EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

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Committee on Herbal Medicinal Products (HMPC)

# Assessment report on *Allium cepa* L., bulbus

Draft

| | |
|---|---|
| Herbal substance(s) (binomial scientific name of the plant, including plant part) | *Allium cepa* L., bulbus |
| Herbal preparation(s) | a) Liquid extract from fresh bulb (DER 1:1), extraction solvent soya-bean oil<br>b) Liquid extract (DER 1:6), extraction solvent ethanol 70% V/V |
| Pharmaceutical forms | Herbal preparations in liquid or solid dosage forms for oral use |

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

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# 1. Introduction

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

- Herbal substance(s)

Allii cepae bulbus consists of fresh or dried thick and fleshy leaf sheaths and leaf approaches from *Allium cepa* L. (Monographie BGA/BfArM, 1986; Blaschek *et al*., 2006).

Bulbus allii cepae is the fresh or dried bulb of *Allium cepa* L. (*Liliaceae*) or its varieties and cultivars (WHO monographs, 1999).

- Herbal preparation(s)

See chapter 1.2.

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

Onion extracts are also used in combination with other herbal substances/herbal preparations. This Assessment report on *Allium cepa* L., bulbus refers exclusively to monopreparations.

- Short overview of main compounds

According to WHO monographs (1999), sulfur- and non-sulfur-containing chemical constituents have been isolated from Bulbus allii cepae; the sulfur compounds are the most characteristic. The organic sulfur compounds of Bulbus allii cepae, including the thiosulfinates, thiosulfonates, cepaenes, *S*-oxides, *S*,*S*'-dioxides, monosulfides, disulfides, trisulfides, and zwiebelanes occur only as degradation products of the naturally occurring cysteine sulfoxides (e.g. (+)-*S*-propyl-L-cysteine sulfoxide). When the onion bulb is crushed, minced or otherwise processed, the cysteine sulfoxides are released from compartments and contact the enzyme alliinase in adjacent vacuoles. Hydrolysis and immediate condensation of the reactive intermediate (sulfenic acids) form the compounds as indicated above. The odorous thiosulphonates occur (in low concentrations) only in freshly chopped onions, whereas the sulfides accumulate in stored extracts or steamdistilled oils. Approximately 90% of the soluble organic-bound sulfur is present as γ-glutamylcysteine peptides, which are not acted on by alliinase. However, on prolonged storage or during germination, these peptides are acted on by γ-glutamyl transpeptidase to form alk(en)yl-cysteine sulfoxides, which in turn give rise to other volatile sulfur compounds. Alongside the sulphur compounds (Lanzotti, 2006), some seleno-derivatives were detected: γ-glutamyl-Se-methylselenocysteine, Se-methylselenocysteine, "Se-alliins", Se-methionine, Se-cystine/Se-cysteine (Arnault and Auger, 2006).

Compounds belonging to the flavonols, the anthocyanins and the dihydroflavonols have been reported to occur in onion bulbs. Flavonols are the predominant pigments of onions. Yellow onions contain 270–1187 mg of flavonols per kilogram of fresh weight (FW), whereas red onions contain 415–1917 mg of flavonols per kilogram of FW. At least 25 different flavonols (mostly as glucosides) have been characterised, with quercetin, kaempferol, isorhamnetin being the most important aglycones in all onion cultivars. Quercetin 4'-glucoside and quercetin 3,4'-diglucoside have been identified as the main flavonols. These two pigments have been shown to account for more than 80–85% of the total flavonoid content in some yellow cultivars, with concentrations from 50 to 1300 and from 36 to 394 mg/kg FW for quercetin 4'-glucoside and quercetin 3,4'-diglucoside, respectively (Slimestadt *et al*., 2007).

Most of the anthocyanins reported to occur in various cultivars of red onion are cyanidin derivatives, although minor amounts of peonidin derivatives have been identified. The main anthocyanins of all cultivars investigated are exclusively glycosylated at the anthocyanidin 3-position (Slimestadt *et al*., 2007). Determinations of absolute quantities of anthocyanins in red and pink onion cultivars are sparse in the literature. According to Fossen *et al*. (1996), the cultivar "Red Baron" contains 51% cyanidin 3-(6''-malonylglucoside), 22% cyanidin 3-(3''-glucosyl-6''-malonylglucoside), 3% cyanidin 3-(3''-glucosylglucoside), and 18% cyanidin 3-glucoside of the total anthocyanin content, respectively. Donner *et al*. (1997) reported on the total content of anthocyanins in four cultivars of red onions. The one with the highest score, "Red Bone", contained 219 mg of anthocyanins per 100 g of dry weight and cyanidin 3-malonylglucoside accounted for 39.4%, cyanidin 3-malonyllaminariobioside 23.5%, cyanidin 3-laminariobioside 17.8%, and cyanidin 3-glucoside 6.8% of the total anthocyanin content.

Studies of onion led to the isolation of different saponins and/or sapogenins. The main sapogenins are: sitosterol, gitogenin, oleanolic acid, amyrin, diosgenin, β-chlorogenin and cepagenin. Corresponding glycosides are: alliospirosides A–D, alliofuroside, tropeosides A1/A2, tropeosides B1/B2, ascalonicosides A1/A2 and ascalonicoside B (Corea *et al.*, 2005; Lanzotti, 2006; Slimestadt *et al*., 2007).

A mixture of 9,12,13-trihydroxy-10-octadecenoic and 9,10,13-trihydroxy-11-octadecenoic acids (Claeys *et al*., 1984).

Variable content (in %) of glucose (1.06-1.79), fructose (1.08-1.75) and sucrose (0.17-0.32) between the studied cultivars from different regions were established (Galdón *et al.,* 2009). The total content of fructans varies even more (Jaime *et al*., 2001; Galdón *et al*., 2009).

A total of 63 elements were found in each of 110 Danish white onion varieties samples. The highest mean values (mg/kg FW): sulphur 1230, phosphorus 440, sodium 210, calcium 197 and kalium 164. Some important elements were detected in very low amounts only: aluminium 0.25, lead 0.099, cadmium 0.022, mercury 0.0125, ... (Bibak *et al*., 1998). In another study, 394 of phosphorus and calcium 126 (expressed as mg/kg FW) were determined in six Spanish samples (Galdón *et al*., 2008).

