

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

CLINICAL TRIAL ON TERAPEUTIC EFFICACY, TOLERABILITY AND SAFETY OF PERORAL SOLUTIONS VARUMIN 1 AND 2 (INTER-EVROGENEKS) IN A COMBINED TREATMENT WITH POLYVITAMIN – ANTIOXIDATIVE THERAPY IN PATIENTS WITH BIOLOGICALLY (PCR) DETECTED HPV INFECTION OF HIGH-RISK TYPE, WITH OR WITHOUT BIOPTICALLY VERIFIED CIN I

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INTRODUCTION

Human Papilloma Virus (HPV) is a DNA virus with an affinity towards epithelial cells. There are over 100 different types, of which 40 are detected into the human anogenital tract and some of them (most frequently types 16, 18 31 and 45) are potentially oncogenic and therefore are named as high-risk types, unlike the rest of them, predominantly types 6 and 11, that are named low-risk types because they always cause benign lesions.

Form	HPV Type
Plantar warts	1, 2, 4
Condylomas	1, 2, 4, 26, 27, 29, 41, 57
Flat warts	3, 10, 27, 28, 41, 49
Genital warts	6, 11, 30, 40-45, 51, 54
Cervical carcinoma	16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58
Precancerous lesions	16, 18, 34, 39, 42, 55
Laryngeal papilloma	6, 11, 30

HPV is transmitted usually by sexual intercourse. That is the most frequent sexually transmitted microorganism. Using a condom in order to hamper the infection is thought to be an unsafe method. The immunity towards HPV is typically-specific, so infection with one HPV type does not prevent infection with another different type. Concomitant infections with more than one HPV type are not rare. At the majority of females the cervical HPV infections remain asymptomatic and transient, and sometimes can not be detected even with the most sensitive methods, such as GAA (gene amplification assay). The same counts for the high-risk HPV, that leads to the conclusion that the majority of the subclinical infections do not mean a risk for cervical cancer development, but that immune uncompetitive (non-responsive) organism allows persistence of the virus, which is the basis of the strong association with the precancerous cervical lesions.

Persistent high-risk HPV infection, depending on the site, is strongly associated with development of cervical, laryngeal and pharyngeal squamous cell cancer (Braakman et al., 2004), vulvar, anal (Carter et al., 2001) and penile squamous cell cancer (Ferreux et al., 2003).

The immune response to HPV basically relies on cellular immunity, no matter of the eventual findings of IgG or IgA antibodies for the antigen fractions, in the cervical mucus, into and around the lesion. After penetrating into the epithelial cell, HPV replicates its DNA and afterwards there is the consequential presentation of the antigens on the infected cell membrane, together with secretion of Interferon type I (alpha and beta), which produces traction of the antigen-presenting cells. The antigen-presenting lymphocytes forward the chain reaction in the regional lymph nodes with the inductor T-lymphocytes (CD4+), which recognize the antigen in the form of peptide associated to the MHC class II molecules (tumor cells do not present MHC class II molecules, only the infected cells that did not turn into neoplastic cells). On the other hand, the infected cells with incorporated viral DNA secrete cytokines with immunosuppressive features and cytokines with inductive features towards the suppressor T-lymphocytes.

In the regional lymph nodes many reactions happen simultaneously – antigen-presenting cells secrete Interferon alpha and Interleukin 12, that stimulates proliferation and cytotoxic effect of the cytotoxic T-lymphocytes, macrophages and NK-cells (large granulated lymphocytes), which migrate towards the lesion. In the mean time, the humoral immunity turns on – memory-

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lymphocytes for HPV antigens E2 (CD4+), E6 and E7 (CD8+) are produced. So, this way the first goal is achieved – localization of the infection.

Antibodies (IgG class III) for the E2, E6 and E7 antigens assist to the mechanisms of cellular cytotoxicity, by opsonising the infected cells, thereby allowing cytotoxic effect (exocytose of granules through transmembrane signalized apoptosis) of the NK-cells (present CD56 and CD16 receptors for binding to the target-cell opsonised by IgG class III).

Spontaneous healing of the lesion can be achieved through the action of activated cytotoxic lymphocytes, via the following mechanisms of action:

1. Granule exocytose via perforins, producing osmotic disbalance, and via serine esterase (granzimes) through a not yet known mechanism, inducing DNA degradation. Theories about the cytotoxic effect of the lymphocytes through lymphotoxins and proteoglycans are still not confirmed.
2. Transmembrane signaling apoptosis: activated cytotoxic T-lymphocyte presents a ligand-molecule on its membrane for Fas-receptor of the infected cell membrane, and their interaction generates a signal for apoptosis
3. Macrophages and NK-cells afterwards clean up the cellular debris

HPV positive neoplasmas have increased expression of viral oncogenes E6 and E7. HPV E7 protein interacts with pRb (retinoblastoma-regulator of the cell-cycle), thus starting the synthetic phase of the cell-cycle of the infected cells. For cancerogenesis, continuous expression of E6 and E7 is needed, and because they are from viral origin, are ideal for immunostimulated targeting. There are data that pointing out to the presence of a life-time subclinical high-risk HPV infections (maybe better term would be colonization), so if there is no moment of immune disregulation, that the subclinical lesion would have never been detected, no matter to the regular investigations, even with the most sensitive detecting methods.

Concerning the latent period from transmission to manifested HPV lesion, hypothesis are diverse and often opposite, ranging from a month to dozens of years. Contrary, the fact that most detectible are the lesion of the relatively younger population, maybe indicates that if after transmission in a relatively short time lesion is not developed, that the risk for developing a late cervical lesion is rather small.

Thoughts differ and frequently are opposite on the issue of duration of latency period to manifest HPV lesion, usually ranging from one month to several tenths of years. Contrary, the fact that most detectible are lesions of relatively young population, indicates that if after transmission a lesion does not develop in a relatively short time, than the risk for developing a late cervical lesion from the same HPV type is relatively small.

Theories of immune disregulation for precancerous lesions that develop into cancer are still not proven because of several issues:

- infected cells do not present co-signalizing molecule B-7 for cytotoxic T-lymphocytes
- theory of antigen modulation (as a result from an adequate immune response antigens from the infected cell are lost because of endocytosis – release of the membrane complex viral antigen / antibody, so for a certain period of time infected cell can not induce immune response)
- immune suppression induced by viral cytokines, etc.

There are no ways yet to prevent cervical, vulvar, anal, laryngeal and oropharyngeal HPV lesions with chemotherapy, so researchers worldwide do not perform chemoprevention trials any more.

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Due to the lesion treatment, depending on HPV type (high or low risk), and according to PAP findings, there are several modalities, such as Podophyllotoxin as 0.15% cream or 0.5% solution (Ib, A), Imiquimod as 5% cream (Ib, A), Interferon beta gel 0.1 million IU/g, 3-chloroacetic acid, cryotherapy, electrocauterization (Ib, A) / laser evaporation (IIb, B), curettage, and lately DNA-free viral particles vaccines.

After many unsuccessful attempts to detect any therapeutic effect on HPV lesions of beta-carotene (Keefe KA et al.), lycopene, ascorbic, folic acid, or any other vitamin or antioxidant, separately investigated or combined, lately several trials reported step forward for non-invasive, but successful treatment:

- Coriolus versicolor extract (Mycology Research Laboratories Ltd (500 mg/tablet). Couto JS and Pereira da Silva D, 2008) presented 72% regression of LSIL lesions compared to 45% placebo, and 90% negativization of HPV PCR typization in a short period of time, compared to 8,5% placebo.
- Combination of aloe vera extract, propolis and β -interferon, presenting several simultaneous actions on the HPV lesions, such as antibacterial, antifungal, antiviral, antineoplastic, trombolitic and immune modulating (Iljazovic et al. 2006).
- Immunotherapy with human leukocyte ultrafiltrate (H. Spitzbart, U.B. Hoyme, 2000) presented 88,7% negativization of HPV PCR typization in a 6 weeks period of time, and after the second administration the rest 11% went negative as well.
- A-007 intravaginal gel (Tigris Pharmaceuticals, Multi-Center Phase II Clinical Trial of A-007 in Patients with High-Grade CIN and Invasive Cervical Carcinoma St.I) – at 77,78% of patients complete response was observed, at 11,22% partial response, 11% non-responders.
- Indole-3-Carbinol – presented dose dependent effect in several trials (Wattenberg, L. W. & Loub, W. D. 1978, Grubbs C. J. et al.1995, Newfield et al. 1993).

Based on many trials and reports for the immunomodulatory action of active substances from mistletoe, St John's wort, aloe and propolis (lectins, viscotoxins, rutine, quercetine, proanthocyanidines, aloe-emedine, aloe-barbaloine, acacetine, acemannan, betamannan, etc), and guided by many well documented and successfully treated cases of HPV lesions with Varumin solutions, and in coordinance with the experts reports for Varumin solutions, we performed this trial.

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DESIGN

Double-blind, placebo-controlled, randomized, longitudinal trial, performed at the University Gynecology and Obstetrics Clinic, University St. Cyrillus and Methodius, Skopje, Macedonia, from October 2007 till October 2008.

Including criteria were: females in which a high risk HPV is detected by PCR on a cervical smear, with or without biotical proven CIN 1, in a good general condition, in which conditions leading to exitus or a significant worsening of the general health condition before the closure of the trial are excluded.

Non-including criteria were previously detected hypersensitivity to some of the components of the investigated agent, patients suffering from severe cardiovascular, respiratory, renal, hepatic and cerebral impairments (decompensated), patients with malignancies, autoimmune diseases, AIDS, diabetes, refractory epilepsy, inflammatory intestinal diseases (Chronn`s disease, Ulcerative colitis), intestinal obstruction, appendicitis or a stomach discomfort by unexplained etiology, patients receiving corticoid or any other immunosuppressive therapy and anti-coagulant therapy, patients with hematuria from improvable etiology, pregnancy and lactation.

Exclusion criteria were: patients who do not submit to the instructions for administration of the investigated agent, patients in which a higher degree of CIN (2, 3) or in situ cervical carcinoma was diagnosed on a biopsy, patients where clinical signs of hypersensitivity occurred during administration of the investigated agent (as monotherapy or in a combined therapy with other, previously used medicines), patients in which gastrointestinal difficulties occurred, or cardiac arrhythmias, albuminuria, hematuria, which were directly associated with the administration of the investigated agent (other genesis of the emerged symptoms was excluded) and pregnancy.

During the trial, as an additional therapy based on indication because of eventual comorbidity, were allowed: NSAIDs, analgesics, diuretics, H2 blockers or proton pump blockers, beta-blockers, sedatives, antidepressants, mood stabilizers, antibiotics, vitamins and antioxidants.

Recruited patients on voluntary basis (n=88) were randomized into two groups:

- Group 1: Varumin 1 (50 ml) and 2 (200 ml) solutions plus vitamin and antioxidant supplements
- Group 2: Placebo 1 (50 ml) and 2 (200 ml) solutions plus vitamin and antioxidant supplements

AIM

Evaluation of therapeutic efficacy towards cervical high-risk HPV lesions, evaluation of safety and tolerability of per oral solutions Varumin 1 and 2. Statistically significant difference between groups in negativization of the PCR HPV findings and diminishing abnormal PAP findings would present therapeutic option of Varumin 1 and 2.

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MATERIAL

CHEMICAL AND PHARMACOLOGICAL PROFILE OF INVESTIGATED AGENT

Qualitative and quantitative composition of Varumin 1:

100 ml solution contains:

- Water extract of herbal drugs * 98.7 g
- Aloe 0.3 g
- Propolis 0.8 g
- Preservatives (MPB 0.18 g and PPB 0.02 g) 0.2 g

*100 ml water extract of herbal drugs contains:

- Visci albi herba (*Viscum album*) 1.0 g
- Aqua purificata ad 100.0 ml

Qualitative and quantitative composition of Varumin 2:

100 ml solution contains:

- Water extract of herbal drugs * 99.0 g
- Aloe 0.8 g
- Preservatives (MPB 0.18 g and PPB 0.02 g) 0.2 g

* 100 ml water extract of herbal drugs contains:

- Inulae helenii radix (*Inula helenium*) 1.6 g
- Visci albi herba (*Viscum album*) 1.2 g
- Corni mas cortex (*Cornus mas*) 0.6 g
- Calendulae flos (*Calendula officinalis*) 1.0 g
- Milefolii herba (*Achillea millefolium*) 0.4 g
- Cynodonii rhizoma (*Cynodon dactylon*) 1.5 g
- Hyperici herba (*Hypericum perforatum*) 1.4 g
- Aqua purificata ad 100.0 ml

MPB – methyl hydroxybenzoate

PPB – propyl hydroxybenzoate

Giving details about the active ingredients of Varumin 1, the following ones are worth mentioning: considering Aloe, German Commission E for evaluation of herbal medicinal products approves the use of Aloe (*Aloe ferox* Mill., *Aloe vera* Mill., Aloaceae former Liliaceae) in the treatment of constipation as ‘potent colon laxative’ for ‘patients for who an easy defecation with a soft stool is desirable’ (German Commission E Monographs, no. 154, 1985). According to the PDR for Herbal Medicines (2000), Aloe shows laxative, antibacterial/antiviral and antineoplastic effects.

Mistletoe herb (*Visci albi herba*, *Viscum album* L., Loranthaceae) is approved by the German Commission E (Monograph no. 228, 1984) for treating of ‘degenerative inflammation of the joints by eliciting cuti-visceral reflexes following local inflammation brought about by intradermal injections’ as well as for ‘palliative therapy for malignant tumors through non-specific stimulation’. According to the PDR for Herbal Medicines (2000), the drug shows hypotensive, cytotoxic and immunostimulative activity. Traditionally, it is used in the folk medicine of the Balkan region for ‘improving metabolism, regulation of high blood pressure, as haemostatic agent and for treatment of CNS disturbances (Dervendzi 1977, Petkov 1982, Tucakov 1984).

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Propolis (bee glue) is a material gained by honeybees, well known and used in folk medicine in the world, on the Balkan Peninsula and on the territory of our country (Lukic 1988, Gorunovic 2001). Propolis is still very popular for medical use in traditional systems because it points out antibacterial, antiviral, antifungal, anti-inflammatory and local anesthetic activity (Evans 2002, Capasso 2003). It has been used for a long time for treatment of laryngitis, gastric disturbances, dermatitis, duodenal and oral ulcers, etc. (Capasso 2003).

Giving details about the active ingredients of Varumin 2, the following ones are worth mentioning: the German Commission E (Monograph no. 85, 1988) approves elecampane root (*Inulae helenii radix*, *Inula helenium* L., Asteraceae) for treating ‘complaints and problems affecting the respiratory and gastrointestinal tract’ as well as ‘for treating problems related to the kidneys and lower urinary tract’. According to the PDR for Herbal Medicines (2000), the drug shows antiphlogistic and antibiotic effects, antifungal, antimicrobial and anthelmintic activity as well as antitumor activity. Traditionally, in the folk medicine of the Balkan people elecampane is used ‘for enhancing the metabolism, treating the illnesses of the respiratory system, as a diuretic and for treating digestive disorders’ (Dervendzi 1977, Petkov 1982, Tucakov 1984).

The bark of the cornel tree (*Corni mas cortex*, *Cornus mas*, L. Cornaceae) is a drug used in Balkans folk medicine for treating abdominal pain and regulating abdominal functions (Petkov 1982, Tucakov 1984).

Marigold flower (*Calendulae flos*, *Calendula officinalis* L., Asteraceae) is approved by the German Commission E (Monograph no. 50, 1986) for ‘topical use in treatment of inflammatory changes in the mucous membranes of the mouth and throat’ and ‘for external use in treatment of wounds, including those healing with difficulty as well as for *ulcus cruris* (leg ulcers)’. According to the PDR for Herbal Medicines (2000), the drug shows antimicrobial and antiviral effects, topically granulation and epithelization of damaged skin. Traditionally, in the folk medicine of the Balkan people marigold is used ‘for healing of wounds and ulcers on the skin, for treating inflammations in large intestine, for decreasing abdominal pain, etc.’ (Dervendzi 1977, Petkov 1982, Tucakov 1984).

Yarrow herb (*Millefolii herba*, *Achillea millefolium*, L. Asteraceae) is approved by the German Commission E (Monograph no. 22a, 1990) for treating of ‘loss of appetite, dyspeptic disorders, such as mild crump-like complaints in the abdominal region, for hepatic and biliard disorders as well as in hip baths for painful, crump-like conditions of psychosomatic origin (in the lower part of the female pelvis)’. According to the PDR for Herbal Medicines (2000), the drug shows holagoge activity, spasmolytic, antiedemic and anti-inflammatory effects. Traditionally, in the folk medicine of the Balkan people yarrow is used for ‘treatment of hepatic and biliar illnesses, as minor bleeding haemostatic agent (uterine, hemorrhoid) and for appetite improvement’ (Dervendzi 1977, Petkov 1982).

Cynodonii rhizoma (*Cynodon dactylon* Pers., Poaceae) is a dried underground part of *Cynodon spp.* mentioned in the folk medicine of the Balkans as diuretic agent and drug used for herbal obesity treatment (Petkov 1982, Tucakov 1984).

St. John’s wort herb (*Hyperici herba*, *Hypericum perforatum* L., Hypericaceae) is approved by the German Commission E (Monograph no. 228, 1984) for internal use in the treatment of ‘psychogenic disturbances, depressive states, anxiety, and/or nervous excitement’. For external use, oily hypericum preparations are recommended for treatment and after - treatment of incised and contused wounds, myalgia, and first degree burns’. According to the PDR for Herbal Medicines (2000) the drug shows antidepressive effect after oral administration and in external use, anti-inflammatory and antimicrobial activity.

Traditionally, in the folk medicine of the Balkans, St. John’s wort is used for regulating of gastric and intestinal functions, as an aperitif and stomachic, for treating hepato-biliary disorders, nervous

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

conditions and tensions, insomnia, and externally for wound healing, hemorrhoids, for massage in cold and flew conditions and for cleaning the skin' (Dervendzi 1977, Petkov 1982, Tucakov 1984).

The following dosage is the recommended dosage by the German commission E and PDR for Herbal Medicines for the above mentioned drugs:

Varumin 1:

- **Aloe*: average daily dose 0.05 - 0.2 g, which suits 10 - 30 mg hydroxyanthraquinone per day or 0.1 g single dose at evening (WHO 1999).
- *Visci albi herba*: recommended single dose for preparation of tea is 2-2.5 g; 2-3 times a day, approximately about 6-7.5 g daily dose.
- *Propolis*: recommended daily dose for oral administration is 3.0 g (Capasso 2003).

Varumin 2:

- *Inulae radix*: average daily dose is about 4 g.
- *Visci albi herba*: recommended single dose for preparation of tea is 2-2.5 g; 2-3 times a day, approximately about 6-7.5 g daily dose.
- *Corni mas cortex*: the drug is used in similar way as *Corni mas fructus* for which recommended single dose is one soup-spoon three times a day (approximately about 30 g per day) (Petkov 1982).
- *Calendulae flos*: single dose for oral administration is 1-2 g (average daily dose 3-6 g).
- *Milefollii herba*: single dose is 2-4 g; average daily dose 6-12 g.
- *Cynodonii rhizoma*: recommended daily dose is 20 g (Petkov 1982).
- *Hyperici herba*: average daily dose is 2-4 g.

*Considering the dosage in which Aloe is taken during the oral administration, it is important to mention that posology of Varumin 1+2 have some specifics. The first day administration starts with the whole content of Varumin 1 solution (50 ml that contain 0.15 g aloe). After 6 h of taking the first solution, the patient is supposed to take one soup-spoon of Varumin 2 solution (10-15 ml which contains 0.10-0.13 g aloe) four times a day.

PHARMACODYNAMICS

ACTIVE INGREDIENTS

Aloe - Aloe extract is the juice from the leaves of two species of Aloe plants (*A. ferox* and *A. vera* Syn. *A. barbadensis*, Liliaceae). Two types of aloe are produced, Aloe capensis or Kap-aloe from *A. ferox* and Aloe barbadensis or Barbados-aloe from *A. vera* (*A. barbadensis*), (Bruneton 1999, Evans 2002, WHO 1999).

Constituents - Aloe is used as crude or purified drug known as Aloe extractum siccum. Quality standards for crude drug require total contents of hydroxyanthracene derivatives not less than 18% for Aloe capensis and 28% for Aloe Barbadensis, calculated as anhydrous aloin (barbaloin) (Ph. Eur., USP).

Besides hydroxyanthracene derivatives aloin A and B, Aloe contains other anthracene derivatives such as aloinosides A and B, free aloe-emodin, 5-hydroxyaloin A and B (for *A. capensis*) and 7-hydroxyaloin A and B (for *A. barbadensis*), chromone derivatives of aloe-resin group and sugar

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

free aloesones, bitter-substance aloenin, glucomannan, acemannan, betamannan, tannins, sterols, gamolenic acid, saponins, traces of essential oil, organic acids, mineral elements, etc. (Bruneton 1999, Evans 2002, WHO 1999).

Viscum album - Mistletoe herb (*Visci albi herba*, *Viscum album* L., Loranthaceae) is dried herbaceous part of the plant mistletoe, which grows as semi-parasite plant on different trees from Rosaceae (such as apple - tree, plum tree, pear - tree, etc).

Mistletoe contains different chemical constituents: triterpenoides and steroids, amines, phenols such as flavonoids (e.g. quercetin derivatives), etc. Most important components that produce the favorable biopharmacological activity are several different groups of specific proteins such as viscotoxins A2, A3 and B, and lectins (glycoproteins) ML I, ML II and ML III, (Bruneton 1999, Evans 2002, WHO 1999, Kulevanova 2004).

Propolis - Propolis is a resinous material, dark - colored collected by honeybees from the buds of living plants mixed with bee wax and salivary secretions (Sljahov 1990, Gorunovic 2001, Evans 2002). Large amount of propolis composition are resins (40 - 60 %) chemically still unknown, wax (about 30 %) and different impurities (up to 20%).

The active constituents of propolis are phenols and phenylpropanoides, mainly free and ester - bounded alcohols (vanilil alcohol), acids (caffeic, ferulic, coumaric), aldehydes and other components such as flavonoids (up to 6.0 %) (Evans 2002), terpenes, lignans, lipids, mineral elements (Mn, Cu, Zn) (Evans 2002), sugars, etc. (Capasso 2003). Depending on the floral characteristics of the geographical origin of propolis, the chemical composition of some constituents varies a lot, qualitatively and quantitatively. Besides other constituents of propolis, this variation mainly refers to flavonoids. Propolis, originated in Europe, is mainly a product obtained by preparation of cuticle waxes from *Populus* spp., Fagaceae, which consequently gives the reason of a specific flavonoid composition consisting of hrizine, galangine, tectohrizine, acacetine, quercetin, etc. (Gorunovic 2001).

Inulae radix (Helenii rhizome) - Elecampane root (*Inulae radix*, *Inula helenium* L., Asteraceae) is dried root of the elecampane plant, which is native for the southern part of Europe. It is naturalized in central Europe, Near East and North America.

