**Uses and examples of NYSTATIN (Nys)**

1. The Structure of Nystatin

Nys, like all polyenes, consists of two different chemical structures, namely a macrolide ring and an amino sugar. The amino sugar, a mycosamine (3-amino-3,6-dideoxy-D-mannose) is glycosidically attached to the macrolide ring. The macrolide ring consists of carbon atoms and is closed by the formation of an internal ester or a lactone. The macrolide ring is a stable rod-shaped structure with a hydrophobic side which is built by the polyene chain (π-electron carrying structure) and an opposite hydrophilic side which is built by the hydroxyl groups. The conjugated double-bonds of the polyene chain are in the trans-position. The sugar moiety is bonded at the one end of the macrolide ring and carries a primary amino group. At the same end, a carboxyl group is present on the macrolide ring. A single strongly polarized and hydrophilic hydroxyl group is positioned at the other end of the macrolide ring and imparts to this part of Nys hydrophilic properties. The amphiphilic character of Nys is determined by the entirety of these groups. They are responsible for the orientation of the molecules in biological membranes. There is strong evidence that the sugar moiety and the carboxyl group are on the extracellular side of the membrane while the strongly polarized hydroxyl group is on the cytoplasmatic side, although flip-flop phenomena cannot be completely precluded. The orientation of Nys in the membrane thus corresponds to that of an integral protein. All membrane proteins are glycoproteins and their glycosidic part is always on the extracellular side. This rigid orientation of the membrane proteins is connected with their specific function.

In the bioenergetic sense, the structure of Nys exhibits all functional groups that can also be found in the membrane proteins and that determine their mode of action as semiconductors or energy transporters. The long macrolide ring corresponds the transmembrane part of the integral protein. Like the α-helix, it possesses a hydrophilic side and a lipophilic side which consists of a long π-electron chain. The amphipathic character of Nys determines its self-organization in the lipid bilayer. The macrolide ring protrudes into the membrane. It is about 22 to 25 Angstrom in length and is about as long as the α-helix of a membrane protein. The sugar moiety determines its orientation. Nys has two charged groups, one positively charged (the NH$_3^+$ group at the sugar moiety) and the other negatively charged (the COO$^-$ group at the macrolide ring) which are in vicinity to the π-electron chain. They act as electron donor and electron acceptor, depending on the voltage polarity. Nys thus possesses a soliton triplet, just as the membrane proteins. It functions like a semiconductor in the membrane potential and allows the ion transport across the membrane. Nys is known as ionophoric. The dipole character of Nys is determined by the strongly polar hydroxyl group at the opposite end of the macrolide ring, the π-electrons of the polyene chain and the positively and negatively charged groups. When starting only from the charge of the π-electrons that can move freely in the polyene chain and can undergo a polarization under the membrane potential, one can estimate the dipole energy of Nys:

$$E_D = q.l.F_2 \approx 10^{-19} \text{J}$$

($q=n.e$, number of π-electrons, n=12 for Nys, n=14 for Amp; l=22×10$^{-10}$ m length of the π-electron chain, $F_p = 4.5\times 10^7$ V/m$^{-1}$, electric field strength of the plasma potential, e=1.6×10$^{-19}$ coulomb). The dipole energy of a Nys molecule is smaller by factor 10$^5$ than the energy of the plasma gradient $E=10^{-14}$ J. The energy turnover between the levels of self-organization proceeds in energy packages, energy
quantums, with fixed energy amounts. A Nys molecule under the given conditions \(F_P\) has a specific energy value. The energy values of such packages have a statistical distribution around the most frequent energy value. The dipole energies of the most cell-stimulating substances are in the range of \(10^{19}\) J. The energy of an electron in \(V_P\) is \(0.19 \times 10^{19}\) J.

It is conventionally believed that Nys binds to cholesterol or ergosterol, with the affinity to the latter being stronger. At present, from this belief a "specific" antimycotic effect is derived. This is a classical example of a reductive-deterministic, and, i.e., mechanistic explanation. For one thing, any biological membrane consists of structural lipids, phospholipids. If the phospholipids are present in an ionic solution, they organize themselves immediately to a bilayer due to their amphipathic properties. The same holds true for cholesterol or related chemical substances such as the steroid hormones but also for the ergosterol of fungal membranes. This phenomenon can also be observed for all membrane proteins. As soon as they are in contact with the lipid bilayer, they organize themselves by forming α-helices and loops. All constituents of biological membranes organize themselves. Nys also organizes itself in the membrane. Due to its amphipathic properties it exhibits a pronounced affinity to biological membranes and binds spontaneously. All these forms of self-organization are energetically controlled. In biological membranes the cholesterol is present in a molar ratio of about 1:1 to the phospholipids. However, its concentration may vary widely—particularly in intracellular membranes. Due to the narrow spatial conditions in the membrane Nys necessarily has to enter into contact with cholesterol. But it just as well enters into contact with all other membrane lipids and protein structures. It is a fundamental error to explain the effect of Nys with a single interaction with a single element of the membrane (see de Kruiff's model in Polyene antibiotic-sterol interactions in membranes of Acholeplasia laidlawii cells and lecithin liposomes, Biochimica and Biophysica Acta 339, 1974, 57–70). Hence, the explanation given so far for the antimycotic effect of Nys is not correct. Due to its specific molecular structure Nys can be considered a universal non-proteinaceous ion channel. Such an interpretation explains its ubiquitous ionophoric properties (see item 2 below). In this capacity it predominantly influences the plasma potential of the cells and thereby enters a global energetic interaction with all membrane and cell constituents. Furthermore, direct bindings to cholesterol may occur.

The energetic interaction between Nys and cholesterol is paramount for its therapeutical effects, such as in arteriosclerosis. Cholesterol is essential for the energy conversion on biological membranes. Its function could be defined anew within the sense of the BP. According to the dipole model, the cholesterol molecule has almost no dipole character. Its dielectric properties distinguish it as strong biological insulator. Cholesterol therefore determines the insulating properties of the biological membranes and not only their fluid character. In view of the extremely high \(F_E\) of \(10^7\) Vm\(^{-1}\) the biological membranes have to be strong insulators. On the basis of this concept the effect of all steroid hormones, such as the sex hormones and the glucocorticoids can be explained. They are built from cholesterol and have a stronger dipole character than cholesterol—the aliphatic moiety of cholesterol is replaced by polar groups. As soon as polar cholesterol derivatives are mixed in small amounts with cholesterol in the membrane their conductivity is slightly increased. In physiological concentrations the sex hormones therefore have a cell stimulating effect and promote cell growth—e.g., in the fetal development and during puberty. If, however, they are administered in considerably higher concentrations, such as during therapy with glycocorticoids, they excessively increase the conductivity of the biological membranes. The original level of the membrane potentials \(V\) and thus of the stored electrical energy \(E\) cannot be maintained due to the unfavorable dielectric properties of
the membranes--V and $E_A$ are inversely proportional to the dielectric constant $\varepsilon$ of the membrane. Consequently, the energy turnover of the cell is reduced. In higher concentrations all glucocorticoids therefore have cell-inhibiting and above all immune-inhibiting effects. This gives an energetical explanation of the etiology of arteriosclerosis/atheromathosis (see below).

2. Pharmacology and Kinetics of Nys within the Meaning of the BP

On the basis of the Nys structure all known pharmacological effects of this polyene macrolide and all others used according to the invention can be explained logically and coherently within the meaning of the BP. On top of that, all new therapeutical effects that are the subject matter of the present invention can be substantiated. First the known ones: Nys is, as already mentioned above, ionophoric. It is therefore widely used in the patch-clamp technique. Nys is said to increase the membrane permeability to ions. In biological membranes an increased permeability to Na$^+$ and K$^+$ ions can be mainly observed. In cell cultures there is a Na$^+$ inflow and a K$^+$ outflow along the ion gradients. Nys leads to a depolarization in all cell types examined so far. Furthermore, it modifies the membrane permeability to other ions. Nys increases, e.g., the intracellular concentration of Ca$^{2+}$ (Wiegand et al, Nystatin stimulates prostaglandin E synthesis and formation of diacylglycerol in human monocytes, Agents and Action, vol. 24, 3/4, 1988). In this case this is a consequence of the global stimulation of the cells by Nys. Any cell stimulation is accompanied by an increase in intracellular concentration of calcium. Therefore, calcium is incorrectly referred to as "second messenger". All intracellular systems examined so far that are stimulated by way of a cell activation and can be considered indicators of such cell stimulation--and not as "second messengers"--are activated by Nys. Nys, just like Amp, stimulates the production of the prostaglandins, the phosphoinositol cascade (Wiegand et al.), the adenylatcyclase cascade (Dipple I & MD Housley, Amphotericin B has very different effect on the glucagon- and fluoride-stimulated adenylat-cyclase activities of rat live plasma membranes, FEBS Letters, 106, 1979, 21-24), DNA- and RNA synthesis and the substrate transport (Kitagawa, T. & Andoh, T. Stimulation by Amphotericin B or uridine transport, RNA synthesis and DNA synthesis in density-inhibited fibroblasts, Experimental cell research, 115, 1978, 37-46), etc. The ubiquitous cell-stimulating properties of Nys and other polyenes can be observed in all cell types--eucaryotes, bacteria and fungi. Nys stimulates all lymphocytes, the killer activity of the T cells, the macrophages, the polymorphonuclear neutrophils (PMN) the oxidative burst of the macrophages, etc. This universal cell-stimulating property of Nys could only be explained by way of the BP. Nys acts like a universal, biological ion channel and leads to cell stimulation by depolarization. This effect can be observed in all eucaryotes. The energetic mechanism of a cell stimulation by Nys is based on the universal equation of the BP: $E=E_A.f$ (see above). As in the physiological membrane proteins that are responsible for the ion transport across the membrane, for Nys, too, an open and a closed state could be observed for its ionophoric activity (Ermischkin L. N., et al., Single ionic channels induced in lipid bilayers by polyene antibiotics amphotericin B and nystatine, Nature 262, 1976, 698-9). From this it can be concluded that in Nys, too, the same quantum effects which are responsible for the formation of solitons bring about specific configurational changes of the Nys structure and thus dynamically control the opening of the Nys channels.

