The current model in drug discovery is based on the ligand-receptor paradigm as the drugable unit. The common analogy of drug action is that of a lock and key, with a drug acting as a selective 'key' that fits into the 'lock' of a specific drug target. And while this selectivity paradigm has yielded efficacious drugs, many drugs act via modulation of multiple proteins: Many effective drugs in oncology, psychiatry and antimicrobials act on multiple, rather than single targets. Systems biology has revealed physiological function as a robust network structure that strongly suggests that exquisitely selective compounds, compared with multitarget drugs, may exhibit lower than desired clinical efficacy and induce more adverse events. Chemically complex botanically-based medicines inherently act on multiple sites and therefore have never been fully understood in a reductionist ligand-receptor model well. However, botanical medicines fit into a network pharmacology paradigm seamlessly. Viewing mixtures of natural products through the lens of network biology may provide insights into how we can improve drug discovery and utilize botanical medicines for complex diseases.

**Key words:** science, chemical complex, natural products, pharmacology
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Exo-methylene-fÁ-butyrolactone natural products are abundant in nature that display broad biological profiles, among them high tumor activity. Despite their generally high cytotoxicity, these compounds are considered to be unsuitable for drug development, owing their high but unspecific reactivity with biological nucleophiles. We started a program that combines the synthesis of exo-methylene-fÁ-butyrolactone natural products focusing on asymmetric catalysis,[1] synthesis of peptide foldamers containing fÁ-amino acids[2] as carriers and evaluation of hybrids derived thereof to selectively recognize specific cancer cells. Thus, following our approach we are confident that a large body of biologically highly potent, but greatly neglected natural products and analogs might become promising for drug development in medicinal chemistry.

Key words: synthesis, evaluation, exo- methylene butyrolactone, natural product, drug
Medicinal plants of the brazilian Caatinga biome: from popular use to scientific studies (Master Lecture)

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The Caatinga biome (semi-arid vegetation) is a highly threatened biome covering a vast area in Northeastern Brazil and is the source of few studied natural resources. Many medicinal plant species from Caatinga are widely known and used in folk medicine and for commercial manufacturing of phytotherapeutic products. Few ethnomedical and pharmacological studies have been undertaken in this region, in spite of the great cultural and biological diversity to be found there. The purpose of this lecture is to present results of research carried out at the Federal University of Vale do São Francisco with the species Annona vepretorum, atemoia (A. cherimola Mill. x A. squamosa L.), Selaginella convoluta, Hymenaeamartiana, Bromelialaciniosa, Encholiriumspectabile and Neoglazioviavariegata, typical species from the Caatinga biome. The fundamentals of the main methods for assessing the antinociceptive, anti-inflammatory, antiulcerogenic and central nervous system activity of a natural substance or plant extracts will be presented. The main chemical constituents isolated from extracts of these species will be presented as well as the chemical composition of some essential oils. Regarding the pharmacological activity, results will be presented on the antinociceptive, anti-inflammatory, antiulcerogenic and on the central nervous system. Furthermore, the in vitro photoprotective and antioxidant activity was also investigated. The exact mechanism involved in the antinociceptive and anti-inflammatory activities is not completely understood but, at least in part there is the participation of opioid receptors and inhibition of cyclooxygenase enzyme. Pharmacological and chemical studies are continuing in order to characterize the mechanism responsible for these effects.

Key words: medicinal plants, phytochemistry, pharmacology, natural products, caatinga
SPN-O-001  Therapeutic effects of policosanol and atorvastatin against global brain ischemia-reperfusion injury in gerbils

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El ictus isquémico constituye la tercera causa de muerte y la primera de discapacidad en la población adulta. El uso de agentes antiplaquetarios en el tratamiento del ictus isquémico ha reducido su recurrencia solo de forma moderada y se asocia a frecuentes eventos adversos por lo que la búsqueda de nuevas estrategias terapéuticas seguras y eficaces constituye una problemática actual. El policosanol, el extracto de semilla de uva y la atorvastatina son sustancias con reportados efectos antiplaquetarios. Este estudio compara los efectos preventivos del policosanol y el extracto de semilla de uva así como los efectos terapéuticos del policosanol con la atovastatina en un modelo de isquemia cerebral en Mongolian gerbils. Los resultados mostraron que policosanol y extracto de semilla de uva presentaron eficacias similares para prevenir la isquemia cerebral mientras que el policosanol y la atorvastatina resultaron similarmente efectivas para tratar la isquemia cerebral previamente inducida en Mongolian gerbils.