The drug contains sesquiterpene lactones (eudesmanolides such as alantolactone, isoalantolactone, dihydroalantolactone and others and germacrene D-lactone). The mixture of alantolactone derivatives is known as helenine. Besides them, the drug contains small amounts of essential oil, oxyacetylene's, triterpens (friedeline, dammarandienol, dammarandienol acetate), sterols (β - sitosterol and stigmasterol), large quantity of inulin (40%), etc (Wichtl 1994).

Corni mas cortex - the drug is represented by dried bark of cornel - tree *Cornus mas* L., Cornaceae. It possesses similar chemical composition as the plant fruits (*Corni mas fructus*), containing organic acids, tannins, etc. (Tucakov 1994, Petkov 1982).

Calendulae floss - Marigold flowers (*Calendulae flos*, *Calendula officinalis* L., Asteraceae) are the ray florets of the completely unfolded, collected and dried capitula of the plant. Marigold is native for Europe. Nowadays is cultivated in the Mediterranean region, the Balkans, Germany.

The drug contains: essential oil (0.12-0.40%) with menthone, isomenthone, γ -terpinene, α -mumulene, γ - and δ -cadinene, caryophyllene, α - and β -ionone, geranylacetone, carvone, caryophyllene ketone etc.; sesquiterpenes; flavonol glycosides (3-O-glycosides of isorhamnetin and quercetin, 1,5%); triterpene saponins based on oleanolic acid (2-10%) (i.e. calendulosides); triterpene alcohols (α - and β -amyrins, taraxasterol, calenduladiol, arnidiol, faradiol, pentacyclic

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triterpene triols, etc.); carotenes, xanthophylls, polyacetylenes, phenol - carboxylic acids, bitter substances, sesquiterpene lactones, tannins, immunostimulative polysaccharides (rhamnoarabinogalactan and two arabinogalactans) (Wichtl 1994).

Milefolli herba - Milefolli herba is dried herbaceous part of yarrow (*Achillea millefolium* L., Asteraceae) collected in flowering stage. Contains essential oil (0.2 - 1.0 %) which depending on the origin, may contain up to 50% chamazulene (azulene chemotype) or no chamazulene (azulene - free chemotype). In azulene type of essential oil large amount of α - and β -pinene are also present, followed by caryophyllene, while in azulene-free type camphor, sabinene, 1.8-cineole, α -pinene and isoartemisiaketone are the major components. Achilicin has been identified as one of the pro-azulenes. Besides this, guaianolides and germacranolides are also present, polyalkynes (pontica epoxide), flavonoids (apigenine, luteoline and their glycosides), phenolic acids, triterpenes, sterols, cyanogenic glycosides, coumarins, tannins, etc. (Wichtl 1994).

Cynodonii rhizoma - The drug is represented by dried underground part of the plant *Cynodon dactylon* (L.) Pers., Poaceae. It contains saponins, mucilage, sugars, triticyne and other components (Petkov 1982).

Hyperici herba - Hyperici herba is dried herbaceous part of St. John's wort (*Hypericum perforatum* L., Hypericaceae) collected in flowering stage.

The drug contains: 0.05-0.30% naphthodianthrones (hypericin, pseudohypericin and related components); 2 - 4% flavonoids, derivatives of quercetin (hyperoside, quercitrin, isoquercitrin, rutin) and apigenine (biapigenine), up to 3% hyperforin and related phloroglucinols, structurally close to the bitter substances of hops, up to 0.3% essential oil, up to more than 10% tannins, small amounts of procyanidins, etc. (Wichtl 1994).

SPECIFIC PHARMACODYNAMICS

Laxative effect

Anthraquinone glycosides from Aloe have the most pronounced laxative activity. After oral administration, anthraquinone glycosides are hydrolyzed to active anthrone which stimulates colonic motility over the inhibition of Na^+/K^+ - pump and chloride channels under the influence of the intestinal bacterial flora and the enzyme glycosidase. This leads to increased liquid and chloride secretion inside the colon (De Witte 1993, Ishii 1990).

The laxative effect of Aloe is tested on rats. Nine hours after aloe administration, diarrhea was provoked by different doses, like 5g/kg (20%) and 20g/kg (100%) (Barnes 2002, PDR for Herbal medicines 2000, WHO 1999, Izzo 1999). Researchers point out that the influence of a large number of anthraquinones and anthrones (23 different constituents) over the Cl^- -channels activation in Ehrlich tumor cells leads to inhibition of the activity, especially aloe - anthrone or emodin - anthrone (Hoenig 1992). In many cases, these anthraquinones reduce the chloride cell permeability even more than the Cl^- -channel block 130B does it. On the other hand, both components do not show inhibiting action over Na^+/K^+ - ATPase (Hoenig 1992). Rhein, frangula - emodin and other anthraquinones with additional phenol function show inhibition also (Hoenig 1992).

Research implied that laxative effect of aloin and 1.8 - anthraquinones can relatively depend on prostaglandin synthesis inside the intestines (Capasso 1983). Possibly, Nitric oxide (NO) may be the mediator of the laxative effect over the anthraquinone products (Izzo 1999).

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

The effect of the rein and aloe - emodin is compared with the effect of the ricinoleic acid and calcium ionophore A23187 over the release of thrombocyte - activating factor (PAF) from human gastrointestinal tissue in vitro. It is confirmed that rhein does not have any activity and that the activity of aloe - emodin is determined by the activity of the other components (Tavares 1996). Later, it was confirmed that the inhibition of Na⁺/K⁺ - ATPase and the release of NO are relevant in stimulating secretion of electrolytes and relaxation of smooth intestinal mucosa and that NO plays significant pathophysiological newly - confirmed role in relation to PAF, which provokes contractions of the smooth muscles (Longo 2002). Although the above mentioned data comes from experimental animal studies, no general conclusion can be made that biological factors PAF and NO have their role in stimulating the colon (Assessment report of HMPC for Aloe 2007).

Anti - neoplastic (cytotoxic) activity

The anthraquinone aglycones (aloe-emodin from Aloe) exert certain antitumor activity in vitro. Aloe-emodin suppresses tyrosine kinase activity of HER-2/neu-encoded p185-neu receptor resulting in antiproliferative activity (Barnes 2002, Leung 1980, Zhang 1996).

Antitumor effects of five Aloe components (aloin A and B, aloesin, aloeresin, aloe - emodin) were studied on human K562 leukemia cells and on the multidrug resistant b(MDR) variant cell line, k562/R. Only the aglycone aloe - emodin produced reproducible antitumor effects. Aloe - emodin caused mainly cytostasis and accumulation of the cells in S and G2 - M phases of the cell cycle during the first 48h of treatment. In another study, where chemopreventive role of aloe - emodin in human promyelocytic leukemia HL - 60 cells in vitro was evaluated, it was concluded that aloe - emodin appeared to exert its antineoplastic properties by inhibiting proliferation and inducing cell cycle arrest, and apoptosis underwent activation of caspase-3 in human leukemia HL-60 cells (Chen 2004).

Cytotoxic activity of mistletoe extract and isolated mistletoe glycoproteins has been investigated in numerous in vitro and in vivo studies. Significant antitumor activity has been observed in vivo against murine Lewis lung carcinoma, colon adenocarcinoma 38 and C3H mammary adenocarcinoma 16/C (Khwaja 1986). Sensitivity to mistletoe extract has been documented for acute lymphoblastic leukemia (Newall 1986).

Cytotoxic activity in vitro and antitumor activity in vivo (against mouse Ehrlich carcinoma) have been documented for marigold extracts. The most active fractions in vivo were saponine - rich fraction (Boucard - Maitrey 1988).

Numerous in vitro studies have demonstrated that hypericin is a potent inhibitor of protein kinase C (Agostinis 1995, De Witte 1993, Zhang 1996). Hypericin treatment of glioma cell lines inhibited growth and induced cell death due to protein kinase C. Receptor tyrosine kinase activity of epidermal growth factor is also inhibited by hypericin and may be linked to its antiviral and antineoplastic effects (De Witte 1993, Panossian 1996). In vitro cytotoxicity against human colon carcinoma cells (CO 115) has been described for hyperforin - related constituents, isolated from *Hypericum calycinum* and *H. revolutum* (Newall 1996).

Immunostimulative and immunomodulatory effects

Extracts of mistletoe herb and isolated polysaccharides of the plant demonstrate non - specific immunostimulative activity, which depends on the frequency and the quantity of the applied extract (Newall 1996). Nowadays, there is no more doubt about the effect of mistletoe extracts on immune reactions in vivo and in vitro and it is clear that different antigens presented in these extracts can modulate various cell types of the innate and adaptive immune system (Klein 2007). Mistletoe lectin (ML-1), viscotoxins, oligo- and polysaccharides exert immunostimulative

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

properties. It is a fact that despite all efforts to analyze the effect of mistletoe extracts on immunocompetent cells during the last 20 - 30 years it is, however, still unknown whether the observed alterations of immunological reactions during mistletoe therapy have any effect indeed on the tumor defense and survival of tumor patients (Klein 2007).

Extracts of propolis and isolated components of propolis demonstrate immunostimulative activity. Ethanol extract of propolis induces antibody production by mice spleen cells. Propolis modulated both in vivo and in vitro C1q production by macrophages as well as the action of complement system receptors on these cells (Dimov 1991, 1992). Some components of propolis (i.e. cinnamic acid) act on host defense, stimulating lymphocyte proliferation and inducing IL-1 and IL-2 production (Ivanovska 1995), and tend to inhibit H₂O₂ release by peritoneal macrophages or induce increased metabolic production. Other study confirmed increased lytic capacity of NK cells (when activated with propolis extract) against tumor cells (Sforcin 1996). Some authors suggest that immunostimulative activity of propolis may be due to macrophage activation and enhancement of macrophage phagocytic capacity (Scheller 1989, Tatefuji 1996).

Extract of marigold (contains specific high molecular weight polysaccharides) possess immunostimulative activity in granulocyte and carbon clearance tests (Wagner 1985).

Agglutinating activity

Lectin fraction of mistletoe herb shows agglutinating activity. Lectins are glycoproteins capable for binding to large number of cells including erythrocytes, lymphocytes, leucocytes, macrophages, glycoproteins and plasma proteins (Newall 1996).

Hypotensive effects

The exact nature of the hypotensive effect of mistletoe herb remains unclear, although it has been reported that the activity is mainly due to inhibitory effect on the vasomotor centre in medulla oblongata (Petkov 1979, Newall 1986). In addition, it has been stated that hypotensive action of mistletoe is mainly a reflexive feature, exerting a normalizing effect on both hypertensive and hypotensive states (Petkov 1979). The effect of different mistletoe plant parts and host - plant on the hypotensive activity has been studied with highest activity reported for mistletoe leaves parasitizing on willow (Petkov 1979).

Sesquiterpene lactones from elecampane root possess hypotensive effect (Newall 1996).

Sesquiterpene lactones from yarrow possess hypotensive activity (PDR for herbal Medicines 2000, Barnes 2002).

Effects on central nervous system

Water extract of elecampane root shows sedative effect on CNS (Newall 1996).

St. John's wort extracts (hypericin, xanthenes and flavonoids) possess antidepressive, antineurotic and anxiolytic activity (Barnes 2002, Evans 2002, Petkov 1982, Newall 1996). The European commission categorizes St. John's wort as MAO - inhibiting plant. Results obtained from in vivo studies revealed that MAO - inhibiting action has a very low intensity and it is due to hyperforin, flavonoids (aglycones and quercitrin), less to hypericin. All of the mentioned components act as anxiolytics by inhibition of type A MAO. Extracts of St. John's wort, due to their CNS activity, are used for treating insomnia, epilepsy, middle neurotic states, etc. (WHO 1999, Barnes 2002, PDR for Herbal Medicines 2000, Bruneton 1999, Dewick 1997, Newall 1996).

A commercial extract of St. John's wort has exhibited psychotropic and antidepressant activities in mice (Okpanyi 1987). It has been suggested that biflavonoids may be sedative principal in the

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

extract (Berghofer 1987, 1989), while for hypericin is stated that in small quantities it has tonic and tranquillizing action in human (Newall 1996).

Antibacterial and antiviral activity

Anthraquinone derivatives from Aloe possess antibacterial activity, which is confirmed in vitro on *Mycobacterium tuberculosis* and *Bacillus subtilis* (MICs 0.125 mg/ml and 0.25 mg/ml respectively). Aloe - emodin is considered a particularly active component that in certain doses inhibited growth of *Helicobacter pylori* and four different species of *Staphylococcus aureus*, which demonstrated resistance to methicillin (Barnes 2002, PDR for Herbal Medicines 2000, Heinrich 2004). Aloe - emodin presents direct antiviral activity on Herpes simplex virus type 1 and 2, Varicella zoster virus, Pseudorabies virus and Influenza virus (Barnes 2002, PDR for Herbal Medicines 2000).

Propolis acts as a strong bacteriostatic, bactericide and fungicide agent. With respect to its antimicrobial activity, propolis inhibits bacterial growth with a major effect on Gram - positive and limited action on Gram - negative bacteria (Grange 1990). Ethanol extracts of propolis show antimicrobial activity on *Bacillus aureus*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Candida albicans* (Kujumgiev 1999, Velikova 2000, Bankova 2005). The effects are probably due to the presence of flavonoids, phenolic acids and their esters (i.e. methyl ester of caffeic acid) (Gorunovic 2001, Evans 2002, Capasso 2003, Bankova 1999, 2000). Ethanol extracts of propolis show antimicrobial activity on methicillin - resistant *Staphylococcus aureus* as well as on vankomycin - resistant *Enterococcus faecium* (Kilic 2005).

Extracts of elecampane root and isolated sesquiterpene lactones (alantolaktone, isoalantolaktone and other lactones) possess antimicrobial and antifungal effects (Newall 1996).

Extracts of marigold (flavonoids and essential oil) possess antibacterial and fungicidal activity (Barnes 2002).

Ethanol extracts of yarrow (sesquiterpene lactones) demonstrate middle antimicrobial activity on *Staphylococcus aureus*, *Bacillus subtilis*, *Mycobacterium smegmatis*, *Escherichia coli*, *Shigella sonnei* and *Shigella flexneri* (Moskalenko 1986, Chandler 1982).

Phenolic components of St. John's wort possess antimicrobial activity (Barnes 2002, Evans 2002, Newall 1996). In the older literature, some authors cited that St. John's wort isolates such as novomanin, water - soluble imanin and imanin were strong antimicrobial agents (Newall 1996). It is also published that novomanin is potent topical antimicrobial agent against *Staphylococcus aureus*, with more potent activity than sulphanilamides (Newall 1996). Extract of St. John's wort are active on staphylococci, shigellae, *Escherichia coli* (Sakar 1986, Koesnikova 1986). The fractions containing flavonoids and catechins possess antiviral activity on Influenza virus in range 83 - 100 % (Mishenkova 1975). Antiviral activity is published for hypericin against HIV and hepatitis (Newall 1996).

Anti - inflammatory effect

Water extract of Aloe inhibits histamine release from peritoneal mast cells in animal models (rats) where histamine release was induced by antigen stimulation (Yamamoto 1993, Barnes 2002, PDR for Herbal Medicines 2000).

Extracts and isolated components of elecampane root possess antiflogistic effects (Evans 2002, Newall 1996).

Some components of propolis like caffeic acid methyl ester and flavonoids (galangin) are pointed out to be responsible for the extensive anti - inflammatory effects, via inhibition of COX II enzymes important for prostaglandin production, NF - kB inhibition and stimulation of

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

leukocytosis (Capasso 2003). Anti - inflammatory and analgesic effects of a standard ethanol extract of propolis were tested on mice (Paulino 2003) - the extract inhibited abdominal contortions with an ID50 = 7.4 mg/kg. In the formalin test the extract caused a significant reduction of pain in mice treated with 100 mg/kg of extract during the neurogenic phase and for the inflammatory phase with all doses of the extract, with an ID50 = 2.5 mg/kg. Ethanol extract of propolis inhibited also the capsaicin - induced ear edema in mice. The analgesic effect of the extract was associated with the inhibition of inflammatory responses and not to a simple irritation of nervous terminals (Paulino 2003). In other study, inhibition of dyhydropholate - reductase was found, due to the activity of caffeic acid (Strehl 1994).

Anti - inflammatory activity has been documented for aqueous extract of yarrow using mouse and rat paw edema models, with inflammation induced by yeast and various inflammatory substances including histamine, caragenan and prostaglandin (Barnes 2002, Newall 1996). In general, anti - inflammatory properties are associated with components of the essential oil of yarrow, particularly for azulenes (Evans 2002, Newall 1996, Miller 1998).

Alcohol extract of marigold acts as an anti-inflammatory agent (Mills and Bone, 2000), probably due to triterpenoid esters (Barnes 2002, Newall 1996). In experimental models on rat marigold extracts were effective in dextran and burn edemas and in acute lymphedema. Activity against lymphedema was primarily attributed to an enhancement of macrophage proteolytic activity (Casley - Smith 1983).

St. John's wort extract prepared with vegetable oil has a durable reputation as an anti-inflammatory and wound-healing agent (Samuelsson 1999, Bruneton 1999, WHO 1999, Bombardeli 1995). The anti - inflammatory effect is probably due to the activity of hypericin which demonstrates protein kinase C inhibition as well as inhibition of releasing of arachidonic acid and leucotriene B4 (Panossian 1996). St. John's wort extract was found to suppress inflammation in mice induced by caragennan and PGE1 (Schipochliev 1981). Anti - inflammatory and antiulcerogenic properties have been documented for amentoflavone, a biapigenine derivative (Berghofer 1989).

INTERACTIONS

Herb-Drug Interactions in simultaneous use of Aloe

- Oral Glyburide: Aloe may increase hypoglycemic effects.
- Topical Hydrocortisone: Aloe may increase anti-inflammatory effects.
- Sevoflurane: Aloe may have additive antiplatelet effect causing excessive bleeding during surgery.
- Long-term use of Aloe with products containing cardiac glycosides or medicines for cardiac disorders (arrhythmia) can lead to excessive loss of potassium and can accentuate the effect of cardiac glycosides and arrhythmia medicines.
- Thiazide diuretics and corticosteroids: Aloe increases the possibility of potassium loss (Newall 1996).

Herb-Drug Interactions in simultaneous use of Viscum Album (Newall 1996, Barnes J, et al. 2001):

- Uterotonics – additive effect, because of the tonic stimulation of tyramine on the uterus
- Antihypertensives – additive hypotensive effect
- Digoxin – antagonistic effect due to negative inotropic effect

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

- Antiarrhythmics - antagonistic effect due to negative inotropic effect
- Antidepressants - additive effect
- Anticoagulants - additive effect

Simultaneous use of products containing elecampane extract with hypoglycemic and antihypertensive medicines may cause interactions (Newall 1996).

Overdose of yarrow may interfere with anticoagulant and antihypertensive therapy (Newall 1996).

No data is published on interactions of marigold, cynodon or cornel - tree.

Herb-Drug Interactions in simultaneous use of St. John's wort extract (Andren L, et al. 2007; Barnes J, et al. 2001; Brinker F. 2001; Eggertsen R, et al. 2007; Frye RF, et al. 2004; Gurley BJ, et al. 2008; Jiang X, et al. 2004; Markowitz JS, et al. 2003; Mathijssen RH, et al. 2002; Schwarz UI, et al. 2007; Smith P, et al. 2004; Xu H, et al. 2008)

Cytochrome P450 3A4: St. John's wort has been shown to induce cytochrome isoenzyme 3A4, therefore affecting metabolism of certain medications and reducing serum concentrations. CYP 3A4 levels return to normal 1 week after discontinuing St. John's wort 21. Drugs metabolized by 3A4 include:

- **Theophylline:** Blood levels of theophylline may be significantly reduced, resulting in decreased efficacy.
- **HIV protease inhibitors:** Blood levels of indinavir, nelfinavir, ritonavir, and saquinavir can be significantly reduced, resulting in increased HIV viral load and development of viral resistance.
- **HIV non-nucleoside reverse transcriptase inhibitors:** Blood levels of efavirenz and nevirapine can be significantly reduced, resulting in increased HIV viral load.
- **Cyclosporin / Tacrolimus:** Blood levels of cyclosporin or tacrolimus can be significantly reduced, resulting in decreased efficacy.
- **Diltiazem / Nifedipine:** Blood levels of diltiazem or nifedipine can be reduced, resulting in decreased efficacy.
- **Irinotecan:** Due to changes in hepatic metabolism caused by St. John's wort, levels of irinotecan metabolite SN-38 may be lowered by as much as 40% for up to 3 weeks following discontinuation of St. John's wort.
- **Warfarin:** May increase or decrease activity when administered concomitantly. Internal normalization ratio (INR) should be monitored routinely. S-isomer may have increased metabolism due to CYP 3A4 induction. S-isomer may have decreased metabolism due to CYP 1A2 inhibition.
- **Digoxin:** Prolonged concurrent administration may result in decreased absorption of digoxin with lowered plasma concentrations.
- **Triptans:** Increased serotonergic effect and possible serotonin syndrome when combined with sumatriptan, naratriptan, rizatriptan, or zolmitriptan.
- **SSRIs:** Increased serotonergic effect and possible serotonin syndrome when combined with citalopram, fluoxetine, fluvoxamine, paroxetine, or sertraline.
- **Tricyclic Antidepressants:** Increased serotonergic effect and possible serotonin syndrome when combined with nefazodone, amitriptyline, or imipramine. Possible reduction in efficacy of antidepressants due to changes in metabolism.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

- **Oral Contraceptives:** May reduce blood levels resulting in decreased efficacy (i.e., breakthrough bleeding or pregnancy).
- **Alcohol:** May result in increased sedation.
- **Anesthetics:** Case report of cardiovascular collapse (hypotension without anaphylactic symptoms) shortly after induction of general anesthesia with fentanyl, propofol, d-tubocurarine, and succinylcholine followed by nitrous oxide, oxygen, and isoflurane.
- **Chemotherapy:** Due to changes in hepatic metabolism caused by St. John's wort, chemotherapy levels may be altered, resulting in increased toxicity or decreased efficacy. Caution should be exercised when administering concomitantly with chemotherapy (i.e., cyclophosphamide, paclitaxel, etoposide, irinotecan).
- **Tamoxifen:** Due to changes in hepatic metabolism caused by St. John's wort, levels of tamoxifen may be lowered, resulting in reduced efficacy.
- **Alprazolam:** May reduce blood levels, resulting in decreased efficacy.
- **Dextromethorphan:** May reduce blood levels, resulting in decreased efficacy.
- **Sympathomimetics:** Concomitant administration may produce increased serotonergic activity and possible serotonin syndrome.
- **Imatinib:** Increase clearance.
- **Simvastatin:** Increase clearance, resulting in increased LDL cholesterol.
- **Atorvastatin:** Increase clearance, resulting in increased LDL cholesterol.