Nys stimulates most cells in concentrations between 5 and 50 $\mu$g/ml without triggering cell lysis. In very high concentrations Nys results in cell lysis (=apoptosis) due to an excess depolarization and dissipation of the LRC. Cell lysis, however, is observed only at very high concentrations of more than 100 $\mu$g/ml. In this case the incubation must be 24 hrs or more. Cell lysis increases with incubation
time. During a short action time, however, cell lysis rarely occurs. The cells quickly recover after the excess stimulation. Resting cells are less sensitive to an excess depolarization by Nys than cells that have undergone an a priori maximum stimulation. This phenomenon, too, can be easily explained from an energetic point of view. Immune cells are resting cells and require maximum stimulation until they are activated. In the acute immune reaction, e.g., the concentrations of the cell-stimulating humoral factors such as the lymphokines in the body go up to the 10,000-fold. In contrast, the tubular cells of the kidneys are maximally stimulated under normal conditions. The i.v. administration of Nys, Amp or any other polyene macrolide leads very quickly to kidney toxicity even at very low concentrations. Therefore, Nys is not admitted i.v. This is in clear contrast to the excellent tolerance of Nys upon oral administration of very high doses (up to 5 g daily) over a prolonged period of time which was surprising found according to the invention.

This discrepancy in the safety profile between the oral and the systemic administration lead to the wrong conclusion that oral Nys is not resorbed. As evidence for this conclusion the low concentrations of nystatin in the serum after oral administration are mentioned. This conclusion is not admissible. Nys is a lipophilic substance and has a very high affinity to both cholesterol and its derivatives such as bile acid with which it forms micellae, and to biological membranes. In the presence of lipid membranes in ionic solution the entire Nys is membrane-bound. In the body the resorbed Nys immediately binds to the cell membranes—it is in the so-called deep compartments—and does not occur in the plasma which corresponds to an ionic solution. After i.v. administration of Nys the substance vanishes from the plasma within only few minutes and distributes in the deep compartments. The entire Nys in the plasma is bound to lipoproteins. However, the Nys concentration on the membranes of the blood cells has not been measured.

The kinetics of the polyene macrolides has been examined very insufficiently. There are only results for Amp available and the corresponding data are highly insufficient. The knowledge about the kinetics of i.v. Amp are based on the data of only 2 patients (A. J. Atkinson & J. E. Bennet, in Antimicrob. Agents & Chemoth. (1978), Vol. 13, p. 271-276). This invention, in contrast, is based, inter alia, on the surprising finding that polyenes, such as Nys and Amp upon oral and intranasal administration are substantially resorbed and evoke systemic therapeutical effects.

It was found according to the invention that oral Nys and Amp are resorbed by the gastrointestinal tract almost completely and are mainly stored in the liver but also in other mesenchymal and immunological organs. Due to its lipophilic properties oral Nys obviously bind to bile acid and is transported to the liver and other mesenchymal organs by the chylomicrons. This kinetic behavior is typical of most lipophilic substances (Koch & Ritschel, Synopsis der Biopharmazie und Pharmakokinetik, Ecomed, 1986). If the presently held belief were true that oral Nys is not or only insufficiently resorbed, the major part of Nys would have to be excreted together with the feces, for there is no indication so far that Nys is degraded in the gastrointestinal tract. However, we succeeded in finding that the share of Nys excreted daily with the feces is less than 1% of the orally taken daily dose. Since a degradation in the intestinal tract therefore has to be excluded, the only possibility remaining is that Nys is substantially resorbed by the intestinal tract.

The resorption of Nys and Amp could first be detected via their therapeutical effects. Since the bioavailability of lipophilic drugs cannot be ascertained via the serum concentration it is recommended that the systemic pharmacodynamic effects be ascertained, e.g. by the challenge and
The dechallenge method. Another possibility is the ascertainment of the strength of the effects in relation to the dose administered (dose-effect relation). A variety of challenge-dechallenge tests and tests with increasing doses in patients suffering from different diseases which respond to Nys and/or Amp was carried out. The average daily dose of Nys was 1 to 1.5 g in the challenge-dechallenge tests. In the tests with increasing doses a daily dose of from 250 mg and 2 g was used. Nys and Amp were administered orally as powders, in gelatin capsules a 250 mg pure substance. The results can be summarized as follows:

a) In menopausal women suffering from CFS (chronic fatigue syndrome) a dose-dependent increase in vaginal discharge after 1 to 2 weeks' treatment with 1 to 1.5 g Nys was observed. After dechallenge, the discharge ceased after 1 to 2 days and during challenge started again. An increase in discharge was observed only at a dose of 500 mg Nys or Amp.

b) Increase in bile secretion after 1 week treatment with Nys. Disappearance of sonographically detected gallstones in 4 patients after 2 months' therapy with 1.2 g Nys and in one patient with 1.5 g Amp after 3 months' therapy.

c) Increase in prostaglandin biosynthesis after 1 to 2 weeks' therapy with 1 to 1.5 g Nys or Amp.

d) Dose-dependent decrease in total cholesterol plasma levels in patients suffering from atherosclerosis after 2 to 8 weeks. The cholesterol-reducing effect occurs only at a daily dose of 500 mg and leads to a sustained decrease of cholesterol levels at a daily dose of 1 to 1.5 g Nys or Amp. Increase in cholesterol levels after dechallenge.

e) Remission of symptoms, such as allergic rhinitis, asthmatic attacks and food intolerances, in therapy-resistant allergies, such as allergies to dust mites, food allergies, etc., after 6 to 8 weeks' therapy with a daily dose of 1 g Nys or Amp.

f) Decrease of prostatic hypertrophy in old male patients after 3 to 6 months' therapy with 1 to 1.5 g Nys or Amp. Increase in hypertrophy about 3 months after termination of the therapy.

g) Improvement of the skin turgor after oral administration of Nys or Amp. Deterioration after dechallenge.

h) Lowering the mortality and infection morbidity (by stimulating the immune response) in intensive care patients suffering from severe multiple traumata after administration of 3 g Amp per day as oral paste, starting 24 hours after the traumata were inflicted.

i) Increase in body weight (5 to 10 kg) in final stage cachectic tumor patients after 2 to 4 weeks' therapy with 1.5 to 2 g Nys or Amp. Decrease in weight after discontinuation of the therapy by external physicians. Renewed increase when the therapy was resumed.

j) Dose-dependent remission (beginning at a daily dose of 1 g Nys or Amp) of the symptoms of CFS after 4 to 8 weeks. No dechallenge made.

Furthermore, kinetic tests were made with Nys. Four patients who had undergone cholecystectomy received one week prior to the operation and one week after the operation daily 1 g Nys orally. During the operation tissue samples from the bile walls, bile and liver were taken. During the first few postoperative days gall samples were taken from the T tube drainage. The tissue concentrations
of Nys were from 80 and 180 µg/g in the liver, from 50 and 120 µg/g in the bile wall and from 30 and 150 µg/ml in the bile. These preliminary results agree with results from other kinetic studies, which, however, were carried out with i.v. Amp (Collette N. et al., in Antimicrob. Agents and Chemother., 33, 1989, 362-68, R. M. Lawrence et al., in J. Antimicrob. Chemother., 6, 1980, 241-49). These studies show that i.v. Amp is predominantly stored in the liver, bile and spleen. It can be assumed that the lipophilic polyenes after resorption from the gastrointestinal tract are mainly distributed in vital secondary immunological organs.

