Progetto di svilppo - area sanitaria

Localizzazione: Africa

Oggetto: Donazione unità sanitaria DIAGNOSTICA-TERAPEUTICA tecnologicamente innovativa ma a basso costo gestionale per la prevenzione e la cura delle malattie infettive e patologie correlate.

Finalità del programma:
Il progetto ha la finalità di **arginare** alcuni problemi endemici del continente africano che vede nelle

- **malattie infettive** in genere (malaria ed HIV in primis) e nel
- **tumore al seno la prima causa di morte femminile**,

avendo questa ultima patologia superato in detto Continente la mortalità per parto. (L'esperienza maturata dalla NATURAL PHARMA in questi anni è tale da poter ipotizzare una **relazione strettissima** tra "**infezioni varie prevalentemente ginecologich**e" e "**tumore al seno**", supposizione confortata da una serie di ricerche bibliografiche scientifiche universitarie internazionali, oramai inconfutabili).

I risultati terapeuti sperimentali, ottenuti nella cura di **patologie infettive** e di conseguenza su **neoplasie mammarie**, mediante un preparato fitoterapico estratto da alghe, denominato ABAMAV di proprietà della NATURAL PHARMA, **nonché con l'indispensabile ausilio delle innovative tecnologie di supporto**, (di seguito elencate) sono tali da poter ipotizzare una sua applicazione, su larga scala per sconfiggere tali problematiche a **costi sanitari molto contenuti!**

La strumentazione di base del ambulatorio che la NP donerà al comune di_____ nel territorio di _____ nello stato africano della COSTA D'AVORIO , per la diagnosi e il trattamento della patologia in oggetto, e più in generale per patologie infettive endemiche (quali Malaria e HIV, oltre a malattie gastrointestinali) necessita di un area non superiore ai 150 mq suddivisi in 4 stanze, dotate di servizi igenici, impianto elettrico, collegamento internet, e alla presenza di personale sanitario (minimo 2 infermieri e 2 medici con specializzazione di base).

Avendone la possibilità si ritiene ottimale che la collocazione dei locali in oggetto sia posizionata/ricavata in strutture sanitarie/ospedaliere già esistenti:

Gli spazi saranno così adeguati e finalizzati

Locale 1
adibito a CENTRO DI FORMAZIONE SPECIALISTICO
In tale unità operativa si permetterà di acquisire e divulgare conoscenze sufficienti per divenire in seguito un centro D'ECCELLENZA nella *diagnostica e nella terapia mediante prodotti naturali e tecnologie diagnostiche e terapeutiche innovative*, (diagnosi mediante apparecchiature di biofisica quantistica, elettroporazione, nanomicroscopia, ecc, ecc,)
Sviluppare tale metodica diagnostica e terapeutica porrà la COSTA D'AVORIO al livello internazionale d'avanguardia sanitaria, potendo diventare una iniziativa da applicare esportandola in altre aree della Nazione e/o altre Nazioni del continente africano.

Locale 2
Adibito a DIAGNOSTICA D'AVANGUARDIA

- **MEDIANTE METATRON o tecnologia similare**
  Grazie alle ultime ricerche in ambito Medico Scientifico ed alle acquisizioni della Fisica Quantistica, con l'apparecchiatura Metatron Hunter e similari, vengono valutati gli stati di salute organo-energetici individuando le presunte patologie a carico del sistema fisiologico, individuando la quota degli agenti patogeni come virus, batteri, amebe, parassiti, il tutto a costi gestionali contenuti.

- **MEDIANTE TERMOANGIOGRAFIA DINAMICA**
  La ***termoangiografia dinamica*** di proprietà della BRESTLIFE, azienda italiana leader mondiale in tale tecnologia (sviluppata su Ipad della Apple) e partner della NPI. La stessa tecnologia potrebbe essere adottata

per uno screening di massa sulla popolazione femminile, avendo un costo contenutissimo, assenza di materiale di consumo e non necessita di punti di alimentazione elettrica (vedi specifica allegata. Sottolineiamo le caratteristiche **non radiologiche** e quindi non dannose della tecnologia diagnostica (rispetto per esempio a quelle di una tradizionale mammografia). Perfetta nella verifica della efficacia terapeutica dell' ABAMAV documentabile già nel giro di 2/3 settimana di trattamento. La regressione/guargione della patologia "cancro al seno" per stati non ancora invasivi, trattata con il fitoterapico ABAMAV potrà essere documentata agevolmente, anche giorno per giorno.

Locale 3
AREA TRATTAMENTI specialistici e area NANO-MICROSCOPIA del sangue in real-time

Area somministrazione del fitoterapico ABAMAV nella formula più opportuna ed adeguata tra quelle disponibili, con particolare attenzione alla terapia elettroporativa. (allegato1).
Area trattamento mediante esposizione del paziente a campi magnetici con frequenze specifiche terapeutiche per la patologia trattata con apparecchiatura di BIOFISICA QUANTISTICA di proprietà della NP. (allegato 1a).
Al fine di poter meglio individuare patologie su base microbica, il centro sarà **in seguito**, dotato di un microscopio a campo oscuro e collegamento internet per la eventuale trasmissione di immagini a fini diagnostici con assistenza in remoto in un centro italiano specializzato e convenzionato. (vedi allegato 3)

Locale 4
Area adibita a magazzino e/o ufficio

Di seguito viene allegato del materiale esplicativo al fine di ulteriori approfondimenti delle aree progettuali presentate.

I tecnici della NATURAL PHARMA sono a disposizione per poter elaborare ed approfondire ogni aspetto scientifico ed operativo di suddetto progetto.