Key words: policosanol, atorvastatin, ischemia-reperfusion
SPN-O-002  Comparison of policosanol and grape seed extract on global cerebral isquemia in Mongolian gerbils

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Key words: policosanol, grape seed extract, ischemia-reperfusion
Champa (Campumanesia lineatifolia), a fruit with high potential to be used as a treatment of chronic diseases due to its chemical composition, is grown in the municipality of Miraflores (Boyaca, Colombia). Nowadays, post-harvest losses reach 97%, being this an opportunity to add value and to give sustainability to this crop. The aim of this work was to extract Champa's phenolic compounds and to assess its antioxidant capacity. Lyophilized fruit was subjected to a solid-liquid extraction (1:10), with water, ethanol and ethanol-water mixtures (3:7) as solvents at three temperature levels, 20, 50 and 70°C. Phenolic content and anti-oxidant activity were determined for the dry extracts. Extraction yields ranged from 26.85% to 50.07%, were the higher numbers were obtained with water as a solvent. At tested conditions, water showed no significant dependency on extraction temperature. Phenolic content increased with extraction temperature, in the range of 1469.32 ± 152.14 to 5272.51 ± 424.89 µg of gallic acid/g of lyophilized pulp, where the higher figures were obtained with ethanol-water mix (7:3). Anti-oxidant activity were found between 20.44 ± 2.57 to 51.01 ± 1.93 mM TEAC/g lyophilized pulp, being the water and the ethanol-water mix (7:3) the solvents with higher values. A significant effect on activity was observed with extraction temperature, with different trends depending on the solvent used. As a result of this research, It is concluded that Campumanesialineatifolia is a natural product with a high content of bio-active compounds, with a great potential to be used in agricultural, pharmaceutical and cosmetic industries.

**Key words:** campumanesialineatifolia, dpph, antioxidants, natural products
The present study investigates the hypoglycemic and hypolipidemic effect of oral administration of aqueous extract of "Estrella dorada" a endemic bush corresponding to the family of the Astaracea that grows in the South of Chile. In this study 36 Male Sprague-Dawley rats, body weight of 250-300 gr were divided into 4 groups. Diabetes was induced by three intra peritoneal injection of 75 mg/kg Alloxan. Four weeks after the first injection, the rats were fasted and blood glucose levels were recorded. Animals were considered to be diabetic when plasma glucose concentrations were higher than 200 mg/dl. The effect of the extract on the fasting serum glucose, total cholesterol, triglycerides levels, Lipid peroxides (TBAS) in plasma and in other biochemical parameters analyzed after a ten week treatment. Results are expressed as mean ± SD. The data were analyzed using multiple Test of Dunet (P <0.05). The infusion supply to diabetic rats decreased the blood glucose level(284.6±31.2 to 170.32±6.3 mg/dl) and values similar to the observed in control rats were found (140.6 ± 12.4 and 111.0 ± 1.7 mg/dl), as well as in diabetic control and diabetic treated rats(140.6 ± 12.4 and 111.0 ± 1.7 mg/dl). The data demonstrated that the aqueous extract of "Estrelladorada" was associated with antidiabetic and hypolipidemic effects on the experimentally induced diabetic rats.

Key words: hypoglycemic, hypolipidemic, diabetic rats
Oxidative stress and hypertension are considered factors precursors of cardiovascular diseases according to WHO are the leading cause of death worldwide, treatment exposes the patient to the continued use of drugs that lead to side effects, therefore it is required new therapeutic sources that decrease. The Passiflora genus comprises about 500 species that have wide use in folk medicine in many countries; including the species Passiflora quadrangularis Linn, properties are attributed to headache, hypertension, and diabetes (Dhawan, K., Dhawan, S & Sharma, 2004). According to the above in this study ACE inhibitory activity in vitro by the method of Simonetta Ronca Testoni (Guerra, C; Peñalosa, G; Mendez, J & Murillo, W. 2014), antioxidant activity by ABTS and preliminary phytochemical analysis (AFP) in the ethanol extract of the leaves of P. quadrangularis was evaluated, which will establish preform if it has effects on hypertension. Regarding the ACE inhibitory activity was established that the extract at a concentration of 0.05 mg / ml inhibited 70%, whereas the pattern (Captopril) to 1.0 mg / ml obtained only 15%; antioxidant activity with ABTS extract behaved similarly to the reference standard, with the PPA was also possible to establish the presence of saponins, flavonoids and alkaloids, of which its effect on hypertension is known (Rojas, 2009). According to the results it follows that the extract has hypotensive effect, which must be confirmed with in vivo studies.