VII. Novel Clinical Indications for Polyene Macrolides

1. Virus Infections

Every virus particle enters the host cell by fusing with the host’s cell membrane and by then releasing its particle content into the cell. In this process, viral membrane proteins remain in the host cell membrane and can be detected as markers. On the basis of the deliberations following from the BP, such virus proteins serve to control the viral genome in the DNA of the cell by the electromagnetic, delocalized coupling of the LRC and to thereby allow viral replication. This energetic coupling has not been recognized so far by genetics. If expression of such viral proteins in the host cell is suppressed, for instance by endocytosis, no viral replication takes place. Replication of the virus requires optimum conditions. In most cases, the viral DNA or RNA is degraded by repair mechanisms and the virus is metabolized in the cell. Only in one in about 10,000 cells which are infected by the HIV virus occurs replication. The effectivity of the repair mechanisms increases with the stimulation of the cells. The efficiency of all cell reactions including the repair mechanisms is increased by an enhanced depolarization of the cells. An increase in depolarization of virus-infected cells, however, also results in an increase in endocytosis of viral membrane proteins. Since they no longer emerge on the surface of the cells, no specific electromagnetic coupling to the proviral DNA takes place. This is a short explanation of the bioenergetic mechanism of virus replication in human cells. Two important conclusions can be drawn: a) during depolarization the viral membrane proteins disappear from the cell surface of infected cells and can no longer be measured as markers; during repolarization they are increasingly expressed; b) depolarizing substances have an antiviral effect because they promote endocytosis of the viral membrane proteins as well as the repair mechanisms in the infected cells and thus inhibit virus replication. These effects can be confirmed for Nys and other polyenes for all viruses examined so far.

a) AIDS

There is a detailed scientific report by Dr. med. G. Stankov on the AIDS etiology and its therapy with Nys (April 1995, Copyright DIAS Institut). It considers the most recent data of AIDS research until April 1995. The essential aspects of the development of AIDS and therapy of AIDS with Nys are briefly summarized in the following section:

The HIV membrane protein gp41-gp120 exhibits structural homology to MHC class II proteins which plays an important role in the MHC-restricted T-cell stimulation in the thymus. The interaction between the MHC molecule of the antigen-presenting cells (APC) and the T-cell receptor as well as other receptors of the CD type proceeds via their soliton triplets and leads both to direct depolarization as well as to the release of lymphokines to stimulate the immune cells involved. The HIV protein imitates the MHC molecule and uses the body’s own immunostimulating mechanisms to
control virus replication by electromagnetic coupling. CD4 is depleted because this T-cell subpopulation plays an important role in the MHC-restricted T-cell stimulation in the thymus and in the lymph nodes. Expression of gp41-gp120 results in an increase in apoptosis of the CD4 cells and is paramount for virus pathogenicity. gp41 corresponds to the transmembrane part of MHC, gp120 to the variable extracellular domain. gp120 changes under the bioenergetic constraint of the immune response. It quickly mutates when subjected to a virostatic (cell inhibiting) therapy and forms a one-step resistance. This is why it is impossible to develop a respective vaccine. Virus replication takes place during the entire duration of the disease. The AIDS-related complex represents only the last manifestation of the disease decompensation. At present, the patients are treated with AZT (ziduvodine) only in the last stage of the disease. According to the dipole model, AZT is a cell-inhibiting substance and increases the mortality of AIDS patients compared to placebo patients. The CONCORDE study, which examined the effect of ATZ in the early stage of HIV patients, confirms this conclusion within the meaning of the BP (Lancet, Vol. 343, 1994, 871-881). Presently, there is no effective therapy of AIDS.

A treatment with Nys and other polyene macrolides according to the model of the invention must inhibit both the expression of gp41-gp120 and the HIV replication in the infected cells. Nys and Amp inhibit in vitro the expression of gp120 and gp41 and p24 in H9 lymphocytes and suppress reverse transcriptase (Selvam M. P. et al. in AIDS Res. & Human Retrovirus, Vol. 9, 1993, 476-481). This in vitro study, however, does not indicate a possible HIV inhibition in vivo. Also, this study does not give any recommendation for a therapy with Nys or other polyenes in HIV patients since it was possible to explain the AIDS pathogenesis only when discovering the BP. Particularly, there is no indication in the art to chronically administer oral nystatin, in the high doses recommended by the invention, already after seroconversion to strengthen the immune system of HIV patients, since it was assumed that polyenes are not resorbed and since the cell-stimulating properties of Nys were not known. It is not known from the literature nor from practice that HIV patients were subjected to a therapy with Nys or another polyene in the early stage of the disease in order to suppress HIV replication by immunostimulation and to thereby prevent outbreak of the disease.

On the basis of the findings of the invention, HIV patients should be treated with polyene macrolides, such as Nys or Amp, preferably immediately after seroconversion. The therapy must be chronic for the duration of the disease. The recommended daily oral dose is about 0.5 to 5 g, preferably about 1 g to 3 g, particularly 1.5 to 2 g polyene macrolide as required.

The antiviral effect of polyene macrolides was confirmed by all viruses which have been examined so far in vitro. In the following, the observations made according to the invention are summarized.

b) Herpes Simplex Virus (HSV) Infections

Polyene macrolides, such as Nys and Amp, inhibit HSV I and HSV II in cell cultures at concentrations between 3 and 25 μg/ml. According to the invention, a rapid improvement of labial HSV infections after topical application of ointments (40 to 200 mg, preferably about 50 to 100 mg polyene macrolide/g ointment) is achieved. The drug must be administered several times. Depending on the concentrations of the active ingredient the ointment can be applied 3 to 10, preferably 5 to 8 times a day. A preferably chronic administration of oral macrolide may prevent recurrence of Herpes. The daily dose for chronic oral administration is in the range from about 0.5 to 5 g, preferably from about 1 to 3 g, preferably from 1 to 1.5 g.
c) Herpes Zoster Varicella (HZV) Infection

Polyene macrolides, such as Nys and Amp, inhibit HZV in cell cultures in concentrations between 3 and 25 μg/ml. After topical administration of highly-dosed ointments (about 40 to 200 mg Nys/Amp, preferably about 50 to 100 mg/g ointment) the efflorescences of shingles remit more rapidly than without therapy. The ointment may be applied several times a day, e.g., 3 to 6 times a day.

d) Hepatitis B Virus (HBV)

Polyene macrolides, such as Nys and Amp, inhibit dose-dependently the production of hepatitis B surface antigen (HbsAg) in human hepatoma cell line PLC/PRF/5.

After chronical oral administration of about 0.5 to 3 g, preferably about 1 to 1.5 g active ingredient daily, a remission of the symptoms and an improvement of the liver function can be achieved.

e) Vesicular Stomatitis (VS) Influenza and Reuscher Leukemia Virions

Polyene macrolides, such as Nys, Amp and filipin inactivate these virions in vitro.

f) Other Virus Infections: Nys inhibits in vitro Sindbis virus and vaccinia virus.

g) Recurrent Aphthous Stomatitis (RAS)

The pathogenesis of RAS is not clear, but there is substantial evidence that it can be triggered by various endogenous viruses. So far there has been no successful therapy for RAS. According to the invention, patients with RAS can be topically treated several times a day with a mucosa-adhering ointment that contains Nys, Amp or any other macrolide. For example, the treatment can be carried out in intervals of 1 to 3 hours 2 to 8 times a day. The content of the active ingredient of the ointment is in the range of from 20 to 200 mg, preferably of from 20 to 50 mg/g ointment. RAS remits after 24 to 48 hours (without therapy 5 to 7 days). The pain was relieved quickly after application of Nys.

h) On the basis of the above-mentioned data and the theoretical conclusions drawn from the BP it has to be assumed that polyene macrolides (topical and oral) are therapeutically effective also in the following virus infections, without, however, being limited to them:

Infections with picornaviridae (e.g., poliovirus), caliciviridae (e.g., Norwalk virus), togaviridae (e.g., Rubella virus), flaviviridae (e.g., yellow fever virus), coronaviridae, rhabdoviridae (rabies virus), filoviridae (Marburg virus), paramyxoviridae (German measles virus), orthomyxoviridae, bunyaviridae (e.g., California encephalitis virus), arenaviridae (lymphocytic choriomeningitis virus), reoviridae (e.g., rotavirus), retroviridae (e.g., HIV-1), hepadnaviridae (e.g., hepatitis A and hepatitis B virus), paroviridae (e.g, human parovirus B-19), papovaviridae (e.g., JG virus), adenoviridae (e.g., human adenovirus), herpesviridae, poxviridae, Epstein-Barr virus (e.g., infectious mononucleosis), cytomegalovirus.

2. Diseases with insufficient immune reaction, caused by a hereditary or acquired energetic impairment of the function of the MHC class I and II molecules and/or other integral proteins.