Il valore della donazione comprensiva di fornitura ABAMAV d'impianto e relative apparecchiature è quantificabile in non meno di 68.000 euro

Elettroporazione

L'Elettroporazione (Dermoporazione o anche Elettroveicolazione) sono tecniche di veicolazione transdermica che agiscono direttamente sui meccanismi cellulari mediante impulsi elettrici modulabili che sfruttano i canali di passaggio nel derma favorendo enormemente il flusso di molecole idrosolubili. Con questo unico sistema (nessuna altra tecnologia può fare altrettanto) si riesce a veicolare Farmaci, Sostanze e Principi attivi senza l'impiego di aghi e senza dolore, la penetrazione, con queste nuove tecnologie, è profonda, rapida e selettiva. L'Elettroporazione di ultima generazione ha da tempo superato le vecchie tecniche di veicolazione quali la Ionoforesi, la Jontoforesi, la Crioelettroforesi che si effettuano con protocolli più invasivi e meno efficaci.

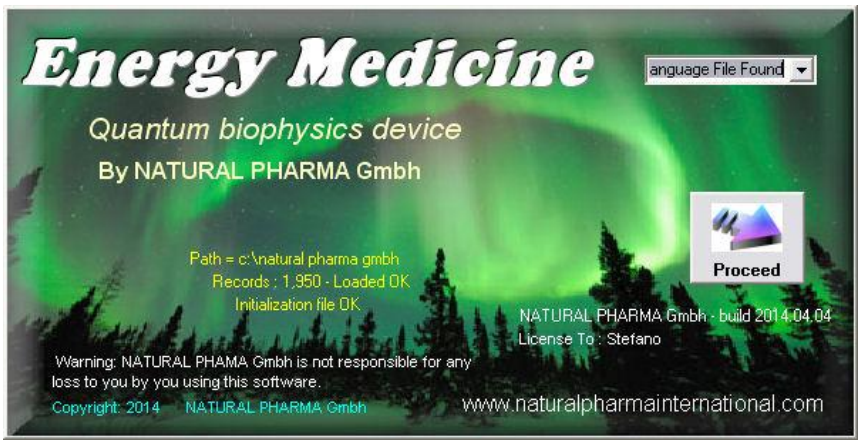
Elettroporazione in oncologia

Impulsi elettrici di breve durata ed alta intensità inducono un aumento della permeabilità della membrana cellulare conseguente ad un ri-orientamento delle molecole polari che la compongono: questo fenomeno è definito ElettroPorazione Quando un tessuto viene sottoposto a EP l'effetto si esercita esclusivamente sulla componente cellulare lasciando inalterato il tessuto connettivo, in assenza di denaturazione proteica. Nell'ambito delle lesioni tumorali primitive e metastatiche, la permeabilizzazione della membrana cellulare crea un accesso diretto al citoplasma per farmaci chemioterapici normalmente non permeanti, ad esempio, la bleomicina, la cui tossicità viene così aumentata di migliaia di volte.
Attualmente, l'ECT è utilizzata nel trattamento locale di tumori primitivi della cute e di metastasi localizzate alla cute ed al tessuto sottocutaneo indipendentemente dall'origine istologica. Viene studiato l'effetto di EP ed ECT su cellule tumorali, tessuto e metastasi ossee attraverso analisi istologiche, istomorfometriche (statiche e dinamiche) e microtomo grafiche.

In genetica molecolare

questo metodo è utilizzato per trasferire molecole di acido nucleico nelle cellule dopo somministrazione di un impulso di corrente ad alto voltaggio.
Le cellule sono immesse in una soluzione contenente DNA e sottoposte a un breve impulso elettrico che produce una transitoria apertura dei pori della membrana, attraverso i quali il DNA entra direttamente nel citoplasma.
L'e. è usata anche nel trasferimento di geni nelle cellule vegetali.

BIOFISICA QUANTISTICA
ELETTROMEDICINA



Oltre al sistema circolatorio del sangue e linfa, alcuni ricercatori suggeriscono che il corpo umano ha un sistema circolatorio supplementare per l'elettricità.

L'agopuntura utilizza aghi metallici inseriti nei punti chiave per influenzare il flusso di energia elettrica lungo i meridiani. La ricerca del Dr. Robert Becker scoperto che molti tessuti del corpo sono semiconduttori e che ci sia una "corrente di pregiudizio" elettrica che deve essere presente per la guarigione

La TENS (stimolazione nervosa elettrica transcutanea) sono usati per trattare le lesioni sportive e dolori. Lo Zapper della dottoressa Hulda Clark utilizza la frequenza elettrica per uccidere i parassiti. Il protocollo del Dr. Bob Beck utilizza corrente elettrica per disattivare virus, batteri, funghi, parassiti e micotossine nel corpo. Il Generatore multi-frequenza del dottor Lakhovsky promuove la salute, stimolando tutte le cellule del corpo. La ricerca del Dott. Lakhovsky è stata continuata dal Dr. Ed Skilling.

Dato i costi operativi minimi tale tecnologia non suscita grandi interessi e stimoli nella diffusione La NP ha messo a punto una delle migliori apparecchiature per poter utilizzare tale Know how non solo come TENS….Se i dispositivi elettro-medicina oggi sono in grado di uccidere tutti i batteri noti patogeni, virus e funghi: in futuro vaccini e antibiotici potrebbero essere oramai superati….

Sono abbastanza sicuri da essere utilizzati anche su bambini.

