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MEXICO – Preliminary Labs:

EPH Technologies, Inc. has entered into clinical trials in Mexico under the Direct supervision of Dr. Luis Barcenas at his facility IIDCA. The purpose of the clinical trials is to gain safety approval and subsequently distribution permission via the authority, COFEPRIS, in Mexico. The strong affiliation between the medical authorities in the United States and Mexico has culminated in progressively higher standards for human safety in products. It is especially strict when it comes to supplements that may be considered treatments for disease in humans. During the course of the clinical trial it has been necessary for strict adherence to protocol. All procedures, documentation, test results, lab observations, patient interactions etc. must be conducted, structured and documented according to protocol. While protocol is important in order to comply with the approval and distribution permitting process it is also important for our own research and development to be assured that all results and interactions with the product/s are uncontaminated.

At the very early stages of the process it was certified that the EPH E3 product was non-toxic. Lab testing (Murine model) was conducted on at least two separate occasions with the assistance of UNAM Veterinary and Chemistry Faculties. this made it possible to represent the molecular data desired by the lab and ultimately the authorities to be collected. Such data cannot be obtained without superior technology. Fortunately, UNAM has state of the art equipment and the E3 product is a new, exciting and unique discovery garnering great intrigue from the Biochemical scientists. The product Toxicology testing concluded without concern. E3 is now officially certified as Non-Toxic. While we have known E3 is non-toxic for a very long time, it is necessary to test using standard medical industry protocol under the strict supervision of a certified and recognized facility for it to be considered credible and reliable.

Following is the report data (Translated to English) from the clinical trial team.

To achieve the goal for product analysis ImmunoFX as a therapeutic agent in humans has been important to identify the chemical components of the compound. We have conducted several preliminary tests by the Institute for Research and Development of Applied Science IIDCA and the results are:

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Instituto de Investigación y Desarrollo de Ciencia Aplicada

Reference Product Physicochemical analysis IIDCA :

Proximal chemical analysis:

Moisture	96%
Ash	0.0%
Excerpt Ethereal	0.8%
Nitrogen Derivatives	2.8%
Carbohydrates	0.4%
Raw Fiber	0.0%

(OMAACH/90)

Microbiological Analysis:

Aerobic Account	720 CFU/ml
Total Count of Yeast	10 CFU/ml

Microbiological Analysis Pathogens:

Staphylococcus Aureus	Absent
Pseudomonas Aeruginosa	Absent
Escherichia Coli	Absent
Salmonella	Absent
Staphylococcus Aureus	Absent

Physicochemical Analysis:

Appearance	Opalescent Aqueous Solution
pH	6.8

After this toxicity test stage we performed the following tests in different faculties of the UNAM:

Microbiological Analysis in the department of preventive medicine and public health at the veterinary faculty of the UNAM.

Results:

Aerobic Bacterial Count In Plate	250 CFU/ml
Molds and Yeast Count	Absent
Total Coliform Bacteria	Absent
Fecal Coliform Bacteria	<3 MPN/ml Absent
Total Coliforms Board	Absent
Salmonella spp	Absent in 25g
S. Aureus	<10 CFU/ml (estimated)

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These results show that the test product, ImmunoFX, does not contain pathogenic bacteria.

Results of the Department of Food and Biotechnology, Faculty of Chemistry UNAM.

Chemical Composition Results:

Parameter (g/100 ml)	
Humidity	99.10%
Ash	0.06%
Protein (factor 6.25)	0.82%
Fat (Ether Extract)	Not Detected
Dietary Fiber	Not Detected
Carbohydrates (by difference)	0.02%

Amino Acid Profile:

Amino Acid g g	aa/100ml Protein	aa/100g Sample
Threonine	7.49	0.067
Aspartic Acid	8.10	0.072
Serina	8.61	0.086
Glutamic Acid	18.91	0.168
Alanine	2.70	0.024
Cystine	Not Detected	Not Detected
Valine	6.64	0.059
Methionine	Not Detected	Not Detected
Isoleucine	3.64	0.032
Tyrosine	0.27	0.002
Phenylalanine	3.81	0.034
Lysine	0.046	5.22
Histidine	0.31	0.003
Arginine	11.35	0.101
TOTAL	87.50	0.779

Assessing the results require analysis mass spectrometry and gas chromatography coupled to mass or liquids, which have asked the Institute of Chemistry of UNAM to determine the detailed composition of the substance. This procedure is awaiting response from the institute