**Key words:** passiflora quadrangularis l, antioxidant activity, hypertension, in vitro, preliminary
Ozone layer hole entails ultraviolet radiation increase on the earth surface, raising the risk of skin diseases, even cancer. The use of vegetal compounds is a new strategy to protect people against UV damages. Substances obtained from natural sources have a huge molecular diversity and biological functions. However, the evaluation of the toxic capacity of these compounds is required before its beneficial properties can be used. Cymbopogon citratus (DC) Stapf is consumed as a popular decoction and has several properties such as: anticancer, antitumor and antimutagenic. Many of these studies were assayed in total extract, while a less portion was assayed in fractioned extract. In this work, we tested the toxicity and the ability to protect against UVC light of the aqueous fractions obtained from Cymbopogon citratus. To do so, SOS Chromotest, RifR and Survival test were used in Caulobacter crescentus. Increasing concentrations of these fractions (0.1, 0.5, 1, 2 and 4 mg/mL) were applied continuously: before, during and after UVC exposure (45 J/m²). Results showed that this fraction was not cytotoxic and don’t possess any genotoxic properties. Aqueous fraction can’t decrease the induction of SOS phenomenon, survival and frequency of mutations in irradiated cells. Finally, the aqueous fraction of Cymbopogon citratus (DC) Stapf, do not protect DNA from primarily damage induced by UVC light.

Key words: cymbopogon citratus, aqueous fractions, sos chromotest, survival test, rifr test, caulobacter
The current investigation focuses attention on the antioxidant properties of aqueous extracts of marine algae Bryothamnion triquetrum and Halimeda opuntia, and the relationship of such properties with total polyphenol content, using different experimental in vitro antioxidant assessment models. We observed following results: Reducing power (concentration 128 mg/mL, B. triquetrum OD=2.798 and H. opuntia OD=0.800, ascorbic acid with 1 mg/mL OD=2.824), DPPH assays (B. triquetrum IC50=1.15 ± 0.06 and H. opuntia IC50=12.34 ± 0.30 mg/mL), inhibition of lipid peroxidation in rat brain homogenates (B. triquetrum CI50=5.09± 025 and H. opuntia CI50=1.25± 031 mg/mL) and the inhibition of red blood cell (RBC) haemolysis induced by AAPH (with 5 mg/mL B. triquetrum inhibited until 100 % and H. opuntia 24%). The contents of polyphenolic compounds was for B. triquetrum 51,21 ± 2,25 and H. opuntia 19,99 ± 1,12 µg GAE/ mg extract. It was demonstrated that B. triquetrum extract was more effective than H. opuntia in free radical scavenge and reducing power while H. opuntia was more effective in inhibition of lipid peroxidation in rat brain homogenates and the inhibition of red blood cell (RBC) haemolysis induced by AAPH. Both extractswerenototoxicin the model of acute toxicity of Artemiasalina. These results supporting itspossible application asphytodrugand /ornutraceuticalfor its antioxidant properties and low toxicity.

Key words: seaweed, natural antioxidants, halimeda opuntia, bryothamnion triquetrum, polyphenol
Los productos de origen vegetal son recursos renovables de múltiples usos para el hombre, entre los cuales se encuentran el uso medicinal y el toxicológico. La información obtenida de la investigación de compuestos de origen vegetal ayuda a comprender la fisiología y bioquímica de los organismos que los producen y poder lograr su mejor aprovechamiento desde el punto de vista científico, médico y económico. Dentro de la composición química de los productos vegetales se pueden considerar dos grupos: Productos del metabolismo primario y Productos del metabolismo secundario. Hay que destacar que muchos metabolitos secundarios desencadenan respuestas fisiológicas muy específicas, en muchos casos uniéndose a determinados receptores con una destacable complementariedad. Este hecho proporciona un importante incentivo y justificación para el estudio de nuevos productos naturales de origen animal o vegetal, como lo son los compuestos que se puedan encontrar en la planta de estudio: Dioscorea bartlettii Morton. El objetivo del trabajo es separar y purificar, en la medida que sea posible, los compuestos que se encuentran en las hojas de Dioscorea bartlettii Morton para posteriormente tratar de elucidar su estructura mediante espectroscopía de infrarrojo y de resonancia magnética nuclear. La separación y elucidación de dichos compuestos nos llevó a la conclusión de que uno de los componentes que más fácilmente se separó de dicha planta fue una saponina. La planta contiene también otros metabolitos pero no se separaron fácilmente.