BREST LIFE

DATG
DYNAMIC ANGIOTHERMOGRAPHY

Società per la e commercializzazione di apparecchiature per la diagnosi precoce del tumore della mammella

LA MISSION:

- **Divulgare nel mondo la Nuova Tecnica per la diagnosi precoce dei tumori della mammella, denominata Angiotermografia Dinamica (DATG).**

L'angiotermografia dinamica (DATG) è una tecnica diagnostica che si è rivelata estremamente valida nella diagnosi delle lesioni tumorali e pre-neoplastiche mammarie in quanto si può affiancare con ottimi risultati, in particolare in giovani pazienti, alle altre metodiche già in uso per lo screening: la mammografia e l'ecografia senza con queste essere in competizione.

E' una tecnica non invasiva che può essere applicata a donne di tutte le età, ed è facilmente ripetibile, indolore, priva di rischi ed i suoi costi molto bassi.

La DATG è uno strumento diagnostico veramente promettente nella valutazione delle giovani pazienti con alta densità della ghiandola mammaria e nella prevenzione del carcinoma della mammella in donne portatrici dei geni BRCA-1 e BRCA-2 correlati con l'insorgenza del carcinoma.

L'angiotermografia dinamica (DATG) è in grado di localizzare anche stati preinvasivi, e di evidenziare chiaramente mediante cambiamenti nel pattern

l'insorgenza di essi. E' evidente che una tale possibilità assicura risultati clinici molto importanti, al limite con la prevenzione secondaria organo-specifica.

La capacità di controllare il flusso funzionale ematico tipica della metodica permette sia di evidenziare localizzazioni multifocali che di controllare in periodo post operatorio l'eventuale comparsa di recidive locali. Esiste cioè un totale controllo dell'evoluzione della malattia e di conseguenza una forte ottimizzazione dei risultati.

La DATG e' considerata una tecnica diagnostica che usa una tecnologia ottica, mediante il rilevamento di un'immagine ottenuta appoggiando una placca di cristalli liquidi micro incapsulati alla mammella.

Quest'immagine che risulta rientrare nella categoria della "termografia mammaria" è basata sull'interpretazione qualitativa dei vasi sanguigni della mammella, mediante lo studio della circolazione e microcircolazione della ghiandola mammaria e della presenza di neo-angiogenesi.

La neoangiogenesi infatti ha un ruolo cruciale nella crescita e progressione del tumore, inoltre la presenza e l'estensione dell'angiogenesi può fornire parametri per la prognosi del tumore ed essere usata per sviluppare nuove strategie diagnostiche e terapeutiche. L'utilizzo del dispositivo per queste applicazioni è limitato esclusivamente a medici.

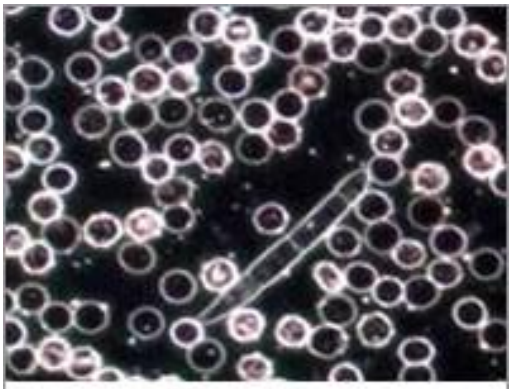
Non essendo una tecnica radiologica non e' obbligatorio un medico specialista in radiologia per redigere il referto, anche se ne consiglia l'uso proprio a questi specialisti, ma possono essere tutti i medici che quotidianamente si occupano di senologia.

Se ne suggerisce l'utilizzo alle seguenti specialità mediche:
- Radiologi
- Ginecologi
- Oncologi
- Chirurghi
- Chirurghi Plastici
- Medici di base (per un primo livello di screening)

Il corretto utilizzo dell'apparecchiatura sarà insegnato da un corso OBBLIGATORIO per tutti i medici che vorranno fare uso della DATG

NANOMICROSCOPIA
in campo oscuro

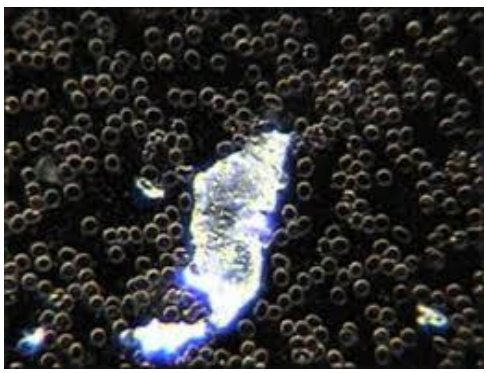
La microspia in campo oscuro permette una visione morfologica molto significativa.
La forma, la dimensione il comportamento delle cellule del sangue possono essere indicative per lo studio dinamico della biologia dell'organismo.



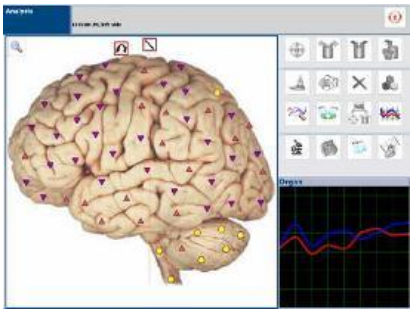
Parassita

Questa tecnica microscopica ci potrà fornire le seguenti informazioni:

- **determinare la presenza di microrganismi simbionti**
- **integrità delle membrane delle cellule sanguigne**
- **mobilità dei granulociti**
- **resistenza vitale del sangue**
- **presenza di tossine metaboliche**
- **presenza di muffe o funghi batteri, micoplasmi, plasmodi**
- **stati infiammatori acuti e cronici**



METATRON



Grazie alle ultime ricerche in ambito Medico Scientifico ed alle acquisizioni della Fisica Quantistica, con l'apparecchiatura Metatron Hunter un'apparecchiatura ideata dal fisico russo Prof. Nesterov Vladimir vengono valutati gli stati di salute organo-energetici individuando le presunte patologie a carico del sistema fisiologico, individuando la quota degli agenti patogeni come virus, batteri, amebe, parassiti. Si possono valutare assieme al paziente gli alimenti più sensibili per il corpo, ed eventualmente cercare le intolleranze vere e proprie.

