretion, pro-migratory and adhesive effects, Saccharomyces boulardii preserves and restores intestinal barrier function. Possibly, a yeast-induced general metabolic activation may enhance barrier function by the acceleration of enterocyte turnover.

Human pharmacokinetic studies show that following oral administration of *Saccharomyces boulardii* steady-state levels are reached within 3 days. With a half-life of 6 h in healthy volunteers, *Saccharomyces boulardii* is eliminated from the gastrointestinal tract within 7 days after cessation of administration. There was no evidence that cells of *Saccharomyces cerevisiae* CBS 5926 cross the gastrointestinal wall in healthy humans when administered orally.

Acute unspecific diarrhoea

The use of *Saccharomyces boulardii* has to be regarded as well-established according to the Directive 2001/83/EC. In addition, the period of 30 years required for the establishment of a Community Herbal Monographs / Entries to the Community List for traditional herbal medicinal products / substances / preparations (EMEA/HMPC/104613/2005) has elapsed for the preparation containing $>1.8 \times 10^{10}$ viable cells/g lyophilisate.

One randomized double-blind, placebo-controlled, multicenter clinical trial was conducted in adults with a positive statistically significant result for the treatment group (Höchter *et al.*, 1990). The daily dose was 3 x 200 mg for the first 2 days and then 3 x 100 mg *Saccharomyces cerevisiae* CBS 5926 *($\geq 1.8 \times 10^{10}$ viable cells/g)*. A second randomized controlled study in adults compared two varying *Saccharomyces* preparations (*Saccharomyces cerevisiae* CBS 5926 (at least 10^{10} live micro-organisms/g according to the reference), however, without a placebo control.

Numerous randomized studies were conducted in children all over the world. Only in the study of Canani *et al.* (2007) *Saccharomyces boulardii* had no effect on duration of diarrhoea, stool outputs and consistency as compared to rehydration therapy alone. In the study of Dalgic *et al.* (2011) *Saccharomyces boulardii* had only an effect on diarrhoea duration when given together with zinc.

Table 27: Summary of clinical studies in children with acute unspecific diarrhoea

Study	**Country**	**Age**	**Patient number under *Saccharomyces***	**Patient number under control**	**Daily dose of *Saccharomyces***
Chapoy, 1985 / 1986	France	2 weeks – 30 months	19	19 only rehydration	2 x 250 mg (*Saccharomyces cerevisiae* CBS 5926 >1.8 x 10^{10} viable cells/g lyophilisate)
Cetina-Sauri and Sierra Basto, 1989 / 1991	Mexico	3 months – 3 years	65	65 placebo	3 x 200 mg (*Saccharomyces cerevisiae* CBS 5926; 2 x 10^{10} cells/g)
Urganci *et al. 2001*	Turkey	2 - 29 months	50	50 placebo	250 mg *Saccharomyces cerevisiae* CBS 5926 (≥1.8 x 10^{10} viable cells/g)
Biloo *et al.*, 2006	Pakistan	2 months - 12 years	50 Mean age 18.3 ± 20.33 months	50 only rehydration mean age 26.01±23.37 months	2 x 250 mg (*Saccharomyces cerevisiae* CBS 5926,about 2 x 10^{10} alive cells/g)
Corrêa, 2011	Brasilia	6 – 48 months	95	91 placebo	2 x 200 mg (*Saccharomyces cerevisiae* CBS 5926, 2 x 10^{10} cells/g according to reference)
Kurugöl and Koturoglu, 2005	Turkey	3 months - 7 years	100	100 placebo	250 mg (*Saccharomyces cerevisiae* CBS 5926, number of cells unknown)

Ozkan *et al.*, 2007	Turkey	6 months – 10 years	16	10 placebo	2 x 250 mg (*Saccharomyces cerevisiae* CBS 5926, number of cells unknown)
Villarruel *et al.*, 2007	? Argentina / Belgium	3 – 24 months	44	44 placebo	<1 year:1 x 250 mg >1 year:2 x 250 mg
Vandenplas *et al.*, 2007	India/ Indonesia	< 3 years (age range 3-33 months)	93	95	500 mg/day
Htwe *et al.*, 2008	Myanmar	3 months - 10 years	50	50 only rehydration	2 x 250 mg
Shen, 2008	China	1 month – 8 years	75	62 dioctahedral smectite	?
Ji *et al.*, 2009	China	2 months – 7 years	46	46 only conventional therapy	?
Grandy *et al.*, 2010	Bolivia	< 2 years	20	20 placebo 20 combination of probiotics	2 x 4 x 10^{10} lyophilized cells/dose
Eren *et al.*, 2010	Turkey	5 months – 16 years (mean age 21,2 ± 28.2 months)	28	27 yogurt fluid	<2 years: 2 x 125 mg >2 years: 2 x 250 mg *Saccharomyces cerevisiae* CBS 5926, number of cells unknown)
Le Luyer *et al.*, 2010	? France / Lebanon	1 month – 9 months	38	39 standard formula	156 mg
Riaz *et al.*, 2012	India	3 months – 5 years	54	54 placebo	2 x 250 mg