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Instituto de Investigación y Desarrollo de Ciencia Aplicada

I I D C A

Instituto de Investigación y Desarrollo de Ciencia Aplicada

Results of the Department of Animal Nutrition and Biochemistry, Laboratory of Animal Nutrition and Biochemistry:

Total Solids	0.079%
pH	8.86-8.93
Urea	Negative
Crude Protein	0.085%

Through tests conducted in this laboratory we could not get much information because the methods used require large volumes of liquid.
In this test however we can see that there are proteins in the compound.
The study of Urea us doubtful as the ImmunoFX compound has a strong smell of ammonia thus conclude that we have urea in its components, though further studies may determine.

Phase II of the research protocol report ImmunoFX test compound in a volunteer population group:

Phase II protocol began with a sample population of 48 people which take from several days ago indicated dose of test product at the same time, between 8 and 9 pm.
In this population sample on day one of the study, we performed complete blood and urine to set parameters before and after treatment, and the evolution of physiological markers during testing.
It is noteworthy that the test product does not have any reference to objectively measurable or perceptible effects so a daily survey is conducted to determine the likely effects noticeable by the patient and can actually result cause – effect and as interact in the body. At the same time it must be kept under close clinical surveillance to prevent health problems.
Similarly we have selected a smaller group of volunteers who are taking a placebo for comparative analysis of the dynamics of physiological markers ImmunoFX allowing us to advance the assessment known as double blind studies in phase. These data are representative when the final statistical analyzes were performed at the conclusion of our study.
The dose should take the volunteers has been arbitrarily determined according to the references of patients seen in EPH Technologies, because we are in a testing phase. To determine the exact dose is important to evaluate the results, so that this compound interacts with the body. Therefore we suggested daily dose of 5ml of patients ImmunoFX volunteer community health club.
In this type of study variables influencing the results emerge, it is for this reason that we have chosen a representative sample size statistically speaking. The results of the laboratory samples require a rigorous medical examination at a comparative level, which is in process.
During this time of trial we applied a questionnaire that allows us to see the adverse or positive reactions of compound ImmunoFX in patients participating in this trial, which gives us important information about how patients regard it feel to making this compound.
The results of these surveys tell us that some people have the first shots in stomach upset, which

goes to the passage of a few days. In this sense we have had minimal dropout rate because the tolerance of these individuals was zero, which is always considered in the protocols in population groups.

Adverse reactions that we report are not the same for all individuals, some of them do not report such reactions. There are people that unlike earlier we reported increased energy capacity decrease fatigue associated with the consumption of this item and a favorable overall health. These data will be useful when we relate with physiological markers in all the tests we do.

In the same way some people report that they have had no apparent change to consume this compound, neither negative nor positive, i.e. have no discernible changes detected.

A very interesting fact is that all people are part of the protocol did not submit pictures related to seasonal respiratory diseases and reported that at this time always have flu pictures.

However to generate conclusions is important to wait at the end of the test to make an assessment of both positive and negative physiological changes in people tested and to determine the effects of compound ImmunoFX in humans.

Comments:

Importantly, the relevance of human labor involved in these studies, dealing with individuals is very complex as a bridge of communication and trust that must be established is not trivial. In this sense we have held information and questions that individuals do not perceive or feel that we are taking profits at their expense and to voluntarily participate in this type of testing generates a long-term welfare in your community, generating sessions therefore a commitment.

Note that the people involved in this phase of the protocol have deep confidence in Dr. Barcenas, that trust has been built on many years of work and genuine perception, which widely exists in these people that work Doctor and his team there is real concern in improving the health of the Mexican population and establishing community ties that alone generate emotional wellbeing which leads to improvements in health.

Report Protocol III. Conducting clinical data recovery installed with various pathologies in 30 patients

At this stage of the protocol 30 patients, 10 with different types of cancer, 10 metabolic diseases and 10 were selected immunocompromised patients.

The development of this test is in progress and the results reflect that the same effects were obtained and the evolution of patients having treatment under this test, it is important to wait for the final results to make an indicative analysis of the effects of product ImmunoFX test in such patients and correlate clinical effectiveness of their drug use, i.e. in which cases are recommended and which are not.

I I D C A

Instituto de Investigación y Desarrollo de Ciencia Aplicada

Protocol II Tox report on the effects of complement – EPHT ImmunoFX being tested in animals.