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

## Regulatory status overview

| **Member State** | **Regulatory Status** | | | | **Comments** |
|---|---|---|---|---|---|
| Austria | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Belgium | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Bulgaria | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Cyprus | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Czech Republic | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Denmark | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Estonia | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Finland | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| France | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |

| Member State | Regulatory Status | | | | Comments |
|---|---|---|---|---|---|
| Germany | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Greece | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Hungary | ☐ MA | ☒ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Iceland | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Ireland | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Italy | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Latvia | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Liechtenstein | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Lithuania | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Luxemburg | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Malta | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| The Netherlands | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Norway | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Poland | ☐ MA | ☒ TRAD | ☐ Other TRAD | ☐ Other Specify: | Two combination products with non-herbal components<br><br>One combination product |
| Portugal | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Romania | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Slovak Republic | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | Only a combination product with non-herbal components |
| Slovenia | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Spain | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| Sweden | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |
| United Kingdom | ☐ MA | ☐ TRAD | ☐ Other TRAD | ☐ Other Specify: | |

MA: Marketing Authorisation

TRAD: Traditional Use Registration

Other TRAD: Other national Traditional systems of registration

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

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

## *1.3. Search and assessment methodology*

Databases: Scopus, Web of Science, Scirus, Medline, Micromedex, and Merck Index were searched using the terms [Allium cepa], [Allii cepae], [Allii cepae bulbus], [Bulbus allii], [Bulbus allii cepae], [onion], [Zwiebel] and [l'oignon]. Handbooks and textbooks were also used.

# 2. Historical data on medicinal use

## *2.1. Information on period of medicinal use in the Community*

Traditional use:

Orally: Cough, bronchitis, asthma, tonsillitis, to stimulate the bile production, digestion problems with flatulence and colic pain, diuresis stimulation, to initiate menstruation, rarely in the case of hypertension, atherosclerosis, and diabetes (Madaus, 1938; Flamm *et al*., 1940; Fischer and Krug, 1980; Poletti *et al*., 1990).
Externally: Insect bites treatment, wounds, minor burns, boils, warts and treatment of bruises (Madaus, 1938; Flamm *et al.*, 1940; Fischer and Krug, 1980; Weiss, 1982).

The efficacy in the above-mentioned routes of administration has not been established.

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

Table 1. Overview of products on the market in the European Member States

| **Member State** | **Medicinal Product** | **Regulatory Status** |
|---|---|---|
| Hungary | 500 mg Allii cepae bulbi extractum fluidum, extraction solvent: soya-bean oil (fresh herbal substance, extraction solvent 1:1)<br>Indications: For prevention or the adjuvant treatment of mild or moderate bacterial upper respiratory airways infections and of hay fever. | TU: since 06.05.1996 |
| Poland | Allii cepae extractum fluidum,<br>extraction solvent ethanol 70% V/V, syrup<br>Indications: Upper respiratory tract infections and of hay fever. | TU: 17 years on the market (stand 01/2011) |

The period of at least 30 years of medical use as required by Directive 2004/24/EC for qualification as a traditional herbal medicinal product is not fulfilled for both monopreparations (See Table 1).

Additionally, the following combination preparations with non-herbal components are available in the following MS:

- Bulgaria, Czech Republic, Estonia, Latvia, Lithuania, Poland and Slovakia:

A gel containing Allantoin (10 mg), Heparinum natricum (8.4 mg), Allii cepae extractum fluidum (100 mg) in 1 g

- Poland only:

A cream containing Allii cepae extractum fluidum, extraction solvent – ethanol + Chamomillae extractum + Heparinum natricum + Allantoinum

A syrup containing Allii cepae extractum fluidum (1:1), extraction solvent – ethanol 70% (V/V) + Allii sativi extractum (1:5), extraction solvent – ethanol 70% (V/V)

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

**Allii cepae bulbi extractum fluidum**, extraction solvent: soya-bean oil (fresh herbal substance – extraction solvent 1:1) in a form of hard capsules (500 mg/capsules)
Posology:
Adults and elderly: 2 capsules three times daily in the first week and one capsule three times daily in the second week
Children above 12 years: one capsule three times daily in the first week and one capsule twice daily in the second week

**Allii cepae extractum fluidum**, (1:6); extraction solvent: ethanol 70% (V/V), in a form of syrup
Posology: Oral use: 5 – 15 ml (corresponding to 0.95 – 2.85 g of extract) 3 times daily

# 3. Non-Clinical Data

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

**Antiallergic activity**
A butanolic fraction of 50% ethanol extract of dried and finely grounded onions (ALC-02) showed a potential mast cell-stabilising property. Chemical standardisation of this particular active fraction was done on the basis of quercetin (6.97%) and luteolin (0.65%). It was found to inhibit the experimental mast cell degranulator compound 48/80-induced histamine burst from rat peritoneal mast cells. Its action was comparable to that of ketotifen (ketotifen is a known mast cell stabiliser that inhibits histamine release in a nonimmunologic manner). Fraction ALC-02 (2.5–20 μg) produced a concentration-dependent inhibitory effect that ranged from 44% to 70%. ALC-02 might interact directly with the lipid bilayer membrane to prevent degranulation of the mast cells by producing a stabilising effect on the membrane integrity. ALC-02 attenuated calcium levels, possibly by preventing peroxidative injury to the plasma membrane, further highlighting the potential membrane-stabilizing activity of this herbal moiety. A restorative effect on the calcium distribution could contribute significantly to the observed antihistaminic property of ALC-02 (Kaiser *et al*., 2009).

In another two studies with guinea pigs (Dorsch *et al.,* 1989; Dorsch, 1996), thiosulfinates and cepaenes have been identified as the antiasthmatic active components and were suggested to mediate their effects through suppression of cyclooxygenase and lipoxygenase mediated reactions. Especially α,β-unsaturated thiosulfinates were the most active (Breu and Dorsch, 1994).

**Allergic activity**
Kawane (1995) correlates this allergic potency with cepaenes content, as they are known as inhibitors of cyclooxygenase and lipoxygenase, causing aspirin-sensitive asthma.

**Antiinflammatory activity**
A butanolic fraction of 50% ethanol extract of dried and finely grounded onions (ALC-02) showed a significant anti-edematogenic effect in the early phase of carrageenan-induced paw edema test in induced rat peritoneal mast cells. A dose of 200 mg/kg of ALC-02 produced a 34% reduction in paw volume at 1 hour, which gradually declined during the next 2–3 hours. In contrast, ibuprofen (100 mg/kg) showed a low (13%) inhibition of paw volume at 1 hour, which increased by 38–41% during the next 2–3 hours. Chemical standardisation of this particular active fraction was done on the basis of quercetin (6.97%) and luteolin (0.65%). In comparison, ibuprofen's action was more pronounced during the late phase. Ibuprofen, a nonsteroidal anti-inflammatory drug, exerts its anti-inflammatory effect by inhibiting synthesis and release of prostaglandins. These finding indicate that an anti-

histaminic property could be responsible for the anti-inflammatory effect of ALC-02 (Kaiser *et al*., 2009).