					(*Saccharomyces cerevisiae* CBS 5926, number of cells unknown)
Erdoğan *et al.*, 2012	Turkey	5 months – 5 years	25	25 *Bifidobacterium lactis* *25 rehydration therapy only*	282.5 mg/d (*S. cerevisae* CBS 5926, number of cells unknown)
Khan *et al.*, 2012	Pakistan	2 months -5 years	210	210 rehydration therapy only	250 mg BD
Burande, 2013	India	<6 months - ?	35	35 rehydration therapy and zinc only	2 x 250 mg
Shaikh *et al.*, 2015	Pakistan	3 months – 5 years	50 + low osmolar ORS, zinc	50 low osmolar ORS and zinc	2 x 250 mg
Das *et al.* 2016	India	3 months – 5 years	30	30 placebo	2 x 250 mg *Saccharomyces boulardii* in lyophilized powdered form
Asmat *et al.* 2018	Pakistan	6 months – 5 years	100 (61: 6 months – 3 years; 39: 4-5 years) in addition to i.v. antibiotics (ceftrioxne) + oral rehydration	100 lactic acid producing probiotics in addition to i.v. antibiotics (ceftrioxne) + oral rehydration	150 – 250 mg *Saccharomyces boulardii*
Canani *et al.*, 2007	Italy	3 – 36 months	87	91 only rehydration 98 *Lactobacillus*	2 x 250 mg (*Saccharomyces cerevisiae* CBS 5926; >2 x 10^{10}

				100 *Bacillus clausii* 94 mix	cells/g lyophilisate)
Dalgic *et al.*, 2011	Turkey	1 - 28 months	240 (60 *S. bouilardii* alone, 60 *S. bouilardii* + zinc 60 *S. bouilardii* + lactose-free formula 60 *S. bouilardii* + zinc + lactose-free formula	60 zinc alone, 60 lactose-free formula alone 60 zinc + lactose-free formula 60 only oral and/or parenteral rehydration	1 x 250 mg

Neglecting Canani *et al.*, 2007 and Dalgic *et al.*, 2011, nine placebo controlled, nine controlled (rehydration or standard therapy) and three active controlled studies were conducted. All together 1293 children between 1 months and 16 years of age received *Saccharomyces boulardii* (485 children between 1 month and 3 years of age; 554 children between 2 months and 7 years; 191 children between 2 months and 12 years; 28 children between 5 months and 16 years, Burande 2013 35 children with unspecified age). The administered daily dose was 150 mg to 500 mg *Saccharomyces.* Predominantly 2 x 250 mg was given.

The meta-analysis (Szajewska and Skórka, 2009) of seven RCTs (treatment group: n=470 – control group: n=474) showed a reduction in the duration of the diarrhoea (WMD -1.08 day, 95% CI -1.64 to -0.53, random effects model) in those treated with *Saccharomyces boulardii* compared with placebo.

A meta-analysis (Pan *et al.*, 2012) of 8 studies showed a reduction in the duration of the diarrhoea (MD: -0.92 day, 95 % CI: -1.32 to -0.52) for those treated with *Saccharomyces boulardii* compared with placebo.

Feizizadeh *et al*., 2014 showed in their review and meta-analysis that *Saccharomyces boulardii* is safe and has clear beneficial effects in children who have acute diarrhoea when administered in addition to rehydration therapy.

In their review Guarino et al. (2015) draw the conclusion that acute gastroenteritis (diarrhoea) is the original and probably the best established indication for probiotics and that they have obtained "conclusive" evidence of efficacy of selected strains including *Saccharomyces boulardii*. The effect is reduction of gastroenteritis by approximately 24 hours. The authors believe therefore that there are now no reasons to omit this active treatment in children with acute gastroenteritis in addition to rehydration. This is supported by solid compelling and authoritative indications by many agencies and institutions.

Recently, Chen et al. (2018) published guidelines on evidence-based indications for the management of children with acute infectious diarrhea in Chinese paediatric population. The guideline was developed by an expert working group composed of paediatric gastroenterology, paediatric infectious disease and epidemiology experts under the organization of Academic Group of Paediatric gastroenterology of Chinese Pediatrics Association. Recommendations were based on a comprehensive thorough literature

review in relevant databases including PubMed, Cochrane, EMBASE, China Biomedical Database (CBM), and Chinese Journal Full-text Database up to June 2013. According to this guideline several probiotics have curative effects on the treatment of acute infectious diarrhoea in children, especially for watery diarrhoea caused by viral infection (evidence level A). *Saccharomyces boulardii* can shorten the duration of acute infectious diarrhoea in children and reduce the duration of hospital stay (evidence level A).

It can be concluded that a moderate clinical benefit exists, mainly a shorter duration of diarrhoea, of *Saccharomyces cerevisiae* therapy in addition to rehydration therapy in otherwise healthy infants and children with acute unspecific diarrhoea even if some studies have methodological limitations and the studies were carried out mainly in non-European countries. These conclusion is supported by recently published reviews / meta-analyses and guidelines based on a consistent amount of evidence in various settings.

Infants from 6 months to 2 years should be treated under the care of a doctor, only. Elder children with unspecific acute diarrhoea can be treated for two days. If the symptoms do not improve or worsen, a doctor has to be consulted.

The use in infants younger than 6 months is not recommended, because an exact analysis of data from existing literature concerning this special age group is not possible. In the first months of life the intestinal flora is changing, especially when nutrition of the infants changes from breast feeding to different nutrition (Lentze, 2013). Therefore, more precise study results concerning efficacy and safety are necessary to recommend a treatment in this age group, too.

Based on the clinical data available (for adults: mainly Höchter et al. 1990; for children: mainly Chapoy 1985 / 1986, Villarruel et al., 2007; supported by studies with Non-European patient groups) and supported by the marketing overviews (for adults and children e.g. Nr. 2, 4, 7, 11, 14, 22) the posology is recommended as follows:

Children (6 months to 11 years): Single dose: 250 mg dried yeast, frequency of administration: 1-2 times

Adolescents, Adults, and Elderly: Single dose: 250 mg dried yeast, frequency of administration: 2 times.