Justification for the trial:

The test plug – EPHT ImmunoFX consists of a nonspecific protein mixture derived from extremophile bacteria reporting , improves the immune response to various diseases , by repairing and renaturation of proteins in diseased cells.
Studies show the qualities of extremophile proteins in the therapeutic application in several diseases in both veterinary medicine and human.
For this reason we have determined the relevant test animal study, as a first approach about the physiological and cellular interaction of this compound.

Effects of Dunkin Hartley guinea pigs ImmunoFX in ICR mice and CD1:

The objective of this test is to analyze the responses of organisms exposed to dietary supplement orally in solution with the reference nomenclature ImmunoFX – EPHT.

Assays were performed in experimental groups of 5 mice, and 5 guinea pigs for acute toxicological analysis consisting of 2 days, sub-acute toxicity tests worked with a sample of 10 mice, and 5 guinea pigs for thirteen days. The administration of serum – EPHT ImmunoFX test solution was orally in drinking water.

Individuals in the sample were selected under the standards required for these tests, considering the weight, sex and age.

At the end time of acute toxicity test organisms were sacrificed for pathological analysis of some organs and tissues, the same for bodies under sub-acute toxicity.
Quantification of dose to be added to the drinking water of the animals was as follows:

Considering the average specimen weight 30g and water consumption of 3 ml, water dilution not acidified in sub-acute sample was 0.6 ml in 1 liter of drinking water, to match the delivered dose to human adult-type (70 kg).
For acute sample was given 10 times the “therapeutic” dose suggested by which 6 ml were added to 1 liter of water, in the case of mice.
In the case of the guinea pigs was calculated by the mean weight in the same manner as in the case of mice.

Tests of Acute and sub-acute
Acute toxicity is to determine the effects of very high doses of the substance to be studied in this case – EPHT ImmunoFX. Usually, the end point of the study is the animal’s death and acute toxicity is expressed by the lethal dose 50, which represents more or less the dose of the substance that causes death in 50% of animals. Upon completion of this phase the animals were sacrificed for necropsy.
In the sub-acute phase observation of the animals was carried out for 13 days by administering the

substance daily to 10 mice, and 5 guinea pigs, after which time the animals were sacrificed and sent to the pathology department of the veterinary faculty at UNAM who undertake the macroscopic examination of the organs and tissues and taking specimens for pathological examination, we'll have the results in 20 days.

Observations:

During the acute toxicity test which consists of two days, the specimens tested both mice and guinea pigs decreased normal consumption of drinking water comprising ImmunoFX serum and non-acidified water.
Showed no other side effects so we believe that this behavior is due to the change in taste and smell normal water, considering that rodents are very sensitive to changes in these parameters.
No changes were observed in physical activity, i.e. neither increased nor decreased the energy values both day and night.
The specimens gained weight normally according to the standards and their food intake was also normal.
Neither did not die during the test specimens so euthanasia was used in all cases. This represents that in the acute phase of the test specimens ImmunoFX 10 (5 mice guinea pigs, and 5) no acute toxic effects causing the death of the individuals tested were presented. The sacrificed specimens were sent to the pathology unit of the Faculty of Veterinary Medicine, UNAM for anatopatológico analysis.
During the time of testing in sub-acute (10 mice guinea pigs, and 5) no behavioral changes or physical activity, only continued with low water consumption were observed apparently showed no change in food intake habits. None of the specimens died of sub-acute toxicity test with ImmunoFX, so were sacrificed.
Almost all specimens continued their normal development was observed that only one of the specimens of mouse did not reach the expected weight.
The bodies were sent to the pathology department and the results will be delivered in 20 days.

Overall results of postmortem studies of acute toxicity test in mice and guinea pigs
In the macroscopic findings of the mice not apparent lesions in any organ was found, however, in the microscopic studies were observed in three mice lesions in the lungs, blood vessels dilated by erythrocytes and rupture of alveolar sacs in the presence of clavas retraction two mice have emphysema. Four mice exhibited in the kidney tubular cells epithelium increased in size with pyknotic nuclei and lightly congested blood vessels.
In the case of the guinea pigs animals had good body condition however the lungs were diffusely dark red varying degrees (congestion), the liver degrees of mild to moderate degeneration observed.
In the case of specimens exposed to sub-acute toxicity have not yet pathological results.