**Key words:** dioscorea, separación, metabolitos secundarios, saponina
Polyphenols are secondary metabolites present in plants, which have gained popularity for its wide antioxidant properties. Numerous investigations have shown that the leaves of Moringa oleifera Lam are rich in this type of substance. The aim of the present work is to study the influence of operating conditions on the extraction of polyphenols by solid-liquid contact from the dried leaves of this plant. The experimental design considered the type of solvent (ethanolic solutions at different concentrations), temperature (50, 60 and 70°C) and the solute-solvent (1:15, 1:25 and 1:35 m/v) ratio. The extraction kinetics were also evaluated. From the investigated temperatures, the activation energy of the process was determined. From the above conditions, those that reported the best results were ethanol solution to 60% (v/v) as solvent, solute-solvent ratio 1:35 (m/v) with an extraction time of 30 minutes. For these conditions, a polyphenol content of 17.5 mg gallic acid equivalent/g dry mass with antioxidant activity of 35 mg Trolox equivalent/g dry mass, was obtained. The temperature effect was not significant in the investigated range. The nature of the leaching is physical considering that the activation energy was 11.47 ± 0.96 kJ/mol.

**Key words:** moringa, total polyphenols content, extraction kinetics, antioxidant activity
Biological control is a tactic to regulate plague populations through the use of natural elements. Tick infestation affects any animal species with important economical and health repercussions; In México, lost about 48 millions of dollars per year had been estimated because of this parasitosis in cattle; that is why the main purpose of this research was to evaluate the in-vitro effect of two organics and a microbial substance in the viability of adults ticks Otobius megnini y Rhipicephalus microplus. 150 adult ticks of each genre were collected from bovines; through those 15 groups with 10 subjects were made to prove three biological agents: essential oil of Juniperus communis, metanolic extract of Annona muricata seeds, purified spore of M. anisopliae Ma14 and distillated water with tween 80 to 0.1% for control groups (3) in four different levels of concentration using the immersion method. The results were established after 15 daily observations through a stereoscopic microscopy getting minimum and maximum mortality ratings of 50% and 100% respectively. According with those results it is possible to design integral plague management systems which consider natural elements like the entomopathogenic fungus Metarhizium anisopliae Ma14, soursop seeds (Annona muricata) and pinus (Juniperus communis) like agents with ixodicide potential against adult ticks of the genres Rhipicephalus microplus y Otobius megnini.

**Key words:** veterinary medicine, biological control, ectoparasites, ticks
SPN-O-014  Antiinflammatory effects of Abexol (D-002)

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La inflamación constituye un factor etiológico importante en el desarrollo de enfermedades crónicas como la osteoartritis, el cáncer, la hiperplasia prostática benigna, entre otras. Los anti-inflamatorios no esteroidales (AINEs) son medicamentos muy efectivos empleados para tratar la inflamación aguda, crónica y el dolor asociado; sin embargo su uso constituye una de las causas principales de úlcera gástrica. Por tal motivo la búsqueda de nuevas sustancias anti-inflamatorios con buen perfil de seguridad gastrointestinal constituye una problemática actual. Este trabajo evaluó los efectos del D-002, mezcla de seis alcoholes alifáticos de alto peso molecular obtenida de la cera de abejas, en modelos experimentales de inflamación y úlcera gástrica así como su posible gastrotoxicidad. La administración oral con D-002 en roedores redujo de forma significativa y moderada la inflamación en diferentes modelos experimentales, mientras que a diferencia de los AINES no produjo gastrotoxicidad y por el contrario protegió la mucosa gástrica.