On the basis of the most recent experimental results it can be shown within the meaning of the BP that almost all chronic diseases are accompanied by an impaired immune system (see also diseases
with immunopathogenesis, Harrison, Laws of Internal Medicine, McGraw Hill, 1992). Usually, these impairments are, as is evident from the deliberations according to the invention, impairments of the energy transmission of immunospecific membrane proteins. One either finds acquired mutations which relate to soliton-specific amino acids or hereditary variants, such as HLA alleles which occur significantly more frequently in certain diseases. Many diseases are associated with an impaired function of the MHC class I and II molecules. The defects frequently relate to the soliton-triplets of the peptide binding site for self-peptides or allo-peptides (HLA association, HLA=Human Leucocyte Association Antigen). This leads to an insufficient presentation of the antigens in the thymus and other secondary immune organs and to an insufficient cell-mediated (T cells) and humoral (B cells) immune response. The MHC-T-cell receptor interaction is energetically impaired and stimulation of the T-cells and B-cells (APC) is insufficient. The entire immune system is insufficient in terms of the energetic law since all immunospecific cell interactions are coupled to one another and are controlled via depolarization (e.g., growth, antibody production, phagocytosis, apoptosis, etc.) or repolarization (e.g., differentiation, chemotaxis, etc.). A detailed description of these complex immunological processes within the meaning of the BP must be omitted here. Individual examples of an HLA association are given below. Rheumatoid arthritis (RA) and multiple sclerosis (MS) are typical examples of chronic diseases with immunopathogenesis which can be put down to an impaired function of MHC class I and II molecules and of other integral proteins.

a) Rheumatoid Arthritis (RA)

A restricted set of genetically determined MHC class II molecules strongly predisposes to the development of RA. Increased risk for RA is associated with HLA-DR and especially with mutations of soliton-specific amino acids (Glu and Lys at position 70/71 on the HLA-DRβ1 chain). Collagen arthritis and myelin-protein-induced experimental allergic encephalomyelitis show a close association with certain MHC class II molecules. The structure of MHC class II proteins has been recently elucidated (Nature, Vol. 368, 1994, 215-220). All 15 hydrogen bonds of the peptide binding site involve soliton-specific amino acids. Mutations of these amino acids which occur quite often in RA (HLA-DR association) lead to a deficient binding of peptides of the body's type II collagen which is incessantly built up and degraded in the bones. The collagen type II peptides are insufficiently presented to the T cells so that the self-tolerance to this collagen cannot be adequately developed. MHC class II molecules are essential for the MHC-restricted T-cell stimulation which is, as already pointed out above, mediated by depolarization. In RA, the activation of the T cells is insufficient leading to a cell-mediated autoreactivity to collagen. Hence, the RA pathogenesis is based on an energetic cause.

In RA, CD4 cells are found in large quantities at the site of inflammation. This is a frequent finding in many cell-mediated autoimmune diseases and clear evidence of the reduced efficiency of the T cell caused by insufficient stimulation. In contrast to the widely held belief in medicine the development of an autoreactivity is not an enhancement of the immune response which should be suppressed by cell-inhibiting drugs such as immunosuppressants but an energetically caused insufficiency of the involved immune cells which can be remedied with cell-stimulating drugs. Rheumatoid factors, mainly autoantibodies of the IgM isotype, are frequently found in RA patients. Ig production of synovial B cells exhibits an anti-type II-collagen activity. This clearly shows that the peptides of this collagen cannot be adequately bound and presented by the MHC class II molecules of the APC cells, particularly the B lymphocytes. This leads to the formation of insufficient T and B cells. Polyene macrolides, such as Nys and Amp, stimulate both the B and T cells by depolarization. Patients
suffering from chronic RA which are treated according to the invention with, e.g., 1 g oral Nys/Amp daily, experience a clear and sustained improvement of the symptoms after about 1.5 to 3 months. Therapy is preferably chronic. The recommended dosage of the macrolide therefore amounts to about 0.5 to 4 g, preferably about 1 to 2 g/day, depending on the condition and progression of RA.

b) Multiple Sclerosis (MS)

MS is characterized by demyelination of the CNS, resulting from an inflammatory process. MS has an intermittent course. As in RA, CD4 cells and B lymphocytes can be found at the site of inflammation. Specific MHC class II alleles (Drw15 and Dqw6) are associated with an increased risk of MS and confirm the same finding as in RA: An insufficient binding of the body's peptides in certain HLA alleles leads to an impaired immune response. The result is also an autoreactivity to CNS proteins because self-tolerance cannot be adequately developed. Polyene macrolides, such as Nys and Amp (about 0.5 to 4 g, preferably about 1 to 1.5 g orally per day) clearly improve the MS symptoms in the patients and can even result in the disease being cured when administered in an early stage. A chronic therapy is preferred.

c) The two diseases, RA and MS, are representative of a group of diseases associated with an impaired function of MHC class I and class II molecules (HLA association) or other integral proteins. They exhibit the same energetically caused pathogenesis even if the symptoms may differ and may be found at different anatomical locations. All these diseases can be successfully treated by preferably chronic administration of oral Nys, Amp or another polyene macrolide. The daily dosage ranges from about 0.5 to 5 g, preferably about 1 to 2 g. For most diseases an HLA association is known. They are sub-classified anatomically (as far as possible, the corresponding HLA allele is indicated in brackets):

Bone diseases: ankylosing spondylitis (B27), Reiter's syndrome (B27), reactive arthritis in yersinia, salmonella or gonococcus (B27), psoriatic arthritis (B27, Bw38), juvenile rheumatoid arthritis (B27, Drw8), rheumatoid arthritis (Dw4, DR4), osteoarthritis.

Gastrointestinal diseases: gluten-sensitive enteropathy (DR3), other food-sensitive enteropathies, chronic active hepatitis (DR3), ulcerative colitis (B5, CD44), acute anterior uveitis (B27), Crohn's disease (CD44), liver cirrhosis.

Skin diseases: dermatitis herpetiformis (Dw3), psoriasis vulgaris (Cw6), pemphigus vulgaris (DR4, A10), Lichen ruber, Behcet's disease (B4), systemic lupus erythematosus (DR3), endogenous eczema and other forms of atopy (e.g., neurodermatitis), all forms of allergy, Sjogren syndrome (Dw3), dermatomyositis, scleroderma, chronic vasculitis (e.g., Raynaud's syndrome), diseases of the hair root.

Hematological diseases: idiopathic hemochromatosis (A3, B14), cold agglutinin disease, cryoglobulinemia, porphyria.

Endocrine diseases: type I diabetes mellitus (DR4, DR3, DR2, BfF1), hyperthyroidism (B8, Dw3), hyperthyroidism in Japanese (Bw35), adrenal insufficiency (Dw3), subacute thyroiditis de Quervain (Bw35), Hashimoto's thyroiditis (DR5), congenital adrenal hyperplasia (Bw47), distress syndrome, chronic fatigue syndrome, postmenopausal syndrome, primary and secondary amyloidosis, gout, cystic fibrosis.
Neurological diseases: myasthenia gravis (B8, DR3), multiple sclerosis (DR2), manic-depressive disorder (Bw16), schizophrenia (A28), polynuromyelitis (Guillain-Barre syndrome), polymyositis, slow-virus diseases of the CNS, other systemic diseases of the CNS (e.g., amyotrophic lateral sclerosis), Alzheimer's diseases.

Renal diseases: idiopathic membranous glomerulonephritis (DR3), Goodpasture's syndrome (anti-GBM, DR2), minimal change disease (B12), polycystic kidney disease (B5), IgA nephropathy (DR4).

Immunological infections: leprosy (B8), paralytic poliomyelitis (Bw16), IgA insufficiency (DR3), sarcoidosis.

3. Diseases with External and Internal Causes which are Associated with a Transitory or Chronic Immunoinsufficiency

a) Sepsis:

In order to reduce the risk of sepsis, polyene macrolides, such as Nys or Amp, may be administered parenterally for selective decontamination of the digestive tract in intensive care patients after severe traumata. Tests have shown that Amp serves to reduce the mortality in these patients. The daily dosage is about 1 to 5 g, preferably about 2 to 4 g, particularly preferred about 3 g. The duration of therapy is several weeks, e.g., for the duration of the intensive care unit stay.

b) Immunodeficiency following Chronic Antibiotic Treatment:

Polyene macrolides, such as Nys or Amp, can be administered orally according to the invention to patients suffering from immunodeficiency and recurring infections caused by chronic administration of antibiotics. The infection rate can be reduced after a 6 week's to 3 month's therapy. The duration of the therapy is at least 6 months, at a dose of about 0.5 to 4 g, preferably about 1 to 1.5 g Nys or Amp/day.

c) Chronic Fatigue Syndrome (CFS):

The diagnosis of CFS was made after the CDC classification and is a frequent disease which is caused by external causes such as infections and administration of antibiotics as well as by internal causes (stress, hormone insufficiency, immunoinsufficiency). CFS is associated with a chronic immunodeficiency. The mutual influence of psychic, humoral and endocrinous factors such as can regularly be observed in CFS, is only explained by BP. All physiological factors have the same effector level: they regulate the cells by changing their membrane potentials. CFS is a classical indication of a therapy with polyene macrolides such as Nys or Amp (see also experimental part). According to the invention, CFS is treated with a daily dose of about 0.5 to 4 g, preferably about 1 to 2 g. The duration of treatment can be 3 to 6 months or longer.

d) Other diseases: any form of acquired or hereditary allergies that are associated with disorders of the respiratory system (e.g., rhinitis, asthma) or of the gastrointestinal tract (e.g., food-sensitive enteropathies).