I I D C A

Instituto de Investigación y Desarrollo de Ciencia Aplicada

Effects of ImmunoFX in New Zealand Rabbits:

Specimens of New Zealand rabbits were achieved by permissions and requirements of the Establishment Unit, Faculty of Veterinary Medicine, UNAM specifically for purposes of our test, which will begin next week.
These specimens will be treated with oral ImmunoFX direct manner, proportional to their weight in relation to the suggested daily dose in humans and not be dissolved in the drinking water dose.
The test will last 15 days at 8 rabbits in which observe the subacute toxicity and then send you the corpses pathology unit as we have done with mice and guinea pigs .

References

Alain , K., & Querellou , J. (2009). Cultivating the uncultured : limits , advances and future challenges . Extremophiles , 13 (4) , 583-594 .
Arakawa , T., and S. N. Timasheff . “The stabilization of proteins by osmolytes . ” Biophysical Journal 47.3 (1985) : 411-414 .
Blomberg A. : Metabolic surprises in Saccharomyces cerevisiaea During adaptation to saline conditions : questions , some answers and a model . FEMS Microbiol Lett 182 , 1-8 (2000) .
Chakravorty , Debamitra , Ashwinee Shreshtha Kumar , VR Sarath Babu, and Sanjukta Patra . ” Molecular Evolution of Extremophiles . ” Extremophiles : Sustainable Resources and Biotechnological Implications (2012) : 1-27 .
Conway de Macario E. & Macario AJL : Heat -shock response in Archaea. Trends Biotechnol 12 , 512-518 (1994) .
Chung N., G. Jenkins, Hannun YA , Obeid LM & J. Heitman : Sphingolipids signal heat stress-induced ubiquitin -dependent proteolysis . J Biol Chem 275 , 17229-17232 (2000) .
A. Ciechanover , A. Orian & Schwartz AL : Ubiquitin -mediated proteolysis : biological regulation via destruction . BioEssays 22 , 442-451 (2000) .
Ferreira , AC , Nobre , MF, Moore , E., Rainey , FA, Battista , JR, & da Costa , MS (1999) . Characterization and radiation resistance of new isolates of Rubrobacter radiotolerans and Rubrobacter xylanophilus . Extremophiles , 3 (4) , 235-238 .
Feller , Georges. “Life at low temperatures : is disorder the driving force ? . ” Extremophiles 11.2 (2007) : 211-216 .
Georlette , D., Blaise , V. , Collins , T., D’ Amico , S., Gratia , E., Hoyoux , A. , & … Gerday , C. (2004) . Some like it cold : biocatalysis at low temperatures . FEMS Microbiology Reviews , 28 (1) , 25-42 .
Sing & B. Gupta RS : Cloning of the HSP70 gene from Halobacterium marismortui : relatedness of archaebacterial HSP70 to its eubacterial homologs and a model for the evolution of the HSP70 gene. J Bacteriol 174 , 4594-4605 (1992) .
Gupta , RS : Protein phylogenies and signature sequences : a reappraisal of evolutionary relationships Among archaebacteria , eubacteria , and eukaryotes . Microbiol Mol Biol Rev 62 , 1435-1491 (1998) .
Jovell RJ , AJL Macario & E. Conway de Macario : ABC transporters in Archaea : two genes

encoding homologs of the nucleotide- binding components in the methanogen Methanosarcina mazei S -6. Gene 174 , 281-284 (1966) .

Klenk , H -P . , R. A. Clayton , J.F. Tomb , O. White, K. E. Nelson , K.A. Ketchum , R. J. Dodson , M. Gwinn , E.K. Hickey , J. D. Peterson , D.L. Richardson , A.R. Kerlavage , D.E. Graham , N.C. Kyrpides , R. D. Fleischmann , J. Quackenbush , N. H. Lee , G.G. Sutton , S. Gill , E.F. Kirkness , B. A. Dougherty , K. McKenney , M.D. Adams , B. Loftus , S. Peterson , C.I. Reich, L. K. McNeil , J.H. Badger, A. Glodek , L. Zhou , R. Overbeek , J.D. Gocayne , J. F. Weidman , L. McDonald , T. Utterback , M.D. Cotton, T. Spriggs , P. Artiach , B. P. Kaine , S. M. Sykes, P. W. Sadow, K. P. D' Andrea , C. Bowman , C. Fujii , SA Garland, T. M. Mason , G.J. Olsen, C. M. Fraser , H. O. Smith , C.R. Woese & J. C. Venter : The complete genome sequence of the hyperthermophilic , sulphate-reducing archaeon Archaeoglobus fulgidus . Nature 390 , 364-370 (1977) .