Metatron Hunter rileva l'alterazione della radiazione elettromagnetica proprio dove essa ha origine, la decodifica e la mostra sullo schermo del computer dove viene riprodotto a colori un modello virtuale dell'organo in esame.

Grazie a questo tipo di esame si può "navigare" all'interno del corpo umano valutando il grado di "salute elettromagnetica" di ogni singolo organo, tessuto o cellula praticamente in tempo reale.

Si possono ottenere risposte circa organi specifici, sull'omeostasi dei liquidi e dei tessuti, sulle certezze e sulle probabilità di una patologia, sugli allergeni, sull'attività di microrganismi (virus, batteri, funghi), sui parassiti intestinali, sui rimedi allopatici o fitoterapici da impiegare, sui cibi consentiti o da evitare, ecc. Ogni risposta è poi correlata da un valore alfa-numerico e da un grafico di riferimento, che consentono all'operatore di comparare in ogni istante la situazione reale con quella virtuale.

Un particolare comando attiva la scansione microscopica di tessuti, cellule, nuclei, cromosomi e DNA, cosicché l'analisi macroscopica di una patologia può trovare riscontro anche a livello citologico o addirittura molecolare.

References

RUSSIAN FEDERATION
MINISTRY OF HEALTH
PUBLIC AGENCY

Hospital of Senior Department of Public Health Services of Administration of Omsk Region

Ref #117, dated August 26, 2002

The nonlinear computer diagnostics (NLS) using the device " OBERON " and the program "Metapathia " have been applied at the Hospital of Senior Department of Public Health Services of Administration of Omsk Region since 1999. During that period 1876 patients have been surveyed. The examinees included 868 men and 1014 women aged from 17 to 83 with most of them (82%) aged 40 and older. The NLS diagnostics method has no contraindications and is comparable in terms of to informational content with other hardware-based research methods (ultrasonic, computer tomography, nuclear magnetic resonance), and in some cases it surpasses them. The outcomes of NLS examination coincided with data of other hardware-based research techniques in 82 % of cases According to our information, NLS accuracy amounted to 81.7 %. Sensitivity to 79.3 %: whereas specificity to 87.2 %. Computer non-linear diagnostics (NLS) as a dynamic non-invasive informative method can be widely used to examine the status of health affected by pathologies of most different origins. In the course of its development and advancement the method may become a foremost instrument of diagnostics and observation with respect to a number of widespread diseases of heart-vascular system, respiratory system, digestive organs, urogenital system and a number of other organs and systems; allowing to promptly collect detailed information about the lesion and its pattern and assess the treatment efficiency. The specific character and working conditions of the therapeutic and clinical institutions in Russia allow to extensively employ the NLS method not only for diagnosing diseases, but also for dispensary observation, and what is more, practically all the patients in need of examination can be placed under observation. So the physicians can now cover patents with

the changes that are latent and can only be verified by means of NLS.

REFERENCES

This is only a part of available numerous references about outcomes of application of the NLS-method by the students of the course of the Institute of Applied Psychophysics provided by V.I.Nesterov in the course of the last few years in Omsk Government Medical Academy. The students having special medical or biological education, among them candidates and doctors of science, professors, physicians, and science officers share their experience. Let us give the floor to those who stepped over the bias barrier, who believed in the suggested method, felt the first pleasures from recovery of themselves and their patients.

G.A. Loppoukhov
Candidate of Medical Science, employee of the Omsk Government Medical Academy, head of Improvement Faculty

I underwent the course of training in V.I.Nesterov's NLS-method in 1999. From then on I have made thousands of experiments that confirmed efficacy, simplicity and safety of utilization of this method in diagnostics and treatment of serious contagions, such as tuberculosis, diphtheria, hepatitis, candidiases, some forms of oncology, invasions (helminthic diseases), forms of some function distresses, etc. My work practice at Omsk GUZO Polyclinic confirms the universality of this method and its huge prospects for the development of the modern medicine with a minimum utilization as a monitoring agent of medical equipment. The method allows a systemic diagnostics of body condition, including identification of type of infection contamination, invasion, dysbacteriosis of intestine, condition of organs and energy channels, endocrine system, presence of allergic status and failures in the central nervous system's operation. The method solves the issue of a precise definition of a body requirement in appropriate medicamental agents both by quantity, and composition. As a result the administered course of treatment yields fast and appreciable effect as does not break the bionomics of the internal human medium, and is extremely sparing. The method allows to

detect the cause of the disease and to remove illness itself, instead of its consequence, as it frequently happens in practice of the orthodox medicine. Besides the method can be widely used for different household needs, for example, for defining the quality of products, drinks, wines, cosmetic creams, etc. Alongside with it I should note that if it has to do with treatment of other people, the operator himself should be substantially free from the above illnesses. I am deeply convinced, that NLS method has a big future. It is medicine of new millenium.