Cytochrome P450 2C9: St. John's wort has been shown to induce cytochrome isoenzyme 2C9, therefore affecting metabolism of certain medications and reducing serum concentrations. Drugs metabolized by 2C9 include:

- **Gliclazide:** Increase clearance.

PHARMACOKINETICS / BIOAVAILABILITY

No pharmacokinetic data is available for water extracts of herbal drugs: aloe, mistletoe herb, propolis, elecampane root, marigold, yarrow, cornel-tree bark, Cynodonii rhisoma, and St. John's wort. Certain number of data could be found considering pharmacokinetics of some active constituents of above mentioned drugs, obtained in animal studies for aloins from aloe, lectins and voscotoxins from mistletoe and hypericin, hyperforin and quercetin from St. John's wort, etc. No pharmacokinetic data is available for the active constituents from elecampane, marigold, cynodon or cornel - tree. The following data may be considered as corresponding to this report:

Aloins A and B, hydroxyaloins and aloinosides A and B are not absorbed in the upper gut. In humans, they pass into the colon unmodified after oral ingestion. Human intestinal flora are able to break down O - glycosides easily but only to some extent C - glycosides of most anthranoides. Experimental studies on rats resulted positively only in gnotobiotic rats mono - associated with the Eubacterium sp. BAR in which administration of barbaloin causes severe diarrhea. Only in those rats, barbaloin was transformed to aloe - emodin athrone, which is responsible for the laxative effect. In contrast, another study showed that aloe - emodin - 9 - anthrone was produced in the rat large intestine, as main active metabolite, which acts specifically on the colon (Assessment report on Aloe barbadensis and aloe HMPC, 2007). After oral administration of 4.5 mg/kg 14C - aloe - emodin to rats, 20 - 30% of the dose was excreted in urine and the rest in faeces. Aloe - emodin was quickly metabolized to rhein, to an unknown metabolite and to conjugates of all three. In the plasma 10% of 14C - activity was identified as free aloe - emodin. Maximum plasma values were

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

reached 1.5 - 3 h p.a. with 248 (male) and 441 (female) ng equivalents aloe - emodin/ml. Maximum concentrations in plasma were about 3 times higher than those in ovaries and 10 times higher than those in testes. Only liver, kidney and intestinal tract showed higher concentrations than plasma. Terminal half - life (for radioactivity) in blood was about 50 h (Assessment report on Aloe barbadensis and aloe HMPC, 2007). The ESCOP monograph mentioned research report of a human pharmacokinetic study, in which after oral administration of aloes (equivalent to 16.4 mg of hydroxyanthracene derivatives) for 7 days, aloe - emodin was detected as a metabolite in the plasma only sporadically and with maximum concentrations of less than 2 ng/ml. In the same study rhein was detected in the plasma in concentrations ranging from 6 - 28 ng/ml after single dose administration. There was no evidence of accumulation of rhein (Assessment report on Aloe barbadensis and aloe HMPC, 2007).

The pharmacokinetics of mistletoe lectins from whole plant mistletoe extract is still unknown. Studies with intravenous application of a recombinant type II ribosome inactivating protein analogous to mistletoe lectin revealed a short half - life of about 13 min in cancer patients (Huber 2007).

Limited summary information is available on the pharmacokinetics of hypericin and pseudohypericin in mice and humans. After oral administration of ¹⁴C - labeled hypericin and pseudohypericin to mice, these substances are absorbed to 80% and 60%, respectively. In humans, after oral administration of hydromethanolic extract of *Hypericum perforatum*, containing 0.1% total hypericin (300 to 1800 mg per person), plasma half - life of approximately 6 h was observed for hypericin. After administration of an extract containing 0.3% total hypericin, plasma half - life was approximately 25 h for hypericin and 16 to 36 h for pseudohypericin (Summary report on *Hypericum perforatum*, EMEA/MRL 1999).

WARNINGS, SIDE EFFECTS, ADVERSE REACTIONS, CONTRAINDICATIONS

Aloe vera gel should not be confused with aloe juice or aloe latex, both of which contain anthraquinone, a cathartic laxative. Aloe vera taken for internal use should be discouraged due to possible adverse effects and inconclusive clinical data. Aloe vera injections for cancer patients have resulted in several deaths. The FDA rules that aloe is not safe as a stimulant laxative.

Topical administration of aloe vera gel is considered safe but oral consumption of aloe can cause gastrointestinal upset and electrolyte abnormalities. Inappropriate use of aloe vera supplements has been linked to thyroid dysfunction, acute hepatitis and perioperative bleeding. Parenteral administration of aloe should be avoided due to potential toxicities and lack of clinical efficacy in humans.

Side effects of Aloe are well documented in literature: abdominal spasms and pain may occur after even one single dose of Aloe. Overdose can lead to abdominal spasms and pain, as well as formation of thin, watery stool. Prolonged use of anthraquinone laxative (aloe) may lead to electrolyte disturbances (hypokalaemia, hypocalcaemia) in more serious cases metabolic acidosis, malabsorption, weight loss, albuminuria and haematuria. Weakness and orthostatic hypotension may be exacerbated in elderly patients when stimulant laxatives are repeatedly used. Melanotic pigmentation of the colonic mucosa has been observed in individuals taking anthraquinone laxatives for extended periods. The pigmentation is clinically harmless and usually reversible within 4 to 12 months after the drug is discontinued (Muller - Lissner 1993). Chronic abuse of anthraquinone laxatives may lead to hepatitis (Beuers 1991). Conflicting data exist on other toxic effects such as intestinal - neuronal damage after long - term use (Muller - Lissner 1993). Products

containing Aloe should not be used by patients that suffer intestinal obstruction or stenosis, dehydration or chronic constipation (WHO 1990), inflammatory intestinal diseases such as appendicitis, Crohn disease, ulcerative colitis, irritable bowel syndrome, etc. (Wichtl 1994). Aloe is not recommended for children under the age of 10. Women are not supposed to take Aloe during pregnancy and lactation unless it is recommended and supervised by a doctor (Wichtl 1994). Patient with cramps, colic, hemorrhoids, nephritis, or any undiagnosed abdominal symptoms such as pain, nausea or vomiting (WHO 1999) should not intake aloe. Prolonged use of products containing Aloe can lead to hypersensitive reactions manifested in dermatitis and eczematous formations, loss of electrolytes (potassium) that may result in hyperaldosteronism, inhibition of intestinal motility, gastrointestinal disorders, cardiac disorders and rarely nephropathy and albuminuria and haematuria (WHO 1999, Gooding 1976).

Viscum album – used raw, without previous extraction of the active substances, the whole leaf of the plant are highly toxic – 3 of the leaf are enough to induce coma.

Adverse Reactions of mistletoe (Schulz V, et al. 2001, Gorter RW, et al. 1998, Foster S, et al. 1999, Newall CA, et al. 1996).

Common: Headache, nausea, abdominal discomfort, constipation, dizziness, confusion, fatigue, dry mouth, sleep disturbances, and sedation.

Infrequent: Photosensitivity or photodermatitis, elevated liver function tests, acute neuropathy, increased prothrombin time (PT).

Case reports: Mania in 3 patients with underlying bipolar disorder. Resolved promptly in 2 patients following discontinuation while the third experienced persistent agitation for several months.

Serotonin syndrome: Hypertension, diaphoresis, agitation, dizziness, and weakness with acute onset following 10 days of St. John's wort. Syndrome resolved following supportive care and discontinuation of St. John's wort.

Erythroderma affecting both light-exposed and non light-exposed areas of skin. Developed 4 days after initiation of St. John's wort and resolved after 5 weeks with concomitant oral steroids.

Sexual dysfunction: Decreased sexual libido that returned following discontinuation of St. John's wort.

Withdrawal syndrome: Nausea, anorexia, dry retching, dizziness, dry mouth, thirst, cold chills, and extreme fatigue in patient within 24 hours of stopping treatment of St. John's wort after 32 days of treatment.

Products that contain extracts or isolated components of mistletoe can cause side effects such as headache, high fever, leucocytosis, diarrhea, vomitus, orthostatic circulatory disorders, bradycardia, hypotension, myosis, epileptic seizures and allergic reactions (German Commission E Monograph no. 228, Foster S, et al. 1999, Newall CA, et al. 1996, Schulz V, et al. 2001, Gorter RW, et al. 1998, Kleijnen J, & Knipschild P. 1994, Mengs U, et al. 2002). In animal studies, some chemical components of mistletoe (tyramine and cardioactive components) stimulate the uterus (Newall 1996). Due to lack of relevant data, taking into consideration the toxicity of the drug, the use of it during pregnancy and lactation is highly restricted (Newall 1996). Products containing extracts from mistletoe are not to be used together with cardiostimulant, immunosuppressive, anti-hypertensive, antidepressant and anticoagulant therapy (Hulsen 1987). The use of mistletoe is contraindicated in cases of protein hypersensitivity and chronic progressive infections such as tuberculosis (German Commission E Monograph no. 228).

Propolis is considered relatively safe for use. However, some data present manifested allergic eczematous contact dermatitis in case of allergic predisposition on bee sting (Capasso 2003). There are some cases when acute oral mycosis was caused by the use of propolis - containing lozenges (Capasso 2003).

Products containing extract from the **root of elecampane** can cause allergic reactions (Newall 1986). The components of essential oil, alantolactone and isoalantolactone can lead to sensitivity

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

(Stampf 1982, Newall 1986). Due to lack of toxicological data, the root of elecampane should not be used during pregnancy and lactation (Newall 1996).

There is no data published indicating side effects from the use of **Cynodoni rhizoma** (Petkov 1982).

There is no data published indicating side effects from the use of **cornel - tree** (Petkov 1982).

There is no data published indicating side effects from the use of **marigold** (Newall 1996). Marigold is traditionally known as drug affecting the menstrual cycle. In vitro studies have shown uterotonic effects of the drug and triterpeoid constituents are reported to be effective as spermatocides (Newall 1996). Due to lack of toxicological data, the use of marigold during pregnancy and lactation is not recommended (Newall 1996).

Yarrow may cause allergic reactions in individuals hypersensitive to other Asteraceae plants (Newall 1996, Mathias 1979). Individuals sensitive to these plants should not take them in any form (Tyler 1993). Traditionally, yarrow is considered as abortive and drug that affects the menstrual cycle. Due to lack of toxicological data, the use of yarrow during pregnancy and lactation is not recommended (Newall 1996).

St John's wort, according to the German Commission E Monograph, is highlighted as possible arouser of photosensitivity that occurs very rarely in people with fair complexion (Wichtl 1994, The Complete German Commission E Monographs 1998). This photosensitivity is demonstrated also in clinical studies in individuals that have been taking hypericin and have been exposed to ultraviolet A and B radiation (WHO 2002). A patient, who was treated with 600 mg of a hydroalcoholic extract of the herb (containing 0.24 - 0.32 % total hypericin) three times a day for 15 days, showed a significant increase in erythema due to the ultraviolet A irradiation. The plasma concentration of hypericin in this subjects was double that seen during normal therapeutic treatment of depression (WHO 2002). Monitoring studies have been done for products based on St John's wort that show that the drug produces rare and mild types of side effects such as gastrointestinal irritation, allergic reactions, fatigue etc. (Linde 1996, WHO 2002, Capasso 2003). Prolonged insolation or exposition to UV - light should be avoided during the use of products containing extracts from St John's wort because it may lead to photosensitive reaction (Wichtl 1994). The use of St John's wort is contraindicated in cases when there is an allergy to the plants from Hypericaceae (WHO 2002).

TOXICITY

Data on the carcinogenicity of Aloe are not available. While chronic abuse of anthranoid - containing laxatives was hypothesized to play role in colorectal cancer, no causal relationship between anthranoid laxative abuse and colorectal cancer has been demonstrated (Siegers 1992, WHO 1999). In animal studies on mice, administration of Aloe 50 mg/kg in a period of 12 weeks did not cause serious pathological disturbances; although it increased the concentration of sorbitol dehydrogenase, which may be connected with eventual hepatic damage (Barnes 2002, PDR for Herbal Medicines 2000).

Intravenous administration of viscotoxin (isolated from mistletoe) to cats (35 µg/kg) resulted in a negative inotropic effect on cardiac muscle, reflex bradycardia, and hypotension. Viscotoxin A3 and B have caused muscle contracture and progressive depolarization in isolated smooth, skeletal or cardiac muscle preparation. Viscotoxin is toxic on parenteral administration and an LD50 value (mice, intraperitoneal injection) has been estimated as 0.7 mg/kg. Mistletoe lectins inhibit protein synthesis in cells and cell - free systems. In common with other known toxic lectins (e.g. ricin), mistletoe lectins bind to plasma proteins, are specific towards D - galactose, possess some cytotoxic

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

activity and have caused macroscopic lesions in rats. An LD50 (mice) value for mistletoe lectin fraction is reported as 80 µg/kg. In animal studies, the stimulative effect on the uterus by tyramin was observed, isolated from mistletoe extract. Because of possible stimulation of the uterus, mistletoe is not recommended for use during pregnancy and lactation (Barnes 2002, PDR for Herbal Medicines 2000, Newall 1996).

Symptoms of toxicity documented following the ingestion of mistletoe include hypotension, coma, seizures, myosis, mydriasis and death (Barnes 2002, PDR for Herbal Medicines 2000, Newall 1996). One case of hepatitis is reported for woman who had ingested a mixed herbal preparation containing mistletoe (Barnes 2002).

Yarrow is considered non - toxic (Newall 1996). In mice LD50 values have been reported of up to 3.65g/kg (by mouth), 3.1g/kg (by intraperitoneal injection), and greater than or equal to 1 g/kg (by subcutaneous injection). In the rat an LD50 (subcutaneous injection) has been recorded as 16.86 g/kg, with corresponding LD0 and LD100 values reported as 12 and 20 g.kg, respectively (Newall 1996). For comparison, an ED25 for anti - inflammatory activity has been estimated as about 0.43 g/kg (Shipochliev 1984). Terpenoide - rich essential oils are defined as irritant agents and the same may be implied for yarrow essential oil (Newall 1996). A toxic constituent of the oil (thujone) is presented in yarrow essential oil in concentration that could not be harmful for patients (Newall 1996).

Mutagenity of hydroalcoholic extracts of St. John's wort containing 0.2 - 0.3 % hypericin and 0.35 % quercetin has been studied in various in vitro and in vivo systems. Although some positive results were observed in vitro, all the in vivo tests were negative, indicating that hydroalcoholic extract was not mutagenic in animals (Schimmer 1988, 1994). In a 26 - week study, intragastric administration of a hydroalcoholic extract to rats and dogs (900 and 2700 mg/kg body weight) had no effect on fertility, development of the embryo, or prenatal or postnatal development (Leuschner 1996). According to EMEA report on St. John's wort, there is no data on acute toxicity, reproductive toxicity or teratogenicity as well as for carcinogenicity for St. John's wort (Summary report on Hypericum perforatum EMEA 1999).

CONTENTS OF PLACEBO SOLUTIONS

Placebo solutions 1 and 2 were made from:

- corn flour,
- ground trifle,
- rye flour,
- water extract of herba apsinti,
- talc,
- microcrystal cellulose,
- magnesium stearate,
- aerosol 200,
- sorbitol
- industrial food color for chocolate LOT 06 E 0019067.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

METHODS

Clinical examinations and laboratory analysis

The following table present the time when investigations were done, shadowed cells mark the check-up when the proper investigation was not done (by the protocol):

examination	Months of the investigation			
	I	IV	VIII	XII
Anamnaesthical data (enlisted in the test list)	X			
Colposcopy	X	X	X	X
Pap test*	X	X	X	X
HPV PCR typization of a cervical smear	X	X	X	X
Side effects		X	X	X

* In case of progression of the CIN findings, patient undergoes biopsy and endocervical curettage, and in case of histological verification, the patient is excluded from the trial and referred to surgical treatment.

Criteria for therapeutic efficacy evaluation

Colpocopic classification findings (Barcelona 2002, IFCPC)	grade
Normal (A findings) Transformation zone Original squamos epithelium ectopy	0
Suspiciuos (B findings) mild leukoplakia, mozaics, punctuation iodine negative zone	1
Abnormal (C findings) severe leukoplakia, mozaics, punctuation acetic-white epithelium atypical transformation zone atypical blood vessels	2
Pap smear (Bethesda cytological classification 2004)	grade
Benign – normal findings with or without accompanying infection	0
Low-grade SIL (HPV or CIN I/HPV)	1
ASC-H or ASC-US	2
High-grade SIL (CIN II, CIN III or CIS HPV)	3
Invasive cancer suspected findings	4
HPV PCR typization	grade
Negativization of the findings	0
Persistence of the findings of identical type of HPV	1
Occurrence of a new type HR HPV	2

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Side effects

The absence of the side effects is evaluated with a grade 0, while the presence of some of the side effects mentioned bellow is evaluated with a grade 1, noting that at the statistical processing collective analysis will be carried out for each category of the side effects.

category		Grade of the side effects	
A	Skin allergic reactions	none	Papulous dermatitis / eczema
		0	1
B	GIT	none	Stomach pain / colic / diarrhea / constipation / nausea / vomiting
		0	1
C	General condition	same	Fatigue /vertigo /sleepiness / orthostatic hypotension
		0	1
D	UGT	none	Hematuria
		0	1

If any side effect is detected, the following chart needs to be fulfilled, bringing out the category of the side effect (A, B, C, D):

category	A	B	C	D
Date of appearance				
Severity ¹				
Duration ²				
Association ³				
Course/outcome ⁴				

Instructions for fulfilling the above chart:

Severity ¹–it is signed with one of the following numerical grades:

1=weak

2=moderate

3=heavy

Duration ²- the number of the days of the duration of the side effects is numerically signed

Association ³ between the emerged difficulties and the medicine –it is marked by the following numerical grades:

0= there is no association

1= assumed

2= probably

Course/out come ⁴ - it is signed with the following numerical grades:

1= administration of the investigated agent is continued, the side effect diminishes

2= administration of the investigated agent is continued, the side effect persists

3= administration of the investigated agent is stopped due to the side effect

RESULTS

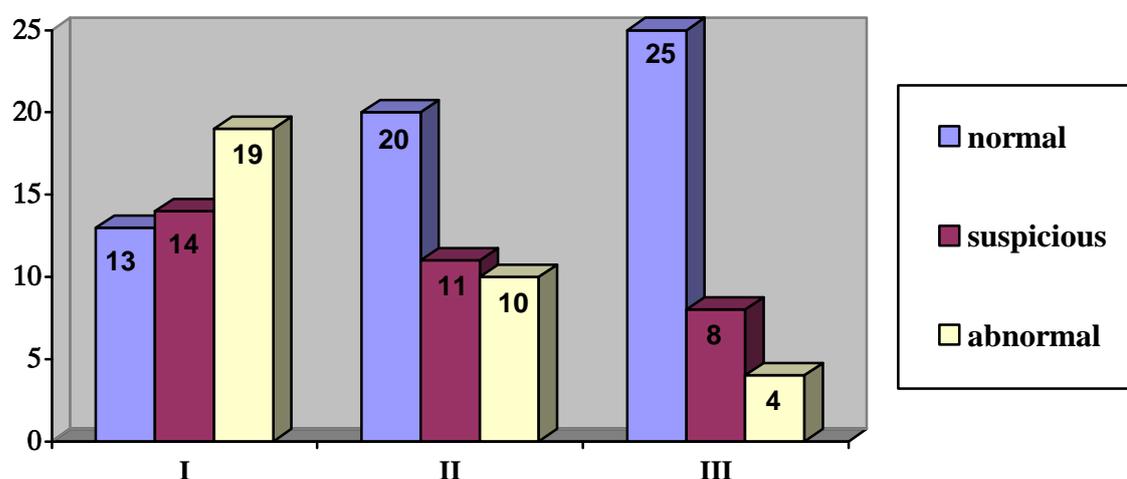
We examined results obtained from the objective variables who are direct indicator of the treatment feasibility (colposcopic examination, PAP, PCR typization) and from the anamnestic variables who can contribute for the evaluation of the treatment rationality (smoking, regularity of vitamin and antioxidants intake, age of first sexual intercourse, number of abortions, number of deliveries, regularity of using condom, marital status, number of active sexual partners, number of ex-sexual partners), as well as from anamnestic variables for who we doubted that have any kind of influence to the treatment (nationality, inhabitation, education).

1. COLPOSCOPIC FINDINGS

Table 1. Distribution of patients treated with the investigated agent according to colposcopic findings on first, second and third check up

colposcopic findings of the group treated with the investigated agent	I		II		III	
	n	%	n	%	n	%
normal findings (0)	13	28.26	20	48.78	25	67.57
suspicious findings (1)	14	30.44	11	26.83	8	21.62
abnormal findings (2)	19	41.30	10	24.39	4	10.81
total	46	100	41	100	37	100

Chart 1. Distribution of patients treated with the investigated agent according to colposcopic findings on first, second and third check up



Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Analysis with Friedman ANOVA test presented **statistical significant differences between colposcopic findings on the first, the second and the third check-up among patients treated with the investigated solutions.** (ANOVA Chi Sqr. = 27,444 df = 2 p = 0,000001)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between colposcopic findings on the first and the second check-up among patients treated with the investigated solutions.** (T = 0,00 Z = 2,9340 p = 0,00334)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between colposcopic findings on the first and the third check-up among patients treated with the investigated solutions.** Differences are more pronounced on the third check-up compared to the first. (T = 0,00 Z = 3,723 p = 0,000196)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between colposcopic findings on the second and the third check-up among patients treated with the investigated solutions.** The difference is statistically significant, but smaller compared to the both previously presented. (T = 0,00 Z = 2,366 p = 0,0179)

Table 21. Distribution of patients treated with placebo according to colposcopic findings on first, second and third check up

colposcopic findings of the group treated with placebo	I		II		III	
	n	%	n	%	n	%
normal findings (0)	12	28.57	14	35.89	20	60.6
suspicious findings (1)	13	30.95	12	30.77	9	27.3
abnormal findings (2)	17	40.48	13	33.33	4	12.1
total	42	100	39	100	33	100

Analysis with Friedman ANOVA test presented **statistical significant differences between colposcopic findings on the first, the second and the third check-up among patients treated with placebo.** (ANOVA Chi Sqr. = 19,500 df = 2 p = 0,00006)

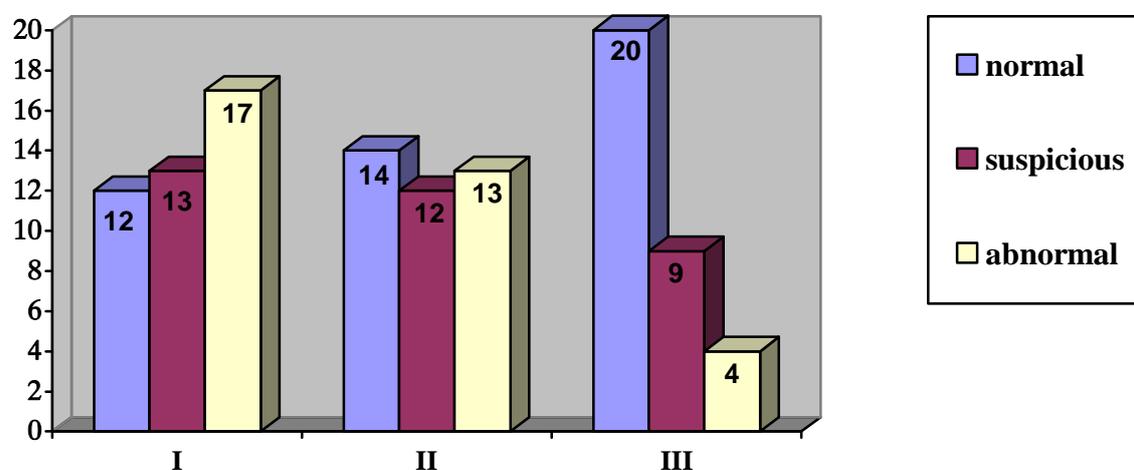
Analysis with Wilcoxon Matched Pairs Test **did not presented statistical significant differences between colposcopic findings on the first and the second check-up among patients treated with placebo.** (T = 0,00 Z = 1,603 p = 0,1088)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between colposcopic findings on the first and the third check-up among patients treated with the investigated solutions.** (T = 0,00 Z = 3,059 p = 0,00221).