According to the information given in section 1.1 the medicinal product should contain at least 1.8×10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g dried yeast, rounded up to 2×10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g dried yeast.

Prevention of antibiotic associated diarrhoea

A variety of clinical studies and meta-analyses has been published with regard to the effects of *Saccharomyces boulardii* in the prevention of AAD in both children and adults. As the selection of clinical studies for the meta-analyses has been made based on different criteria (e.g. age group, study design, indication) different assessments on the efficacy of *Saccharomyces boulardii* in this indication were given by the authors.

Many of the clinical studies, which have been published so far, indicate that the incidence of AAD tends to be lower when *Saccharomyces boulardii* is administered concomitantly. Nevertheless, it has to be emphasized that the study designs employed show a great variety with regard to e.g. the dosage of *Saccharomyces boulardii*, study duration and follow-up, selection of patients (age, in- or out-patient), choice of antibiotic, or definition of diarrhoea. Therefore, the data available are very inhomogeneous and give no evidence to support a general efficacy of *Saccharomyces boulardii* in the prevention of

AAD. The effective dosage and the duration of treatment as well as the patient groups at a particular risk are not sufficiently investigated.

A reliable assessment of efficacy is only possible, if risk factors for patients for the development of AAD have been identified. Although the risk for AAD seems to be increased in hospitalized patients (26-60% according to McFarland, 2006) the clinical study by Pozzoni *et al*. (2012) reported a diarrhoea rate of 13.3% only in elderly, hospitalized patients receiving placebo during treatment with various antibiotics. Thus, it does not seem justified to recommend *Saccharomyces boulardii* for general administration in the prevention of AAD, since in about 90% of the patients diarrhoea does not occur while on antibiotic therapy.

This assessment is in agreement with Suardi *et al*. (2013). In their review on probiotics in the prevention of AAD in adults, they summarize as follows: *In spite of some evidence consensus statements favoring the use of probiotics in the prevention of AAD is lacking due to the difficulties in analyzing the available studies. The differences of sample sizes with heterogeneous populations, the differences in the given dose of probiotics agents, the differences in the amounts of viable organisms administered represent the main factors impacting the correct interpretation of the promising results found in these studies. Moreover, study designs are not homogeneous as well as endpoints and objectives that do not include assessments of possible adverse events. In conclusion, albeit some promising results on the efficacy of probiotics in the prevention of AAD and C. difficile associated diarrhoea, there are no evidence-based guidelines regarding probiotics for this use. Adequately powered, double blind, randomized controlled trials are needed to assess the efficacy of specific probiotic strains.*

In contrast to acute gastroenteritis (diarrhoea) Guarino *et al.* (2015) also state that AAD is a less clear indication for probiotics, and even if efficacy is supported by several randomized clinical trials and meta-analysis, there is no official recommendation. The question, if probiotics should be prescribed to 7 children to prevent an episode of diarrhea in children remaining antibiotics, has no answer as yet. Maybe a selective use of active intervention based on the antibiotic involved and especially the child's age and underlying condition may be considered as the best advisable approach.

The latest Cochrane review (Guo *et al.* 2019) concerning children also demands, the benefit of high dose probiotics (e.g. *Lactobacilus rhamnosus* or *Saccharomyces boulardii*) needs to be confirmed by a large well-designed multi-centered randomized trial. Most of the included studies were categorized as high risk of bias.

Prevention of antibiotic associated diarrhoea caused by triple therapy in *Helicobacter pylori* infection

The use of the strain *Saccharomyces cerevisiae* CBS 5926 in prevention of diarrhoea caused by triple therapy in *H. pylori* infection cannot be regarded as well-established according to the Directive 2001/83/EC and the Guideline on the assessment of clinical safety and efficacy in the preparation of the 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).

So far, the administration of *Saccharomyces cerevisiae* for the prevention of diarrhoea during triple therapy for infection with *H. pylori* has not been established in Europe. Medicinal products for prevention of diarrhoea due to triple therapy are not available in European Union Member States.

Thus, although a great number of clinical studies have included patients undergoing triple therapy and have indicated a tendency of a reduction of diarrhoea the requirements for a well-established use are not met (at least one decade of wellestablished medicinal use).

In addition, the studies included in this assessment report have several methodological shortcomings. Only two studies (Cremonini *et al.,* 2002a, Cindoruk *et al.,* 2007) have a double-blind study design, however, the number of patients included was low (43 resp. 124 patients). Differences were also observed with regard to the duration of triple therapy (Cremonini *et. al.*, 2002a: one week; Cindoruk *et al.,* 2007: 2 weeks) and the dosage of *Saccharomyces boulardii* administered which ranged from 500 mg to 1000 mg *Saccharomyces cerevisiae* daily (2 x 500 mg, 3 x 250 mg, 2 x 250 mg) in the studies assessed. Besides, most of the studies did not investigate diarrhoea as primary parameter and thus a definition of diarrhoea was not given. Accordingly, the rates of diarrhoea incidence under triple therapy are very divergent ranging from 6% (Song *et al*., 2010) to 43.1% (Lee *et al*., 2011). Thus, there is a great uncertainty about the true incidence and definite factors have not been described for the group of patients who may benefit from concomitant treatment with *Saccharomyces cerevisiae* during *H. pylori* eradication therapy so that a general recommendation cannot be given in this indication. Another important aspect is that that the efficacy of standard triple treatment for *H. pylori* eradication has decreased and many new treatments have been introduced to improve eradication rates. (Li *et al*., 2015) Geographical differences have to be taken into account.