E.V.Silinna
Candidate of Biological Science, Senior Researcher of Natural Nidus Infections:

Since in medicine I am on a calling, I have always felt some dissatisfaction with my capabilities, because most often you face a barrier which you cannot cross, though the intuition hints you, that behind the barrier something else hides. This explains the infinite searches in the nonconventional areas of medicine in hope to find something, which removes doubts and gives definite positive results. And here after more than forty years in medicine I come to grips with the methodology developed by V.I.Nesterov, the NLS-diagnostics. It appears, that all knowledge of ourselves, of our bodies, and our illnesses we wear in ourselves, even not suspecting it, and all you have to do is to learn how to read it. At times you do not believe your eyes and even get scared of its opportunities to find the shortest and optimal way to implement the basic postulate of medicine - do not cause harm, but cure!

K.N.G.
Doctor of Medicine, professor, Omsk Government Medical Academy:

After a complex operation in the field of small pelvis I had a serious condition, daily in the evenings in the course of 5-6 days the temperature rose to 39 degrees and higher. The cause of the postoperative complication remained obscure. The newest antibiotics of broad range of action appeared inefficient. I decided to define the etiology of the disease using the NLS method and pick up the antibiotic required for treatment. "Oberon" indicated a

staphylococcal infection. It was important to pick up a drug for treatment of this infection. Out of more than 30 antibiotics, specified in the program "Metapathia ", the device indicated Oxacillinum, an old antibiotic used in Russia more than 25 years ago. In two days after the intramuscular injection of Oxacillinum the temperature dropped to subfebrile, my condition considerably improved. In a satisfactory condition I was discharged home. In the course of the next four months I had batches of fever, delicacy, sweating, and tussiculation. I had to find out the diagnosis. Out of different diseases specified in the"Oberon" process I chose one - which was radical pneumonia. With the help of the device I chose an antibiotic for treatment which was doxycycline. After the treatment, the phenomenon that had disturbed me disappeared. It became clear to me, that after the operation and narcosis I developed a two-sided pneumonia which was not timely recognized. I shall notice, that the monographies on pneumonia issued by Academician V.I.Pokrovski et all. in 1995, stated that the lethality at intrahospital pneumonias reached 40-50 %, that is, nearly every second patient dies. Quite often these pneumonias take a lingering course. Summarizing the above, with all definiteness I can declare, that I owe my life both to the surgeons who operated me, and to Vladimir Igorevich Nesterov, the enlightened talented person who had trained me in NLS method and given me the skills how to operate "Oberon", shortly before I was hospitalized. Have I not learned how to operate the device I would not survive after the operation at the neglected pneumonia. The NLS-method is expedient for use to define whether the cure fits to the patient. With the help of the device I checked the effect of medicines on the systems of biological adaptation of my organism. It appears, that approximately 60-65 % of the drugs, prescribed to me once, considerably increase the adaptable reserves, 20-25 % sharply reduce them, the other 10-15 % react sort of neutrally . It was in particular found out, that Riboxinum, which is prescribed to improve trophicities of cardiac muscle, and Cinnorizin prescribed to improve brain blood circulations, in my case, dramatically reduce adaptive reactions. It became clear why at the long-course treatment, prescribed to me repeatedly for a number of years, these drugs did not improve my condition. I believe, that utilization of the NLS method can increase efficacy of the traditional methods

of treatment, will allow to individualize treatment, picking up just those drugs, which salutarily affect this or that organ, increase bioenergetics of the body in general.

S.K.Petrova
Candidate of Biological Science, Senior Researcher, head of virus vaccines laboratory of Natural Nidus Infections Institute:

Mrs. G.K., an employee of Natural Nidus Infections Institute was operated on in May, 2002 concerning the extradural compression of the celiac axis. Before that time, since 1998, she had begun to feel bad, felt pains after meals; delicacy emerged; her weight dropped from 62 to 36 kg. The operation did not bring convalescence and her health condition became critical. Because of delicacy she ceased to move. It was at this very time we found a possibility to examine her with the help of NLS method. The employees of the institute, who had completed V.I.Nesterov's course in April 2002, established the following: using virtual model they detected lesions of many organs, such as liver (right lobe), pancreas, and esophagus, trachea of bronchuses, ascending and descending departments of colon. Examination of the contagious spectrum detected presence of several "dozing" infections, such as: Coxsackie virus B-4, cytomegalovirus. It has to be noted, that indicated lesions were registered independently from each other by two students of V.I.Nesterov's course. Treatment was carried out on the grounds of the detected damages. The infection contaminations and illnesses detected using Nesterov's method, were fought against with the help of metasods, biologically active food supplements, phytopreparations. In 11 day nearly all dozing infections disappeared. In 4 months the patient's condition considerably improved: he developed a healthy complexion, ability to move without efforts including walking in the street, his appetite improved, the NLS method defined food that was good to her. During the last period her weight grew up to 43 kg. Currently the patient's condition is satisfactory.

N.L. Ogluzdina
attending physician:

Having completed in 2001 V.I.Nesterov's course, I immediately joined in treatment of my mother who lay in hospital with the diagnosis "kidneys polycystosis". The patient developed urosepsis of blood, penetrating trophic ulcers on the legs, low haemoglobin, constant vomiting, there was no urination. After examination on a tomograph physicians said, she would only last for 2 or 3 days, and it was necessary to urgently connect the patient to the artificial kidney, the haemodialysis. The patient was transfered to haemodialysis - 3 times a week. Collaterally under observation of the attending physician we started examination and treatment with NLS method. A complete systemic diagnostics of health was carried out. As a result the following was found: damages of kidneys, liver, thick intestine, and heart. During treatment involving NLS method all damages were eliminated using custom-made metazodes. After that, the urinalyses came back to normal. During therapy of liver with administration of tansy selected by NLS method, and metasods, swirling started in intestine, diarrhea, evacuation of black colour, and vomiting. After a while vomiting stopped and appetite developed. Analysis of blood in terms of urea and creatinine improved. Soon the patient was removed from haemodialysis and in two months she was discharged from hospital.