Key words: Abexol, antiinflammatory effect, drug, natural product
SPN-O-015  Gastrotoxic and gastroprotector effects of Abexol  (D-002)

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Key words: Gastrotoxic, gastroprotector effects, Abexol  D-002
La enfermedad de reflujo gastroesofágico (ERGE), es una patología crónica, donde alrededor del 40 % de la población mundial la sufre. La ineficacia de los tratamiento con inhibidores de la bomba de protones y la aparición de reacciones adversas, hacen que sea este hecho una problemática de salud en la actualidad. Con el objetivo de evaluar el efecto protector esofágico del D-002 en diferentes modelos experimentales de esofagitis, se emplearon en el estudio ratas SD macho, a las cuales se les indujo, la esofagitis aguda, en la cual se evaluó el efecto citoprotector 5 h después de la inducción; el modelo de esofagitis gastro-duodeno-esofágico, el que fue evaluado 48 h después de la inducción y el modelo de reflujo esófago-duodenal con gastrectomía con una duración máxima de 14 días post operatorio. Los resultados demostraron el efecto protector esofágico del D-002 en los modelos experimentales, con excepción del modelo de reflujo gastro-duodeno-esofágico. Dicho efecto protector estuvo mediado por la inhibición de la peroxidación lipídica y proteica sin interferir en el volumen de secreción ni la acidez del jugo gástrico. Se concluye, que se demostró el efecto esófago protector del D-002 en la esofagitis inducida por reflujo gastro-esofágico y esófago-duodenal.

**Key words:** Abexol, D-002, esofagitis, gastroesophageal reflux, rats
Background and purpose: Osteoarthritis (OA) is a degenerative joint disease characterized by joint pain and dysfunction caused by a progressive and irreversible loss of articular cartilage, that affects hundreds of millions worldwide, mainly the elderly. The management of OA included a combination of non-pharmacological interventions and pharmacologic agents. Non selective non-steroidal anti-inflammatory drugs and cyclooxygenase 2 inhibitors provide symptoms relief in OA, but produce several adverse effects (AE), which supports the search for other options. Abexol, a mixture of higher aliphatic beeswax alcohols, has been shown to produce anti-inflammatory effects experimentally and clinically. This double-blind study investigated the effects of Abexol (50-100 mg/day) on patients with OA symptoms randomized to Abexol or placebo for 6 weeks. Methods: The primary efficacy outcome was the reduction of the total Western Ontario and McMaster Osteoarthritis Index (WOMAC) score. The secondary efficacy outcome was the reduction on pain, joint stiffness, physical activity scores, and the reduction of the VAS scores. The collateral efficacy outcome was reduction the use of rescue medications. Treatment tolerability was assessed. Results: At study completion Abexol (50-100 mg/day) significantly reduced the total WOMAC score versus baseline (p<0.00001, 77.1%) and placebo (p <0.0001, 65.4%), and also decreased the pain (54.9%), joint stiffness (76.8%) and physical activity (66.9%) scores versus placebo. After 1 week on therapy all benefits were significant already. In addition, Abexol significantly reduced the VAS score versus baseline (p<0.00001, 60.2%) and placebo (p <0.001, 46.8%). The use of rescue medication in Abexol (6/30, 20%) was lower (p <0.01) than in placebo (17/30; 56.7%). Treatment was well tolerated. Seven patients (2 Abexol, 5 placebo) reported moderate AE. Conclusions: Abexol (50-100 mg/day) given for 6 weeks improved OA symptoms and was well tolerated. Abexol could be a suitable option for managing OA symptoms, but this appreciation requires extensive further research.