Paediatric population

Up to now, the data available for the use in children and adolescents are insufficient, too.

Some open randomized clinical studies in children were performed.
Hurduc *et al*. (2009) only treated 48 children (range from 3-18 years) with *Saccharomyces cerevisiae* in addition to triple therapy and the incidence of AAD resp. diarrhoea is not stated; only adverse reactions in general are reported.
Bin *et al.* (2015) conducted their study in China and it remains open, if the data presented also apply for European children.

The publications of further studies conducted in China mentioned in Feng *et al.* (2017) are not available.

In conclusion, a well-established use for *Saccharomyces cerevisiae* CBS 5926 in the prevention of diarrhoea due to triple therapy in *H. pylori* infections is given neither in adults nor in children.

***Clostridium difficile*-associated disease (CDD)**

The data available in literature at that time do not support a well-established use of *Saccharomyces boulardii* as an adjunct to standard antibiotic treatment with vancomycin or metronidazole for the secondary prevention of CDD. Statistically significant results, which demonstrate a reduction of CDD recurrences in patients with active CDD have only been observed in a low number of highly selected patients and thus cannot be used as a basis for a general recommendation. Consequently, clinical studies with a sufficiently high number of patients receiving different categories of antibiotic drug regimen and with an adequate duration of follow-up are needed.

Prevention of diarrhoea associated with tube-feeding

So far, clinical experience with the use of *Saccharomyces boulardii* in prevention of nutrition-related diarrhoea in tube-fed patients is very limited. Only a low number of patients has been included in clinical studies. The dosages applied ranged from 1 g/d (Bleichner *et al*., 1997) to 2 g/day (Tempé *et al*., 1983, Schlotterer *et al*., 1987). Thus, a well-established use of *Saccharomyces boulardii* in tube-fed patients cannot be supported, especially since up to now the recommended daily dosage is 750 mg/1.5 l nutrient solution per day. This posology which is recommended in current SPCs, is not supported by adequate data.

Furthermore it has to be taken into account that a risk for immunocompromised patient cannot be excluded when administered *Saccharomyces cerevisiae* (see below). Tube-fed patients are often

immunocompromised patients. Therefore a well performed benefit-risk evaluation is needed, which is only possible when well designed studies are available concerning also dose response.

Irritable bowel syndrome (IBS)

A WEU of *S. bouilardii* in the treatment of IBS is not recommended. The clinical data available do not comply with the recommendations of the CHMP guideline on the evaluation of medicinal products for the treatment of irritable bowel syndrome (CHMP, 2014).

There is only one elder double-blind randomized clinical study from 1983 (Maupas *et al*.) which showed a superiority of treatment with *Saccharomyces boulardii* regarding the following items: opinion of physician/patient, number and consistency of stools). Other clinically relevant signs of IBS such as abdominal pain, distension, dyspepsia, however, were not improved by *Saccharomyces boulardii* as compared to placebo.

Another study (Choi *et al*., 2011) showed that *Saccharomyces boulardii* was superior to placebo only with regard to quality of life. According to the guidelines of the CHMP (2014), the primary outcome in clinical studies for the evaluation of medical products for the treatment of IBS consists of abdominal pain and consistency/frequency of stool. This evidence of efficacy for the indication irritable bowels syndrome still has to be provided.

Since there is no medicinal product available on the European market with a tradition of 30 years in this indication a traditional use is neither possible. Furthermore, IBS cannot be diagnosed by the patients themselves, as it is mainly based on the exclusion of underlying organic causes which can be done only by a medical practitioner.

Prevention of travellers' diarrhoea

Two randomized double-blind clinical trials have been performed, in order to assess the efficacy of *Saccharomyces boulardii* for the prevention of traveler's diarrhoea. There seems to be a dose-dependent preventive effect and the administration of high-dose *Saccharomyces boulardii* (1000 mg/d) showed the best anti-diarrhoea results. Nevertheless, the prophylactic efficacy of *Saccharomyces boulardii* was influenced by the patient's strict adherence to the dosage instructions. Furthermore, regional differences were observed. Concluding from this, further investigations are needed to find out which individuals have the highest benefit from the preventive administration of *Saccharomyces boulardii*. A general recommendation cannot be given for this indication, as the preventive efficacy is also affected by geographical factors, which are not completely understood at present. Another question which still remains relates to optimal dosing. Different dosages have been administered and varying effects have been observed. With regard to the clinical study by Kollaritsch *et al*. (1993) another point of criticism is the high rate of drop outs. From 3000 subjects included into the study only 1016 questionnaires could be in the analysis of efficacy, so that there are doubts, if the results reported are representative.

In conclusion, more information is needed for recommending a WEU for the prevention of traveller's diarrhoea. Based on the data and medicinal products available, the period requested for the traditional use has elapsed for the medicinal use of the preparation containing $>1.8x10^{10}$ viable cells/g lyophilisate in adolescents and adults. The recommended posology is as follows: single dose: 250 mg dried yeast; daily dose: 1-2 times daily equivalent to 250-500 mg. Posology of 3 x 100-150 mg does not have a tradition of 30 years, because the medicinal product is only available since 1995. The treatment should start 5 days before departure. The treatment should be consequently maintained during the travel.

Acne

None of the clinical studies performed confirms an efficacy of *Saccharomyces cerevisiae* in the treatment of acne. Controlled clinical studies which demonstrate efficacy of *Saccharomyces cerevisiae*

as compared to other therapies using objective parameters for assessment are not available. Thus, a well-established use would not be accepted here.