A.Ya.Shvak
Candidate of Medical Science, senior researcher of Omsk Government Medical Academy:

I heard a course lectures on NLS method in June - July, 2001. During this period I acquired rich and extensive information on application of the NLS method. This knowledge let me comprehend myself and the world around in a new fashion. I became a different person, who tries to control her words and deeds. I express deep gratitude to V.I.Nesterov for that titanic labor that he had laid on his shoulders, for his disinterested activity and the knowledge that he gave to his students. The NLS method has entered my life and became an integral part of my existence. The application field of NLS method is so multi-sided, that it is hard to imagine a field, where this method cannot be used. For example, I am constantly using the NLS method for defining the prescribed food articles. My practical studies enabled me to make myself a diagnosis of an obstructive bronchitis using the NLS method. With the help of the

device I have selected the necessary drugs and nutriceuticals. I monitored my condition by the device. In two days the temperature dropped from 39.5 to 37 degrees, and gradually my conditions started to improve without application of antibiotics. I will give you another example of NLS method application. I found in two my relatives, that their thick intestine did not function by itself, and made the following diagnosis with the help of Oberon: dyskinesia of thick intestine. I provided a course of meta-therapy to my patients. With the help of the NLS method I saw my patient's condition gradually improving. Upon completion of the course of therapy my patients joyfully told me, that their thick intestine had started to function by itself and was getting back to normal, and the torment that they had felt for several months, has passed.

V.T.Sokhnova
Microbiologist, Natural Nidus Infections Institute:

I extensively use the device "Oberon" for detecting "hidden" infections, as well as diagnostics and selection of medical agents and herbs by NLS method. I have repeatedly and successfully applied the acquired knowledge, helping myself and my folks. I'll give you some examples. Last August when in country house, I felt cutting pains in intestine. Using the device I detected an inflammation in cholic bladder, as well as an infection contamination: golden staphylococcus. I was takin immortelle, which removes infection contamination and recovers operation of cholic bladder, for 1.5 days. Everything passed. My daughter complained to acute severe pains in intestine. She paid a visit to a physician, who detected a contagious colitis; the recommendation was to do seeding in order to select a medicine. We didn't do it. We defined the infection on "Oberon" instead, and picked up the therapy. The daughter was taking the selected drug for a day. As a result she is healthy. My daughter's friend complained of frequent illnesses of her child. On the device I detected presence of an infection contamination in the boy. I recommended dog-rose alongside with biologically active food supplements. Everything came to normal.

L.V.Sorokina
physician, researcher of Laboratory of Encephalitises of Natural Nidus Infections Institute:

I graduated in 2000 V.I.Nesterov's courses on NLS diagnostics technique and therapy and have been examining patients (basically my relatives, acquaintances, employees of the Institute) ever since in the course of the last two years. In Mrs. K.N. an employee of my Institute who had the official diagnosis "multiple sclerosis", the NLS method detected a cytomegalovirus, Epstein Barr's virus, and mononucleosis was diagnosed. After a treatment with nosodes picked up by NLS method, her condition normalized. In 4 days the patient's depression was replaced by excellent mood, almost euphoria, and the desire to overcome everything. Based on the results of examination on "Oberon" improvements have been noted. The treatment is proceeding. NLS method has investigated ancestral peculiarities in operation of different organs and systems, and detected predisposition to the same pathology in family members. Thus in one family, grandmother, son, daughter and the 15-year-old grand daughter - all had changes in pancreas, and an intestinal dysplasia. In other family a 67 years old mother had some lesion on her leg. She was taken to hospital. Using the NLS method I detected a Kaposi's sarcoma in her, and the diagnosis proved to be true. The woman was operated concerning a malignant tumour. In her son, who was apparently healthy I found a predisposition to this pathology and a number of similar signs. Using the NLS method information drugs were manufactured, which helped suppressing an "epidemy" in Nizhnevartovsk where I worked in summer of 2002 (high temperature and nausea were noted all at once in several families among children and adults). The successful cupping of the disease was performed overnight with the help of custom-made metasods, with a complete convalescence in two or three days. At this "epidemy" NLS method detected a staphylococcus in all patients.

T.G.Kuznetsova
Deputy Head physician of the clinic of the Main Department of Public Health Services of Omsk Region Administration:

In June, 2001 I completed the course of NLS diagnostics and therapy and was very soon convinced of the efficacy of the method. Here are some examples of my application of the acquired knowledge. Suddenly I had urodynias. Using the Meta-therapy method I got rid of the pains within a very short time. It was pretty much the same thing with the heart. I have an idiopathic hypertensia, an exertional angina. After the Meta-therapy my pressure normalized, pains in the field of heart disappeared completely. There were problems with my throat, such as constant anginas. Using the NLS method I detected the contagious agent, and treated it with nosodes. As a result the pains passed, anginas seized. My daughter used some cosmetics, and as a result had developed red eyes and pain. With the help of "Oberon" I found out that these cosmetics negatively affected her. The cosmetics were replaced by a patient-fir kind, and a week later everything passed. Many thanks to Vladimir Igorevich Nesterov for the knowledge, which I acquired.