MT Madigan , JM Martinko and J. Parker: Brock Biology of Microorganisms , Ninth Edition. Prentice Hall , NJ (2000) .

AJL Macario & E. Conway de Macario : Heat shock response: overview. In: The encyclopedia of stress. Ed : G. Fink . Academic Press, CA . pp . 350-357 (2000) .

Mayerhofer LE, E. Conway de Macario & Macario AJL : Conservation and variability in Archaea : protein antigens with tandem repeats encoded by a cluster of genes with common motifs in Methanosarcina mazei S -6. Gene 165 , 87-91 (1995) .

Mayerhofer LE, E. Conway de Macario , AJL Macario R. & Yao : Structure , organization , and expression of genes coding for envelope components in the archaeon Methanosarcina mazei S -6. Arch Microbiol 169 , 339-345 (1998) .

AJL Macario & E. Conway de Macario : Chaperone proteins . In: The encyclopedia of stress. Ed : G. Fink . Academic Press, CA . pp . 429-435 (2000) .

Macario AJL Macario & E. Conway : The archaeal molecular chaperone machine: Peculiarities and paradoxes . Genetics 152 , 1277-1283 (1999) .

Macario AJL , Dugan CB & E. Conway de Macario : A dnaK homolog in the archaebacterium Methanosarcina mazei S6. Gene 108 , 133-137 (1991)

AJL Macario & E. Conway de Macario : Stress and molecular chaperones in disease . Intl J Clin Lab Res 2000 (in press)

Morel Y & R. Barouki : Repression of gene expression by oxidative stress. Biochem J 342 , 481-496 (1999) .

Nakasone , K., Ikegami , A. , Kato , C., Usami , R., & Horikoshi , K. (1998) . Mechanisms of gene expression controlled by pressure in deep -sea microorganisms . Extremophiles , 2 (3) , 149-154 .

Oger , Philippe M., and Anaïs Cario . "Adaptation of the membrane in Archaea. " Biophysical chemistry (2013).

Patel GB & GD Sprott : Archaeobacterial ether lipid liposomes (archaeosomes) as novel vaccine and drug delivery systems . Crit Rev Biotechnol 19 , 317-357 (1999) .

GB Patel , BJ Agnew, L. Deschatelets , Fleming LP & Sprott GD : In vitro assessment of stability for archaeosome develop developing oral delivery systems . Intl J Pharmaceuticals 194 , 39-49 (2000) .

Pikuta , E. V. , Hoover , R. B. , & Tang, J. (2007) . Microbial extremophiles at the limits of life .

Critical reviews in microbiology , 33 (3) , 183-209 .

Szilágyi A. & P. Zádodszky : Structural Differences Between mesophilic , moderately thermophilic and extremely thermophilic protein subunits : results of a comprehensive survey . Structure 8 , 493-504 (2000) .

T. Schindler , PL Graumann , D. Perl, FX Schmid S. Ma & Marahiel MA : The family of cold shock proteins of Bacillus subtilis. J Biol Chem 274 , 3407-3413 (1999) .

van de Vossenberg , JL , Driessen AJ & Konings WN (1998) . The essence of being extremophilic : the role of the unique archaeal membrane lipids . Extremophiles , 2 (3) , 163-170 .

Van Den Burg , B. (2003) . Extremophiles as a source for novel enzymes . Current Opinion in Microbiology , 6 (3) , 213-218 .

N. Wang , K. & Yamanaka M. Inouye : CSPI , the ninth member of the CspA family of Escherichia coli , is induced upon cold shock. J Bacteriol 181 , 1603-1609 (1999) .

Sengupta , D., & KChattopadhyay , Madhab (2013) . Metabolism in bacteria at low temperature: A recent report . J. Biosci , 38 (2) , 1-4.

DOES WHAT PCR. ” Polymerase Chain Reaction. ” Journal of Investigative Dermatology 133 (2013) .

Yao R., AJL Macario & E. Conway de Macario : An archaeal S -layer gene homolog with repetitive subunits . Biochim Biophys Acta 1219 , 697-700 (1994) .