Based on the data available, however, the period requested for the traditional use of *Saccharomyces cerevisiae* for the adjuvant treatment of chronic acne has elapsed for the preparation containing $>1.8x10^{10}$ viable cells/g lyophilisate. The indication should be restricted to uncomplicated mild acne. If acne is accompanied by a high rate of inflammatory lesions and scarring occurs, a doctor should have to be consulted.

Supported by the marketing overviews and the available clinical experience the posology is recommended as follows: 250 mg 3 times daily. The use is recommended for a period of 3 months at least considering the precautions and warnings mentioned.

5. Clinical Safety/Pharmacovigilance

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

Saccharomyces cerevisiae has been used as a probiotic for a long period of time. Numerous clinical trials have shown that the yeast has expressed a positive safety profile in adults and children.

5.2. Patient exposure

In her review on *Saccharomyces boulardii* in adults McFarland (2010) summarizes that 2963 patients were included in clinical trials with different gastrointestinal indications and thus provide data on safety. According to her the following adverse events were associated with *Saccharomyces boulardii*: thirst (n=5 patients), constipation (n=8 patients). These adverse events were reported in a clinical study with CDI patients (McFarland *et al*., 1994). In their review on the efficacy and safety of *Saccharomyces boulardii* for the prevention and treatment of gastrointestinal disorders Kelesidis and Pothoulakis (2012) confirm that adverse effects have not been observed in any of the clinical trials performed.

On the basis of the longstanding use in many Member States a significant exposure can be expected.

According to information received following the call for marketing overview, preparations containing *Saccharomyces boulardii* are on the European market at least since 1968.

5.3. Adverse events, serious adverse events and deaths

In the abovementioned clinical investigations, no signs of acute toxicity and serious adverse events have been observed.

Hypersensitivity reactions (pruritus, urticaria, localized or generalized exanthema, angioneurotic oedema, dyspnoea, anaphylactic shock) have been reported in the German pharmacovigilance database. The frequency is not known.

Furthermore, in some cases gastrointestinal complaints like flatulence, nausea and abdominal pain were reported. As these symptoms could also be signs of the underlying disease diarrhoea, it is difficult to assess, if these symptoms are adverse events.
During the single PSUR assessment (PSUSA/00009284/201702), the lead assessor listed constipation and flatulence as frequently reported adverse drug reactions. The MAH reported, that in assessed studies constipation occurred significantly more frequently with *Saccharomyces boulardii* in comparison to placebo. The actual frequency cannot be objectively estimated due to fact that the majority of cases were reported in the self-medication context, and due to the very low reporting of such effect in the clinical studies.

Based on this, flatulence and constipation are included as undesirable effects with unknown frequency.

5.4. Laboratory findings

No data available.

5.5. Safety in special populations and situations

According to literature, there is a risk of fungemia which is of clinical relevance especially in patients with severe general or intestinal disease having an indwelling catheter. According to the review of Enache-Angoulvant and Hennequin (2005) 92 cases of invasive infections with *Saccharomyces* have been identified in literature, 40% of them were caused by *Saccharomyces boulardii* (n=37). They also found out that patients infected with Saccharomyces boulardii were more likely than patients infected with *Saccharomyces cerevisiae* to have digestive tract disease (58% vs. 6%; $p<0.01$), to have intravenous catheters (83% vs. 29%; $p<0.0001$), and to be hospitalized in an intensive care unit (32% vs. 0.05%, $p<0.01$). As from the 37 patients affected by *Saccharomyces boulardii* infection 5 did not take a probiotic containing *Saccharomyces boulardii* at the time of diagnosis, the authors conclude that nosocomial acquisition is probable with catheters being a likely portal of entry due to possible contamination through hand transmission. Simulation tests performed by

Hennequin *et al*. (2000) showed that opening a packet of *Saccharomyces boulardii* lead to massive air contamination and consecutively contaminated surrounding inert surfaces and the skin of a simulated patient. Moreover, the hands of the person who opened the packet were highly and persistently contaminated despite vigorous hand washing. According to the authors *Saccharomyces boulardii* fungemia is an underestimated nosocomial and iatrogenic infection and the potential benefit has to be evaluated for each patient.

Venugopalan *et al*. (2010) mention the following factors which constitute excessive and undue risk for development of *Saccharomyces* fungemia during probiotic administration: the patient's immunocompromised state during critical illness, the potential for live yeast spore contamination of healthcare workers' hands during preparation of the probiotic capsule for administration, and introduction of live yeast from contaminated hands to catheter sites.

Accordingly, in the Clinical Practice Guidelines for *Clostridium difficile* infection in adults (Cohen *et al*., 2010) it is stated: "The administration of *Saccharomyces boulardii* has, however, been associated with fungemia in immunocompromised patients and in patients with central venous lines, and it should be avoided in critically ill patients."

The review by Didari *et al*. (2014) revealed that although probiotics are helpful, they do not always seem to be safe. Through interference with commensal microflora, they can result in opportunistic performances in the host due to bacteraemia and fungaemia. Provided conclusion of the expert is that the main observed adverse effects of probiotics were sepsis, fungaemia and GI ischemia. Generally, critically ill patients in intensive care units, critically sick infants, postoperative and hospitalized patients and patients with immune-compromised complexity were the most at-risk populations. While the overwhelming existing evidence suggests that probiotics are safe, complete consideration of risk-benefit ratio before prescribing is recommended. With regard to *Saccharomyces boulardii* the authors mentioned 30 case reports of fungemia in preterm infants and adults with underlying disease. One of the reported fungemia occurred in an infant who had not received *Saccharomyces boulardii* but was in the cot adjacent to an infant with fungemia after probiotics digestion. In four controlled clinical trials (Costalos *et al.* 2003, Bleichner *et al.*, 1997, Schneider *et al.*, 2005, McFarland *et al.*, 1994) no significant adverse effects such as fungemia or sepsis were reported. *Saccharomyces boulardii* was well

tolerated. Didari's et al recommendation is to avoid the administration of *Saccharomyces boulardii* in patients with central venous catheter and patients with synthetic cardiac valve replacement.