Key words: Abexol, osteoarthritis
SPN-O-019 Carcinogenicity of D-004

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El D-004 es un extracto lipídico del fruto de la Roystonea regia (palma real), constituido por una mezcla de ácidos grasos, principalmente oleico, palmítico, láurico y mirístico, que ha mostrado prevenir la hiperplasia prostática inducida con testosterona, no con dihidrotestosterona, en roedores. El objetivo del presente estudio consistió en determinar si el tratamiento oral a largo plazo (24 meses) con D-004 (500-1500 mg/kg) muestra evidencias de toxicidad y/o carcinogenicidad en ratas SD adultas jóvenes de ambos sexos. Los animales se distribuyeron aleatoriamente en cuatro grupos (50/sexo/grupo), un grupo control al cual sólo se le administró el vehículo Tween 65/H2O y tres grupos tratados con D-004 a las dosis de 500, 1000 y 1500 mg/kg. Los tratamientos se administraron oralmente por intubación gástrica una vez al día durante 24 meses. La mortalidad (análisis de supervivencia), tiempo de aparición de tumores, signos clínicos, peso corporal, consumo de alimentos, parámetros hematológicos y de bioquímica sanguínea, peso de órganos e incidencia de lesiones neoplásicas y no neoplásicas no mostraron diferencias significativas ni tendencias con las dosis entre animales tratados y controles en ambos sexos. Por tanto, el D-004 no incrementa la frecuencia o acelera la aparición de lesiones neoplásicas benignas o malignas, ni induce la aparición de tumores inusuales a la especie, comportándose muy similar la frecuencia de tumores entre los grupos tratados y controles. Los resultados del estudio permiten concluir que el D-004 no es tóxico o carcinogénico en ratas SD tratadas durante 24 meses y que la dosis mayor (1500 mg/kg) se comportó como el nivel donde no se producen efectos tóxicos observables.

Key words: Carcinogenicity, D-004
Benign prostatic hyperplasia (BPH), common among elderly men, often causes lower urinary tract symptoms (LUTS). BPH/LUTS pharmacological therapies include 5α-reductase inhibitors, 1-adrenoreceptors blockers and phytotherapies, like saw palmetto (SP) lipid extracts. D-004, a lipid extract of Roystonea regia fruits, reduced experimental prostate hyperplasia in rodents and the International Prostate Symptoms Score (IPSS) as effectively as SP in a pilot trial in men with BPH. This work includes two clinical trials conducted in order to compare the effects of D-004 and SP in patients with BPH. In the first phase II study sixty-one patients with moderate BPH were double-blindly randomised to D-004 or SP (320 mg/day both) capsules for 4 months. Decrease on IPSS was the primary efficacy variable. Effects on prostate size, residual volume post-voiding and serum prostate specific antigen (PSA) were secondary outcomes. After 2 months on therapy, D-004 and SP lowered mean IPSS values versus baseline and this effect increased at study completion, when IPSS decreased 36.0% with D-004 and 34.1% with SP, without differences between groups. Both treatments did not modify prostate size, residual volume post-voiding. Two SP-treated patients withdrew from the study, one due to an adverse experience. One D-004- and 2 SP-treated patients experienced adverse experiences. These results were confirmed in a second phase III study. One hundred (100) eligible patients with mild to moderate BPH were double-blindly randomised to D-004 or SP (320 mg/day both) capsules for 24 weeks. After 2 months on therapy, D-004 and SP lowered mean IPSS values. This effect increased over the study, IPSS decreasing 70.3% with D-004 and 69.0 % with SP after 6 months on treatment, without differences between groups. Both treatments decreased significantly the residual volume post-voiding as compared to baseline. D-004, not SP, reduced significantly, but modestly, the prostate size and PSA values versus baseline. No significant between group differences were found. Summarizing D-004 (320 mg/day) was as effective as SP (320 mg/day) for decreasing LUTS in men with moderate BPH. Both treatments were well tolerated.

Key words: D-004, patient, benign prostatic hyperplasia
Scutellaria havanensis Jacq, species, known as Escudo de la Habana (Havana skullcaps), is a Cuban endemic plant and it is ethnomedically used for the treatment of scabies, and as diaphoretic and febrifuge. It is also cultivated for ornamental purposes. Several pharmacologic researches have confirmed that total extracts or flavonoids of Scutellaria genus have shown antiinflammatory, anxiolytic, antithrombotic, neuroprotective, anticonvulsivant, antioxidant, anticancer, antitumor, antiarthritic, antiviral, and antimicrobial properties. Little is known about the chemical composition of this plant. In this regards, the aim of this work was to carry out a phytochemical study of the ethereal, methanolic and aqueous extracts from aerial parts (leaves and stems) of S. havanensis species. Saponins, sugars, bitter principles, amino groups, alkaloids, flavonoids, coumarins, triterpenes and steroids, quinones and resins, in the extracts were qualitatively identified. The wogonin flavone was crystallized from the ethereal extract, which was structurally characterized by their GC-MS and NMR spectra. The identified groups of compounds in this plant, mainly flavonoids, could be of potential pharmacological interest.