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between colposcopic findings on the second and the third check-up among patients treated with placebo.** (T = 0,00 Z = 2,665 p = 0,0076).

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Chart 1. Distribution of patients treated with placebo according to colposcopic findings on first, second and third check up



2. PAP findings

Table 3. Distribution of patients treated with the investigated agent according to PAP findings on first, second and third check up

PAP findings of the group treated with investigated agent	I		II		III	
	n	%	n	%	n	%
normal findings (0)	20	43.5	22	53.66	28	75.7
Low-grade SIL (1)	19	41.3	14	34.15	7	18.9
ASC-H / ASC-US (2)	7	15.2	4	9.76	1	2.7
High grade SIL (3)	0	0	1	2.43	1	2.7
Cais suspicious (4)	0	0	0	0	0	0
total	46	100	41	100	37	100

Analysis with Friedman ANOVA test did not present **statistical significant differences between PAP findings on the first, the second and the third check-up among patients treated with the investigated agent.** (ANOVA Chi Sqr. = 5,096 df = 2 p = 0,0782)

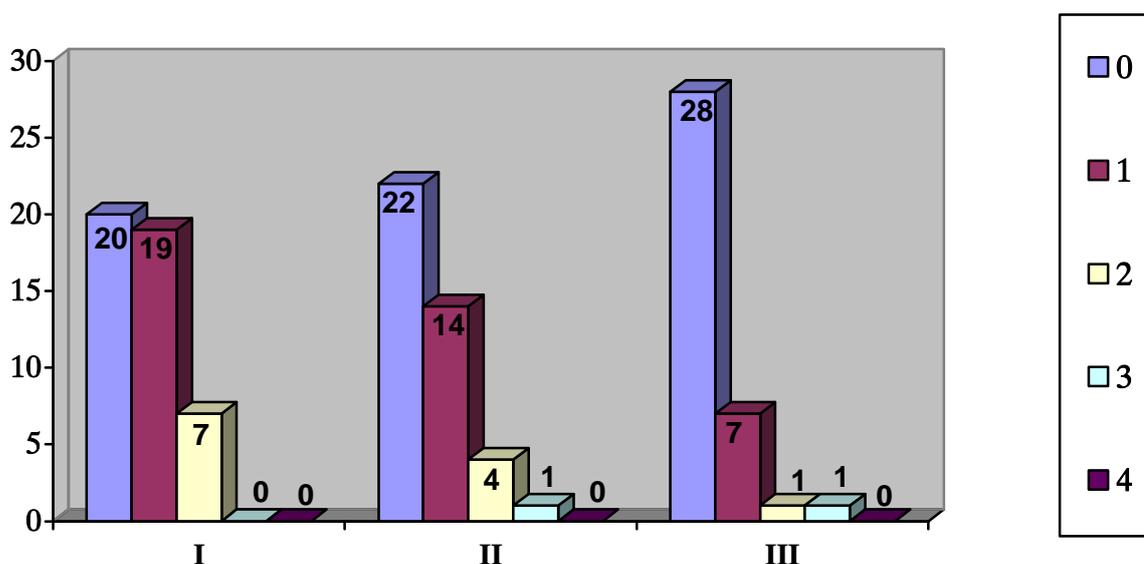
Analysis with Friedman Wilcoxon Matched Pairs Test did not present **statistical significant differences between PAP findings on the first and the second check-up among patients treated with the investigated agent.** (T = 7,00 Z = 0,733 p = 0,4630)

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Analysis with Friedman Wilcoxon Matched Pairs Test did not present **statistical significant differences between PAP findings on the first and the third check-up among patients treated with the investigated agent.** (T = 15,00 Z = 1,274 p = 0,2026)

Analysis with Friedman Wilcoxon Matched Pairs Test did not present **statistical significant differences between PAP findings on the second and third check-up among patients treated with the investigated agent.** (T = 9,00 Z = 1,260 p = 0,2075)

Chart 3. Distribution of patients treated with the investigated agent according to PAP findings on first, second and third check up



Analysis with Friedman ANOVA test presented **statistical significant differences between PAP findings on the first, the second and the third check-up among patients treated with placebo.** (ANOVA Chi Sqr. = 8,00 df = 2 p = 0,01832)

Analysis with Friedman Wilcoxon Matched Pairs Test did not present **statistical significant differences between PAP findings on the first and the second check-up among patients treated with placebo.** (T = 5,00 Z = 0,000 p = 1,00)

Analysis with Friedman Wilcoxon Matched Pairs Test did not present **statistical significant differences between PAP findings on the first and the third check-up among patients treated with placebo.** (T = 2,00 Z = 1,782 p = 0,0747)

Analysis with Friedman Wilcoxon Matched Pairs Test presented **statistical significant differences between PAP findings on the second and the third check-up among patients treated with placebo, thus presenting statistically significant deterioration among patients treated with placebo according to PAP findings between the second and the third check-up.**

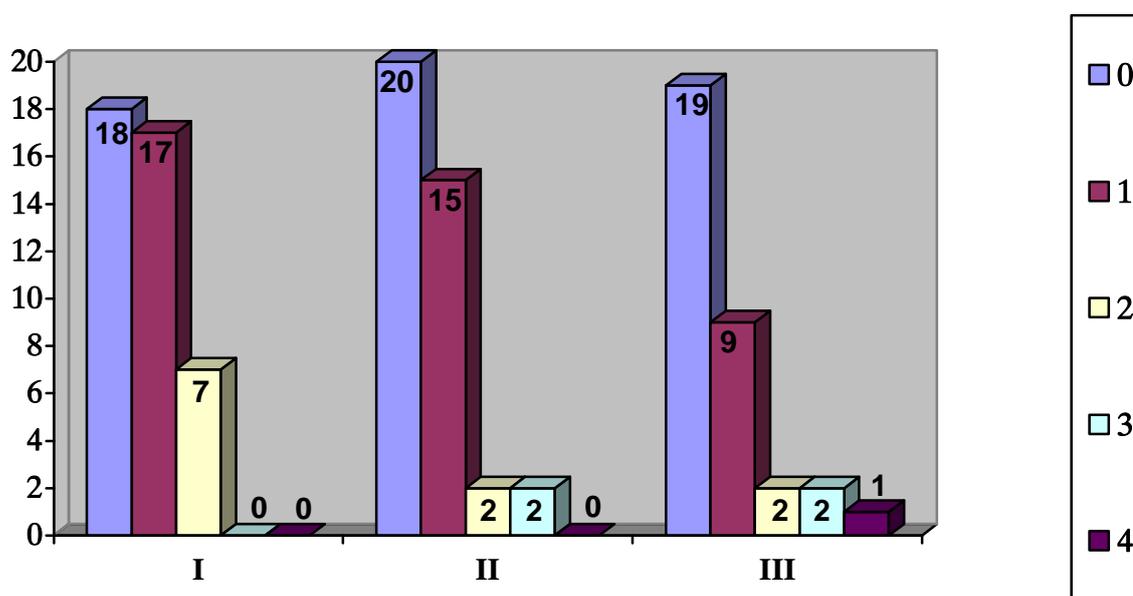
(T = 0,00 Z = 2,201 p = 0,0277)

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Table 4. Distribution of patients treated with placebo according to PAP findings on first, second and third check up

PAP findings of the group treated with placebo	I		II		III	
	n	%	n	%	n	%
normal findings (0)	18	42.86	20	51.28	19	57.68
Low-grade SIL (1)	17	40.48	15	38.46	9	27.28
ASC-H / ASC-US (2)	7	16.67	2	5.13	2	6.06
High grade SIL (3)	0	0	2	5.13	2	6.06
Cais suspicious (4)	0	0	0	0	1	3.03
total	42	100	39	100	33	100

Chart 4. Distribution of patients treated with placebo according to PAP findings on first, second and third check up



3. HPV PCR TYPISATION

Analysis with Friedman ANOVA test presented **statistical significant differences between HPV PCR typization on the first, the second and the third check-up among patients treated with the investigated agent.** (ANOVA Chi Sqr. = 51,058 df = 2 p = 0,000001)

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between HPV PCR typization on the first and the second check-up among patients treated with the investigated agent.** (T = 14,00 Z = 4,204 p = 0,000026)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between HPV PCR typization on the first and the third check-up among patients treated with the investigated agent.** The difference is more significant on the third check-up compared to the first.

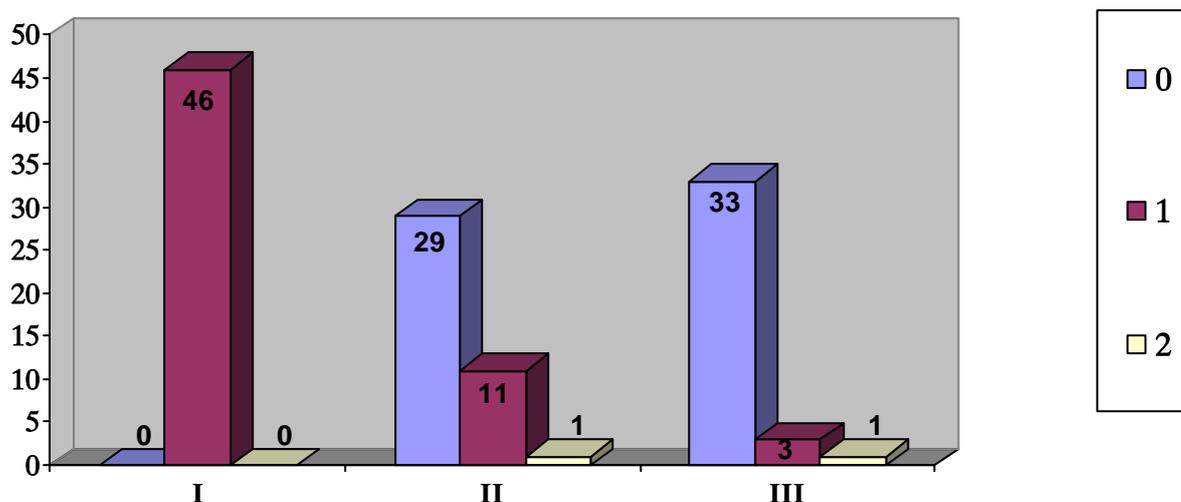
(T = 17,50 Z = 4,787 p = 0,000002)

Analysis with Wilcoxon Matched Pairs Test did not present **statistical significant differences between HPV PCR typization on the second and the third check-up among patients treated with the investigated agent.** (T = 4,50 Z = 1,890 p = 0,0587)

Table 5. Distributions of patients treated with the investigated agent according to HPV PCR typization on first, second and third check up

HPV PCR typization of the group treated with the investigated agent	I		II		III	
	n	%	n	%	n	%
Negative findings (0)	0	0	29	70.73	33	89.2
Persistence of the same HPV type (1)	46	100	11	26.83	3	8.1
Occurrence of a new high-risk HPV type (2)	0	0	1	2.44	1	2.7
total	46	100	41	100	37	100

Chart 5. Distributions of patients treated with the investigated agent according to HPV PCR typization on first, second and third check up

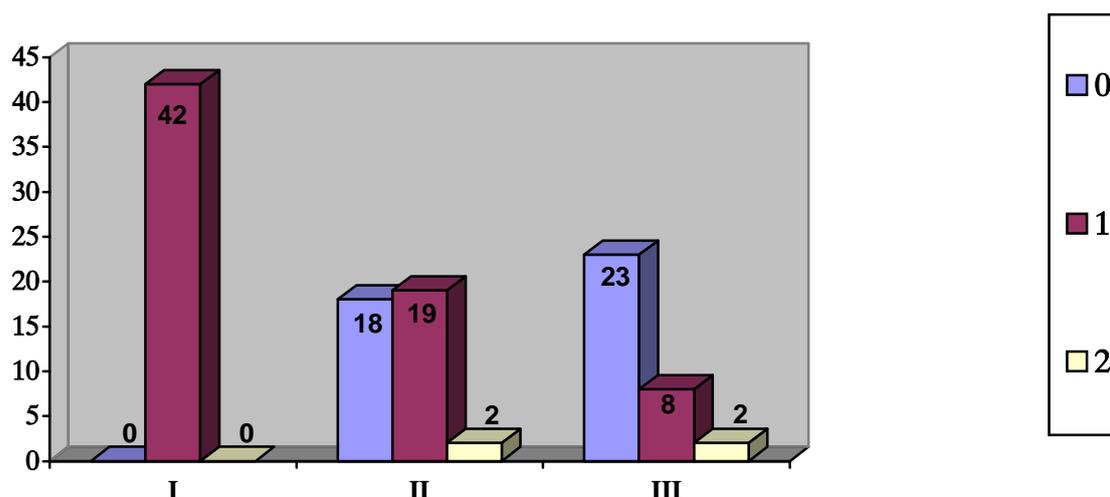


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Table 6. Distributions of patients treated with placebo according to HPV PCR typization on first, second and third check up

HPV PCR typization of the group treated with the investigated agent	I		I		I	
	n	%	n	%	n	%
Negative findings (0)	0	0	18	46.15	23	69.70
Persistence of the same HPV type (1)	42	100	19	48.72	8	24.24
Occurrence of a new high-risk HPV type (2)	0	0	2	5.13	2	6.06
total	42	100	39	100	33	100

Chart 6. Distributions of patients treated with placebo according to HPV PCR typization on first, second and third check up



Analysis with Friedman ANOVA test presented **statistical significant differences between HPV PCR typization on the first, the second and the third check-up among patients treated with placebo.** (ANOVA Chi Sqr. = 32,240 df = 2 p = 0,00001)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between HPV PCR typization on the first and the second check-up among patients treated with placebo.** (T = 22,00 Z = 3,249 p = 0,00115)

Analysis with Wilcoxon Matched Pairs Test presented **statistical significant differences between HPV PCR typization on the first and the third check-up among patients treated with placebo.** The difference is more significant at the third check-up compared to the first check-up.

(T = 26,00 Z = 3,672 p = 0,00024)

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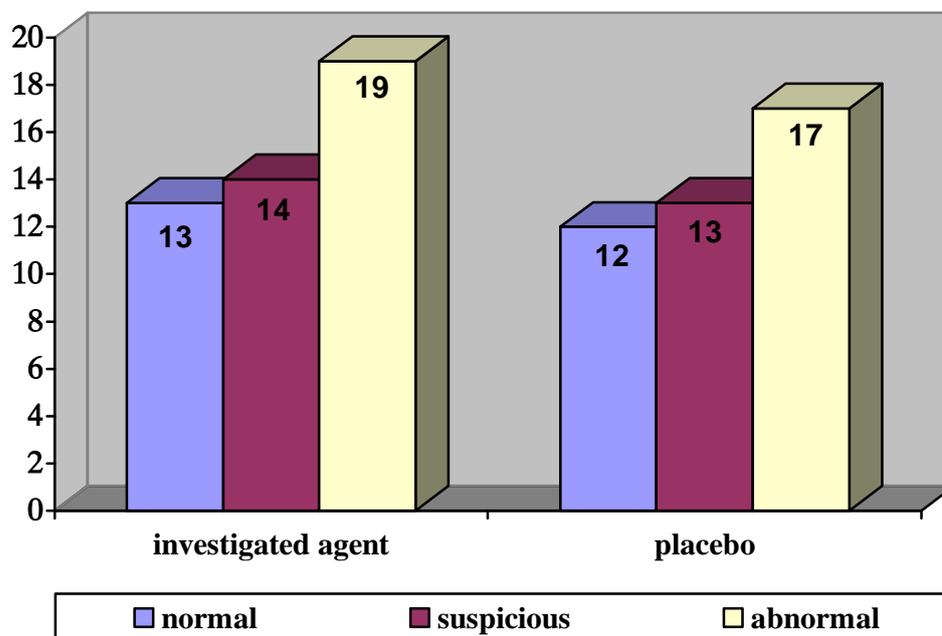
Analysis with Wilcoxon Matched Pairs Test did not present **statistical significant differences between HPV PCR typization on the second and the third check-up among patients treated with placebo.** (T = 7,00 Z = 0,733 p = 0,4630).

4. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO COLPOSCOPIC FINDINGS ON FIRST CHECK-UP

Table 7. Distribution of patients according to colposcopic findings on first check up

colposcopic findings	investigated agent		placebo	
	A	%	A	%
normal findings (0)	13	28.26	12	28.57
suspicious findings (1)	14	30.44	13	30.95
abnormal findings (2)	19	41.30	17	40.48
total	46	100	42	100

Chart 7. Distribution of patients according to colposcopic findings on first check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to colposcopic findings between groups on the first check-up.** (Z = - 0,062 p = 0,950)

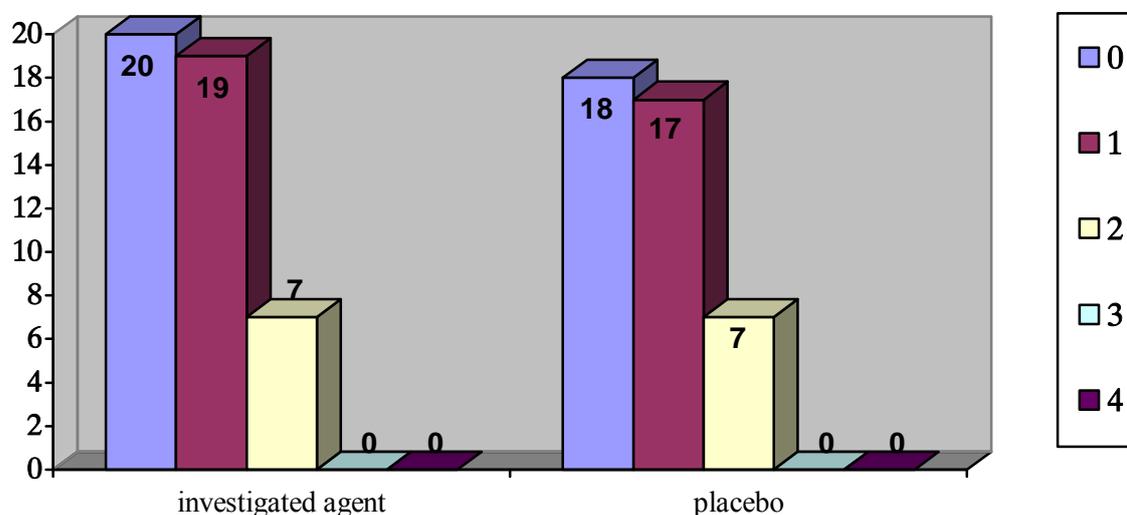
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5. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO PAP FINDINGS ON FIRST CHECK-UP

Table 8. Distribution of patients according to PAP findings on first check up

PAP findings	Treated with investigated agent		placebo	
	A	%	A	%
normal findings (0)	20	43.5	18	42.86
Low-grade SIL (1)	19	41.3	17	40.48
ASC-H / ASC-US (2)	7	15.2	7	16.67
High grade SIL (3)	0	0	0	0
Cais suspicious (4)	0	0	0	0
total	46	100	42	100

Chart 8. Distribution of patients according to PAP findings on first check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to PAP findings between groups on the first check-up.** (Z = - 0,1086 p = 0,9135).

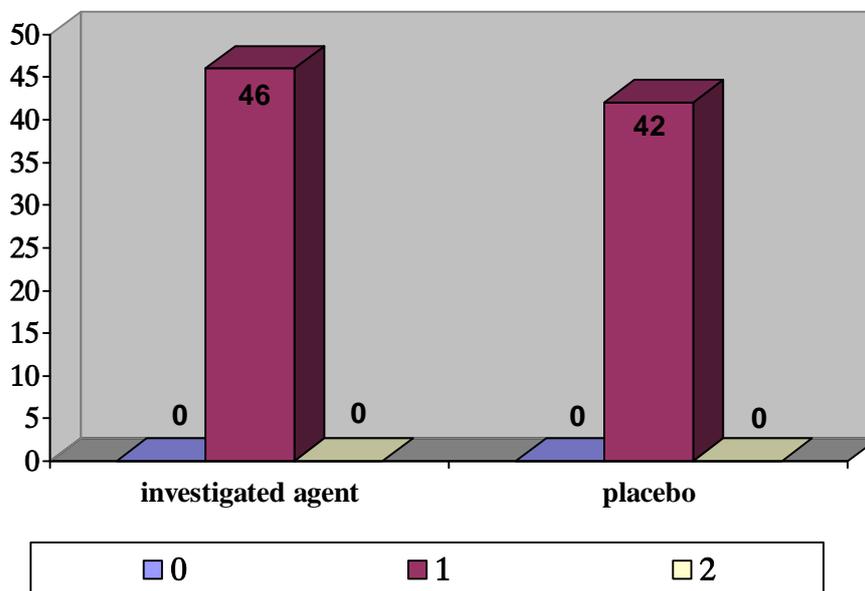
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evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically
(PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

6. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO HPV PCR TYPISATION
FINDINGS ON FIRST CHECK-UP

Table 9. Distribution of patients according to HPV PCR typization findings on first check up

HPV PCR typization	Treated with investigated agent		Treated with placebo	
	A	%	A	%
Negative findings (0)	0	0	0	0
Persistence of same HPV type (1)	46	100	42	100
Occurrence of new high-risk HPV type (2)	0	0	0	0
total	46	100	42	100

Chart 9. Distribution of patients according to HPV PCR typization findings on first check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to HPV PCR typization findings between groups on the first check-up.** ($Z = 0,000$ $p = 1,00$).