**Antimicrobial activity**

Benkeblia (2004) carried out a disc diffusion test for an antibacterial activity of the essential oils (EOs) isolated by steam distillation. Activity of different concentrations (50, 100, 200, 300 and 500 ml/l) of three type of onions (green, yellow and red) against two bacteria *Staphylococcus aureus*, *Salmomella enteritidis*, and three fungi, *Aspergillus niger*, *Penicillium cyclopium* and *Fusarium oxysporum*, was investigated. Low concentrations (50 and 100 ml/l) inhibited weakly the development of bacteria; however *S. enteritidis* was more sensitive than *S. aureus*. At high concentrations (200, 300 and 500 ml/l), EOs exhibited marked inhibition activity against bacteria. *A. niger* was less inhibited by low concentrations (50 and 100 ml/l) of EOs of green and yellow onions. However, higher concentrations exhibited marked inhibition. *F. oxysporum* showed the lowest sensitivity to green, yellow and red EOs extracts except at 300 and 500 ml/l concentrations of red onion which inhibited markedly its development.

Antimicrobial activity of the ethyl acetate and water subfractions of methanolic extracts of three Spanish onion varieties were assayed by Santas *et al*. (2010). Grano de Oro and Calcot de Valls varieties (ethyl acetate subfraction of crude onion extracts) were efficient in inhibiting gram-positive bacteria (minimal inhibition concentration MIC = 80 mg of dried onions/ml), but not gram-negative bacteria and *Candida albicans*. Fuentes de Ebro variety was only efficient in inhibiting *Listeria monocytogenes* (MIC = 80 mg of dried onions/ml), which was the most sensitive bacteria. No inhibitory effect of the water subfraction of the extracts was observed (MIC > 100 mg of dried onions/ml). These results corresponded to the quercetin and kempferol, and total flavonoids content in all varieties.

Ramos *et al*. (2006) isolated two new constituents of water onion extract, 2-(3,4-dihydroxyphenyl)-4,6-dihydroxy-2-methoxybenzofuran-3-one (1) and 3-(quercetin-8-yl)-2,3-epoxyflavanone (2). Compound 1 showed specific activity against the gram-negative *Helicobacter pylori* strains (inhibitng zone = 12 mm for 10 μg/disc), but not against the gram-positive multidrug-resistant *Staphylococcus aureus* (MRSA). This compound also showed no activity against the gram-negative *Salmonella typhimurium* IFO13245 and DT104-26 or *Pseudomonas aeruginosa.* Compound 2 showed comparatively the highest activity against MRSA strains (inhibiting zone = 15 mm for 10 μg/disc) and also high activity against *H. pylori* strains (inhibitng zone = 12 mm for 10 μg/disc).

Antilisterial activity of water extracts (10% w/v) from different spices was tested on 11 *Listeria monocytogenes* strains (Zouhir *et al*., 2008). Onion showed antilisterial inhibitory activity against all tested strains at 2.5 – 5.0 mg/ml, and bactericidal activity at 5.0 mg/ml, determined by the microdilution method. For comparison, garlic inhibitory activity was 0.6 – 1.2 mg/ml and > 5.0 mg/ml for bactericidal activity. The antimicrobial activity has been attributed to ability to inhibit RNA synthesis and to disrupting cell membranes by allicin.

**Antifungal activity**

The inhibitory effect of onion essential oil against nine different species of dermatophytic fungi were studied. Onion oil (200 ppm) completely inhibited the growth of *Microsporum canis*, *M. gypseum* and *Trichophyton simii* while the growth of both *Chrysosporium queenslandicum* and *Trichophyton mentagrophytes* was completely inhibited by 500 ppm of onion oil. The growth of four other species of dermatophytic fungi was gradually reduced by increasing the concentrations of onion oil. The inhibitory effect (expressed as invasive mold infections = IMI) of onion oil was also tested against four toxigenic isolates of fungi. Onion oil at different concentrations (100, 200 and 500 ppm) tested gradually reduced fungal growth and aflatoxin production by *Aspergillus flavus* (IMI 89,717) and *Aspergillus parasiticus* var. *globosus* (IMI 120,920). Fungal growth and production of sterigmatocystin and

rubratoxin A by *Aspergillus versicolor* (IMI 16,139) and *Penicillium rubrum* (IMI 136,127) were completely inhibited by the addition of 200 ppm onion oil (Zohri *et al*., 1995).

By using an agar dilution assay, the antifungal activity of aqueous extract prepared from onion (AOE) was evaluated against *Malassezia furfur* (25 strains), *Candida albicans* (18 strains), other *Candida* species (12 strains) as well as 35 strains of various dermatophyte species and compared with the activity of a known antifungal drug, ketoconazole (KTZ). The AOE was found to be able to inhibit growth of all fungi tested in a dose-dependent manner e.g. $MIC_{50}$ (µg/ml) against *Malassezia furfur=2, Candida albicans = 0.125, Trichophyton mentagrophytes* = 0.5 and *Candida parapsilosis* = 0.25 (Shams-Ghahfarokhi *et al.,* 2006).

Wang and Ng (2004) isolated allicepin, a novel antifungal peptide. It exerted an inhibitory activity on mycelial growth in several fungal species including *Botrytis cinerea*, *Fusarium oxysporum*, *Mycosphaerella arachidicola* and *Physalospora piricola.* The $IC_{50}$ values of its antifungal activity against the four aforementioned fungal species were respectively 14.4 µM, 11.7 µM, 10.3 µM and 4.2 µM.

**Antiprotozoal activity**

Antileishmanial effects of aqueous onion extracts (AOE) towards leishmanial promastigotes have also been reported. Five strains of *Leishmania* including *L. major*, *L. major* (Pakistan), *L. tropica*, *L. mexicana* ssp. *mexicana* and *L. donovani* were found to be sensitive. Seventy two hour inoculation of AOE gave an average $IC_{50}$ values of 1.25 mg/ml and 0.376 mg/ml, respectively, against all leishmanial strains tested. A susceptibility order of *L. major* (Pak.) > *L. major* > *L. donovani* > *L. tropica* > *L. mexicana* ssp. *mexicana* was determined. The authors (Saleheen *et al*., 2004) suggest that the susceptibility of the Leishmania strains is a likely consequence of the sulfur compounds present within the onion extract.

**Antinematodicidal activity**

Two unknown oligosaccharides (with 29 carbon atoms) were identified as active principles against *Meloidogyne exigua* Goeldi nematode in a water fraction of methanolic onions extract (Amaral *et al.,* 2003). When exposed to 500 ppm aqueous solutions of each of substances, 55% and 38% of the second-stage juveniles of *M. exigua*, respectively, were dead after 24 h.