**Key words:** scutellaria havanensis, phytochemical, flavonoids, wogonin, lamiaceae, gc-ms
On the islands of Palau, traditional plant medicines have been vital in the survival of the small isolated populations for generations, pre and post-foreign contact. This traditional medicinal knowledge has mostly been passed on through oral tradition. Through ethnobotanical and ethnomedical surveys, a Palauan panacea, that most widely used for the treatment of diabetes mellitus type II was analyzed for phytochemical content. Mangiferin, a compound found in significant quantities in this Palauan Traditional medicine with diabetes inhibition activity, was measured in aqueous preparations of traditional medicine and compared to those of organic solvent extracts. Aqueous extracts were most effective in isolating mangiferin. The insulin stimulation of the extracted mangiferin was measured using a -cell stimulation assay. Insulin secretion was induced by the extracted mangiferin in a glucose concentration dependent manner. Further clinical mechanistic trials were performed to assess blood sugar levels and insulin quantities in human serum with and without traditional medicine preparations during an Oral Glucose Tolerance Test. Insulin secretion and blood sugar levels showed potential of this Palauan traditional medicine as a therapeutic for treatment of type II Diabetes Mellitus.

Key words: palau, mangiferin, diabetes mellitus type ii
Artemisia afra is a shrub indigenous to South Africa and has been used for medicinal purposes. This study investigated the effects of leaf extracts on inflammatory/anti-inflammatory activity using RAW264.7 macrophages. Curcumin, a known anti-inflammatory phytochemical was also analysed concurrently to compare the relative effectiveness of these two phytoproducts. Interleukin 6 and nitric oxide were used as biomarkers for inflammation. Artemisia was extracted with ethanol. RAW264.7 macrophages were cultured to confluence in 96 well culture trays. Extracts were tested for anti-inflammatory activity by the following method: Medium was replaced with medium containing various concentrations of leave extracts in the presence and absence of 1 µg/ml LPS. After 24 hour incubation culture supernatants were removed and tested for nitric oxide production (Griess reagent) and interleukin 6 (DAS ELISA). The cells were tested for viability using a WST1 assay. The results of this study show that Artemisia afra is very effective in inhibiting the inflammatory activity due to LPS exposure (IC50 at 170 ug/ml). Curcumin, which is a highly purified component of turmeric, has an IC50 of 3 ug/ml. Data is also presented on NFκB and sex steroid receptor inhibitors on the anti-inflammatory activity of Artemisia afra.

Key words: anti-inflammatory activity, Artemisia afra
Introducción: La Revista Cubana de Plantas Medicinales (RCPM) fundada en 1996, ofrece a los profesionales del campo de la salud y sus afines, la posibilidad de publicar los resultados de sus investigaciones acerca del uso de las plantas medicinales. Expertos reconocidos median el proceso de arbitraje. La RCPM pretende contribuir a la prevención de enfermedades y al mejor manejo de los pacientes, y al mejoramiento de la calidad de vida. Objetivo: Analizar el trabajo de la revista durante los últimos 5 años. Métodos: se revisaron todos los números publicados en los últimos 5 años. Con los cuales de analizaron estadísticamente los principales datos de la revista, los autores más publicados, países y la producción científica de la misma. Los datos fueron analizados en una base de datos Access. Resultados: los cubanos son los autores que más publican en la revista seguida por Colombia y Brasil. En el año 2013 se publicaron 60 artículos en total siendo el año con mayor producción científica. La temática con mayor número de publicaciones es Farmacología y los Master en Ciencias son los que más publican en la revista. Conclusiones: Los resultados demuestran la importancia de RCPM y que se debe incrementar la visibilidad y elevar el número de citas por artículo. Dedicado a: DrC. Francisco José Morón Rodríguez