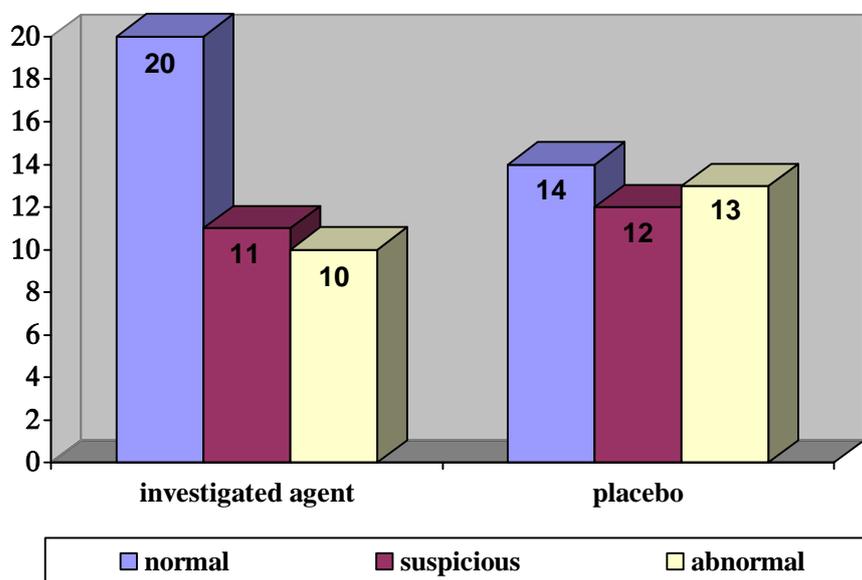
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7. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO COLPOSCOPIC FINDINGS ON SECOND CHECK-UP

Table 10. Distribution of patients according to colposcopic findings on second check up

colposcopic findings	Treated with investigated agent		Treated with placebo	
	A	%	A	%
normal findings (0)	20	48.78	14	35.89
suspicious findings (1)	11	26.83	12	30.77
abnormal findings (2)	10	24.39	13	33.33
total	41	100	39	100

Chart 10. Distribution of patients according to colposcopic findings on second check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to colposcopic findings between groups on the second check-up.** (Z = - 1,102 p = 0,2704)

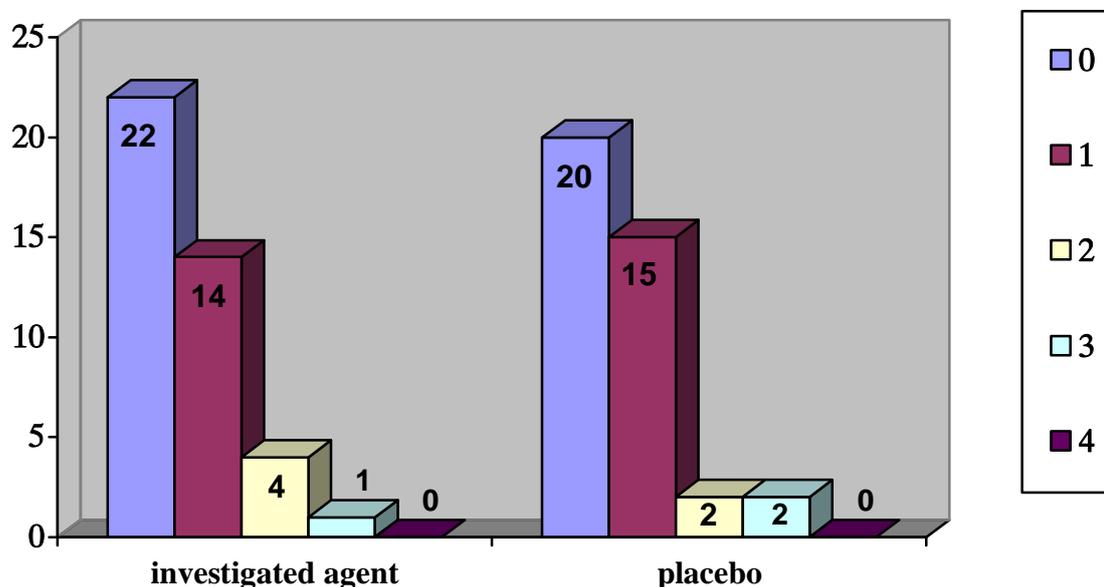
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8. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO PAP FINDINGS ON SECOND CHECK-UP

Table 11. Distribution of patients according to PAP findings on second check up

PAP findings	Treated with investigated agent		Treated with placebo	
	A	%	A	%
normal findings (0)	22	53.66	20	51.28
Low-grade SIL (1)	14	34.15	15	38.46
ASC-H / ASC-US (2)	4	9.76	2	5.13
High grade SIL (3)	1	2.43	2	5,13
CAis suspected (4)	0	0	0	0
total	41	100	39	100

Chart 11. Distribution of patients according to PAP findings on second check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to PAP findings between groups on the second check-up.** ($Z = - 0,120$ $p = 0,9042$).

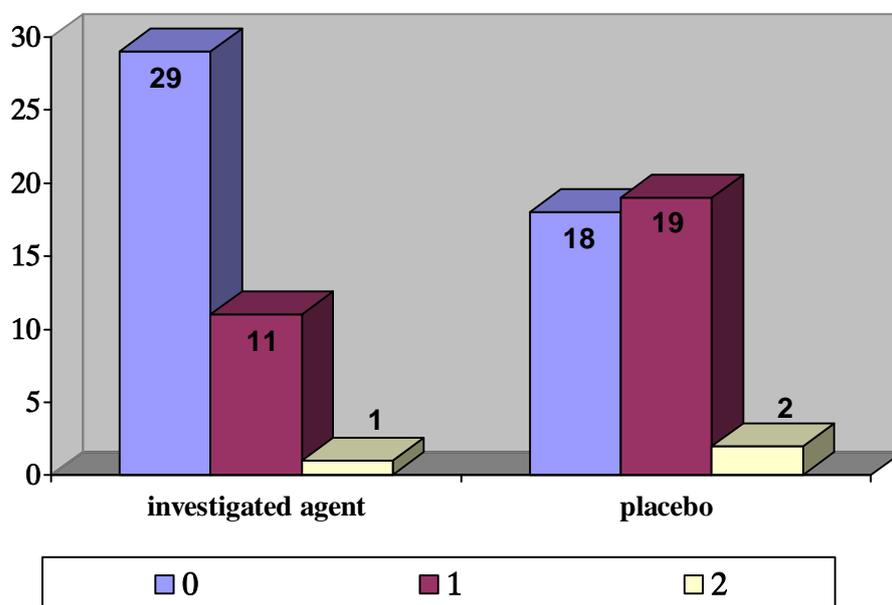
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9. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO HPV PCR TYPISATION FINDINGS ON SECOND CHECK-UP

Table 12. Distribution of patients according to HPV PCR typization findings on second check up

HPV PCR typization	Treated with investigated agent		Treated with placebo	
	A	%	A	%
Negative findings (0)	29	70.73	18	46.15
Persistence of same HPV type (1)	11	26.83	19	48.72
Occurrence of new high-risk HPV type (2)	1	2.44	2	5.13
total	41	100	39	100

Chart 12. Distribution of patients according to HPV PCR typization findings on second check up



Analysis with Mann-Whitney U Test presented **statistical significant differences according to HPV PCR typization findings between groups on the second check-up.**
(Z = - 2,208 p = 0,0272).

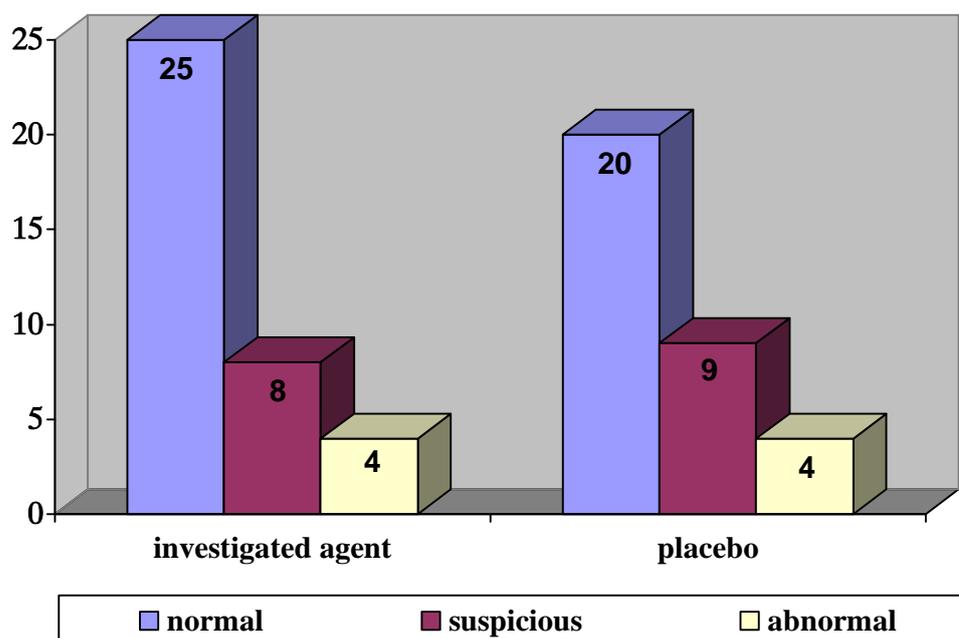
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10. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO COLPOSCOPIC FINDINGS ON THIRD CHECK-UP

Table 13. Distribution of patients according to colposcopic findings on third check up

colposcopic findings	Treated with investigated agent		Treated with placebo	
	A	%	A	%
normal findings (0)	25	67.57	20	60.6
suspicious findings (1)	8	21.62	9	27.3
abnormal findings (2)	4	10.81	4	12.1
total	37	100	33	100

Chart 13. Distribution of patients according to colposcopic findings on third check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to colposcopic findings between groups on the third check-up.** ($Z = -0,476$ $p = 0,6337$)

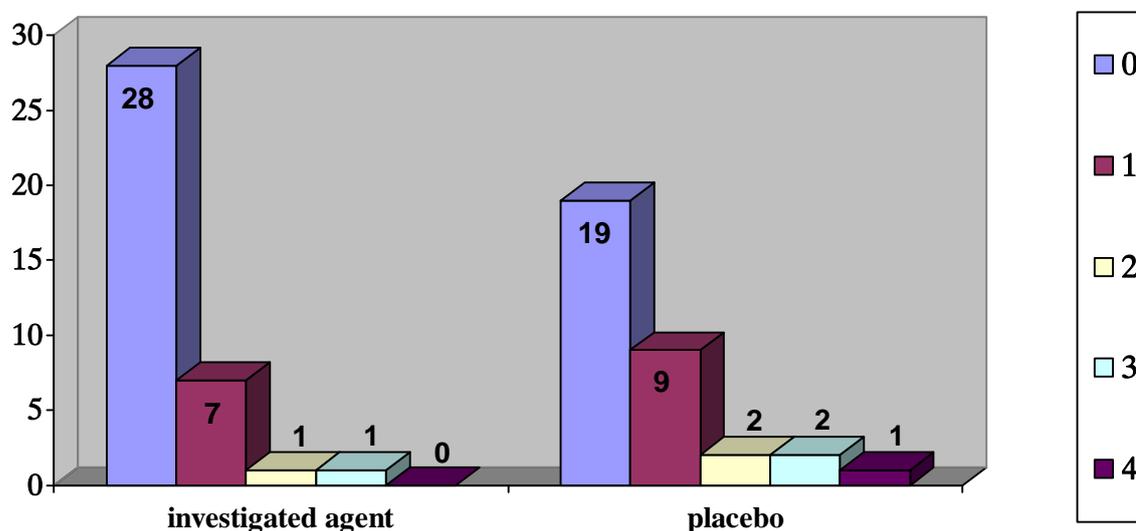
Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

11. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO PAP FINDINGS ON THIRD CHECK-UP

Table 14. Distribution of patients according to PAP findings on third check up

PAP findings	Treated with investigated agent		Treated with placebo	
	A	%	A	%
normal findings (0)	28	75.7	19	57.68
Low-grade SIL (1)	7	18.9	9	27.28
ASC-H / ASC-US (2)	1	2.7	2	6.06
High grade SIL (3)	1	2.7	2	6.06
CAis suspected (4)	0	0	1	3.03
total	37	100	33	100

Chart 14. Distribution of patients according to PAP findings on third check up



Analysis with Mann-Whitney U Test did not present **statistical significant differences according to PAP findings between groups on the third check-up.** ($Z = - 1,705$ $p = 0,0881$).

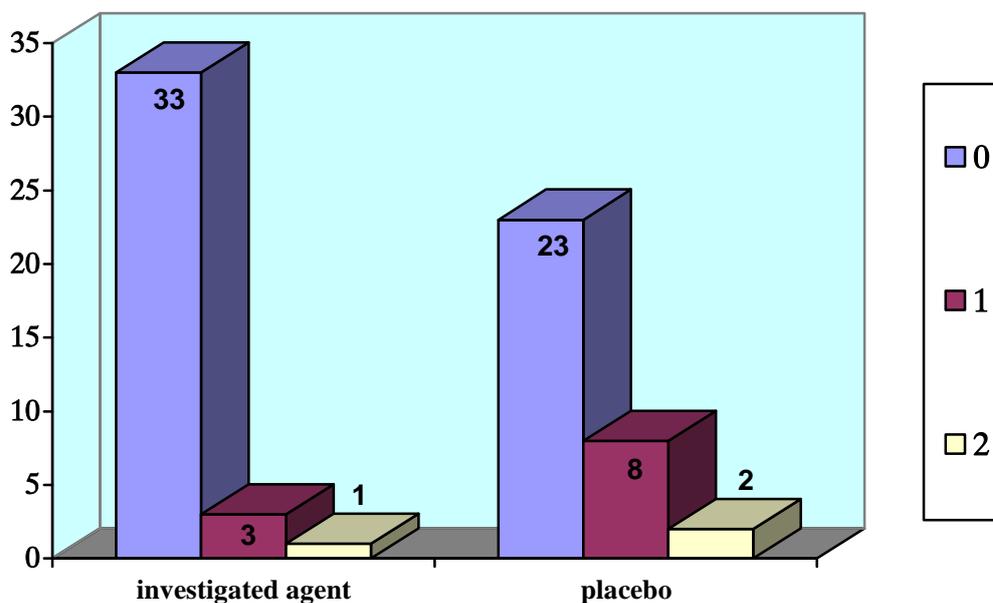
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12. CROSS ANALISYS BETWEEN GROUPS ACCORDING TO HPV PCR TYPISATION FINDINGS ON THIRD CHECK-UP

Table 15. Distribution of patients according to HPV PCR typization findings on third check up

HPV PCR typization	Treated with investigated agent		Treated with placebo	
	A	%	A	%
Negative findings (0)	33	89.2	23	69.7
Persistence of same HPV type (1)	3	8.1	8	24.2
Occurrence of new high-risk HPV type (2)	1	2.7	2	6.1
total	37	100	33	100

Chart 15. Distribution of patients according to HPV PCR typization findings on third check up



Analysis with Mann-Whitney U Test presented **statistical significant differences according to HPV PCR typization findings between groups on the third check-up. The statistical significance is smaller on the third check-up compared to the second check-up.**

(Z = - 1,995 p = 0,0460)

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

13. CORELATION BETWEEN SMOKING AND HPV PCR NEGATIVIZATION

Analysis with Fisher exact Test presented association (correlation) between HPV persistence and smoking among patients ($p = 0,0408$).

Smoking – when compared to all up-to-date trials investigating this variable, in our trial smoking significantly influenced on treatment outcome. Benzen-pyrene from the inhaled smoke, through lungs and blood stream, like in other organs, concentrates on the cervical mucus, where inhibits the immunologic features of the Langerhans cells, preventing secretion of adequate quantities interleukins needed for proper immunologic response.

The largest trial ever performed (Anthony Gunnell et all) among 105760 examinees, presented several conclusions:

- Women who smoked and had a high HPV-16 load during their first exam had a 27-fold increased risk of later cancer than women who smoked but did not have an HPV infection.
- Women who were positive for HPV-16 (irrespective of amount of viral load) and were smokers had a 14-fold increased risk over women who were HPV-16 negative and smoked.
- Nonsmoking women with high HPV-16 loads had just a 6-fold risk compared to HPV-negative nonsmokers

The researchers also found a statistically significant multiplicative interaction between the duration of smoking and HPV presence causing cervical cancer. One explanation for this interaction could involve the influence of smoking on persistence of HPV infection, probably due to localized immune suppression. Conversely, it could be related to the progression of neoplastic growth, since HPV and smoking appear to alter the levels of certain cytokines, which are involved in controlling abnormal cell growth and differentiation. More likely, both mechanisms combined are contributory factors.

In our trial examinees were globally distributed on smokers and non-smokers, variables such as average daily smoking or duration of smoking (in years) were not examined. Our conclusions are:

Slower HPV PCR negativization among smokers

Moderate statistically significant difference of HPV PCR negativization among smokers between both groups

14. CORELATION BETWEEN OTHER ANAMNESTICAL VARIABLES AND TREATMENT OUTCOME (HPV PCR NEGATIVIZATION ON THIRD CHECK-UP)

Age – average age of patients was $30,2 \pm 6,6$ years (min = 18, max = 47), which is excellent for research because of the correlation with the worldwide detected, age-related frequency of HPV infections. Meta-analyses confirm that HPV infections are most frequent between 18 and 30 years of age, which also corresponds to the age when sexual intercourses are most frequent. Our trial did not confirm significant correlation between age and treatment outcome (HPV PCR negativization) on the third check-up. ($\chi^2 = 2,14$ $df = 1$ $p = 0,1432$)

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Table 16. Distribution of patients according to age and HPV PCR typization findings on third check up

Age	HPV PCR typization on third check-up		total
	Positive findings	Negative findings	
below 30	6	36	42
over 31	8	20	28
total	14	56	70

Age of first sexual intercourse – nevertheless the level of discretion was very high (on the test lists only randomization number and initials were displayed, not full names nor ID numbers) we doubted the honesty of the anamnestic data on this topic, like the issues about active sex partners, number of ex-sex partners, number of abortions, as well as the frequency of sexual intercourses during the trial. Because trials confirmed increased relative risk for cervical carcinoma correlated to age of first sexual intercourse younger than 18 years (maybe better term would be age of start of personal sexual history), we have performed statistical analyses that did not present significant correlation between the age of first sexual intercourse and the treatment outcome (HPV PCR negativization) on the third check-up. ($\chi^2 = 1,16$ df = 1 p = 0,2819)

Table 17. Distribution of patients according to age of first sexual intercourse and HPV PCR typization findings on third check up

Age of first sexual intercourse	HPV PCR typization on third check-up		total
	Positive findings	Negative findings	
below 20	9	27	36
over 21	5	29	34
total	14	56	70

Number of births – according to published results, number of births and number of abortions impact on the risk of cervical carcinoma, especially in developing countries: woman with seven and more births have two fold increased risk than woman with one or two births. Young age of first birth, younger than 17 years of age, doubles the risk compared to 25 years of age. Also, risk of squamos cell cervical carcinoma doubles with 3 or more births compared to nullipara, but among nullipara the incidence of cervical adenocarcinoma is greater. In our trial, there were 56 nulliparas, 16 uniparas, 15 biparas and one multipara. Therefore, we analyzed if this variable would affect treatment outcome only comparing patients with no births to patients with any number of births. We did not detect statistical significant correlation between births and treatment outcome (HPV PCR negativization) on the third check-up. ($\chi^2 = 0,92$ df = 1 p = 0,3373)

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Table 18. Distribution of patients according to number of births and HPV PCR typization findings on third check up

births	HPV PCR typization on third check-up		total
	Positive findings	Negative findings	
yes	8	24	32
no	6	32	38
total	14	56	70

Current comorbidity therapy – because of the small cohort interactions between eventual comorbidity therapy and investigated agent / placebo was not analyzed.

Inhabitanace – most of the patients (88.63%) are inhabited in large communities, so this kind of prevalence did not give us the basis for statistical analysis of this variable to the treatment outcome (much bigger cohort and multicentric dispersion were needed). As an opinion, the advantage of a large urban community would be based on better access to condoms, better medical service and due to mentality (level of discretion), anamnestic data would be more precise.

Nationality – the cohort was homogenic, 90.90% patient are Macedonian, the cohort is not representative compared to the real demographic dispersion of minorities, so this variable was not analyzed for impact on treatment outcome. Multicentric trials are needed, especially from centers with multinational structure to establish any kind of correlation of nationality to treatment outcome. According to published results, there is no association of spontaneous regression of the lesions with nationality, but there are differences due to life style, sexual behavior, usage of condoms and the policy of screening. Due to interracial differences, some trials present lower risk among Black woman compared to Caucasian. In regions where men are circumcised there is lower incidence of HPV infections.

Number of abortions – as already mentioned above, because of suspicion on the honesty of anamnestic data, we did not analyze this variable. As a country, due to abortions per capita, we hold the second place in the region (Albania holds the first), but as expected, data from our trial do not correlate with the published data of abortions, per capita and total: in 2001 there were 8427 legal abortions in Macedonia (total, including illegal would be around 30000), and correlation between births and abortions is 3,2 : 1. Published results present increase relative risk for cervical carcinoma with one abortion of 2,3 fold, and two and more abortions increase the risk 4,92 fold compared to woman with no abortion. Also, it is very interesting that besides the expected increased risk for ovarian, endometrial and breast cancer among woman that have abortion, there is increased relative risk for hepatocellular carcinoma.

Condom use – 65 of 88 patients reported active sex life during the trial, and all of them reported regular usage of condoms, but occurrence of new HPV types (2 of 65) lead us to the conclusion not to investigate this variable, due to doubts on honesty reports. According to published data on this matter, usage of condoms shortens the time for spontaneous regression of the lesions, so condom use (mechanic barrier for infection, reinfection and co-infection) would be an important predictor.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Oral hormone contraceptive use – 5 of 88 patients were using oral hormone contraceptives during the trial. We did not analyze this variable due to the huge difference. According to meta-analysis (Berrington A. Cancer Research UK Epidemiology Unit, Radcliffe Infirmary, Oxford), where 12500 examinees from 24 countries were investigated, it was concluded that oral hormone contraceptive use no longer than two years does not increase the risk for cervical carcinoma, but longer than five years increase the risk by 10% and longer than ten years by 60%. Before this study it was taught that only the risk for breast cancer increases with the duration of oral hormone contraceptive use.

Marital status – 35 of 88 patients are single, 47 of 88 are married (first marriage), 6 of 88 are divorced after the first marriage and single during the trial. There is no significant correlation between marital status and treatment outcome.

Number of active sex partners – 65 of 88 patients reported one active sex partner, and 23 claimed no sexual intercourse at all during the trial. None reported more than one active sex partner. Among the newly detected high-risk HPV lesions, none patient reported new active sex partner, so there was no significance for the treatment outcome.

Number of ex-sex partners - the variable was not analyzed because we suspected the honesty of the anamnestic data – only 7 of 88 (7.95%) patients reported more than two ex-sex partners, but none reported more than four ex-sex partners.