**Anticarcinogenic and antimutagenic activities**

Antiproliferative activity of methanolic extracts from red onion, white onion and yellow onion on human cancer cell lines (Calu-6 for human pulmonary carcinoma and SNU-601 for human gastric carcinoma) were measured using MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay (Gorinstein *et al.,* 2009). The antiproliferative activity on SNU-601 was 86.3 ± 4.3% for white onion in concentration of 1000 µg/ml, while other onion samples were inactive. The antiproliferative activity on Calu-6 was 85 ± 3.2% for red onion in a concentration of 1,000 µg/ml, while other onion samples were inactive.

Another study investigated the effect of onion on the chemical induction of preneoplastic lesions in rat liver. Male Sprague-Dawley rats were fed with control or onion powder diet for 9 weeks. Hepatocellular carcinogenesis was induced by a single intraperitoneal injection of diethylnitrosamine (DEN) (200 mg/kg body weight) and 2/3 partial hepatectomy (Bang and Kim, 2010). Dietary supplementation of onion suppressed the formation of placental glutathione-S-transferase positive foci in numbers ($p<0.01$) and area ($p<0.05$). Cytosolic activity of glutathione-S-transferase (GST) was increased by DEN in the rats fed control diet. However, onion significantly decreased GST activity in DEN-treated rats. Glutathione peroxidase activity showed a similar tendency to GST activity. Glutathione reductase activity and microsomal thiobarbituric acidreactive substance value, however, did not show noticeable difference among the groups. These results suggest that onion has anti-tumor activity that suppresses oxidative stress and the formation of preneoplastic foci in the rat liver.

Methanol extracts of onion powder dried by hot air (60°C), vacuum (35°C), and lyophilization (35°C) were used to study the effects of drying method on the flavonoids composition. HPLC analyses showed that freeze- and vacuum-dried onions contained more quercetin glycosides, whereas hot air-dried onion dominated in aglycone. A strong cell proliferation inhibition activity in hot air-dried onion was observed for leukemia cell lines CEM and U937, whereas freeze- and vacuum-dried onions gave comparatively moderate inhibition. Low cell proliferation inhibition was obtained with dried onions in 3 leukemia cell lines K562, P3HR-1, and Raji (Fu, 2004).

Dipropyl sulfide (DPS) and dipropyl disulfide (DPDS) can inhibit both early and late stages of carcinogenesis in male SPF Wistar rats (the corresponding compounds (1 mmol/kg body weight) administered daily by gavage for 4 days) inducing microsomal epoxide hydrolase, glutathione *S*-transferase and UDP-glucuronosyltransferase activities in the liver, and decreasing renal glutathione *S*-transferase activity (Guyonnet *et al.*, 1999). ,

Contrary to this previous work, Srivastava *et al.* (1997) could not detect any induction of microsomal epoxide hydrolase in female A/J mice by DPS and DPDS. The hepatic GST activity toward *anti*-7β,8α-dihydroxy-9α,10α-oxy-7,8,9,10-tetrahydrobenzo(a)pyrene was also not affected by either DPS or DPDS administration.

Phase II xenobiotic metabolizing enzymes confer amelioration of risk arising from potentially carcinogenic chemicals. Xiao and Parkin (2007) isolated and identified potentially cancer preventive constituents from methanolic extracts of green onion (*Allium cepa*) directed by the quinone reductase (QR) induction bioassay using murine hepatoma (Hepa 1c1c7) cells. 5-(Hydroxymethyl) furfural, 1-(4-hydroxy-3-methyl-phenyl)-ethanone, 5-hydroxy-3-methyl-4-propylsulfanyl-5H-furan-2-one, methyl 4-hydroxyl cinnamate and Ferulic acid methyl ester were found active at inducing QR. In a previous study by Xiao and Parkin (2006), other isolates from ethyl acetate extract were shown to possess potent QR-inducing activities *in vitro*: ferulic acid, *p*-hydroxyphenethyl-*trans*-ferulate, *N*-*trans*-feruloyl 3-O-methyldopamine, 5,6-dimethyl-2-pyridinecarboxylic acid, and 1-(6-hydroxy-[3]pyridyl-propan-1-one).

Theoretically, onion and some of its constituents may exert an anticarcinogen action of indirect way by different mechanisms: alteration of carcinogen metabolism either increasing the detoxification enzymatic systems' activity that increases the carcinogen polarity, facilitating its excretion from the body (Guyonnet *et al.*, 1999) or inhibition of oxidative damage due to their antioxidant action or inhibition of cellular proliferation by induction of apoptosis and inhibition of cell division or inhibition of the lipoxygenase and cyclooxygenase activities (Perchellet *et al.*, 1990).

**Antihyperglycemic activity**

Kook *et al.* (2009) investigated the antidiabetic effects of onion and garlic supplementation by meta-analysis. The factors considered were body weight and the levels of glucose, plasma triglycerides (TG), plasma total cholesterol (TC), plasma high-density lipoprotein cholesterol (HDL-C), and liver glycogen (LG). The studies used in meta-analysis showed statistically significant results for most factors. The glucose, HDL-C, TC, TG, and LG levels were significantly different between the untreated diabetic group and the treated diabetic group in single-component onion supplement studies. In the extracts of onion supplement studies, the glucose level of the untreated diabetic group was significantly lower than that of the treated diabetic group. The body weight was significantly different between the untreated diabetic group and the treated diabetic group. The data analysis in this study showed that onion reduced glucose levels and that single component S-methyl cysteine sulfoxide from onion has better antidiabetic effects than the extracts of onion.

Jung *et al.* (2011) examined the hypoglycemic and insulin-sensitizing capacity of onion peel extract in 60% ethanol (OPE) containing high quercetin in high fat diet/streptozotocin-induced diabetic male Sprague-Dawley rats. OPE (0.5% or 1% of OPE, respectively fed *ad libitum* for 8 weeks) improved

glucose response and insulin resistance associated with type 2 diabetes by alleviating metabolic dysregulation of free fatty acids, suppressing oxidative stress, up-regulating glucose uptake at peripheral tissues, and/or down-regulating inflammatory gene expression in liver.

Jelodar *et al.* (2005) induced diabetes mellitus in adult male albino rats, using intraperitoneal injection of 185 mg/kg body weight alloxan. Diabetic rats were fed a diet containing 12.5% body weight onion (ground and mixed with standard pellet) for 15 days. Onion diet was not able to reduce blood glucose significantly compared with the control group ($p<0.05$). In the control positive group, morphometric factors (volume density of B cells, volume density of islets, percent of B cells, number of islets per square millimeter, average area of islets and average volume density of B cell in whole pancreas) were significantly changed in comparison with the control negative (normal health) group, but the same did not show significant change between treated and untreated diabetics.