Electroporation

Electroporation, or **electropermeabilization**, is a significant increase in the electrical conductivity and permeability of the cell plasma membrane caused by an externally appliedelectrical field. It is usually used in molecular biology as a way of introducing some substance into a cell, such as loading it with a molecular probe, a drug that can change the cell's function, or a piece of coding DNA.[1]

Electroporation is a dynamic phenomenon that depends on the local transmembrane voltage at each point on the cell membrane. It is generally accepted that for a given pulse duration and shape, a specific transmembrane voltage threshold exists for the manifestation of the electroporation phenomenon (from 0.5 V to 1 V). This leads to the definition of an electric field magnitude threshold for electroporation (E_{th}). That is, only the cells within areas where $E \geqq E_{th}$ are electroporated. If a second threshold (E_{ir}) is reached or surpassed, electroporation will compromise the viability of the cells, *i.e.*, irreversible electroporation (IRE).[2]

In molecular biology, the process of electroporation is often used for the *transformation* of bacteria, yeast, and plant protoplasts. In addition to the lipid membranes, bacteria also have cell walls which are different from the lipid membranes and are made of peptidoglycan and its derivatives. However, the walls are naturally porous and only act as stiff shells that protect bacteria from severe environmental impacts. If bacteria and plasmids are mixed together, the plasmids can be transferred into the cell after electroporation. Several hundred volts across a distance of several millimeters are typically used in this process. Afterwards, the cells have to be handled carefully until they have had a chance to divide producing new cells that contain reproduced plasmids. This process is approximately ten times as effective as *chemical transformation*.[1][3]

This procedure is also highly efficient for the introduction of foreign genes in tissue culture cells, especially mammalian cells. For example, it is used in the process of producingknockout mice, as well as in tumor treatment, gene therapy, and cell-based therapy. The process of introducing foreign DNAs into eukaryotic cells is known as transfection. Electroporation is highly effective for transfecting cells in

suspension using electroporation cuvettes. Electroporation has proven efficient for use on tissues in vivo, for in utero applications as well as in ovo transfection. Adherent cells can also be transfected using electroporation, providing researchers with an alternative to trypsinizing their cells prior to transfection.

Contents

[hide]

Laboratory practice[edit]

Cuvettes for electroporation. These are plastic with aluminium electrodesand a blue lid. They hold a maximum of 400 µl.

Electroporation is done with **electroporators**, appliances that create an electro-magnetic field in the cell solution. The cell suspension ispipetted into a glass or plastic cuvette which has two aluminum electrodes on its sides.

For bacterial electroporation, typically a suspension of around 50 microliters is used. Prior to electroporation it is mixed with the plasmid to be transformed. The mixture is pipetted into the cuvette, the voltage and capacitance are set, and the cuvette is inserted into the electroporator. Immediately after electroporation, one milliliter of liquid medium is added to the bacteria (in the cuvette or in an eppendorf tube), and the tube is incubated at the bacteria's optimal temperature for an hour or more to allow recovery of the cells and expression ofantibiotic resistance, followed by spreading on agar plates.

The success of the elecroporation depends greatly on the purity of the plasmid solution, especially on its salt content. Solutions with high salt concentrations might cause an electrical discharge (known as arcing), which often reduces the viability of the bacteria.

For a further detailed investigation of the process more attention should be paid to the output impedance of the porator device and theinput impedance of the cells suspension (e.g. salt content). As the process needs direct electrical contact between the electrodes and the suspension, and is inoperable with isolated electrodes, obviously the process involves certain electrolytic effects, due to small currents and not only fields.

Electroporators[edit]

Benchtop electroporators are generally used as common lab equipment, residing atop a central bench or hood. Some units offer the possibility of electroporating multiple samples at the same time with special electrode assemblies that fit into multi-well cell culture dishes. Benchtop electroporators can

also be set to different operating parameters, allowing researchers to optimize their field strengths depending on the cell type and whether or not the cell has a cell wall.

Electroporators have been used on a wide range of cells - including *E. coli* (for transformation) and mammalian cells such as neurons, astrocytes, neuroglia, lymphocytes, monocytes, fibroblasts, epithelial and endothelial cells from humans, mice, rats and monkeys (for transfection).

Medical applications[edit]

The first research looking at how electroporation might be used on human cells was conducted by researchers at Eastern Virginia Medical School and Old Dominion University, and published in 2003.[4]

The first successful treatment of malignant cutaneous tumors implanted in mice was completed in 2007 by a group of scientists who achieved complete tumor ablation in 12 out of 13 mice. They accomplished this by sending 80 pulses of 100 microseconds at 0.3 Hz with an electrical field magnitude of 2500 V/cm to treat the cutaneous tumors.[5]

A higher voltage of electroporation was found in pigs to irreversibly destroy target cells within a narrow range while leaving neighboring cells unaffected, and thus represents a promising new treatment for cancer, heart disease and other disease states that require removal of tissue.[6] IRE has since proven effective in treating human cancer, with surgeons at Johns Hopkins and other institutions now using the technology to treat pancreatic cancer previously thought to be unresectable.[7]

Electroporation can also be used to help deliver drugs or genes into the cell by applying short and intense electric pulses that transiently permeabilize cell membrane, thus allowing transport of molecules otherwise not transported through a cellular membrane. This procedure is referred to as electrochemotherapy when the molecules to be transported are chemotherapeutic agents or gene electrotransfer when the molecule to be transported is DNA. Scientists from Karolinska Institute and the University of Oxford use electroporation ofexosomes to deliver siRNAs, antisense oligonucleotides, chemotherapeutic agents and proteins specifically to neurons after inject them systemically (in blood). Because these exosomes are able to cross the blood brain barrier this protocol could solve the issue of poor delivery of medications to the central nervous system and cure Alzheimer's, Parkinson's Disease and brain cancer among other diseases.[8]

A recent technique called non-thermal irreversible electroporation (N-TIRE) has proven successful in treating many different types of tumors and other unwanted tissue. This procedure is done using small electrodes (about 1mm in diameter), placed either inside or surrounding the target tissue to apply short, repetitive bursts of electricity at a predetermined voltage and frequency. These bursts of electricity increase the resting transmembrane potential (TMP), so that nanopores form in the plasma membrane. When the electricity applied to the tissue is above the electric field threshold of the target tissue, the cells become permanently permeable from the formation of nanopores. As a result, the cells are unable to repair the damage and die due to a loss of homeostasis.[9] N-TIRE is unique to other tumor ablation techniques in that it does not create thermal damage to the tissue around it.