Roy *et al.* (2017) described seven cases of *Saccharomyces* fungaemia in two hospitals in India between July 2014 and September 2015. Two patients were premature neonates and five were adults. They were admitted in intensive care unit and were on probiotics containing *Saccharomyces boulardii* (except one adult patient).The probiotic intake for the two neonates was part of routine protocol for premature babies. In adult patients, probiotics were prescribed to either treat or prevent diarrhoea. The adult patient, who had no clear history of probiotics intake, might had acquired the agent by cross-contamination. Several hypotheses have been postulated on the acquisition offungaemia from probiotics: translocation across the intestinal barrier or contamination of central or peripheral vascular lines from the hands of the healthcare workers when the sachets of probiotics are opened.The authors recommend avoiding this probiotic in critically ill or vulnerable patients, especially those with central venous catheter.

During the single PSUR assessment (PSUSA/00009284/201702), the CMDh reached the position that the marketing authorization(s) of products in the scope of this PSUSA should be varied (CMDh scientific conclusions and grounds for variation, amendments to the product information and timetable for implementation – PSUSA/00009284/201701, last updated: 01/12/2017).
This scientific conclusion was based on the data presented within the periodic safety update report (PSUR) under review, on the data in EudraVigilance database and available literature. The benefit-risk balance for use of *Saccharomyces boulardii* containing products in critically ill or immunocompromised patients was considered changed.
"There were 19 cases reported with preferred term (PT) fungaemia during the interval period and 61 cases cumulatively. The search in EudraVigilance database overall revealed 10 fatal cases of fungaemia/fungal infection and sepsis associated with administration of Saccharomyces boulardii containing medicinal products where the causal association could not be ruled out. Moreover, there was also 1 fatal case of fungal infection and sepsis reported in a 48-year old patient, however no case narrative was provided, therefore the causality could not be established properly. Approximately half of the fatal fungaemia cases were reported in patients with central venous catheter (CVC) which has been already contraindicated. However, in rest of the fatal cases no CVC insertion was reported. In 1 fatal case of fungaemia insertion of CVC was explicitly ruled out by the reporter. Considering the known potential risk of fungaemia in critically ill patients and reported fatal cases in patients with no CVC insertion, the use of *Saccharomyces boulardii* in critically ill or immunocompromised patients should be contraindicated and relevant sections of the SmPC (sections 4.2, 4.3, 4.4 and 4.8) and patient information leaflet (PIL) should be updated accordingly."

5.5.1. Use in children and adolescents

According to the data available, treatment of acute diarrhoea with *Saccharomyces cerevisiae* is adequate and well tolerated in infants >6 months, children and adolescents. However, the use in infants from 6 months to 2 years of age requires medical advice due to the disease itself. Severity of the diarrhoea with possible loss of water and electrolytes has to be supervised by a doctor.

If the symptoms persist longer than 2 days during the use of the medicinal product, a doctor or a pharmacist should be consulted for children above 2 years of age.

The use in infants below 6 months of age is not recommended.

5.5.2. Contraindications

Hypersensitivity to yeast, especially *Saccharomyces cerevisiae*.

Critically ill patients or immunocompromised patients (e.g. HIV-infection, organ transplantation, leukaemia, malignant tumours, radiotherapy, chemotherapy, long-term high dosage glucocorticoids treatment), and patients with central venous catheter because *Saccharomyces cerevisiae* CBS 5926 is a living microorganism, which can cause systemic fungaemia under adverse circumstances.

5.5.3. Special Warnings and precautions for use

In case of diarrhea, the most important therapeutic measure is rehydration therapy particularly in infants, children and elderly.

There have been very rare cases of fungaemia (and blood cultures positive for *Saccharomyces* strains) reported mostly in patients with central venous catheter, critically ill or immunocompromised patients, most often resulting in pyrexia. In most cases, the outcome has been satisfactory after cessation of treatment with *Saccharomyces boulardii,* administration of antifungal treatment and removal of the catheter when necessary. However, the outcome was fatal in some critically ill patients (see sections 4.3 and 4.8).

As with all medicines made from living micro-organisms, special attention must be paid to the handling of the product in the presence of patients mainly with central venous catheter but also with peripheral catheter, even if not treated with *Saccharomyces boulardii*, in order to avoid any contamination by hands and/or the spread of microorganisms by air (see section 4.2).

As *Saccharomyces boulardii* is frequently used in self-medication respective information has to be included in the informative texts (section 4.2): The risk of airborne contamination should also be considered in the presence of other critically ill or immunocompromised patients.

If during or shortly after the treatment with *Saccharomyces cerevisiae* CBS 5926 a microbiological examination of faeces is made, the results might be false positive. Therefore, the research laboratory should be informed adequately.

In case of traditional use for acne, a doctor has to be consulted, if acne is accompanied by a high rate of inflammatory lesions and scarring occurs.