4. Cancerous Diseases (Neoplasias of any Kind)
Cancer has four characteristic properties: a) it develops from single transformed cells (clonality); b) cell stimulation, growth and proliferation of the tumor cells elude the physiological supracellular regulation and are controlled by autostimulation (autonomy); c) there is no normal cell differentiation (anaplasia); d) the tumor cells can leave the cell aggregate and disseminate (metastasis).

All these phenomena can be energetically explained according to the invention. The mechanisms of quantum mechanics will be briefly explained. The transformation into carcinogenic cells is always a product of insufficient cell stimulation. Environmental toxins, such as nicotine, tar substances or asbestos (inhibition of the energy conversion along the membranes due to the dipole character of these carcinogens), chronic therapy with cell-inhibiting substances (e.g., with cyclosporin or other immunosuppressants) or radiation (direct energetic impact on the DNA and inhibition of the DNA solitons) result in a higher entropy in the cells. In all these cases the conversion of the LRC during an action potential is inhibited. An increased entropy leads to an increase in the degrees of freedom of the molecules in the cells (dS=R.lnZ₁ : Z₂). This holds also true for the transcription of the biological structures involved (DNA, RNA, DNA-controlling proteins, etc.). The probability increases that incorrect nucleotides are inserted and that mutations occur. Some of these mutations prove energetically advantageous for the cells. In many cancer cells a mutation-caused over-production of growth factors or receptors of growth factors, leading to an excess depolarization of the cell, can be observed. The result is an autostimulation of the cancer cells. In this manner the cancer cells become independent of supracellular regulation. The transformation into a cancer cell is always the result of insufficient cell stimulation and represents an evolutionary process (=adaptive process) of the cell at the expense of the self-organization of the organism. The over-production of growth factors or the transformation of protooncogenes into oncogenes is often the result of acquired mutations in the cancer cells, although genetically-inherited mutations can also contribute to the transformation. Usually, more than a single energetically caused modification (transformation threshold) is required for the cell to mutate. Cancer cells are, in contrast to the normal cells, immortal in vitro since their stimulation is autosufficient. The one-sided stimulation of the cancer cells results in a reduced differentiation and an increased endocytosis of integral proteins. Important membrane proteins, particularly those that are responsible for the formation of "tight junctions", do not emerge on the cell surface. The cancer cells lose their capability of adhering to neighboring cells and increasingly leave the cell aggregate. Due to the excess autostimulation they increasingly proliferate. However, the growth rate of cancer cells is often highly overestimated. Most cancer cells do not grow substantially faster than many normal body cells, they just grow in a more uncoordinated manner, beyond physiological supracellular regulation.

There are two possibilities of destroying cancer cells: by inhibiting the energy exchange on the plasma membrane or by apoptosis.

The first-mentioned mechanism is the only one presently used in cancer therapy. According to the dipole model, all cytostatics are potent cell-inhibiting substances which unspecifically inhibit both the healthy and the cancer cells. Since immune cells proliferate rapidly, the cytostatics substantially inhibit the immune system. The immune system is the strongest natural defense of the organism against the cancer cells. It controls the entire cell transformation and destroys above all the newly produced cancer cells. Since cancer cells are developed in our body with a certain degree of
probability, an intact immune defense is an indispensable prerequisite for the prevention of cancer. Cytostatic therapy deliberately accepts a suppression of the immune defense.

The second, physiological mechanism used by the immune cells to destroy cancer cells is apoptosis. Immune cells can often be found in the vicinity of tumor cells. They increasingly produce lymphokines and other depolarizing or repolarizing substances and trigger apoptosis of the cancer cells by an excess de- or repolarization. They use for this purpose cell-bound proteins with which they enter into contact with the cancer cells (e.g., killer cells). Since the cancer cells are a priori more stimulated than the normal cells, the result is a selective apoptosis of the tumor cells.

The anticarcinogenic effect of the polyene macrolides according to the invention can thus be based on the following effects which may occur simultaneously:

The immune cells are stimulated by the macrolide. They increasingly destroy the cancer cells by apoptosis.

The macrolide directly stimulates the cancer cells and increasingly brings about their apoptosis by depolarization. This effect is particularly pronounced in liver metastases since the macrolide is stored in the liver in high concentrations. The macrolide results in a remission of the liver metastases. Presently, there is no drug available for the treatment of liver metastases.

The macrolide improves the supracellular regulation of healthy cells which are about to be transformed and thus prevents their transformation into cancer cells.

The macrolide stimulates all body cells and thereby strengthens the unspecific defense of the organism.

The treatment of cancer with the macrolides according to the invention is a fundamentally new approach which discards the conventional cytostatic therapy for being inappropriate and which is based on natural defense mechanisms. They result in the remission of metastases and clearly improve quality of life in patients suffering from solid tumors. There is evidence that life expectancy is substantially prolonged. They cannot cause large solid tumors to disappear completely. Therefore, a combination from surgical removal and medicamentous treatment with macrolides appears to be more promising than a purely medicamentous treatment. However, depending on the case, the operative risk has to be evaluated, too. The polyene macrolides will be paramount in cancer prophylactics. The sooner therapy with macrolides is started, the better the prognosis. Cancer therapy with macrolides, such as Nys or Amp, is preferably chronic. The daily dose is about 0.5 to 5 g, preferably about 1 to 3 g, depending on the patient’s condition and the stage of the disease.

5. Wound Healing, Diseases with Impaired Wound Healing

The therapeutical effect of polyene macrolides, such as Nys and Amp, can be observed particularly well in the enhanced wound healing (predominantly after topical application). A rapid cell growth is caused by depolarization. In the same manner physiological healing is brought about. Any wound is surrounded by immune cells which energetically promote the growth of the damaged tissue cells by releasing stimulating lymphokines, eucosanoids and other humoral factors. Polyene macrolides, such as Nys and Amp, can be used according to the invention in high dosages (500,000 to 1,000,000 I.U./g basic ointment or, e.g., in 5 to 10% DMSO solution) several times a day, e.g., 3 to 8 times a day.
Following diseases can be topically treated according to the invention with polyene macrolides, such as Nys and Amp (cf. experimental part):

any kind of wounds

ulcus cruris
decubitus or other chronic-trophic disorders

burns. Burns must be immediately be treated with Nys. The effect can be observed within only few minutes. Nys prevents blisters. The administration of polyene macrolides, such as Nys, is particularly useful in the treatment of sun burn.

6. Diseases Caused by Disturbances of the Cholesterol Metabolism

a) Atherosclerosis (AS) and Hypercholesterinemia

The pathogenesis of AS has been unknown. There are several hypotheses and attempts at explanation which concentrate on individual known aspects of AS. The development of AS can only be correctly explained based on the teaching of the present invention. This explanation requires a correct understanding of the part cholesterol plays in the energy conversion along the biological membranes.

As already mentioned above, cholesterol is present in the membranes in a molar ratio of 1:1 to the other phospholipids which build the lipid bilayer. Its energetic function is to regulate the insulating properties of the membranes. The cholesterol molecule is almost apolar and is therefore a good insulator. Since the electric field strength of the membranes \( F_E = 10^7 \text{ V m}^{-1} \) is very high, the cell membranes must exhibit excellent insulating properties as biological capacitors in order to build such strong electric field strengths. Any modulation of the dielectric properties of the membranes results in a modification of the plasma potential \( V \) and thus of the stored electrical energy \( E_{el} \). As is known, the dielectric constant \( \varepsilon \) between the plates of a capacitor is inversely proportional to \( V \) and \( E_{el} \). The modification of \( \varepsilon \) in the membranes allows infinitely many possibilities for influencing the energy conversion in the cells by modifying \( V \) and \( E_{el} \). The biological regulation of the cell is based on this law. Cholesterol plays a decisive part in it. It is the universal biological regulator of \( \varepsilon \) in the membranes and thus also of the energy conversion in the cells. Therefore, its fundamental importance: on the one hand cholesterol is vital, on the other hand hypercholesterinemia results in several diseases and an increased mortality (the Janus molecule).