Contrastingly, reversible electroporation occurs when the electricity applied with the electrodes is below the electric field threshold of the target tissue. Because the electricity applied is below the cells' threshold, it allows the cells to repair their phospholipid bilayer and continue on with their normal cell functions. Reversible electroporation is typically done with treatments that involve getting a drug or gene (or other molecule that is not normally permeable to the cell membrane) into the cell. Not all tissue has the same electric field threshold; therefore careful calculations need to be made prior to a treatment to ensure safety and efficacy.[10]

One major advantage of using N-TIRE is that, when done correctly according to careful calculations, it only affects the target tissue. Proteins, the extracellular matrix, and critical structures such as blood vessels and nerves are all unaffected and left healthy by this treatment. This allows for a quicker recovery, and facilitates a more rapid replacement of dead tumor cells with healthy cells.[11]

Before doing the procedure, scientists must carefully calculate exactly what needs to be done, and treat each patient on an individual case-by-case basis. To do this, imaging technology such as CT scans and MRI's are commonly used to create a 3D image of the tumor. From this information, they can

approximate the volume of the tumor and decide on the best course of action including the insertion site of electrodes, the angle they are inserted in, the voltage needed, and more, using software technology. Often, a CT machine will be used to help with the placement of electrodes during the procedure, particularly when the electrodes are being used to treat tumors in the brain.[12]

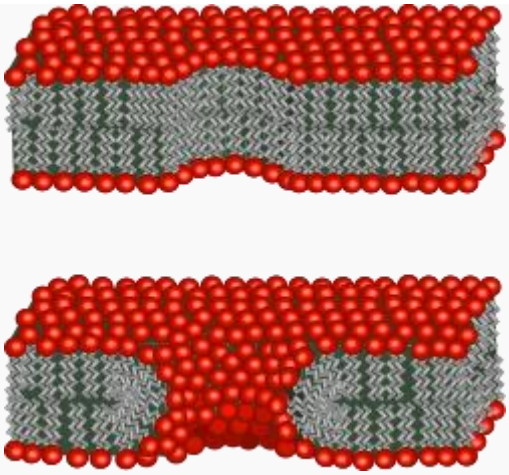
The entire procedure is very quick, typically taking about five minutes. The success rate of these procedures is high and is very promising for future treatment in humans. One disadvantage to using N-TIRE is that the electricity delivered from the electrodes can stimulate muscle cells to contract, which could have lethal consequences depending on the situation. Therefore, a paralytic agent must be used when performing the procedure. The paralytic agents that have been used in such research are successful; however, there is always some risk, albeit slight, when using anesthetics.

A more recent technique has been developed called high-frequency irreversible electroporation (H-FIRE). This technique uses electrodes to apply bipolar bursts of electricity at a high frequency, as opposed to unipolar bursts of electricity at a low frequency. This type of procedure has the same tumor ablation success as N-TIRE. However, it has one distinct advantage, H-FIRE does not cause muscle contraction in the patient and therefore there is no need for a paralytic agent.[13]

Physical mechanism[edit]

Further information: Lipid bilayer mechanics

Electroporation allows cellular introduction of large highly charged molecules such as DNA which would never passively diffuse across the hydrophobic bilayer core.[1] This phenomenon indicates that the mechanism is the creation of nm-scale water-filled holes in the membrane. Although electroporation and dielectric breakdown both result from application of an electric field, the mechanisms involved are fundamentally different. In dielectric breakdown the barrier material is ionized, creating a conductive pathway. The material alteration is thus chemical in nature. In contrast, during electroporation the lipid molecules are not chemically altered but simply shift position, opening up a pore which acts as the conductive pathway through the bilayer as it is filled with water.



Schematic showing the theoretical arrangement of lipids in a hydrophobic pore (top) and a hydrophilic pore (bottom).

Electroporation is a multi-step process with several distinct phases.[14] First, a short electrical pulse must be applied. Typical parameters would be 300-400 mV for < 1 ms across the membrane (note- the voltages used in cell experiments are typically much larger because they are being applied across large distances to the bulk solution so the resulting field across the actual membrane is only a small fraction of the applied bias). Upon application of this potential the membrane charges like a capacitor through the migration of ions from the surrounding solution. Once the critical field is achieved there is a rapid localized rearrangement in lipid morphology. The resulting structure is believed to be a “pre-pore” since it is not electrically conductive but leads rapidly to the creation of a conductive pore.[15] Evidence for the existence of such pre-pores comes mostly from the “flickering” of pores, which suggests a transition between conductive and insulating states.[16] It has been suggested that these pre-pores are small

(~3 Å) hydrophobic defects. If this theory is correct, then the transition to a conductive state could be explained by a rearrangement at the pore edge, in which the lipid heads fold over to create a hydrophilic interface. Finally, these conductive pores can either heal, resealing the bilayer or expand, eventually rupturing it. The resultant fate depends on whether the critical defect size was exceeded[17] which in turn depends on the applied field, local mechanical stress and bilayer edge energy.

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ABAMAV - watch attached

Breastlife - watch attached