5.5.4. Drug interactions and other forms of interaction

Interaction with concomitant administration of monoamine oxidase inhibitors (MAOI)

Several SPCs indicate that concomitant use of MAOI with *Saccharomyces cerevisiae* can cause an increase of blood pressure. Probably this is based on the old preclinical data mentioned above and on theoretical considerations concerning the effects of these inhibitors.

MAOIs increase the central concentration of serotonin, norepinephrine and dopamine, but due to the unselective mechanism these inhibitors also have peripheral effects. When ingested orally, MAOIs inhibit the catabolism of dietary amines. When food containing tyramine is consumed concomitantly, the individual may suffer from increased blood pressure / hypertensive crisis.

Laux and Ulrich (2006) describe that 1000 to 1600 mg tyramine caused a 30 mm Hg increase of blood pressure in eight probands. These amounts are not reached with normal diet. But after administration of 20 mg tranylcypromine (MAOI) for 2 weeks, already 20-50 mg tyramine causes the same increase. These amounts can be reached with normal diet, exceptionally. The authors consider an amount of tyramine of 6 mg per meal as safe for most of the patients even if they are treated with tranylcypromine.

Microbial fermentation during manufacturing of foodstuff can lead to augmentation of biogenic amines. This kind of food should be avoided (e.g. Marmite (yeast extract) Blackwell and Marley (1966).

Up to now there is no evidence that the specific strain of *Saccharomyces cerevisiae* CBS 5926 covered by this assessment report causes such interactions.

Interaction with concomitant administration of antimycotics

Elmer *et al*. (1995) studied the concomitant use of *S. bouilardii* in 8 healthy volunteers receiving antifungals. *Saccharomyces boulardii* (*Saccharomyces cerevisiae* CBS 5926), was administered at 2 daily doses of 500 mg for 2 weeks. On days 7-14 an additional dose of either 50 mg/d or 100 mg/d fluconazole or 1,500,000 units/d nystatin was given. The authors showed that fluconazole with a high bioavailability did not affect steady-state levels of *S. bouilardii* when the ingestion of the two drugs was separated by 3 h. In contrast, the concomitant use of nystatin which is not absorbed resulted in *Saccharomyces boulardii* levels below the limits of detection.

Therefore, a possible interaction with antimycotics when administered orally or systemically is to be considered.

5.5.5. Fertility, pregnancy and lactation

There are no data from use during pregnancy and lactation. Safety during pregnancy and lactation has not been established. In the absence of sufficient data, the use during pregnancy and lactation is not recommended.

No fertility data available.

5.5.6. Overdose

No case of overdose has been reported.

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

Not relevant.

5.5.8. Safety in other special situations

The use in critically ill, immunocompromised patients (e.g. HIV-infection, organ transplantation, leukaemia, malignant tumours, radiotherapy, chemotherapy, long-term high dosage glucocorticoids treatment), and patients with central venous catheter is contraindicated. These contraindications are based on case reports of fungaemia in such patients, often caused by nosocomial and iatrogenic infections. It is not evident whether cells of *Saccharomyces cerevisiae* CBS 5926 can cross the gastrointestinal wall in this special patient population, when administered orally.

5.6. Overall conclusions on clinical safety

In summary, the use of *Saccharomyces cerevisiae* could be evaluated as safe and well tolerated treatment under the conditions mentioned in this assessment report. Hypersensitivity reactions (pruritus, urticaria, localized or generalized exanthema, angioneurotic oedema, dyspnoea, anaphylactic shock) and gastrointestinal disorders (flatulence, constipation) have been reported. Therefore, patients with hypersensitivity to the active substance have to be excluded form the use.

Treatment with *Saccharomyces cerevisiae* is contraindicated in immunocomprised patients and patients with central venous catheters.

6. Overall conclusions (benefit-risk assessment)

Acute unspecific diarrhoea

The use of the strain *Saccharomyces cerevisiae* CBS 5926 in acute unspecific diarrhoea could be regarded as well-established according to the Directive 2001/83/EC.

One randomized double-blind, placebo-controlled, multicenter clinical trial was conducted in adults with a positive statistically significant result for the treatment group (Höchter *et al.*, 1990). The daily dose was 3 x 200 mg for the first 2 days and then 3 x 100 mg *Saccharomyces boulardii*. A second randomized controlled study in adults compared two varying *Saccharomyces* preparations, however without a placebo control.
The recommended posology is as follows: single dose: 250 mg dried yeast containing at least 2 x 10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g; daily dose: 2 times daily equivalent to 500 mg.

Numerous randomized studies were conducted in children all over the world.
The recommended posology is as follows: single dose: 250 mg dried yeast containing at least 2 x 10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g; daily dose: 1-2 times daily equivalent to 250-500 mg.

In general, the use of *Saccharomyces cerevisiae* CBS 5926 is safe and well tolerated under the conditions described above. The use in critically ill or immunocompromised patients (e.g. HIV-infection, organ transplantation, leukaemia, malignant tumours, radiotherapy, chemotherapy, long-term high dosage glucocorticoids treatment) and patients with central venous catheter is contraindicated.

The use is not recommended in infants below 6 months of age as well as in pregnant and breast-feeding women due to insufficient data on safety and efficacy.

In summary, the benefit-risk assessment has to be regarded as positive.

Prevention of travellers' diarrhoea

No sufficient clinical evidence is available for the well-established use of the strain *Saccharomyces cerevisiae* CBS 5926 in prevention of travellers' diarrhoea. Two randomized double-blind clinical trials have been performed, in order to assess the efficacy of *Saccharomyces boulardii* for the prevention of traveler's diarrhoea. The results do not justify a general recommendation for this indication.