El-Demerdash *et al.* (2005) studied the effect of onion juice (dose of 1 ml of onion juice/100 g body weight (equivalent to 0.4 g/100 g body weight) orally administered daily to alloxan-diabetic rats for four weeks. Treatment of the diabetic rats with repeated doses of onion juice could restore the changes of many parameters (glucose, urea, creatinine and bilirubin, aspartate aminotransferase, alanine aminotransferase, lactate dehydrogenase, and alkaline and acid phosphatases activities in plasma, as well as glutathione S-transferase in plasma, liver, testes, brain, and kidney) to their normal levels. Results showed that onion juice exerted antioxidant and antihyperglycemic effects and consequently may alleviate liver and renal damage caused by alloxan-induced diabetes.

In another study by Campos *et al.* (2003), male Wistar STZ-diabetic rats were fed with water onion (40 g/100 ml) liquid extract for 30 days. Onion increased the fasting serum high-density lipoprotein levels and demonstrated alleviation of hyperglycaemia in STZ diabetic rats. The hypoglycaemic and hypolipidaemic actions of onion were associated with antioxidant activity, since onion decreased superoxide dismutase activities while no increased lipid hydroperoxide and lipoperoxide concentrations were observed in diabetic rats treated with onion.

The high dietary fat (HF) may impair the anti-diabetic effects reported with dietary onion intake as shown in a current study (Shahidul *et al.*, 2008). Five-week-old male Sprague-Dawley rats were fed a HF-diet for 2 weeks. Normal control (NC), diabetic control (DBC), onion low (ONL, 0.5% of total dietary intake), and onion high (ONH, 2.0% of total dietary intake) groups were created. Diabetes was induced by an intra-peritoneal injection of streptozotocin (40 mg/kg body weight) in all groups except the NC group. After 4 weeks of free access to the allocated diets, animals were sacrificed and fasting blood glucose levels, total cholesterol, LDL-cholesterol, triglycerides, and HDL-cholesterol were measured. Results suggest that onion intake worsens hyperglycaemia and hyperlipidemia induced by the diabetic condition when taken in combination with a HF-diet.

The inhibitory activity of onion extracts against porcine pancreatic α-amylase and rat intestinal α-glucosidase was also investigated by Kim *et al.* (2010). Ethyl alcohol extract of onion skin had the higher α-glucosidase inhibitory activity, ORAC (Oxygen Radical Absorbance Capacity) value and total phenolic content than water extract of skin. The α-glucosidase inhibitory activity of the onion extracts correlated to the phenolic content and antioxidant activity of the extracts. These results suggest that onion, which has high quercetin content, has the potential to contribute as a dietary supplement for controlling hyperglycemia and oxidative stress-linked diabetes complications.

**Effects on skin**

Skin-whitening effects of the methanolic extract from dried red onions skins and some flavonoids were recently described in a mouse melanoma B 16 cell line (Arung *et al.*, 2011). The dried skin extract dose-dependently inhibited melanin formation in B16 melanoma cells. The inhibition of 40–50% of melanin formation was evident at concentrations of 50 and 100 μg/ml without any cytotoxicity. In contrast, the extract of the flesh of the onion did not lead to melanin inhibition, even at concentrations

up to 250 and 500 μg/ml. Notably, isolated compounds quercetin and its 4'-O-glucoside were found to be more-potent inhibitors of melanin formation in B16 melanoma cells than was the positive control, arbutin (198 μM). It should be noted, that the opposite results were reported previously, namely that quercetin enhanced the total melanin content in B16 melanoma cells (Kubo *et al.,* 2007). The reason for the difference of the effect of quercetin on melanin production in cells remains unclear.

**Antiaggregatory activity**

Claeys *et al.* (1984) and Üstünes *et al.* (1985) studied PGE-like bioactivity of ethanolic onion extract as an *in vitro* vascular smooth muscle relaxing and non-vascular smooth muscle stimulating activity in the cascade superfusion system. By monitoring the bioactivity profile throughout the extraction and separation procedures, the active mixture of 9,10,13-trihydroxy-11-octadecenoic acid and 9,12,13-trihydroxy-10-octadecenoic acid was obtained (150 and 410 μg/3 kg FW of onions, respectively).

Atherosclerosis and thrombosis, most common vascular complications of diabetes mellitus, are usually associated with the platelet activation and the release of eicosanoids, which contribute to initiation and aggravation of thrombosis. The aim of another study (Jung *et al.,* 2002) was to investigate whether onion had an antithrombotic effect in diabetic rat. Authors showed elevated serum $TXB_2$ level in streptozotocin-induced diabetic rats. This elevation of serum $TXB_2$ level in diabetes was significantly inhibited by the treatment with aqueous extract of onion (0.5 g/ml/kg/day, i.p.) for 4 weeks. On the other hand, these results (that onion had no effect on the serum $TXB_2$ level in normal rats) are different from others, that onion has inhibitory effect on serum $TXB_2$ levels in normal female rats (Bordia *et al.,* 1996). The reason for this discrepancy might include the difference in animal sex, male vs female, and the difference in cultivar of onion. The results suggest that onion can produce beneficial antithrombotic effect in diabetes.

Briggs *et al.* (2001) studied the *in vivo* effect of onion on platelet aggregation in 11 dogs with mechanically damaged and stenosed coronary arteries. In five dogs, 0.09 6 0.01 ml/kg onion juice administered intravenously abolished cyclic flow reductions (CFR) within 20 min. This was followed by a 60±14% (p=0.002) reduction in collagen-induced ex vivo whole-blood platelet aggregation. Six dogs were given 2.0 g/kg raw onion homogenate intragastrically. CFR were eliminated within 2.5–3 h in five of the dogs. This was accompanied by a 44±24% (p=0.04) reduction in *ex vivo* aggregation. These findings suggest that the consumption of raw onion may help prevent platelet-mediated cardiovascular disorders. However, *in vitro* incubations of onion juice demonstrated that the platelet inhibitory response was significantly greater in dog blood than in human blood.

Makheja and Bailey (1990) have investigated the components of onion that have the greatest effect on arachidonic acid *ex vivo* metabolism in platelets. They reported that the polysulfides, particularly dimethyl- and diallyl trisulfides, found in onion extracts inhibit thromboxane synthesis in platelets. In addition, they observed that incubation of onion oil with washed platelets caused the induction of new lipoxygenase metabolites, identified as 10-hydroxy-11-12-epoxyarachidonic acid and 8,11,12-trihydroxy-5,9,14-eicosatrienoic acid. The total antiplatelet activity of 50% inhibition value of ADP-induced aggregation in 1 ml of human platelet-rich plasma was 3350 units per 100 g of fresh onion, with 36% contribution of adenosine, 22% one of allicin and 42% of polysulfides.