Cholesterol is exclusively produced by de-novo synthesis in the cells. Cholesterol is quickly metabolized in the lipid metabolism of the plasma membranes. The turnover rate of the cell membranes is extremely high. It is estimated that the lipid content of the membranes is regenerated completely within a few hours. Cell stimulation results in an increased membrane turnover. Any endocytosis of integral proteins is accompanied by, e.g., a consumption of lipid content (see, e.g., the inositol cascade). More than 95% of the cholesterol are located in the cell membranes. The cholesterol turnover takes place almost exclusively in the cell membranes and is proportional to the cell stimulation. The more cells are stimulated the more cholesterol they consume and the more cholesterol is newly produced in the mevalonic acid synthesis. This is because any cell stimulation leads to an adequate increase in all reactions in the cells via the LRC. The cell membranes can only
take up certain amounts of cholesterol because its concentration determines the dielectric properties of the lipid bilayer. The excess cholesterol circulates in plasma or is stored in certain cells, e.g., in the macrophages, the so-called foam cells in the atheroma.

Cholesterol is transported in the body as LDL and VLDL. VDL and VLDL are large spherical particles that carry cholesterol and phospholipids in a ratio of 1:1 and contain apolipoproteins (apoA and apoE). apoA and apoE are polymorphous glycoproteins which interact with the apo receptors of the cells via their soliton triplets. The uptake of LDL and VLDL in the cell is coupled to its action potential or to the discrete potential fluctuations and increases during stimulation with depolarizing substances. Via this active energy transport the lipids are taken up by the cell. All cells have apo receptors. Liver cells have particularly many apo receptors because the uptake of alimentary fats and the de novo synthesis of cholesterol in the organism mainly takes place in this organ. Cholesterol is a precursor of steroid synthesis in the ovary, in the adrenal gland, in the prostate and in other endocrine and sexual organs. This fact is essential for the understanding of the therapeutical effect of Nys, Amp and other macrolides in prostate hyperplasia, postmenopausal syndrome, cholelithiasis and other diseases associated with disorders of the cholesterol metabolism.

Based on these facts, the development of AS can be explained logically and coherently. For energetic reasons, the cholesterol concentration in the membranes is maintained in a very narrow range. In this range the dielectric properties of the membranes and the energy conversion is optimal. The cholesterol metabolism is a very dynamic process which depends on the organism’s whole metabolism. According to the universal equation of the BP the cholesterol metabolism directly depends on $E=E_A f$ (4). If the turnover is increased, more cholesterol is produced but there is no excess cholesterol in the body because it is increasingly consumed along the membranes. If the body’s turnover--for whatever reasons--decreases, there is either an excess of cholesterol in the body--all the more so if more calories are taken up via the food than are consumed. This excess cholesterol cannot be taken up by the membranes and the cholesterol-storing cells and increasingly circulates in plasma (LDL and VLDL increase). If the supply is higher than the demand, cholesterol is deposited in the intima of the vessels. Here it is removed from the macrophages, the foam cells, and other immune cells. When the capacity of the immune system is exhausted, cholesterol is deposited in the intima. Atheromas are formed which lead to histologic degenerations of the vessels which are known for AS. This is the visible part of the AS pathogenesis. The consequences are hypertonia, coronary heart disease (CHD), ischemia, renal insufficiency, etc. The differences in metabolism are great between the individuals--there are faster and slower metabolizers among the population. The metabolic turnover is age-dependent and decreases in old age. Hypercholesterinemia is a particular disease of old age. But also other interindividual factors such as immobility are the cause for a predisposition. A reduced body turnover can also be genetically predisposed and can become manifest already in childhood. Familial hypercholesterinemia (FH) is caused by hereditary mutations in the apoA and apoE receptors which, according to the most recent findings, relate to soliton-specific AS. LDL receptors in FH homozygotes do not bind sufficiently apoA and apoE. The lipid transport into the cell and thus energy conversion is reduced. Cholesterol accumulates in circulation. This excess causes AS in early childhood.

The therapy of AS can also be explained with the energetic law. Nys reduces the serum values of cholesterol in hypercholesterinemic patients via a global cell stimulation by depolarization. Particularly, the hepatocytes are stimulated: both the de-novo synthesis and the cholesterol
consumption increase. The effect results in an increase in the cell frequency \( f \) according to the energy
balance equation \( E = E_A f \). According to the dipole model, all anticholesterolemic known from the art
as MHG-CoA reductase inhibitors are cell-stimulating substances, such as Nys, however they have a
less pronounced dipole character than the latter. Nys, Amp and other polyene macrolides according
to the invention as well as the MHG-CoA reductase inhibitors result in an increase in MHG-CoA
reductase (Goodman & Gilman, The Pharmacological Basis of Therapeutics, 1991, page 883) via a
stimulation of the cells. At the same time, they enhance the cholesterol turnover in the cell
membranes and thereby reduce the share of the circulating cholesterol. The anticholesterolemic
effect can be observed with all lipid-lowering substances only after several weeks to months--the life-
sustaining effects, such as reduction of mortality and sever cardiovascular events, however, only
after 2 to 3 years (the Scandinavian Simvastatin Survival Trial, The Lancet, Vol. 344, Nov. 19, 1994,
1383-89). Patients suffering from hypercholesterinemia can be chronically treated according to the
invention with an oral dose of about 0.5 to 4 g, preferably about 1 to 1.5 g of a polyene macrolide,
such as Nys and Amp, per day. They show a substantial reduction of the overall cholesterol level after
1 to 2 months after start of the therapy. A lipid-reducing therapy therefore should preferably be
chronic.

b) Alzheimer's Disease (Alz)

The apoE4 allele is significantly associated with Alz. This apolipoprotein is substantially involved in
cholesterol metabolism. ApoE can be found in the plaques of dystrophic neuritis in Alz. apoE binds
tightly to the soluble and insoluble forms of the \( \beta \)-amylloids in Alz patients. The amino acid position
112 (Cys) of the apoE allele is occupied by Arg+, which is a mutation variant which, as predicted
theoretically, relates to a soliton-specific amino acid and causes an impaired energy conversion (The
chronic therapy with macrolides, such as Nys, improves the Alzheimer symptoms.

c) Prostatic Hyperplasia

The prostate is an important organ for the synthesis of sex hormones, with cholesterol functioning as
precursor. A prostatic hyperplasia in old age reflects a reduced function of the organ. An organ
hyperplasia typically develops compensatory if cell capacity of the organ is reduced (e.g., struma in
iodine deficiency or hypothyreodism, adrenal hypertrophy in endocrinal insufficiency, etc.). Organ
hyperplasias usually are reversible. Polyene macrolides, such as Nys and Amp, improve the energy
turnover in the prostate and promote the cholesterol metabolism, thereby improving the production
of steroid hormones. Remission of prostatic hyperplasia takes place after 3 to 6 months and is
reversible--after termination of the Nys therapy, prostatic hyperplasia recurs within 1/2 to 1 year.
This fact confirms the necessity of a chronic therapy with cell-stimulating substances for this and
many other indications. The treatment according to the invention is carried out by administration of
the active ingredient in a daily dose of about 0.5 to 4 g, preferably about 1 to 2 g.

d) Cholecystolithiasis (Gallstones)

Gallstones are caused by an insufficient production of bile acid. Bile acids are cholesterol derivatives
with a stronger dipole character than cholesterol. They form micellae with the alimentary lipids in
the gastrointestinal tract and thus facilitate their resorption. Nys stimulates the production of bile
acids of the hepatocytes and leads to the degradation of gallstones. According to the dipole model,
all effective cholagogics are cell-stimulating substances. The cholesterol acids chenodeoxycholic acid and Ursodeoxycholic acid exhibit, e.g., due to the COO⁻ group at the aliphatic part a stronger dipole character than cholesterol and are successfully used as drugs against cholecystolithiasis. The treatment according to the invention is carried out optionally by chronic administration of the active ingredient in a daily dose of about 0.5 to 4 g, preferably about 1 to 1.5 g.

e) Acne

Acne is closely associated with the cholesterol metabolism via the steroid hormone synthesis. Polyene macrolides, such as Nys and Amp, have a beneficial effect on acne both in topical and systemic application. Topical treatment is carried out by optionally repeated (2 to 4 times) application of an ointment (about 20 to 200 mg, preferably about 50 to 100 mg active ingredient/g of ointment). Oral treatment is carried out by daily administration of about 0.5 to 4, preferably about 1 g of the active ingredient.

7. Various Skin Diseases

a) neurodermatitis

b) dermatological allergies (contact allergies)

c) eczema

d) psoriasis

e) aphthous stomatitis

f) any kind of stomatitis

In most of these diseases, the active ingredient must be administered both orally and topically (see experimental part).