Education – although according to published data the level of education plays a role in the manner of more eager attitude towards condom use, more regular check-ups and more regular hygienic and dietary regime, in our trial this variable was not analyzed because the cohort was homogenic (most of the patients had university degree).

Regularity of vitamin and antioxidant consumption – all patients (in both groups) were obliged on regular vitamin and antioxidant daily consumption (beta-carotene, folic acid, ascorbic acid, tocopherol and selenium). Doses were prescribed according to RDA for each substance. Because besides the RDA as a general prophylactic recommendation there is no data, or trial, or guideline for a daily intake that can establish this administration as an evidence-based treatment, the daily vitamin and antioxidant intake as variable influencing on the outcome was not statistically analyzed. According to published results on this matter, there is no significant correlation of vitamin and antioxidant supplements compared to normal, everyday food intake, although the only significant difference makes the inadequate caloric and qualitative food intake.

Other aspects of hygienic-dietary regime – many researchers confirmed two fold decreased risk of progression of cervical high-risk HPV lesions into cervical carcinoma, especially adenocarcinoma, among woman that take a shower more than six times a week, compared to woman that take a shower one to five times a week. Because of the small cohort, we did not analyze this variable.

ADVERSE EFFECTS

Of total 88 patients, adverse effects associated to administration of the investigated agent / placebo, we obtained the following results:

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

Among patients treated with the investigated agent there were two reports of abdominal discomfort and diarrhea, the treatment was discontinued in one of the patients because we confirmed that the adverse effect was directly provoked from the investigated agent. The other patient lowered the dose of ascorbic acid from 6000 mg to 1000 mg daily, the adverse effect vanished and patient continued with the treatment.

Among patients treated with placebo there were five reports of abdominal discomfort – epigastric pyrosis, the treatment was discontinued and H2-blocker or inhibitor of proton pump was suggested to be administered. We think that the difference of the ingredients between the investigated agent / placebo, the extract from herba absynthi, was the reason of such effect.

CONCLUSIONS

Although we had small number of patients (of 88 recruited, 70 completed the trial), our trial concluded that treatment of high-risk HPV lesions with extracts from aloe vera, viscum album, hypericum perforatum and propolis has significant favorable effect. We state this four extracts because so far, they are most extensively examined, but we feel the need of performing large multicentric trials, as well as the need of in vitro, animal model based and human studies for investigating effects of active substances from herbal extracts that are widely used into traditional medicine worldwide. The problem of exactness for stating conclusion for the therapeutic effect is obvious. One explanation is the large number of active substances, whose inter-reactions and pharmacokinetics when administered altogether, is almost impossible to determine. Second issue is the concentration constancy of the investigated agent – quality control for each serial of the agent must be performed, in order to determine if the agents from different series has the necessary concentration of active substances, because many investigations claim not only seasonal, but also endemic and geographic variations due to the concentration of a same active substance into an extract of a same herb species.

During the trial patients were administered with investigated agent and placebo from a same series. We can not give an exact answer on which active substances the favorable effect was accomplished, thus according to the literature, most serious candidates would be lectins and viscotoxins for viscum album, rutin, quercetin and proanthocyanidines from hypericum perforatum, aloe-emodin, aloe-barbaloin, acemannan or betamannan from aloe vera, acacetin from propolis or some other active substance from the other herbal extracts not yet properly investigated.

Our conclusion is that the investigated agent has favourable effect on the treatment of high-risk HPV lesions, especially among non-smokers. Time to HPV (PCR typization) negativization was significantly shortened. Percentage of normalized colposcopic and PAP findings is greater among the group treated with the investigated agent. Concerning safety, we conclude that the investigated agent is safe, administered as monotherapy or combined with vitamin and antioxidant therapy, remarking that because of the small cohort we did not examine correlation between eventual current therapy for different indication and adverse effects.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Table 19. Distribution of patients according to colposcopic, PAP and HPV PCR typization findings on third check up

colposcopic findings	CONTROL GROUP						PLACEBO GROUP					
	0		I		II		0		I		II	
	n	%	n	%	n	%	n	%	n	%	n	%
normal (0)	13	28.26	20	48.78	25	67.57	12	28.57	14	35.89	20	60.6
suspicious (1)	14	30.44	11	26.83	8	21.62	13	30.95	12	30.77	9	27.3
abnormal (2)	19	41.30	10	24.39	4	10.81	17	40.48	13	33.33	4	12.1
total	46	100	41	100	37	100	42	100	39	100	33	100
pap findings	CONTROL GROUP						PLACEBO GROUP					
	0		I		II		0		I		II	
	n	%	n	%	n	%	n	%	n	%	n	%
normal (0)	20	43.5	22	53.66	28	75,7	18	42.86	20	51.28	19	57.68
low-grade sil (1)	19	41.3	14	34.15	7	18.9	17	40.48	15	38.46	9	27.28
asc-h / asc-us (2)	7	15.2	4	9.76	1	2.7	7	16.67	2	5.13	2	6.06
high grade sil (3)	0	0	1	2.43	1	2.7	0	0	2	5,13	2	6.06
suspicious for invasive ca (4)	0	0	0	0	0	0	0	0	0	0	1	3.03
total	46	100	41	100	37	100	42	100	39	100	33	100
HPV PCR findings	CONTROL GROUP						PLACEBO GROUP					
	0		I		II		0		I		II	
	n	%	n	%	n	%	n	%	n	%	n	%
negative (0)	0	0	29	70.73	33	89.2	0	0	18	46.15	23	69.70
perzistence of the same HPV type (1)	46	100	11	26.83	3	8.1	42	100	19	48.72	8	24.24
new high-risk HPV type occurence (2)	0	0	1	2.44	1	2.7	0	0	2	5.13	2	6.06
total	46	100	41	100	37	100	42	100	39	100	33	100

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

LITERATURE

Ali SW, Evans AS, Monaghan JM. Results of CO₂ laser cylinder vaporization of cervical intraepithelial disease in 1234 patients. An analysis of failures. Br J Obstet Gynecol 1986;93(1):75-8.

Akihisa T, Yasukawa K, Oinuma H, et al. Triterpene alcohols from the flowers of compositae and their anti-inflammatory effects. Phytochemistry. Dec 1996;43(6):1255-1260.

Andren L, Andreasson A, Eggertsen R. Interaction between a commercially available St. John's wort product (Movina) and atorvastatin in patients with hypercholesterolemia. Eur J Clin Pharmacol. Oct 2007;63(10):913-916

Anon. License revoked for aloe vera use. Nat Med Law 1998;1:1-2.

Anthony Gunnell et al, Karolinska Institutet, Stockholm, High HPV Concentrations Combined with Cigarette Smoking Significantly Raise Risks of Later Cervical Cancer, Cancer Epidemiology, Biomarkers & Prevention, November 2006

Antony S, Kuttan R, Kuttan G. Effect of *Viscum album* in the inhibition of lung metastasis in mice induced by B16F10 melanoma cells. J Exp Clin Cancer Res. 1997;16:159-162.

Assessment report on *Aloe barbadensis* and aloe (various species, mainly *Aloe ferox* Mill. and its hybrids), EMEA/HMPC, Doc. Ref. 76313 /2007.

Baggish MS. Treating viral venereal infection with the CO₂ laser. J Reprod Med 1982;27(12):737-42.

Bako E, Deli J, Toth G. HPLC study on the carotenoid composition of *Calendula* products. J Biochem Biophys Methods. Oct-Nov 2002;53(1-3):241-250

Bankova V., Recent trends and important developments in propolis research, eCAM 2 (1) 29 - 32, 2005.

Bantel E and Breuninger H. In vitro studies of the effect of a mistletoe preparation on melanoma cells. Z Hautkr. 1990;65:562-564, 567

Barnes J, Anderson LA, Phillipson JD. Herbal Medicines. London: Pharmaceutical Press; 2002.

Barnes J, Anderson LA, Phillipson JD. St John's wort (*Hypericum perforatum* L.): a review of its chemistry, pharmacology and clinical properties. J Pharm Pharmacol. May 2001;53(5):583-600

Bayazit V. Cytotoxic effects of some animal and vegetable extracts and some chemicals on liver and colon carcinoma and myosarcoma. Saudi Med J. 2004;25:156-163

Becker H. Botany of European mistletoe (*Viscum album* L.). Oncology. 1986;43(Suppl 1):2-7

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Benrubi G, Nuss RC, Holmes K, Lammert N. Efficacy of cryotherapy in the treatment of human papillomavirus infection of the uterine cervix. *J Fla Med Assoc* 1986;73(3):188-91.

Benrubi GI, Shannon J, Glazer J, Nuss RC. Cervical human papilloma virus infection: is treatment possible? *J Fla Med Assoc* 1988;75(12):799-800.

Berger M, Schmahl D. Studies on the tumor-inhibiting efficacy of Iscador in experimental animal tumors. *J Cancer Res Clin Oncol.* 1983;105:262-265

Berghofer R., Holz J., Biflavonoids in *Hypericum perforatum*; Part 1. Isolation of 13,II8 - biapigenin, *Planta Med.* 53, 216 - 17, 1987.

Berghofer R., Holz J., Isolation of 13',II8 - biapigenin (amentoflavone) from *Hypericum perforatum*, *Planta Med.* 55, 91, 1989.

Beuers U., Spengler U., Pape G.R., Hepatitis after chronic abuse of senna, *Lancet*, 337 - 472, 1991.

Beta Carotene Cancer Prevention Study Group, (). "The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers." 1994 *N Engl J Med* 330 (15): 1029–35. doi:10.1056/NEJM199404143301501. PMID 8127329.

Bhopal JS. St John's wort-induced sexual dysfunction. *Can J Psychiatry.* Jun 2001;46(5):456-457

Bocci V. Mistletoe (*Viscum album*) lectins as cytokine inducers and immunoadjuvant in tumor therapy. A review. *J Biol Regul Homeost Agents.* 1993;7:1-6

Boothby RA, Carlson JA, Rubin M, Morgan M, Mikuta JJ. Single application treatment of human papillomavirus infection of the cervix and vagina with trichloroacetic acid: a randomized trial. *Obstet Gynecol* 1990;76(2):278-80

Boothby RA, Benrubi GI, Nuss RC, Lammert N. Laser treatment of Human papillomavirus infections of the uterine cervix. *Colposc Gynecol Laser Surg* 1986;2(1):25-9.

Bombardeli E., Morazzoni P, *Hypericum perforatum*, *Fitoterapija*, 66, 43 - 68, 1995.

Bostick RM, Potter JD, McKenzie DR, Sellers TA, Kushi LH, Steinmetz KA, Folsom AR. Reduced risk of colon cancer with high intakes of vitamin E: The Iowa Women's Health Study. *Cancer Res* 1993;15:4230-17

Bosch FX, Manos MM, Munoz N et al. Prevalence of human papillomavirus in cervical cancer: a worldwide perspective. *J Natl Cancer Inst,* 1995;87:796-802

Boucaud-Maitre Y, Algernon O, Raynaud J. Cytotoxic and antitumoral activity of *Calendula officinalis* extracts. *Pharmazie.* Mar 1988;43(3):220-221

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Boucard - Maitre Y. et al, Cytotoxic and antitumoral activity of *Calendula officinalis* extracts, Pharmazie, 43, 220, 1988 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Bradley GW, Clover A. Apparent response of small cell lung cancer to an extract of mistletoe and homeopathic treatment. Thorax. 1989;44:1047-1048

Bradley PR (ed.): British Herbal Compendium, vol. 1. British Herbal Medicine Association, Bournemouth, UK; 1992

Braakman et al., 2004 Treatment of HPV positive penile and cervical cancer by induction of HPV E7-specific T cell immunity through DNA vaccination

Brenner R, Azbel V, Madhusoodanan S, et al. Comparison of an extract of hypericum (LI 160) and sertraline in the treatment of depression: a double-blind, randomized pilot study. Clin Ther. Apr 2000;22(4):411-419

Brinker F. Herb Contraindications and Drug Interactions. 3rd ed. Sandy (OR): Eclectic Medical Publications; 2001

British Herbal Pharmacopoeia (BHP), British herbal medicine association, 1996.

Brodman M, Dottino P, Friedman Jr F, Heller D, Bleiweiss I, Sperling R Human papillomavirus-associated lesions of the vagina and cervix. Treatment with a laser and issueal 5-fluorouracil. J Reprod Med 1992;37(5):453-6.

Bruneton J., Pharmacognosy, Phytochemistry and Medicinal Plants, 2nd edition, Intercept Ltd., Londres, Paris, New York, 1999.

Bruseth S, Enge A. Mistletoe in the treatment of cancer. Tidsskr Nor Laegeforen. 1993;113:1058-1060

Bussing A, Suzart K, Bergmann J, et al. Induction of apoptosis in human lymphocytes treated with *Viscum album* L. is mediated by the mistletoe lectins. Cancer Lett. 1996;99:59-72

Bussing A, Schaller G, Pfuller U. Generation of reactive oxygen intermediates (ROI) by the thionins from *Viscum album* L. Anticancer Res. 1998;18(6A):4291-4296

Caballero F, Baez J, Troya CI. La crioterapia en el tratamiento de las lesiones premalignas del cervix uterino. Rev Med Caja Seguro Social 1985;17(2):140-3

Cancer Research UK : Jury still out over vitamin E supplements and increased lung cancer risk

Capasso F., Gaguinella T., Grandolini G., Izzo A., Phytotherapy, Springer, Berlin, Heidelberg, New York, Hong Kong, London, Milan, Paris, Tokyo, 2003.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Capasso F. et al., 1983 in Assessment report on Aloe barbadensis and aloe (various species, mainly Aloe ferox Mill. and its hybrids), EMEA/HMPC, Doc. Ref. 76313 /2007.

Carter JJ, Madeleine MM, Shera K, Schwartz SM, Cushing-Haugen KL, Wipf GC, Porter P, Daling JR, McDougall JK, Galloway DA. Human papillomavirus 16 and 18 L1 serology compared across anogenital cancer sites. Program in Cancer Biology, Fred Hutchinson Cancer Research Center, Seattle, Washington 98109-1024, USA

Casley - Smith J.R., The effect of Unguentum lymphaticum on acute experimental lymphoedema and other high - protein edemas, Lymphology, 16, 150 - 6, 1983 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Cazacu M, Oniu T, Lungoci C, et al. The influence of Isorel on the advanced colorectal cancer. Cancer Biother Radiopharm. 2003;18:27-34

Chan JM, Stampfer MJ, Giovannucci EL. What causes prostate cancer? A brief summary of the epidemiology. Semin Cancer Biol 1998;8:263-73

Chandler R.F. et al., Ethnobotany and phytochemistry of yarrow, Achillea millefolium, Compositae, Economic Bot., 36, 203 - 23, 1982 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Chandran PK, Kuttan R. Effect of Calendula officinalis Flower Extract on Acute Phase Proteins, Antioxidant Defense Mechanism and Granuloma Formation During Thermal Burns. J Clin Biochem Nutr. Sep 2008;43(2):58-64

Chen H.C. et al., 2004 in Assessment report on Aloe barbadensis and aloe (various species, mainly Aloe ferox Mill. and its hybrids), EMEA/HMPC, Doc. Ref. 76313 /2007.

Coeugniet EG, Elek E. Immunomodulation with Viscum album and Echinacea purpurea extracts. Onkologie. 1987;10(3 Suppl):27-33 Cinel A, Wittenberg L, Minucci D. β -interferon issueal treatment in low and high-risk cervical lesions. Clin Exp Obstet Gynecol 1991;18(2):91-7.

Costa S, Poggi MG, Palmisano L, Syrjänen S, Yliskoski M, Syrjänen K. Intramuscular β -interferon treatment of Human papillomavirus lesions in the lower female genital tract. Cervix 1988;6:203-12

Davila GW, Shroyer KR. Issueal 5-fluorouracil in the treatment of cervical Human papillomavirus infection. Gynecol Obstet Invest 1996;41(4):275-7.

Dean AJ, Moses GM, Vernon JM. Suspected withdrawal syndrome after cessation of St. John's wort. Ann Pharmacother. Jan 2003;37(1):150

Decker G. Patients are talking about--Iscador (mistletoe). Clin J Oncol Nurs. 2001;5:183-184

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Diakomanolis E, Rodolakis A, Simiakaki H, Frangopoulou E, Efstratiadou M. Randomized control trial of laser vaporization in the management of Human papillomavirus infection in the uterine cervix. *Rec Adv Cerv Pathol Colposc*, 1990;3:505-10

Diakomanolis E, Rodolakis A, Dailianas S, Kalpaktsoyiou K, Sakellaropoulos G, Aravantinos D. Long term results of CO₂ laser treatment of low-grade squamous intraepithelial lesions. *Cervix* 1995;13:149-52

Dervendzi V., Nasi lekoviti rastenija i nivna upotreba, Prosvetno delo, Skopje, 1977.

De Witte P., Metabolism and pharmacokinetics of antranoids, *Pharmacology*, 47, 86 - 97, 1993.

Dewick Paul, Medicinal Natural Product, John Wiley and Sons, Chichester, New York, Weinheim, Brisbane, Singapore, Toronto, 1997.

Dietrich JB, Ribereau-Gayon G, Jung ML, et al. Identity of the N-terminal sequences of the three A chains of mistletoe (*Viscum album* L.) lectins: homology with ricin-like plant toxins and single-chain ribosome-inhibiting proteins. *Anticancer Drugs*. 1992;3:507-511

Dimov V., Ivanovska N., Manolova N., Bankova V., Nikolov N., Popov S., Immunomodulatory action of propolis. Influence on anti - infectious protection and macrophage function, *Apologie*, 22, 155 - 62, 1991.

Dimov V., Ivanovska N., Bankova V., Popov S., immunomodulatory action of propolis: IV. Prophylactic activity against Gram - negative infections and adjuvant effect of the water - soluble derivative, *Vaccine*, 19, 817 - 23, 1992.

Duong Van Huyen JP, Delignat S, et al. Comparative study of the sensitivity of lymphoblastoid and transformed monocytic cell lines to the cytotoxic effects of *Viscum album* extracts of different origin. *Chemotherapy*. 2003;49:298-302

eCanadaNow.com - Vitamin E Linked To Increased Risk of Lung Cancer

Eggertsen R, Andreasson A, Andren L. Effects of treatment with a commercially available St John's Wort product (Movina) on cholesterol levels in patients with hypercholesterolemia treated with simvastatin. *Scand J Prim Health Care*. Sep 2007;25(3):154-159

Evans AS, Monaghan JM, Beattie AB. Carbon dioxide laser treatment of cervical warty atypias. *Gynecol Oncol* 1984;17:296

Evans W.C., Trease and Evans Pharmacognosy, W. B. Saunders, edonburg, London, New York, Philadelphia. St Luis, Sydney, Toronto, 2002.

Fairley CK, Tabrizi SN, Chen S, et al. A randomized trial of beta-carotene vs. placebo for the treatment of cervical HPV infection. *Int J Gynecol Cancer* 1996;6:225-30

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

FDA. Status of Certain Additional Over-the-Counter Drug Category II and III Active Ingredients. <http://www.fda.gov/ohrms/dockets/98fr/050902a.htm> (Accessed July 25, 2008)

Fetrow CW, Avila JR. Professional's Handbook of Complementary & Alternative Medicine. Springhouse (PA): Springhouse; 1999

Finall AI, McIntosh SA, Thompson WD. Subcutaneous inflammation mimicking metastatic malignancy induced by injection of mistletoe extract. *BMJ* 2006;333:1293-4

Fischer S, Scheffler A, Kabelitz D. Oligoclonal in vitro response of CD4 T cells to vesicles of mistletoe extracts in mistletoe-treated cancer patients. *Cancer Immunol Immunother.* 1997;44:150-156

Foster S, et al. Tyler's Honest Herbal: A Sensible Guide to the Use of Herbs and Related Remedies, 4th ed. New York: Hawthorn Herbal Press; 1999

Frank U, Engels I, Wagner A, et al. Influence of mistletoe (*Viscum album*) extracts on phagocytosis/burst activity of human phagocytes. *Eur J Clin Microbiol Infect Dis.* 2003;22:501-503

Franz H, Ziska P, Kindt A. Isolation and properties of three lectins from mistletoe (*Viscum album* L.). *Biochem J.* 1981;195:481-484

Fritz P, Dippon J, Kierschke T, et al. Impact of mistletoe lectin binding in breast cancer. *Anticancer Res.* 2004;24:1187-1192

Frye RF, Fitzgerald SM, Lagattuta TF, et al. Effect of St John's wort on imatinib mesylate pharmacokinetics. *Clin Pharmacol Ther.* Oct 2004;76(4):323-329

Gal D, Friedman M, Mitrani-Rosenbaum S. Transmissibility and treatment failures of different types of human papillomavirus. *Obstet Gynecol* 1989;73(3 Pt 1):308-11.

Gall SA, Constantine L, Koukol D. Therapy of persistent human papillomavirus disease with two different interferon species. *Am J Obstet Gynecol* 1991;164(1):130-4.

Giuliano AR, Gapstur S Arizona Prevention Center, University of Arizona, Tucson 85724, USA; Can cervical dysplasia and cancer be prevented with nutrients? *Nutr Rev* 1998 Jan; 56 (1 Pt 1):9-16

Godding E.W., Therapeutics of laxative agents with special reference to the anthraquinones, *Pharmacology* 14, 78 - 101, 1976.

Goebell P, et al. Evaluation of an unconventional treatment modality with mistletoe lectin to prevent recurrence of superficial bladder cancer: a randomized phase II trial. *J Urol.* 2002 Jul;168(1):72-5

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Gori J, Castano R, Dominguez J, et al. Laser vaporization, cryotherapy and trichloroacetic acid application in the therapy of ectocervical condylomatous lesions: a comparison of clinical results. *Cervix* 1992;10(4):217-9

Gorter RW, van Wely M, Stoss M, Wollina U. Subcutaneous infiltrates induced by injection of mistletoe extracts (Iscador). *Am J Ther.* 1998;5:181-187

Gorunovic M., Lukic P., Farmajkognozija, Univerzitet u Beogradu, Farmaceutski fakultet, Beograd, 2001.

Graham S, Sielezny M, Marshall J, Priore R, Freudenheim J, Brasure J, Haughey B, Nasca P, Zdeb M. Diet in the epidemiology of Postmenopausal Breast Cancer in the New York State Cohort. *Am J Epidemiol* 1992;136:3127-37

Grange J. M., Davey R. W., Antibacterial properties of propolis (bee glue), *J. R. Soc. Med.*, 83, 259 - 60, 1990.