It has been reported by Goldman *et al.* (1996) that the anti-platelet activity from four different genotypes of onion in human blood was significantly correlated with the genotypically-determined sulfur content of the bulb.

Antiplatelet actions of aqueous extract of onion were investigated in rat and human platelets (Moon *et al.,* 2000). IC50 values of onion extract for collagen-, thrombin-, arachidonic acid (AA)-induced aggregations and collagen-induced thromboxane $A_2$ ($TXA_2$) formation were 0.17 ± 0.01, 0.23 ± 0.03, 0.34 ± 0.02 and 0.12 ± 0.01 g/ml, respectively. [3H]-AA release induced by collagen (10 mg/ml) in rat platelet was decreased by onion compared to control (22.1 ± 2.13 and 5.2 ± 0.82% of total [3H]-

AA incorporated, respectively). In fura-2 loaded platelets[1], the elevation of intracellular $Ca^{2+}$ concentration stimulated by collagen was inhibited by onion. Onion had no cytotoxic effect on platelets. Onion significantly inhibited $TXA_2$ synthase activity without influence on COX activity. Platelet aggregation induced by U46619, a stable $TXA_2$ mimetic was inhibited by onion, indicating its antagonism for $TXA_2/PGH_2$ receptor. These results suggest that the mechanism for antiplatelet effect of onion may, at least partly, involve inhibition of AA metabolism, $TXA_2$ synthase inhibition and $TXA_2/PGH_2$ receptor blockade.

**Cardiovascular and lipid-lowering effects**

The effects of onion were studied in rabbits fed a diet containing cholesterol (0.5 g/day). A steady increase in serum cholesterol was observed in the control group (82 mg to 1028 mg at 8 weeks and 2150 mg at 16 weeks). The group supplemented with onion (25 g raw/day) showed increases similar to those of the control, reaching a maximum of 2120 mg at 16 weeks (Jain, 1976).

The lipid lowering action of S-methyl cysteine sulfoxide (SMCS) isolated from *Allium cepa* was investigated in Sprague–Dawley rats fed on 1% cholesterol diet (Kumari and Augusti, 2007). Administration of SMCS at a dose of 200 mg/kg body weight for 45 days ameliorated the hyperlipidemic condition. The lipid profile in serum and tissues showed that concentrations of cholesterol, triglyceride and phospholipids were significantly reduced when compared to their untreated counterparts. The total lipoprotein lipase activity in the adipose tissue was decreased as well as the free fatty acid levels in serum and tissues. The activities of the lipogenic enzymes glucose 6-phosphate dehydrogenase and malic enzyme as also of 3-hydroxy-3-methyl-glutaryl-CoA reductase in the tissues remained low on treatment indicating that SMCS did not favor lipogenesis and cholesterogenesis in the hyperlipidemic animals. The fecal excretion of bile acids and sterols was further increased upon treatment with SCMS.

Yanagita *et al.* (2003) investigated the effects of cycloalliin (present at lower levels in fresh onions but increases during cooking and accounts for about 50% of all sulfur-containing compounds) on lipid metabolism in Sprague-Dawley rats. When supplemented at the 0.1% and 0.3% levels to the atherogenic diet, cycloalliin reduced serum triacylglycerol (TAG) concentration by approximately 40% compared to the control. Serum cholesterol ester level also showed a tendency to decrease in cycloalliin groups. Hepatic lipid levels were comparable among the groups, although TAG and phospholipid contents were slightly higher in both cycloalliin groups. Dietary cycloalliin had no significant effect on hepatic enzyme activities responsible for TAG synthesis (phosphatidate phosphohydrolase, malic enzyme, and glucose-6-phosphate dehydrogenase). In conclusion, dietary cycloalliin has serum TAG-lowering effect without affecting hepatic TAG synthesis and content in rats, suggesting an alteration of lipoprotein assembly and secretion processes in the liver.

White and red varieties of onion were processed by a variety of culinary methods, and the bioactive compounds then determined. During the 30-day trial, the basal diet of the male Wistar rats was supplemented with 1% cholesterol and raw or processed onion. Both raw red onion and red onion subjected to blanching for 90 seconds hindered the rise in plasma lipids more than the other samples studied in the supplemented diets. The decrease in antioxidant activity (ABTS (=2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid), FRAP (= ferric reducing ability of plasma) and DPPH (= 2,2-diphenyl-1-picrylhydrazyl) tests) compared to the cholesterol-supplemented control group was significantly less for the group fed with red onion subjected to blanching for 90 seconds. No histological changes were detected in the studied organs of rats that had been fed with cholesterol. Alkaline

[1] **Fura-2-acetoxymethyl ester**, often abbreviated **Fura-2AM**, is a membrane-permeable derivative of the ratiometric calcium indicator Fura-2 used in biochemistry to measure cellular calcium concentrations by fluorescence. When added to cells, Fura-2AM crosses cell membranes and once inside the cell, the acetoxymethyl groups are removed by cellular esterases. Removal of the acetoxymethyl esters regenerates "Fura-2", the pentacarboxylate calcium indicator. Measurement of $Ca^{+2}$-induced fluorescence at both 340 nm and 380 nm allows for calculation of calcium concentrations based 340/380 ratios

phosphatase levels correlated with classical atherosclerosis indices and determination of alkaline phosphatase is suggested as an additional index in atherosclerosis testing (Gorinstein *et al.*, 2010).

The effect of the methanol-soluble extract of onion on ischemic injury in heart-derived H9c2 cells *in vitro* and in rat hearts *in vivo* were investigated (Park *et al.*, 2009). The onion extract (0.05 g/ml) inhibited the elevation of the reactive oxygen species, mitochondrial membrane depolarization, cytochrome c release and caspase-3 activation during hypoxia in H9c2 cells. In the *in vivo* rat myocardial infarction model, onion extract (10 g/kg) significantly reduced the infarct size, the apoptotic cell death of the heart and the plasma malondialdehyde level. In conclusion, the results of this study suggest that the methanolic extract of onion attenuates ischemia/hypoxia-induced apoptosis in heart-derived H9c2 cells *in vitro* and in rat hearts *in vivo*.