VIII. Galenic Preparations for Polyene Macrolides

Depending on the specific indication the polyene macrolides according to the invention will be administered orally, topically or intranasally, e.g., as inhalation. Oral or topical administration is preferred.

According to the invention, the oral daily dose of the active ingredient will be in the range of about 1 to 200 mg/kg body weight, preferably about 10 to 100 mg/kg body weight, particularly preferred about 15 to 30 mg/kg body weight. The specific daily dose depends on the active ingredient used, the disease and the patient’s condition and must be chosen by the attending physician. The preferred oral daily dose for adults for Nys and Amp is about 100 mg to 5 g, particularly about 0.7 to 2 g, e.g., 1.0 to 1.5 g. The maintenance dose of Nys and Amp is about 200 mg to 2 g. The frequency of application will be 1 to 6 times per day and preferably should be 1 to 4 times per day. 1 mg nystatin corresponds to about 5,000 I.U.

Appropriate preparations include tablets, capsules, lozenges, powder for emulsions, solutions and suspensions. Care is taken that all preparations contain the polyene as pure active ingredient. The share of other ingredients will preferably be minimal. For example, the tablets may contain other
galenic ingredients such as ethyl cellulose, lactose, corn starch, magnesium stearate, talc, saccharose, paraffin, gelatin, wax, vanillin, etc. Lozenges may contain flavors, d-mannitol, polyvinyl alcohol or other alcohols, magnesium stearate, talc, etc. Capsules will, if possible, contain only the pure substance. All other necessary additives are reduced to a minimum. In emulsions conventional emulsifiers will be used. Dimethylsulfoxide (DMSO) can for instance be used for preparing solutions.

Topical applications will include the polyene macrolide in a dose of about 10 to 200 mg, preferably about 40 to 200, 50 to 100 mg/g of composition are particularly preferred. Examples of topical preparations include creams, ointments, pastes, lotions, emulsions, solutions, etc. The following additives may be used: soft and liquid paraffins, cetyl or stearyl alcohol and other alcohols, stearic acid, sorbic acid, sodium hydroxide, propyl and methyl hydrobenzoate, propylene glycol, glycercyl monostearate, scents, solvents such as DSMO, etc.

For inhalations with aerosol and nasal sprays appropriate inhalation solutions will be used. The concentrations of the polyene will be between 2 and 200 μg/ml, preferably about 10 to 20 μg/ml of the composition.

The compositions to be applied topically and intranasally may be applied 1 to 8 times per day.

Commercial Nys contains two components designated as nystatin A1 and nystatin A2. Nystatin A1 is considerably more stable than A2 and the ratio of A1 to A2 determines the stability of the material. Although not critical for the present invention, a commercial preparation is preferred which shows suitable stability. The choice of suitable commercial preparations is a routine matter for the person skilled in the art.

IX. Experimental Part

EXAMPLE 1 Treatment of AIDS Patients

At present, 3 HIV-positive symptom-free patients receive chronic oral Nys treatment (1.5 to 2 g/day). More patients are being recruited. 4 more patients with the AIDS-related complex (ARC) not showing systemic mycoses receive the same therapy. These patients showed an improvement of the ARC-symptoms after receiving treatment for 4 to 6 weeks.

The same results were obtained in 4 patients (2 without symptoms, 2 in the ARC phase) with a corresponding Amp treatment.

EXAMPLE 2 Treatment HSV Infections

a) 25 patients suffering from Herpes labialis were topically treated with Nys ointment (50 mg Nys/g ointment) immediately after occurrence of the disease. Application frequency: 6 to 8 times a day. Relief from pain and remission of the pustulae was achieved within 24 to 28 hours. Treatment up to curing took 1 week.

The same results were obtained in 8 patients with a corresponding Amp treatment.

b) 7 patients with chronic recurrent HSV (3 to 4 HSV episodes per year) were prophylactically treated with orally administered Nys (1 g/day) for 3 to 6 months. The rate of incidence could be markedly
lowered. 6 patients did not suffer from episodes for 6 months and more. 1 patient experienced a HSV infection only once and this was a mild form.

The same results were obtained in 5 patients with a corresponding Amp treatment.

EXAMPLE 3 Treatment of Herpes Zoster Varicella (HZV)

3 patients, including 2 children, suffering from HZV infections and shingles at the trunk were topically treated with Nys ointment (50 to 100 mg Nys/g ointment) for the duration of the disease. An intraindividual comparison with all patients showed a more rapid and marked remission of the efflorescences that had been treated as compared to sites that had not been treated.

The same results were obtained in 4 patients with a corresponding Amp therapy.

EXAMPLE 4 Treatment of Hepatitis B Virus (HBV) Infections

4 patients with chronic hepatitis B were treated with oral doses of Nys (1 to 1.5 g/day) for 6 months. Complete remission of the symptoms could be obtained. The liver values (GPT, GOT, gamma-GT) were back to normal towards the end of the treatment.

The same results were obtained with a corresponding Amp treatment.

EXAMPLE 5 Treatment of Recurrent Aphthous Stomatitis (RAS)

18 patient with RAS were treated with mucosa-adhering Nys ointment (20 to 50 mg/g of ointment) prepared according to the German patent application P 44 34 929.7. The ointment was applied every 2 hours. Pain was already markedly relieved after the first application. Depending on the size of the lesion, RAS disappeared within 24 to 48 hours, while RAS not being treated took 5 to 7 days to disappear. None of the patients showed RAS after a four-day therapy.

The same results were obtained in 12 patients with a corresponding Amp treatment.

EXAMPLE 6 Treatment of Rheumatoid Arthritis (RA) and Osteoarthritis (OA)

9 patients with chronic RA and 3 patients with OA were treated with oral doses of Nys (1 g/day). The RA symptoms disappeared after 6 to 8 weeks and did not reappear during the maintenance therapy (same dose). In 2 female patients therapy was temporarily discontinued and the symptoms reappeared. After the Nys treatment was resumed, the RA symptoms disappeared again (challenge-dechallenge).

The same results were obtained in 5 patients with a corresponding Amp treatment.

EXAMPLE 7 Treatment of Multiple Sclerosis (MS)

3 patients with MS received chronic treatment with oral doses of Nys (1 g/day). A marked improvement of the symptoms was observed as early as 2 months after commencement of therapy.

The same results were obtained in 2 patients with a corresponding Amp treatment.

EXAMPLE 8 Treatment of Allergies
a) 7 patients with food allergies (3 with gluten-sensitive enteropathy, 2 with milk allergy and 2 with fruit allergies) were treated with daily oral doses of 1 g Nys for 6 months. The intolerance phenomena largely disappeared after 2 to 3 months and the patients could eat the respective food.

The same results were obtained in 5 patients with a corresponding Amp therapy.

b) 15 patients with pollen allergies and 8 patients with dust allergies were treated with daily oral doses of 1 g Nys for 6 months. Before commencement of therapy, the allergies of all patients had resisted treatment for at least 5 years or since birth. In all patients an almost complete disappearance of the symptoms (allergic rhinitis, asthma attacks, blocked nose, etc.) could be seen after 6 to 8 weeks. After discontinuance of therapy for experimental purposes (dechallenge) the symptoms slowly reappeared in about half the patients. After resumption of therapy (challenge) the symptoms could again be cured/improved.

The same results were obtained in 7 patients with a corresponding Amp therapy.

EXAMPLE 9 Treatment of Morbus Crohn and Ulcerative Colitis

Both diseases show CD44-mutants in the crypts of the mucosa. CD44 takes part in the MHC-restricted T-cell-receptor interaction. 1 patient with morbus Crohn and 2 patients with ulcerative colitis received chronic treatment with oral doses of 1 to 1.5 g Nys/day. Progression of the disease could be stopped and the symptoms disappeared. Endoscopic investigations confirmed these findings.

The same results were obtained with a corresponding Amp therapy (2 patients with morbus Crohn, 1 patient with ulcerative colitis).

EXAMPLE 10 Treatment of Psoriasis

4 patients suffering from therapy-resistant psoriasis on the entire body, whose condition had remained unchanged for many years received chronic treatment with oral doses of 1 to 1.5 g Nys/day. In addition, Nys was applied topically. All patients showed improvement after 2 to 3 months. Remission of the marginal inflammation and reduction of the efflorescences was noted. There was a remission of hyperkeratosis within the psoriasis efflorescences and normal epithelialization isles formed. Scales and itching diminished after treatment for 3 to 4 weeks.

The same results were obtained in 2 patients with a corresponding Amp treatment.

EXAMPLE 11 Treatment of Liver Cirrhosis

3 patients with alcohol-induced liver cirrhosis received chronic treatment with oral doses of 1 to 1.5 g Nys/day. Their liver values and general well-being improved.

The same results were obtained in 4 patients with a corresponding Amp therapy.