Grimaudo S. et al., 1997 in Assessment report on *Aloe barbadensis* and aloe

Grossarth-Maticcek R, et al. Use of Iscador, an extract of European mistletoe (*Viscum album*) in cancer treatment: prospective nonrandomized and randomized matched-pair studies nested within a cohort study. *Altern Ther Health Med* 2001;7:57-78

Grubbs C. J. et al., "Chemoprevention of Chemically-Induced Mammary Carcinogenesis by Indole-3-Carbinol," Vol. 15, *Anticancer Res.*, 709-716, 1995

Gurley BJ, Swain A, Williams DK, et al. Gauging the clinical significance of P-glycoprotein-mediated herb-drug interactions: comparative effects of St. John's wort, Echinacea, clarithromycin, and rifampin on digoxin pharmacokinetics. *Mol Nutr Food Res.* Jul 2008;52(7):772-779

Guyatt GH, Sackett DL, Cook DJ. Users' guides to the medical literature. II. How to use an article about therapy or prevention. A. Are the results of the study valid? *JAMA* 1993;270(21):2598-601

Hajto T, Hostanska K, Weber K, et al. Effect of a recombinant lectin, *Viscum album* agglutinin on the secretion of interleukin-12 in cultured human peripheral blood mononuclear cells and on NK-cell-mediated cytotoxicity of rat splenocytes in vitro and in vivo. *Nat Immun.* 1998;16:34-46.

Hajto T, Hostanska K, Gabius HJ. Modulatory potency of the beta-galactoside-specific lectin from mistletoe extract (Iscador) on the host defense system in vivo in rabbits and patients. *Cancer Res.* 1989;49:4803-4808

Hajto T, Lanzrein C. Natural killer and antibody-dependent cell-mediated cytotoxicity activities and large granular lymphocyte frequencies in *Viscum album*-treated breast cancer patients. *Oncology.* 1986;43:93-97

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Hall AH, Spoerke DG, Rumack BH. Assessing mistletoe toxicity. *Ann Emerg Med.* 1986; 15:1320-1323

Happle R. Alternative medicine: really an alternative to academic medicine? *Hautarzt.* 2000;51:439-443

Hassauer W, Gutsch J, Burkhardt R. What prospects of success does Iscador therapy offer in advanced ovarian cancer? *Onkologie.* 1979;2:28-36

Heggie S, Bryant GP, Tripcony L, Keller J, Rose P, Glendenning M, et al. A Phase III study on the efficacy of topical aloe vera gel on irradiated breast tissue. *Cancer Nurs.* 2002 Dec;25(6):442-51

Helzlsouer K, Huang H, Alberg A, Hoffman S, Burke A, Norkus E, Morris J, Comstock G (2000). "Association between alpha-tocopherol, gamma-tocopherol, selenium, and subsequent prostate cancer". *J Natl Cancer Inst* 92 (24): 2018–23. doi:10.1093/jnci/92.24.2018. PMID 11121464

Heinrich M., Barnes J., Gibbons S., Williamson E., *Fundamentals of Pharmacognosy and Phytotherapy*, Churchill Livingstone, Edinburg, London, New York, Oxford, Philadelphia, St Luis, Sydney, Toronto, 2004.

Hoening J. et al., 1992 in Assessment report on *Aloe barbadensis* and aloe (various species, mainly *Aloe ferox* Mill. And its hybrids), EMEA/HMPC, Doc. Ref. 76313/2007.

Holme SA, Roberts DL. Erythroderma associated with St John's wort. *Br J Dermatol.* Nov 2000;143(5):1127-1128

Hording U, Rygaard C, Ruge S, Felding C, Lundvall F, Junge J. Cervical koilocytosis and high risk HPV types: the benefit of laser vaporization. *Eur J Obstet Gynecol Reprod Biol* 1993;51(2):125-30.

Huber R., Eisenbraun J., Miletzki B., Adler M., Gleiter C.H., *Pharmacokinetics of mistletoe lectins – a phase I study*, *int. J. Phytoth. Phytopharm.*, 2007 (<http://findarticles.com>).

Huber R, Klein R, Berg PA, et al. Effects of a lectin- and a viscotoxin-rich mistletoe preparation on clinical and hematologic parameters: a placebo-controlled evaluation in healthy subjects. *J Altern Complement Med.* 2002;8:857-866.

Huber R, et al. Effects of a mistletoe preparation with defined lectin content on chronic hepatitis C: an individually controlled cohort study. *Eur J Med Res* 2001;6:399-405

Hulsen H., Mechelke F., *In vitro* effectiveness of a mistletoe preparation on cytostatic - drug - resistant human leukemia cells, *Naturwissenschaften*, 74, 144 - 145, 1987.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Hulsen H, Doser C, Mechelke F. Differences in the in vitro effectiveness of preparations produced from mistletoes of various host trees. *Arzneimittelforschung*. 1986;36:433-436

Husseinzadeh N, Guoth JG, Jayawardena DS. Subclinical cervicovaginal human papillomavirus infections associated with cervical condylomata and dysplasia. Treatment outcomes. *J Reprod Med* 1994;39(10):777-80

Iljazovic E, Ljuca D, Sahimpasic A, Avdic S. Pathology Department, Polyclinic for Laboratory Diagnostic, University Clinics Center Tuzla, Trnovac 1, Gradina, 75,000 Tuzla, Bosnia and Herzegovina; PMID: 17177657 [PubMed - indexed for MEDLINE] Efficacy in treatment of cervical HRHPV infection by combination of beta interferon, and herbal therapy in woman with different cervical lesions. *Bosn J Basic Med Sci*. 2006 Nov;6(4):79-84

Imai H, Kotegawa T, Tsutsumi K, et al. The recovery time-course of CYP3A after induction by St John's wort administration. *Br J Clin Pharmacol*. May 2008;65(5):701-707

Instituto Fernandes Figueira, Fundação Oswaldo Cruz, Rio de Janeiro, Brazil, Efficacy in treatment of subclinical cervical HPV infection without intraepithelial neoplasia: systematic review; indexed for Medline, LILACS, Excerpta Medica

Ishii O., Tanizawa H., Tqakino Y., Studies of Aloe III: Mechanism of laxative effect., *Chemical and pharmacological bulletin*, 38, 197 - 200, 1990.

Ivanovska N., Stefanova Z., Valeva V., Neychev H., immunomodulatory action of propolis. VII. A comparative study on cinnamic and caffeic acid lysine derivatives, *Biol. Immun.*, 46, 115 - 7, 1993.

Ivanovska N., Neychev h.m, Stefanova Z., Nankova V., Popov S., Influence of cinnamic acid on lymphocyte proliferation, cytokine release and Klebsiella infection in mice, *Apologie*, 26, 73 - 81, 1995.

Izzo A.A. et al., 1996 and 1999 in Assessment report on Aloe barbadensis and aloe (various species, mainly Aloe ferox Mill. And its hybrids), EMEA/HMPC, Doc. Ref. 76313/2007.

Jiang X, Williams KM, Liauw WS, et al. Effect of St John's wort and ginseng on the pharmacokinetics and pharmacodynamics of warfarin in healthy subjects. *Br J Clin Pharmacol*. May 2004;57(5):592-599.

Jose Silva Couto and Daniel Pereira da Silva, Use Of Coriolus Versicolor As Immunonutrition In HPV Patients With Cervical Lesions (LSIL) 20th European Congress of Obstetrics and Gynaecology 2008

Johne a. et al., Interaction of St John's wort extracts with digoxin, in WHO selected monographs of medicinal plants, WHO, Geneve, 2002.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Jung ML, Baudino S, Ribereau-Gayon G, Beck JP. Characterization of cytotoxic proteins from mistletoe (*Viscum album* L.). *Cancer Lett.* 1990;51:103-108

Kalvatchev Z, Walder R, Garzaro D. Anti-HIV activity of extracts from *Calendula officinalis* flowers. *Biomed Pharmacother.* 1997;51(4):176-180.

Kasper S, Volz HP, Moller HJ, et al. Continuation and long-term maintenance treatment with *Hypericum* extract WS((R)) 5570 after recovery from an acute episode of moderate depression - A double-blind, randomized, placebo controlled long-term trial. *Eur Neuropsychopharmacol.* Nov 2008;18(11):803-813.

Kasper S, Anghelescu IG, Szegedi A, et al. Placebo controlled continuation treatment with *Hypericum* extract WS 5570 after recovery from a mild or moderate depressive episode. *Wien Med Wochenschr.* 2007;157(13-14):362-366.

Kast A, Hauser SP. Helixor--mistletoe preparation for cancer therapy. documentation No. 19. *Schweiz Rundsch Med Prax.* 1990;79:291-295

Keefe KA, Schell MJ, Brewer C, McHale M, Brewster W, Chapman JA, Rose GS, McMeeken DS, Lagerberg W, Peng YM, Wilczynski SP, Anton-Culver H, Meyskens FL, Berman ML. Division of Gynecologic Oncology, and Chao Family Comprehensive Cancer Center, University of California, Irvine, Medical Center, Orange, California 92868, USA) A randomized, double blind, Phase III trial using oral beta-carotene supplementation for women with high-grade cervical intraepithelial neoplasia; *Cancer Epidemiol Biomarkers Prev.* 2001 Oct;10(10):1029-35

Kleeberg UR, et al. Final results of the EORTC 18871/DKG 80-1 randomised phase III trial. rIFN-alpha2b versus rIFN-gamma versus ISCADOR M versus observation after surgery in melanoma patients with either high-risk primary (thickness >3 mm) or regional lymph node metastasis. *Eur J Cancer.* 2004 Feb;40(3):390-402

Kilic A., Baysallar M., Besirbellioglu B., salih B., Sorkun K., Tanyuksel M., In vitro antimicrobial activity of propolis against methicillin - resistant *Staphylococcus aureus* and vancomycin - resistant *enterococcus faecium*, *Annals of Microbiology*, 55, 113 - 117,2005.

Klein R., Effects of mistletoe extracts on immunocompetent cells in vitro and in vivo, *Int. J. Phytotherapy. Phytopharmac.*, 2007 (<http://findarticles.com>).

Kleijnen J, Knipschild P. Mistletoe treatment for cancer: Review of controlled trials in humans. *Phytomedicine* 1994;1:255-60

Klett CY, Anderer FA. Activation of natural killer cell cytotoxicity of human blood monocytes by a low molecular weight component from *Viscum album* extract. *Arzneimittelforschung.* 1989;39:1580-1585

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Klutke JJ, Bergman A. Interferon as an adjuvant treatment for genital condyloma acuminatum. *Int J Gynecol Obstet* 1995;49:171-4

Kjaer M. Mistletoe (Iscador) therapy in stage IV renal adenocarcinoma. A Phase II study in patients with measurable lung metastases. *Acta Oncol.* 1989;28:489-494.

Kovacs E. The in vitro effect of *Viscum album* (VA) extract on DNA repair of peripheral blood mononuclear cells (PBMC) in cancer patients. *Phytother Res.* 2002;16:143-147

Konopa J, Woynarowski JM, Lewandowska-Gumieniak M. Isolation of viscotoxins. Cytotoxic basic polypeptides from *Viscum album* L. *Hoppe Seylers Z Physiol Chem.* 1980;361:1525-1533

Krenzelok EP, Jacobsen TD, Aronis J. American mistletoe exposures. *Am J Emerg Med.* 1997;15:516-520.

Kristin A. Keefe, Michael J. Schell, Cheryl Brewer, Michael McHale, Wendy Brewster, Julia A. Chapman, G. Scott Rose, D. Scott McMeeken, Westley Lagerberg, Yei-Mei Peng, Sharon P. Wilczynski, Hoda Anton-Culver, Frank L. Meyskens and Michael L. Berman A Randomized, Double Blind, Phase III Trial Using Oral β -Carotene Supplementation for Women with High-Grade Cervical Intraepithelial Neoplasia: *Cancer Epidemiology Biomarkers & Prevention* Vol. 10, 1029-1035, October 2001 American Association for Cancer Research

Kolesnikova A.G., Bactericidal and immunocorrective properties of plant extract, *Zh. Mikrobiol. Epidemiol. Immunobiol.*, 3, 75 - 8, 1986. In Newall C., Anderson L., Phillipson D., *Herbal Medicines, A guide for health - care professionals*, The Pharmaceutical Press, London, 1996.

Kujungiev A., Tsvekova I., Serkedjieva Yu., Bankova V., Christov R., Popov S., Antibacterial, antifungal and antiviral activity of propolis of different geographic origin, *J. Ethnopharmacol.*, 64, 235 - 240, 1999.

Kuehn JJ. Favorable long-term outcome with mistletoe therapy in a patient with centroblastic-centrocytic non-Hodgkin lymphoma. *Dtsch Med Wochenschr.* 1999;124:1414-1418.

Kulevanova S., *Farmakognozija, Kultura*, Skopje, 2004.

Kunze E, Schulz H, Ahrens H, Gabius HJ. Lack of an antitumoral effect of immunomodulatory galactoside specific mistletoe lectin on N-methyl-N-nitrosourea-induced urinary bladder carcinogenesis in rats. *Exp Toxicol Pathol.* 1997;49:167-180

Kuo PL, Lin TC, Lin CC. The antiproliferative activity of aloe-emodin is through p53-dependent and p21-dependent apoptotic pathway in human hepatoma cell lines. *Life Sci.* 2002 Sep 6;71(16):1879-92

Kuttan G, Vasudevan DM, Kuttan R. Effect of a preparation from *Viscum album* on tumor development in vitro and in mice. *J Ethnopharmacol.* 1990;29:35-41

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Lee A, Chui PT, Aun CS, Gin T, Lau AS. Possible interaction between sevoflurane and Aloe vera. *Ann Pharmacother.* 2004 Oct;38(10):1651-4

Lee JK, Lee MK, Yun YP, Kim Y, Kim JS, Kim YS, et al. Acemannan purified from Aloe vera induces phenotypic and functional maturation of immature dendritic cells. *Int Immunopharmacol.* 2001 Jul;1(7):1275-84

Lee KH, Kim JH, Lim DS, Kim CH. Anti-leukaemic and anti-mutagenic effects of di(2-ethylhexyl)phthalate isolated from Aloe vera Linne. *J Pharm Pharmacol.* 2000 May;52(5):593-8

Lee JK, Lee MK, Yun YP, Kim Y, Kim JS, Kim YS, et al. Acemannan purified from Aloe vera induces phenotypic and functional maturation of immature dendritic cells. *Int Immunopharmacol.* 2001 Jul;1(7):1275-84).

Leroi R. Postoperative Viscum album therapy after surgery of breast neoplasms. *Helv Chir Acta.* 1977;44:403-414

Leuner K, Kazanski V, Muller M, et al. Hyperforin--a key constituent of St. John's wort specifically activates TRPC6 channels. *FASEB J.* Dec 2007;21(14):4101-4111

Leung A.Y., *Encyclopedia of common natural ingredients used in food, drugs and cosmetics*, Wiley, New York, Chichester, 1980.

Leuschner J., Preclinical toxicological profile of Hypericum extract LI 160., second International Congress of Phytomedicine, Munich, 1996 in WHO selected monographs of medicinal plants, WHO, Geneve, 2002.

Linde K, Knuppel L. Large-scale observational studies of hypericum extracts in patients with depressive disorders--a systematic review. *Phytomedicine.* Jan 2005;12(1-2):148-157

Linde K, Mulrow CD, Berner M, et al. St John's wort for depression. *Cochrane Database Syst Rev.* 2005(2):CD000448.

Linde K. et al., St John's wort for depression – an overview and meta - analysis of randomized clinical trials, *British Medical Journal*, 313, 253 - 258, 1996, in WHO selected monographs of medicinal plants, WHO, Geneve, 2002.

Linde K, Berner MM, Kriston L. St John's wort for major depression. *Cochrane Database Syst Rev.* 2008(4):CD000448

Lomgo R. 2002 in Assessment report on Aloe barbadensis and aloe (various species, mainly Aloe ferox Mill. and its hybrids), EMEA/HMPC, Doc. Ref. 76313 /2007.

Lukic P., *Farmakognozija*, 4 dopuweno izdawe, Univerzitet u Beogradu, Farmaceutski fakultet, Beograd, 1988.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

Luesley MD, Cullimore J, Redman CWE, et al. Loop diathermy excision of the cervical transformation zone in patients with abnormal cervical smears. *BMJ* 1990;300:1690-3

Luczkiewicz M, Cisowski W, Kaiser P, et al. Comparative analysis of phenolic acids in mistletoe plants from various hosts. *Acta Pol Pharm.* 2001;58:373-379

Luther P, Sehr I, Bergmann KC, Reutgen H. Allergy and lectins: action between IgE-mediated histamine release and glycoproteins from *Viscum album* L. (mistletoe). *Acta Biol Med Ger.* 1978;37:1623-1628

Mackerras D, Irwig L, Simpson JM, Weisberg E, Cardona M, Webster F, Walton L, Ghersi D Department of Public Health and Community Medicine, University of Sydney, NSW, Australia; Randomized double-blind trial of beta-carotene and vitamin C in women with minor cervical abnormalities, *Br J Cancer* 1999 Mar;79(9-10):1448-53

Madej J, Basta A, Madej Jr JG, Strama M. Colposcopy staging and treatment of papillomavirus infection of the cervix. *Clin Exp Obstet Gynecol* 1992;19(1):34-9

Mahabir S., Schendel K., Dong Y.Q., Barrera S.L., Spitz M.R., Forman M.R. (2008). "Dietary alpha-, beta-, gamma- and delta-tocopherols in lung cancer risk". *International Journal of Cancer* 123: 1173–1180. doi:10.1056/NEJM199404143301501. PMID 8127329

Malviya VK, Deppe G, Pluszczynski R, Boike G. Trichloroacetic acid in the treatment of human papillomavirus infection of the cervix without associated dysplasia. *Obstet Gynecol* 1987;70:72-4

Mansky PJ, Grem J, Wallerstedt DB, et al. Mistletoe and gemcitabine in patients with advanced cancer: a model for the phase I study of botanicals and botanical-drug interactions in cancer therapy. *Integr Cancer Ther.* 2003;2:345-352

Markowska J, Nowak M, Niecewicz R, Breborowicz J, Wiese E, Zengteler G. Results of issueal treatment of HPV infection in the uterine cervix using interferon beta, 13-cis-retinoic acid and TFX. *Eur J Gynecol Oncol* 1994;15(1):65-9.

Markowitz JS, Donovan JL, DeVane CL, et al. Effect of St John's wort on drug metabolism by induction of cytochrome P450 3A4 enzyme. *JAMA.* Sep 17 2003;290(11):1500-1504.

Mathijssen RH, Verweij J, de Bruijn P, et al. Effects of St. John's wort on irinotecan metabolism. *J Natl Cancer Inst.* Aug 21 2002;94(16):1247-1249.

Menendez-Velazquez JF, Gonzalez-Sanchez JL, Rodriguez-de-Santiago JD, Munoz-Reyes R, Bailon-Uriza R. Tratamiento de la infeccion cervical por el virus del papiloma humano (VPH) con acido tricloroacetico. *Ginecol Obstet Mex* 1993;61:48-51

Mengs U, Gothel D, Leng-Peschlow E. Mistletoe extracts standardized to mistletoe lectins in oncology: review on current status of preclinical research. *Anticancer Res* 2002;22:1399-407

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Merkur H, Baird PJ. Papillomavirus infection of the female genital tract before and after treatment: a cytological, colposcopic and histological study. *Aust NZJ Obstet Gynecol* 1989;29(3 Pt 1):185-8

Miller L., Murray W., *Herbal Medicinale, A clinician's guide*, Pharmaceutical Product Press, New York, London, 1998.

Mills S., Bone K., *Principles and Practice of Phytotherapy*, Churchill livinstone, Edinburg, London, New York, Philadelphia, St Luis, Sydney, Toronto, 2000.

Mills SY: *The Dictionary of Modern Herbalism*. Thorsons, Wellingborough, UK; 1985

Minucci D, Cinel A, Insacco E. Diathermic loop treatment for CIN and HPV lesions. A follow-up of 130 cases. *Eur J Gynecol Oncol* 1991;12(5):385-93.

Mishenkova E.L. et al., Antiviral properties of St John's wort and preparations produced from it, in Newall C., Anderson L., Phillipson D., *Herbal Medicines, A guide for health - care professionals*, The Pharmaceutical Press, London, 1996.

Mitchell H, Medley G. Evidence against diathermy as a beneficial treatment for HPV infection of the cervix. *Aust N Z J Obstet Gynecol* 1989;29(4):439-42.

Moskalenko S. A., Preliminary screening of far - Easteren ethnomedicinal plants for antimicrobial activity, *J. Ethnopharmacol.*, 15, 231 - 59, 1986.

Mohanty KC, Lowe JW. Cryotherapy in the management of histologically diagnosed subclinical HPV infection of the cervix. *Br J Clin Pract* 1990;44(12):574-7.

Mohanty KC, Lowe JW. Cold coagulation therapy in the treatment of histologically diagnosed subclinical human papillomavirus infection of the cervix. *Br J Clin Pract* 1991;45(2):102-4
Maier G, Fiebig HH. Absence of tumor growth stimulation in a panel of 16 human tumor cell lines by mistletoe extracts in vitro. *Anticancer Drugs*. 2002;13:373-379.

Mueller EA, Anderer FA. A *Viscum album* oligosaccharide activating human natural cytotoxicity is an interferon gamma inducer. *Cancer Immunol Immunother*. 1990;32:221-227

Mueller SC, Majcher-Peszynska J, Mundkowski RG, et al. No clinically relevant CYP3A induction after St. John's wort with low hyperforin content in healthy volunteers. *Eur J Clin Pharmacol*. Sep 3 2008

Muller - Lissner S. A., Adverse effects of laxatives: facts and fiction, *Phatrmacology*, 47, 138 - 145, 1993.

Nebel A. et al., Potential metabolic interaction between St John's wort and theophylline, *Annal of Pharmacotherapy*, 33, 502, 1999.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Newall CA, Anderson LA, Phillipson JD: Herbal Medicines. A Guide for Health-care Professionals. Pharmaceutical Press, London, UK; 1996

Newfield et al., "Estrogen Metabolism & Human papillomavirus-Induced Tumors of the Larynx: Chemo-Prophylaxis with Indole-3-Carbinol," Vol. 13, Anticancer es., 337-341,1993

Nieminen P, Aho M, Lehtinen M, Vesterinen E, Vaheri A, Paavonen J. Treatment of genital HPV infection with carbon dioxide laser and systemic interferon alpha-2b. Sex Transm Dis 1994;21(2):65-9

Nikolai G, Friedl P, Werner M, et al. Effect of a mistletoe extract (Iscador QuFrF) on viability and migratory behavior of human peripheral CD4+ and CD8+ T lymphocytes in three-dimensional collagen lattices. In Vitro Cell Dev Biol Anim. 1997;33:710-716

Nursing in Practice - Vitamin E "linked to cancer"

Olsen DL, Raub W, Jr., Bradley C, Johnson M, Macias JL, Love V, et al. The effect of aloe vera gel/mild soap versus mild soap alone in preventing skin reactions in patients undergoing radiation therapy. Oncol Nurs Forum. 2001 Apr;28(3):543-7

Olsnes S, Stirpe F, Sandvig K, Pihl A. Isolation and characterization of viscumin, a toxic lectin from *Viscum album* L. (Mistletoe). J Biol Chem. 1982;257:13263-13270

Okpanyi S.N., Weischer M.L., Animal experiments on the psychotropic action of a Hypericum extract, *Arzneimittelforschung*, 37, 10 - 13, 1987.