The aim of the next study (Naseri *et al.*, 2008) was to investigate the effects of onion peel 70% ethanolic extract (OPE) on rat aorta contraction and on the hypertension induced by high-fructose diet. Results showed that OPE (0.1, 0.2 and 0.4 mg/ml) reduces aorta contractions induced by KCl or phenylephrine in a concentration-dependent manner, and possibly via inhibition of calcium influx without involving NO, cGMP, endothelium and prostaglandins. OPE did not change the heart rate but reduced the hypertension after 3 weeks of rats supplementation due antioxidant activity and vascular smooth muscle cells calcium influx.

The effects of ethanolic extracts of onion on blood pressure and plasma fatty acids composition were studied in spontaneously hypertensive rats (Kivirantat *et al.*, 1989). Oral administration of the extract for up to 7 weeks (onion extract was added at a level of 4 ml per rat per day during the first 3 weeks, and afterwards at a level of 8 ml per day, 1 ml of the extract corresponded to 1 g of onion) during a normal salt diet or during a high salt diet did not influence the blood pressure. The decrease in arachidonic acid level was statistically significant, no changes in linoleic acid levels were observed. The changes in lipid metabolism could be responsible for the apparent beneficial effects of onion on the inhibition of platelet aggregation.

**Neuroprotective activity**

The antioxidative activity and ameliorative effects of sulfur-containing compounds which occur in Allium vegetables such as onion and garlic on memory impairment were investigated by Nishimura *et al.* (2006). The antioxidative activities of S-alk(en)yl-L-cysteines and their sulfoxides, volatile alk(en)yl disulfides and trisulfides, and vinyldithiins were examined by using human low-density lipoprotein. It was elucidated that the alk(en)yl substituents and the number of sulfur atoms in the compounds were important for the antioxidative activities. To demonstrate the ameliorative effects on memory impairment, onion extract (5 ml/kg/day, concentration of di-*n*-propyl trisulfide (DPTS) in the onion extract (OE) is approximately 0.5 to 0.75 mg/ml) and its constituent DPTS (25 mg/kg/day) were administered to senescence-accelerated mouse P8. The behavioral experiments showed that onion extract and di-*n*-propyl trisulfide had a highly ameliorative effect on memory impairment. Furthermore, it was found that the hippocampus lipid hydroperoxide in senescence-accelerated mouse P8 was decreased by the administration of DPTS. These results suggest that DPTS contained in onion ameliorates memory impairment in SAMP8 mouse by its antioxidant effect.

Global cerebral ischemia was induced by bilateral carotid artery occlusion in Swiss albino mice of either sex for 10 min followed by reperfusion for 24 h in a study by Shri and Bora (2008). Pretreatment with methanolic extract of outer scales (100 mg/kg and 200 mg/kg) and edible portions (100 mg/kg and 200 mg/kg) of onion bulb markedly reduced cerebral infarct size and attenuated impairment in short-term memory and motor coordination. The protective effect of the methanolic extract of outer scales and edible portions of onion bulb was accompanied by a marked decrease in mitochondrial thiobarbituric acid reactive substances.

Hwang *et al.* (2009) prepared onion extract (OE) in 80% ethanol containing 2.19 mg of quercetin in 1 g of OE and found that above 50 mg/kg OE showed a significant neuroprotective effect in male Mongolian gerbils. However, 5 mg/kg quercetin did not show any significant neuroprotective effect. This result may indicate that the neuroprotective effect of OE was not induced by quercetin alone, although a very high dose of quercetin did exert a significant neuroprotective effect.

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

No specific data available.

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

The toxic effects of oral and intraperitoneal administration of onion aqueos extracts on lung and liver tissue of female adult Sprague-Dawley rats were investigated (Thomson *et al.,* 1998). Oral or intraperitoneal administration of low doses of onion (50 mg/kg) to rats had little effect on lung and liver tissues when compared to control animals. There was no significant change in serum and liver protein levels in animals which were given low doses of onion. In contrast, administration of high doses of onion (500 mg/kg) resulted in apparent histological changes in lung and liver tissues of rats and it was lethal to two out of eight rats. All the rats that received the high dose of onion exhibited signs of dehydration and consumed large quantities of water during the experimental period. Macroscopic examination of the livers and lungs of the rats revealed that the livers of several rats treated with high intraperitoneal doses of onion had abscesses in the peritoneum in the vicinity of the livers. Intraperitoneal administration of high doses of onion to rats caused marked changes in lung tissue. In lungs of these animals, alveolar walls were very thick and disrupted. Considerable numbers of red blood cells aggregated together in some alveoli and edema was noticeable in several areas of the lung. Intraperitoneal administration of the high dose of onion resulted in a 25% rate of mortality in this treatment group. These results suggest that low doses of onion are nontoxic.

Tests on reproductive toxicity, genotoxicity and carcinogenicity have not been performed.

### *3.4. Overall conclusions on non-clinical data*

Numerous *in vitro*, *ex vivo* and *in vivo* (rats, guinea pigs, mice, dogs, rabbits, gerbils) studies were published with essential oil, different extracts (water, diluted ethanol, butanolic fraction, …), thermally elaborated water extracts, extracts from thermally elaborated onions, and with some isolated constituents of onions. All have confirmed some positive activities related to the traditional use of onions, *e.g*. antimicrobial effects in high concentrations of onion essential oil. However, all results are mutually inhomogenous and incomparable to make the final positive conclusion and/or statement on the unambiguous non-clinical effects.

# 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

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

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

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## *4.2. Clinical Efficacy*

## 4.2.1. Dose response studies

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## 4.2.2. Clinical studies (case studies and clinical trials)

**Effects on skin**

The effectiveness of topical use of crude onion juice in the treatment of patchy alopecia areata in comparison with tap water was tested (Sharquie and Al-Obaidi, 2002). The patients were divided into two groups. The first group [onion-juice treated] consisted of 23 patients, 16 males (69.5%) and 7 females (30.5%). Their age ranged between 5-42 years with a mean of 22.7 years. The second group [control; tap-water-treated] consisted of 15 patients, 8 males (53.3%) and 7 females (46.6%). Their age ranged between 3-35 years with a mean of 18.3 years. The two groups were advised to apply the treatment twice daily for two months. Re-growth of terminal coarse hairs started after two weeks of treatment with crude onion juice. At four weeks, hair re-growth was seen in 17 patients (73.9%), and, at six weeks, the hair re-growth was observed in 20 patients (86.9%) and was significantly higher among males (93.7%) compared to females (71.4%) $p<0.0001$. In the tap-water treated-control group, hair re-growth was apparent in only 2 patients (13%) at 8 weeks of treatment with no sex difference.

**Anticarcinogenic and antimutagenic activities**

Onion consumption is associated with reduced risk of developing brain cancer, as confirmed in a hospital-based case-control study conducted in the North East China, between May 1993 and May 1995, on a total of 129 histologically confirmed brain cancer cases (Hu *et al.,* 1999).