Based on the data available, the period requested by the Directive 2001/83/EC for the traditional use of *Saccharomyces cerevisiae* CBS 5926 in the prevention of travellers' diarrhea has elapsed for the preparation containing >1.8x10^{10} viable cells/g lyophilisate.

Corresponding medicinal products have been in medicinal use throughout a period of at least 30 years, in European Union member states and *Saccharomyces cerevisiae* CBS 5926 can be used in this indication without the supervision of a medical practitioner for diagnostic purposes or for prescription or monitoring of treatment. The pharmacological effects or efficacy are plausible on the basis of longstanding use and experience.

The recommended posology is as follows: single dose: 250 mg dried yeast containing at least 2 x 10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g; daily dose: 1-2 times daily equivalent to 250-500 mg. The treatment should start 5 days before departure. The treatment should be consequently maintained during the travel.

In general, the use of *Saccharomyces cerevisiae* CBS 5926 is safe and well tolerated under the conditions described above. The use in immunocompromised patients (e.g. HIV-infection, organ transplantation, leukaemia, malignant tumours, radiotherapy, chemotherapy, long-term high dosage glucocorticoids treatment) and patients with central venous catheter is contraindicated.

The use is not recommended in children under 12 years of age as well as in pregnant and breast-feeding women due to insufficient data.

Uncomplicated mild acne

No sufficient clinical evidence is available for the well-established use of the strain *Saccharomyces cerevisiae* CBS 5926 in the treatment of acne. Thus, a well-established use according to the Directive 2001/83/EC could not be accepted here.

Based on the data available, the period requested by the Directive 2001/83/EC for the traditional use of *Saccharomyces cerevisiae* CBS 5926 as an adjuvant in the treatment of uncomplicated mild acne in adolescents and younger adults has elapsed for the preparation containing $>1.8x10^{10}$ viable cells/g rounded up to 2×10^{10}. If acne is accompanied by a high rate of inflammatory lesions and scarring occurs, a doctor has to be consulted. For such a traditional use, the requirements of the Directive 2001/83/EC are fulfilled.

The recommended posology is as follows: 250 mg dried yeast containing at least 2×10^{10} viable cells *Saccharomyces cerevisiae* CBS 5926 / g, daily dose: 3 times daily equivalent to 750 mg. The use is recommended for a period of 3 months at least considering the precautions and warnings mentioned.

In general, the use of *Saccharomyces cerevisiae* CBS 5926 is safe and well tolerated under the conditions described above. The use in immunocompromised patients (e.g. HIV-infection, organ transplantation, leukaemia, malignant tumours, radiotherapy, chemotherapy, long-term high dosage glucocorticoids treatment) and patients with central venous catheter is contraindicated.

Due to indication, the use in children under 12 years and elderly is not relevant.

The use is not recommended in pregnant and breast-feeding women due to insufficient data on safety.

Discussions on the classification of *Saccharomyces cerevisiae*

Intensive discussions took place in the HMPC, especially concerning on the nature and classification of *Saccharomyces cerevisiae* CBS 5926 as an herbal or a biological product and then its eligibility to the traditional use registration procedure.

The HMPC sent a request for legal interpretation about the application of Directive 2004/24/EC to living yeast cells (fungi) such as *Saccharomyces cerevisiae* (strain CBS 5926) to the European Commission, explaining the divergent views in the Committee.

Some HMPC members considered *Saccharomyces cerevisiae* CBS 5926 as a "herbal substance" on the basis of the definition from Directive 2001/83/EC which mentions fungi as herbal substances.

Other members considered *Saccharomyces cerevisiae* CBS 5926 as a "biological active substance" since

- the current herbal guidelines on quality are not fully suitable and applicable, moreover GACP or GMP annex 7, and Ph. Eur. herbal monographs do not cover unicellular living yeast or freeze-dried living fungal cells,

- such medicines are assessed in many Regulatory Agencies in accordance with guidelines for biologicals when used as medicinal products and the Live Biotherapeutic Products general monograph published by EDQM in the European Pharmacopoeia monograph 3053

- their specific manufacturing process is evaluated as per the quality requirements for biological medicinal products and inspected according to the GMP annex 2 for biological medicinal products.

The European Commission clarified that the classification of medicinal products falls under the responsibility of Member States based on all the characteristics of a particular product. Based on data provided by applicants, Member States may authorise a particular medicine under a traditional herbal medicine registration, 'well-established use' application or full marketing authorisation application and the product may be authorized differently in different Member States.

Differences in medical approach, regulatory classifications, healthcare customes and patient self-management in the EU were noted with regard to the use of *Saccharomyces cerevisiae.* In the absence of absolute majority in favour of adopting a monograph, the HMPC issued a public statement.

HMPC overall conclusion taking into account the classification, definition and nature of the active substance

Following intensive discussions in the Committee taking into account that 'fungi' in general are included in the herbal substance definition of the Directive 2001/83/EC and on the other hand living yeast are covered by the Live Biotherapeutic Products general monograph of the European Pharmacopoeia, no absolute majority required for adoption of the monograph was achieved.

Therefore, having regard of its Rules of Procedure, the HMPC concluded that the following requirement for the establishment of a European Union herbal monograph on traditional and well-established herbal medicinal products containing Saccharomyces cerevisiae CBS 5926 *is not fulfilled:*

- the requirement laid down in Article 1 of Directive 2001/83/EC on the definition of the herbal substance despite the existence of data on the safety, efficacy and historical data on the medicinal uses within the European Union of products containing Saccharomyces cerevisiae CBS 5926.

Annex

List of references