Orrus S, Scaloni A, Giannattasio M, et al. Amino acid sequence, S-S bridge arrangement and distribution in plant tissues of thionins from *Viscum album*. Biol Chem. 1997;378:989-996

Panossian A.G., Immunosuppressive effects of hypericin on stimulated human leucocytes: inhibition of the arachidonic acid release, leukotriene B4 and interleukin - 1 α production and activation of nitric oxide formation, *Phytomedicine*, 3, 19 - 28, 1996.

Parker V, Wong AH, Boon HS, et al. Adverse reactions to St John's Wort. *Can J Psychiatry*. Feb 2001;46(1):77-79 Pigatto PD, Guzzi G. Aloe linked to thyroid dysfunction. *Archives of medical research*. 2005 Sep-Oct;36(5):608

Paulino N., dantas A.P., Bankova V. et al., Bulgarian propolis induced analgesic and anti-inflammatory effects in mice and inhibits in vitro contraction of airway smooth muscle, *J. Pharmacol. Sciences* 93, 307 - 313, 2003.

PDR for Herbal Medicines, second edition, Thomson Physicians Desk References, 2000.

Petkov V., Plants with hypotensive, antiatheromatous and coronarodilatating action, *Am. J. Chin. Med.*, 7, 197 - 236, 1979.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Petkov V., *Sovremena fitoterapija, Medicina i fizkultura*, Sofia, 1982.

Piscitelli S.C. et al., Indinavir concentrations and St John's wort, *Lancet*, 355, 547 - 548, 2000.

Pommier P, Gomez F, Sunyach MP, et al. Phase III randomized trial of *Calendula officinalis* compared with trolamine for the prevention of acute dermatitis during irradiation for breast cancer. *J Clin Oncol*. Apr 15 2004;22(8):1447-1453

Pugh N, Ross SA, ElSohly MA, Pasco DS. Characterization of Aloeride, a new high-molecular-weight polysaccharide from *Aloe vera* with potent immunostimulatory activity. *J Agric Food Chem*. 2001 Feb;49(2):1030-4

Puttemans P, van Pelle Y, De Muylder E. Carbon dioxide laser vaporization of cervical subclinical papillomaviral infection neoplasia: short-term effectiveness. *Eur J Obstet Gynecol Reprod Biol* 1986;23:167-80

Qian ZW, Mao SJ, Cai XC, et al. Viral etiology of chronic cervicitis and its therapeutic response to a recombinant interferon. *Chin Med J* 1990;103(8): 647-51

Rabe C, Musch A, Schirmacher P, Kruis W, Hoffmann R. Acute hepatitis induced by an *Aloe vera* preparation: a case report. *World J Gastroenterol*. 2005 Jan 14;11(2):303-4.

Reid R, Stanhope CR, Herschman BR, Booth E, Phibbs GD, Smith JP. Genital warts and cervical cancer: I. Evidence of an association between subclinical papillomavirus infection and cervical malignancy. *Cancer* 1982;50:377-87

Reider N, Komericki P, Hausen BM, et al. The seamy side of natural medicines: contact sensitization to arnica (*Arnica montana* L.) and marigold (*Calendula officinalis* L.). *Contact Dermatitis*. 2001 Nov;45(5):269-72

Renske D.M. Steenbergen, Jillian de Wilde, Saskia M. Wilting, Antoinette A.T.P. Brink, Peter J.F. Snijders and Chris J.L.M. Meijer, HPV-mediated transformation of the anogenital tract, doi:10.1016/j.jcv.2004.11.019

Ribereau-Gayon G, Jung ML, Di Scala D, Beck JP. Comparison of the effects of fermented and unfermented mistletoe preparations on cultured tumor cells. *Oncology*. 1986;43(Suppl 1):35-41

Risberg T, Wist E, Melsom H, Kaasa S. Use of alternative medicine among Norwegian hospitalized cancer patients. *Tidsskr Nor Laegeforen*. 1997;117:2458-2463

Riva JM, Sedlacek TV, Cunnane MF, Mangan CE. Extended carbon dioxide laser vaporization in the treatment of subclinical papillomavirus infection of the lower genital tract. *Obstet Gynecol* 1989;73(1):25-30.

Robbers JE, et al. *Pharmacognosy and pharmacobiotechnology*. Baltimore: Williams & Wilkins; 1996.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Rogstad KE, Dixon CA, Ahmed IH. The management of subclinical wart virus infection of the cervix in a genitourinary clinic. *Geniturin Med* 1992;68(5):307-8.

Ruge S, Felding C, Skouby SO, Lundvall F, Hording U. CO₂ laser vaporization of human papillomavirus (HPV)-induced abnormal cervical smears. A simple and effective solution to a recurrent clinical problem. *Clin Exp Obstet Gynecol* 1991;18(2):99-101.

Ruge S, Felding C, Skouby SO, Lundvall F, Hording U, Norrild B. CO₂ laser vaporization in the treatment of cervical Human papillomavirus infection in women with abnormal Papanicolaou smears. *Gynecol Obstet Invest* 1992;33(3):172-6.

Sakar M.K. et al., Antimicrobial activities of some hypericum species growing in Turkey, *Fitoterapia*, 59, 49 - 52, 1988.

Salzer G, Havelec L. Prevention of recurrence of bronchial carcinomas after surgery by means of the mistletoe extract Iscador. Results of a clinical study from 1969-1971. *Onkologie*. 1978;1:264-267.

Salzer G. Investigation to improve the survival of patients with bronchial carcinomas "radically operated." *Z Erkr Atmungsorgane*. 1975;142:127-131

Sardella A, Lodi G, Demarosi F, et al. Hypericum perforatum extract in burning mouth syndrome: a randomized placebo-controlled study. *J Oral Pathol Med*. Aug 2008;37(7):395-401

Sarrell EM, Mandelberg A, Cohen HA. Efficacy of naturopathic extracts in the management of ear pain associated with acute otitis media

Sarrell EM, Cohen HA, Kahan E. Naturopathic treatment for ear pain in children. *Pediatrics*. May 2003;111(5 Pt 1):e574-579. *Arch Pediatr Adolesc Med*. Jul 2001;155(7):796-799

Samuelsson Gunnar, *Drugs of Natural Origin*, Apotekar societeten, Swedish Pharmaceutical Press, Stockholm, 1999.

Scheller S., Gadza G., Pietsz G., Gabrys J., Szumlak J., Eckert L., Shani J., The ability of ethanol extract of propolis to stimulate plaque formation in immunized mouse spleen cells, *Pharmacol. Res. Commun.*, 20, 323 - 8, 1998.

Scheer R. The effect of mistletoe lectins on the limulus amoebocyte lysate test. *Arzneimittelforschung*. 1993;43:795-800

Scheller S., Gadza G., Crol W., Czuba Z., Zajus A., Gabrys J., Shani J., The ability of ethanolic extract of propolis (EPP) to protect mice against gamma irradiation, *Z. Naturforsch. Sect. C. Biosc.*, 44, 1040 - 52, 1989.

Schimmer O. et al., An evaluation of 55 commercial plant extracts in the Ames mutagenicity test, *Pharmazie*, 49, 448 - 451, 1994.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

Schimmer O. et al., The mutagenic potencies of plant extracts containing quercetin in salmonella typhimurium TA 98 I TA 100, Mutation Research, 206, 201 - 2008, 1988, in WHO selected monographs of medicinal plants, WHO, Geneva, 2002.

Schipochliev T. et al., Anti - inflammatory action of a group of plant extracts., Vet. Med. Nauki, 18, 87 - 94, 1981 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Shipochliev T., Fournadjiev G., spectrum of the anti - inflammatory effect of *Arctostaphylos uva ursi* and *Achillea millefolium*, Probl. Vutr. Med., 12, 99 - 107, 1984 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Schneider A, Papendick U, Gissmann L, De Villiers E-M. Interferon treatment of human genital papillomavirus infection: importance of viral type. Int J Cancer 1987;40:610-4

Schrader E. Equivalence of St John's wort extract (Ze 117) and fluoxetine: a randomized, controlled study in mild-moderate depression. Int Clin Psychopharmacol. Mar 2000;15(2):61-68.

Schulz V, et al. Rational Phytotherapy: A Physician's Guide to Herbal Medicine, 4th ed. New York: Springer; 2001.

Schwarz UI, Hanso H, Oertel R, et al. Induction of intestinal P-glycoprotein by St John's wort reduces the oral bioavailability of talinolol. Clin Pharmacol Ther. May 2007;81(5):669-678.

Seitz W. The effect of Iscador (*Viscum praeparatum M.*) on the Walker carcinosarcoma of the rat. Wien Klin Wochenschr. 1975;87:131-132

Semprini AE, Stillo A, Marcozzi S, Castagna C, Fiore S, Radaelli U. Treatment with interferon for genital HPV in HIV-positive and HIV-negative women. Eur J Obstet Gynecol Reprod Biol 1994;53(2):135-7.

Sesti F, De-Santis L, Farne C, Mantenuto L, Piccione E. Efficacy of CO₂ laser surgery in treating squamous intraepithelial lesions. An analysis of clinical and virologic results. J Reprod Med 1994;39(6):441-4.

Sforcin J.M., Efeito da sazonalidade sobre as propriedades imunomoduladora e antibacteriana da propolis e perfil bioquímico de ratos, Veterinaria e Zootecnia, 1996.

Shljahov P., Pcelarstvo, Nasha kniga. Skopje, 1990.

Siegers C.P., Anthranoid laxatives and colorectal cancer, Trends in pharmacological sciences, 13, 229 - 231, 1992.

Smith P, Bullock JM, Booker BM, et al. The influence of St. John's wort on the pharmacokinetics and protein binding of imatinib mesylate. Pharmacotherapy. Nov 2004;24(11):1508-1514.

University Gynecology and Obstetrics Clinic, Vodnjanska bb, University St. Cyrillus and Methodius, Skopje, Macedonia

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Spiller HA, Willias DB, Gorman SE, Sanfletban J. Retrospective study of mistletoe ingestion. *J Toxicol Clin Toxicol.* 1996;34:405-408.

Spitzbart H., Hoyme U.B., Department of Obstetrics and Gynaecology, Frauenklinik, Erfurt, Germany, Immunotherapy of Gynaecological High-Risk Human Papilloma Virus Infection With Human Leukocyte Ultrafiltrate, *Infect. Dis. Obstet. Gynecol.* 8:120-123, 2000. © 2000 Wiley-Liss, Inc.

Stampf J.L. et al. The sensitizing capacity of helenin and two of its main constituents the sesquiterpene lactones alantolactone and isoalantolactone: a comparison of epicutaneous and intradermal sensitizing methods in different strains of guinea pig., *Contact Dermatitis*, 1982 in

Stein GM, Berg PA. Mistletoe extract-induced effects on immunocompetent cells: in vitro studies. *Anticancer Drugs.* 1997;8(Suppl 1):S39-S42

Stein GM, Berg PA. Evaluation of the stimulatory activity of a fermented mistletoe lectin-1 free mistletoe extract on T-helper cells and monocytes in healthy individuals in vitro. *Arzneimittelforschung.* 1996;46:635-639.

Stein GM, Schaller G, Pfüller U, et al. Thionins from *Viscum album L.*: influence of the viscotoxins on the activation of granulocytes. *Anticancer Res.* 1999;19(2A):1037-1042

Stefanon D. Activity of interferon- β in small condylomatous lesions of the uterine cervix. *Cervix* 1983;1:23-6.

Stentella P, Frega A, Di-Renzi F, Palazzetti PL, Pachi A. Issue and systemic administration of natural alpha interferon in the treatment of female and male HPV genital infections. *Clin Exp Obstet Gynecol* 1996;23(1):29.

Stettin A, Schultze JL, Stechemesser E, Berg PA. Anti-mistletoe lectin antibodies are produced in patients during therapy with an aqueous mistletoe extract derived from *Viscum album L.* and neutralize lectin-induced cytotoxicity in vitro. *Klin Wochenschr.* 1990;68:896-900.

Steuer-Vogt MK, et al. The effect of an adjuvant mistletoe treatment programme in resected head and neck cancer patients: a randomised controlled clinical trial. *Eur J Cancer* 2001;37:23-31

Stevinson C, Ernst E. A pilot study of *Hypericum perforatum* for the treatment of premenstrual syndrome. *BJOG.* Jul 2000;107(7):870-876

Strand A, Wilander E, Zehbe I, Rylander E. High risk HPV persists after treatment of genital papillomavirus infection but not after treatment of cervical intraepithelial neoplasia. *Acta Obstet Gynecol Scand* 1997;76(2):140.

Stirpe F, Legg RF, Onyon LJ, et al. Inhibition of protein synthesis by a toxic lectin from *Viscum album L.* (Mistletoe). *Biochem J.* 1980;190:843-845

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

Stoss M, Gorter RW. No evidence of IFN-gamma increase in the serum of HIV-positive and healthy subjects after subcutaneous injection of a non-fermented *Viscum album* L. extract. *Nat Immun.* 1998;16:157-164

Strehl E., Volpert R., elstner E.F., Biochemical activities of propolis extract, *Z. Naturforsch C.*, 49, 39 - 43, 1994.

Szakiel A, Ruszkowski D, Grudniak A, et al. Antibacterial and Antiparasitic Activity of Oleanolic Acid and its Glycosides isolated from Marigold (*Calendula officinalis*). *Planta Med.* Nov 2008;74(14):1709-1715.

Szegedi A, Kohnen R, Dienel A, et al. Acute treatment of moderate to severe depression with hypericum extract WS 5570 (St John's wort): randomised controlled double blind non-inferiority trial versus paroxetine. *BMJ.* Mar 5 2005;330(7490):503

Summary report on *Hypericum perforatum*, EMEA/MRL/690/99, 1999.

Surwit EA, Graham V, Droegemueller W, et al. Evaluation of issueally applied trans-retinoic acid in the treatment of cervical intraepithelial lesions. *Am J Obstet Gynecol* 1982;143(7):821-3

Sydiskis RJ, Owen DG, Lohr JL, Rosler KH, Blomter RN (Department of Microbiology, University of Maryland, Baltimore Antimicrobe Agents Chemother 1991 Dec; 35(12):2463-6

Tanvetyanon T, Bepler G (July 2008). "Beta-carotene in multivitamins and the possible risk of lung cancer among smokers versus former smokers: a meta-analysis and evaluation of national brands". *Cancer* 113 (1): 150–7. doi:10.1002/cncr.23527. PMID 1842900

Tatefuji T., Izumi N., Ohta T., Arai S., Ikeda M., Kurimoto M., Isolation and identification of compounds from Brazilian propolis which enhance macrophage spreading and mobility, *Biol. Pharm. Bull.*, 19, 966 - 70, 1996.

Tavares I.A. et al., 1996 in Assessment report on *Aloe barbadensis* and aloe (various species, mainly *Aloe ferox* Mill. and its hybrids), EMEA/HMPC, Doc. Ref. 76313 / 2007.

Tigris Pharmaceuticals, Multi-Center Phase II Clinical Trial of A-007 in Patients with High-Grade CIN and Invasive Cervical Carcinoma St.I

Timoshenko AV, Gabius HJ. Efficient induction of superoxide release from human neutrophils by the galactoside-specific lectin from *Viscum album*. *Biol Chem Hoppe Seyler.* 1993;374:237-243

Torrise A, Cinel A, Wittemberg L, Minucci D. Therapy with β -interferon cream in the cervical-vaginal infections by Human papilloma virus (HPV). *Eur J Gynecol Oncol* 1987;7(4-5):441.

Trofatter Jr. Interferon treatment of anogenital papillomavirus-related diseases. *Dermatol Clin* 1991;9(2):343-52

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Tucakov J., Lecenje biljem, IRO RAD Beograd, 1984.

Tusenius KJ, Spoek JM, Kramers CW. Iscador Qu for chronic hepatitis C: an exploratory study. Complement Ther Med 2001;9:12-6

Tyler, V. Herbs of Choice, the Therapeutical Use of Phytomedicinals. Binghamton, New York: Pharmaceutical Press; 1994.

Tyler V.E., The honest herbal, 3rd edition, Philadelphia, Strickley, 1993.

Ukiya M, Akihisa T, Yasukawa K, et al. Anti-inflammatory, anti-tumor-promoting, and cytotoxic activities of constituents of marigold (*Calendula officinalis*) flowers. J Nat Prod. Dec 2006;69(12):1692-1696

Wagner H et al., immunostimulating polysaccharides (heteroglycans) of higher plants, Arzaemittelforsch, 35, 1069, 1985 in Newall C., Anderson L., Phillipson D., Herbal Medicines, A guide for health - care professionals, The Pharmaceutical Press, London, 1996.

Van Huyen JP, Bayry J, Delignat S, et al. Induction of apoptosis of endothelial cells by *Viscum album*: a role for anti-tumoral properties of mistletoe lectins. Mol Med. 2002;8:600-606

van Wely M, Stoss M, Gorter RW. Toxicity of a standardized mistletoe extract in immunocompromised and healthy individuals. Am J Ther. 1999;6:37-43

Ward BG, Thomas IL. Randomized prospective intervention study of Human cervical wart virus infection. Aust N Z J Obstet Gynecol 1994;34(2):182-5

Wattenberg, L. W. & Loub, W. D Inhibition of Polycyclic Aromatic Hydrocarbon-Induced Neoplasia by Naturally Occurring Indoles", Vol. 38, Cancer Res., 1410-1413, (1978);

Weber W, Vander Stoep A, McCarty RL, et al. Hypericum perforatum (St John's wort) for attention-deficit/hyperactivity disorder in children and adolescents: a randomized controlled trial. JAMA. Jun 11 2008;299(22):2633-2641

Weck PK, Buddin DA, Whisnant JK. Interferon in the treatment of genital Human papillomavirus infections. Am J Med 1988;85(Suppl 2A):159-64

Wenzel-Seifert K, Lentzen H, Seifert R. In U-937 promonocytes, mistletoe lectin I increases basal $[Ca^{2+}]_i$, enhances histamine H1- and complement C5a-receptor-mediated rises in $[Ca^{2+}]_i$, and induces cell death. Naunyn Schmiedebergs Arch Pharmacol. 1997;355:190-197

WHO monographs on selected medicinal plants, Volume 1, WHO, Geneva, 1999.

WHO monographs on selected medicinal plants, Volume 2, WHO, Geneva, 2002.

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without biologically verified CIN I

Wichtl M., herbal drugs and Phytopharmaceuticals, Medpharm, scientific Publisher, Stuttgart 1994.

Wildfeuer A, Mayerhofer D. The effects of plant preparations on cellular functions in body defense. *Arzneimittelforschung*. 1994;44:361-366

Villa LL. Human papillomaviruses and cervical cancer. *Adv Cancer Res*, 1997;71:321-41

Villani C, Nobili F, Stentella P, Pace S. The viral lesion (HPV) of the lower genital tract in teenagers: clinical and therapeutic aspects. *Eur J Gynecol Oncol* 1989;10(4):267-71.

Villani C, Vecchione A, Pace S, Carenza L. The rationale for treatment of cervical lesions (HPV +/- CIN). *Clin Exp Obstet Gynecol* 1989;16(1):44-7

Williams MS, Burk M, Loprinzi CL, Hill M, Schomberg PJ, Nearhood K, et al. Phase III double-blind evaluation of an aloe vera gel as a prophylactic agent for radiation-induced skin toxicity. *Int J Radiat Oncol Biol Phys*. 1996 Sep 1;36(2):345-9

Wincelous SJ, Brown C, Goh BT. Managing women with human papillomavirus changes in cervical cytology. *Genitourin Med* 1988;64(4):259-62

Woodman CBJ, Byrne P, Kelly KA, Hilton C. A randomized trial of laser vaporization in the management of cervical intraepithelial neoplasia associated with Human papillomavirus infection. *J Public Health Med* 1993;15(4):327-31.

Wojnarowski JM, Konopa J. Interaction between DNA and viscotoxins. Cytotoxic basic polypeptides from *Viscum album* L. *Hoppe Seylers Z Physiol Chem*. 1980;361:1535-1545.

Wu K, Willett WC, Chan JM, Fuchs CS, Colditz GA, Rimm EB, Giovannucci EL. A prospective study on supplemental vitamin E intake and risk of colon cancer in women and men. *Cancer Epidemiol Biomarkers Prev* 2002;11:1298-304

Xu H, Williams KM, Liauw WS, et al. Effects of St John's wort and CYP2C9 genotype on the pharmacokinetics and pharmacodynamics of gliclazide. *Br J Pharmacol*. Apr 2008;153(7):1579-1586

Yagi A, Kabash A, Okamura N, Haraguchi H, Moustafa SM, Khalifa TI. Antioxidant, free radical scavenging and anti-inflammatory effects of aloesin derivatives in *Aloe vera*. *Planta medica*. 2002 Nov;68(11):957-60

Yliskoski M, Syrjänen K, Syrjänen S, Saarikoski S, Nethersell A. Systemic alpha-interferon (Wellferon) treatment of genital Human papillomavirus (HPV) Type 6,11,16 and 18 infections: a double-blind, placebo-controlled trial. *Gynecol Oncol* 1991;43:55-60

Clinical trial on therapeutic efficacy, tolerability and safety of peroral solutions Varumin 1 and 2 (Inter-evrogeneks) in a combined treatment with polyvitamin – antioxidative therapy in patients with biologically (PCR) detected HPV infection of high-risk type, with or without bioptically verified CIN I

Yliskoski M, Saarikoski S, Syrjänen K, Syrjänen S, Castrén O. Cryotherapy and CO₂ laser vaporization in the treatment of cervical and vaginal HPV infections. *Acta Obstet Gynecol Scand* 1989;68(7):619-25

Yoshida T, Zhang M, Chen C, et al. Enhancement of the cytotoxicity of mistletoe lectin-1 (ML-1) by high pH or perturbation in golgi functions. *Pharmazie*. 1991;46:349-351

Zarcone R. Treatment of genital HPV-infections with natural alpha interferon from normal human leukocytes. *Clin Exp Obstet Gynecol* 1995;22(1):14-9.

Zarcone R, Bellini P, Cardone G, Cardone A. Treatment of cervix condylomata with alpha-IFN leucocytar. *Clin Exp Obstet Gynecol* 1995;22(4):326-9.

Zarkovic N, Zarkovic K, Grainca S, et al. The *Viscum album* preparation Isorel inhibits the growth of melanoma B16F10 by influencing the tumor-host relationship. *Anticancer Drugs*. 1997;8(Suppl 1):S17-S22.

Zhang L, Tizard IR. Activation of a mouse macrophage cell line by acemannan: the major carbohydrate fraction from *Aloe vera* gel. *Immunopharmacology*. 1996 Nov;35(2):119-28