EXAMPLE 12 Treatment of Lichen ruber

2 patients with Lichen ruber were treated with oral doses of 2 g Nys/day for 3 months. The efflorescences disappeared completely.
EXAMPLE 13 Treatment of Neurodermatitis

8 children with atopic neurodermatitis were topically treated with Nys ointment (20-50 mg/g of ointment). The ointment was applied 3 to 6 times a day. The neurodermatitis sites that were treated, were completely healed after a few days (3 to 7 days). By contrast, the efflorescences that were not treated persisted.

The same results were obtained in 5 children with a corresponding Amp therapy.

EXAMPLE 14 Treatment of Cold Hemagglutinin Disease (CHD)

1 female patient with chronic CHD and a very high level of monoclonal IgM antibodies was orally treated with 1 to 1.5 g Nys/day for 6 months. Acrocyanosis, which regularly already appeared at 16° to 18° C. of air temperature, largely disappeared. Intensity decreased and exposure time at temperatures below 16° C. increased. Contrary to the condition before treatment, acrocyanosis could not be produced in repeated provocation tests with cold water. Moreover, the antibody level was lowered as well.

EXAMPLE 15 Treatment of Polyneuroradiculitis (Guillain-Barre Syndrome)

1 patient with polyneuroradiculities and neurological pareses received chronic treatment with oral doses of 1 g Nys/day. The pareses and pain symptoms clearly diminished.

EXAMPLE 16 Prophylactic Treatment of Sepsis

405 intensive care patients suffering from grave multiple traumata were orally treated with 3 g/day amphotericin B or Nys for the duration of intensive care (2 to 6 weeks). Compared to the standard group without therapy, the mortality and morbidity rate of nosocomial infections could be lowered in these patients. As the patients showed no mycoses, these effects are to be attributed to the immunostimulation by the polyenes.

EXAMPLE 17 Treatment of Immunodeficiency after Chronic Administration of Antibiotics

9 patients that had been chronically treated with antibiotics for diverse infections and showed reduced immune response received oral doses of 1 to 1.5 g Nys/day over a period of 3 to 6 months. None of the patients showed a mycosis. In this period, the therapy with antibiotics was discontinued. During and after the Nys treatment the conditions of the patients improved significantly with the result that a further treatment with antibiotics became unnecessary. The infection rate dropped and the immunological parameters went back to normal.

The same results were obtained in 5 patients with a corresponding Amp treatment.

EXAMPLE 18 Treatment of Chronic Fatigue Syndrome (CFS)

12 patients with CFS according to the CDC classification were orally treated with 1 to 1.5 g Nys/day for 3 to 6 months. In 8 patients the CFS syndrome disappeared completely and in the other 4 patients the symptoms were clearly improved as was their well-being.

The same results were obtained in 7 patients with a corresponding Amp therapy.
EXAMPLE 19 Treatment of Postmenopausal Syndrome (PMS)

5 patients with PMS who had not been treated with hormones underwent treatment with 1 g Nys/day during 3 to 6 months. The PMS symptoms disappeared completely.

The same results were obtained in 3 patients with a corresponding Amp therapy.

EXAMPLE 20 Treatment of Cancer

a) 3 patients with pancreas carcinoma and distant metastases (liver and lymphatic nodules) and cachexia were orally treated with 1.5 to 2 g Nys/day. Remission of the liver metastases was observed in 1 patient (confirmed by radiographs). All 3 patients showed weight increases and their well-being improved noticeably after only 2 weeks of treatment. In 1 patient, the Nys therapy was replaced with chemotherapy and radiotherapy. The patient died shortly afterwards. The 2 other patients continued to received the Nys therapy and have already reached an age above the statistical age.

b) 4 patients with carcinoma of the breast who had undergone ablation received chronic treatment with oral doses of 1 to 1.5 g Nys/day. No metastatic formation was noted. The same results were obtained with a corresponding Amp therapy (1 patient with pancreas carcinoma, 4 patients with carcinoma of the breast).

c) Two patients with oat-cell lung carcinoma in the final stage and cachexia were treated with 1.5 to 2 g Nys. It was possible to stop the cachexia.

d) 1 patient with liver carcinoma and cachexia was orally treated with 1.5 to 2 g Amp/day. There was a remission of the liver metastases (detected by radiographs). The patient showed a weight increase and a distinct improvement in his well-being after only 2 weeks of treatment.

e) 3 patients with colon cancer (Duke's carcinoma) in the final stage and cachexia were orally treated with 1.5 to 2 g Amp/day. Distinct relief from the gastrointestinal symptoms could be brought about and cachexia could be stopped.

EXAMPLE 21 Use of Nys in Wound-healing

32 patients with soft tissue wounds were topically treated with Nys ointments (50 to 100 mg Nys per g of ointment). The ointment was applied several times a day. An intraindividual comparison with wounds that had not been treated showed a significantly faster healing. The healing time could be reduced with Nys by 35 to 50%.

The same results were obtained in 14 patients with a corresponding Amp therapy.

EXAMPLE 22 Treatment of Burns

a) 19 patients with second degree and third degree skin burns were treated with Nys ointment immediately after suffering the burns. The ointment was applied 6 to 8 times per day. Nys prevents the formation of blisters and brings relief from pain within few minutes. Nys prevents the formation of cicatrices and accelerates healing. The Nys ointment must be applied several times a day.
b) Several persons with sun burns were also topically treated with Nys ointment. The sun burns disappeared completely within 24 to 48 hours.

The same results were achieved with a corresponding Amp therapy (14 patients with wounds, 11 with burns, several ones with sun burns).

EXAMPLE 23 Treatment of Ulcus Cruris and Decubitus

8 patients with chronic ulcus cruris and 9 patients with decubitus were topically treated with Nys. The medication was topically applied 3 to 6 times per day. Depending on their size, the open wounds were epithelized almost completely after about 2 to 4 weeks. After discontinuance of therapy, relapses occurred. Therapy was continued chronically.

The same results were obtained with a corresponding Amp treatment (3 patients with ulcus cruris, 5 patients with decubitus).

EXAMPLE 24 Treatment of Hypercholesterinemia

Both the sepsis study (see example 16) and the individual explorative studies showed markedly lower levels of total cholesterol and LDL fraction in a number of hypercholesterinemic patients that had been treated with oral doses of 1 to 2 g Nys/day. HDL remained unchanged. A decrease occurred after 10 to 14 days, in some patients only after 3 to 4 weeks. The higher the initial value of plasma cholesterol, the greater the lipid-lowering effect of Nys. In patients with reduced cholesterol values (for instance after severe traumata), cholesterol was found to increase. Nys puts the cholesterol metabolism in the body back to normal.

The same results were obtained with a corresponding Amp therapy.

EXAMPLE 25 Treatment of Prostatic Hyperplasia

6 patients with prostatic hyperplasia were orally treated with 1 mg Nys/day for at least 6 months. Hyperplasia already started to progressively disappear after 3 months. The urea flow improved.

The same results were obtained in 4 patients with a corresponding Amp therapy.

EXAMPLE 26 Treatment of Gallstones

In 4 patients with sonographically ascertained cholecystolithiasis, who were orally treated with 1 to 1.5 g Nys/day, gallstones could no longer be detected (by sonography and in radiographs with X-ray contrast agents) at the end of therapy.

The same results were obtained in 1 patient with a corresponding Amp treatment.

EXAMPLE 27 Treatment of Acne

Several patients with acne, including acne conglobata, were treated topically with Nys ointment (20 to 100 m g Nys/g ointment) and orally with Nys (1 g/day). It was possible to achieve a marked acne remission.

The same results were obtained with a corresponding Amp therapy.
The daily doses indicated in the present description can be administered depending on the individual case, either as a single dose or divided into several partial doses, for instance 2, 3 or 4 partial doses, for example before or after the meals.

The term "chronic administration" as used herein includes both lifetime administration of the active ingredient and the administration up to a possibly permanent disappearance or remission of symptoms. Depending on the severity of the disease, the symptoms can disappear in the course of the chronic therapy of the invention, for instance within 3 to 6 months, sometimes earlier, but in many cases only later, for instance within 12 months. It may furthermore include an intermittent chronic therapy, such as for instance a therapy for 3 to 6 months, followed by a therapy pause for 6 to 12 months or resumption of the therapy when the disease reappears.

A therapy according to the invention usually begins once the macroscopic or histological findings are available. In special cases, for instance in the treatment of AIDS or cancer, early therapy is preferred. Early therapy should begin immediately after the diagnosis is made (for instance by immunological evidence) but before the availability of macroscopic or histological findings. In the case of a known predisposition, for instance due to age or hereditary factors, a prophylactic chronic administration of polyene macrolides is also conceivable.

Starting from the general teaching given in the description and on the basis of the specific teaching of the examples, a person of average skill in the art can select other suitable polyene macrolides as therapy for the afore-mentioned indications, without this requiring him to make inventive efforts of his own.