In the Netherlands Cohort Study, comprising 120 852 Dutch men and women aged between 55 and 69, Dorant *et al.,* (1996) reported a strong association between onion consumption and a reduction in the incidence of stomach carcinoma. However, in the same study group, the authors found no evidence for a protective effect of *Allium* vegetable consumption and reduced risk of developing colon, rectum carcinoma, lung or female breast cancer.

**Cardiovascular effects**

In an open, randomized placebo-controlled, double-blind, cross-over phase I study (Kalus *et al.,* 2000) in 10 apparently healthy volunteers (8 men and 2 women), a decrease in arterial blood pressure, a reduction in plasma viscosity and haematocrit were observed 5 hours after administration of an onion-olive-oil macerate (corresponding to a mean daily dose of 2.5 g fresh onion). There was a tendency to reduce by 5-8% in systolic and by 5-10% in diastolic blood pressure. There was still a minimal reduction of blood pressure detectable 8 hours after administration. The mean reduction of 1.6% in haematocrit and 0.04 mPa/s in plasma viscosity were measured.

**Antiaggregatory activity**

Twenty-two ambulant patients aged from 19 to 78 years were selected for the study. They were convalescing from bleeding peptic ulcer, free of symptoms and not on any drug therapy at the time of examination. The patients were divided into two groups.

Group 1 (14 patients): After withdrawal of the blood samples, the first seven patients were given a

breakfast containing 98 g of fat and new samples of blood were collected after two and three hours. The procedure was repeated the following day, but this time 60 g of fried onions were added to the meal. For the remaining seven patients the routine was reversed and they were given the fried onions with their breakfast on the first day.
Group 2 (8 patients): The method was identical to that outlined above, the only difference being that four patients in this group had boiled onions with their breakfast on the first day and none on the second. The opposite was the case with the remaining four patients. The results show that after ingestion of a fat-enriched breakfast a decrease of fibrinolytic activity occurs. The addition, onions, whether fried or boiled, not only prevented this reduction but also caused a marked increase of fibrinolytic activity. Recalcified clotting-times, thrombotests, and cholesterol and fibrinogen levels were not significantly changed (Menon *et al.*, 1968).
The effect of onion juice consumption on some blood properties, influenced by consumption of 100 g butter, has been studied in 10 healthy adult males, between 35 and 50 years old (Bordia et *al.*, 1975). The juice was prepared freshly from 50 g of onion and it was administered orally as a single dose. After administration of onion juice, the blood coagulation time (which had decreased by 13.3% after ingestion of butter) was increased by 9.8%. Analogically, fibrinolytic activity (decreased by 48.6%) increased by 15.7%.

Srivastava (1989) examined the effects of onion consumption (ca. 70 g of raw onion daily for a period of 7 days) on platelet thromboxane (TXB2) production in five women in the age range of 25 to 65 years. Onion feeding slightly but not significantly increased amount of TXB2, from mean 910 to 1005 pmol/ml of serum.
The effect of cycloalliin on fibrinolytic activity and platelet aggregability was tested in venous blood from 18 male volunteers aged 19 to 77 years in a randomised double-blind cross-over study (Agarwal *et al.*, 1977). Cycloalliin was given in a dose of 0.25 g per subject with an interval of not less than 48 h and not more than a week between tested substance consumption. Fibrinolytic activity values measured as by the euglobulin lysis time (ELT, in seconds) were: for cycloalliin before treatment – 34.7, after treatment – 61.2; for placebo before treatment – 36.8 and after treatment – 39.2. The results of the platelet aggregation tests did not show any consistent effects.

**Antidiabetic activity**
A comparative study, using a crossover design, was carried out on 20-well controlled diabetic out-patients to investigate the metabolic effects of the diet (68% cal carbohydrate, 20% cal fat, 12% cal protein) plus 3 x 20 g fresh onion per day during two weeks. Study resulted in significant decrease in blood sugar level (4.37 mg%, $p<0.05$). No blood-lipid changes changes occurred (Tjokroprawiro *et al.*, 1983).

**Lipid-lowering effects**
The effect of onion on alimentary hyperlipemia, induced by feeding 100 g butter, has been studied in 10 healthy adult males, between age of 35 and 50 years (Bordia *et. al*., 1975). The juice was prepared freshly from 50 g of onion and was administered orally as a single dose. Administration of onion juice completely prevented the rise in serum cholesterol (in fact slightly lowered it).

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

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## *4.3. Overall conclusions on clinical pharmacology and efficacy*

No data on a medicinal monopreparation are available.

All clinical studies were performed with a small number of patients; in some cases the study design was not sufficiently described. In others, onion bulb (raw or heated) or juice from raw bulb was used. In the case when an herbal preparation was used, it was not sufficiently characterised. Therefore, the clinical studies cannot be considered acceptable to support the WEU.

# 5. Clinical Safety/Pharmacovigilance

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

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## *5.2. Patient exposure*

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## *5.3. Adverse events and serious adverse events and deaths*

**Allergic activity**
Case reports of bronchial asthma (one case), contact dermatitis (one case) and three cases of rhinoconjunctivitis are described. In order to study the prevalence of allergy to onion, skin prick tests were performed. Valdivieso *et al.* (1994) were able to detect IgE-specific antibodies directed against the onion water extract, with both skin and *in vitro* tests. They were encountering respiratory allergic reactions - IgE-mediated in three patients and also cell-mediated (type IV) in the case of contact dermatitis in another patient.

## *5.4. Laboratory findings*

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

## *5.5. Safety in special populations and situations*

No specific data are available on *Allium cepa* L., bulbus (onion).

## *5.6. Overall conclusions on clinical safety*

No specific data are available on *Allium cepa* L., bulbus (onion) based monopreparations.

# 6. Overall conclusions

The period of at least 30 years of medical use as requested by Directive 2004/24/EC for qualification as a traditional herbal medicinal product is not fulfilled for two onion monopreparations as reported by national competent authorities (Table 1). The first declared monopreparation (Hungary) was registered in 1996, the second one (Poland) in 1994; both registered as TU preparations. Only inhomogenous non-clinical data were published for different onion extracts or their constituents. No clinical data for monopreparations were published. Clinical data are available only for fresh onion or fresh onion juice and for onion after thermal treatment (boiled/fried). In one study, the onion-olive oil macerate was used but the herbal preparation is not satisfactorily described. In the case when an herbal preparation was used, it was not sufficiently characterised. Therefore, the clinical studies cannot be considered acceptable to support the WEU.

Based on these facts, a monograph and a Community list entry cannot be established.

# Annex

## *List of references*