Key words: Cuban Medicinal Plants Journal
Plant-based molecular farming using transgenic plants, and plant cell, tissue, and organ cultures have been investigated as alternative production platforms for heterologous proteins produced in mammal, microbial, and insect cell systems. Now commercial plant cell culture processes include tobacco and carrot cell suspension cultures in bioreactors with defined synthetic medium, and a number of plant-derived pharmaceutical heterologous proteins are close to the market. Plant systems can be grouped into three categories: seed-based systems, leaf-based systems, and bioreactor-based systems. In this work, plant biotechnology systems are discussed for the production of therapeutic proteins. Hairy root cultures of *Brassica oleracea* var. *italica* (broccoli) are used as example to evaluate the production of the human growth hormone (hGH1) or somatotropin, insulin, and Human Papilloma Virus L1 protein. The major advantages of transgenic roots are that they are genetically homogeneous, morphologically stable, and the heterologous protein production trait is inheritable. Therefore, they are capable of synthesizing the heterologous proteins for long periods of time. One of the most important limitations for the commercial exploitation of hairy roots-based systems for production of secondary metabolites and heterologous proteins has been the low yields, productivity and the design of proper bioreactors that permit both growth of the root tissues, normally unevenly distributed through the bioreactor, and expression of the transgenic product. In any case, the economic viability of the production process seems to be ensured by eliminating low yields and low recovery by selecting elite lines with high expression levels, and by developing scalable manufacturing processes.

**Key words:** transgenic protein, agrobacterium rhizogenes, pcambia, gusplus
SPN-O-026 Isolation and characterization of antiretroviral proteins from Curcuma Longa L.
extracts. Results of three multicentric clinical trials in patients with HIV and Hepatitis B and C

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The Curcuma longa L. is a promissory medicinal plant with anti retroviral properties. A new I.J. protein with 19Kd M.W. and two isomers is isolated carry through Capilar Zone Electrophoresis, HPLC, IRFT and Circular Dichroism performing the Convex Algorithm to test the Secondary sequential structure of the ARV protein. The IJ protein was purified and freeze dried for further studies. Two isomers or 8 Kd each M.W. appeared seen as intrachain bonded Diketopyperazins. A generic 250 mg Natural Drug Softgels is performed for Clinical Trials in HIV, Hepatitis B, Hepatitis C. The performed trials showed the reduction of the viral charge in HIV, from 100,000 by ml to 0 performing the PCR test. The results was 89.90% of VCR by ml. In patients with Hepatitis B and C the results were 96.75% VCR respectively. An In Vitro′ Action Mechanism is Done. The IJ protein shows a fusion inhibition of the virus over the Hep-2 liver and HIV Lymphocytes cells surface in Tissue Culture.

Key words: Isolation, characterization, antiretroviral proteins, Curcuma Longa L. clinical trials, patients, HIV, Hepatitis B
Potentialities of the use of lipidic extract from Cuban Royal palm fruits in China population

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La Hiperplasia Prostática Benigna (HPB) es una patología que puede afectar a los hombres fundamentalmente 50 años y cuya frecuencia aumenta con la edad. En china se estima que tengan esta afectación más de 25 millones de hombres hasta el 2020. Esta patología consiste en un agrandamiento benigno de la próstata que puede causar obstrucción y propicia el desarrollo de síntomas del tracto bajo urinario entre los que destacan disminución del volumen de orina y la presión de la micción, retención urinaria y aumento de la nicturia. De acuerdo al grado de obstrucción, la intensidad de los síntomas puede variar, llegando a afectar la calidad de vida de los hombres que la padecen. Se plantea que su desarrollo responde a cambios hormonales y no hormonales que ocurren durante el envejecimiento. El aumento de la transformación de la testosterona (T) en dihidrotestosterona (DHT), a través de la enzima 5α-reductasa prostática, es el principal cambio hormonal asociado y el principal componente no hormonal está dado por el aumento del tono de la musculatura lisa de la vejiga y la próstata, regulado por los adrenoreceptores (ADR)-1. La principal terapia farmacológica de la HPB la constituyen los inhibidores de la 5α-reductasa prostática, los antagonistas de ADR-1, y la terapia combinada con ambos, todos ellos asociados a un número importante de efectos adversos como la disminución de la libido, impotencia, trastornos de la eyaculación, la hipotensión ortostática. En china, con larga tradición del empleo etnomédico de productos naturales, existen diferentes formulaciones como baishu, dangsheng, danggui, etc., son muy variables, pero el éxito de su uso es bajo, mientras que en el mundo occidental el extracto lipídico de Saw palmetto ha sido la alternativa fitoterapéutica más comúnmente usada. Recientemente se han encontrado reportes de un nuevo extracto lípido obtenido de la palma real cubana (Roystonea regia), con amplia información preclínica que avalan su eficacia en modelos experimentales y su seguridad toxicológica a la par que los ensayos clínicos muestran resultados prometedores. Este extracto está constituido por una mezcla de ácidos grasos, principalmente oleico, palmitico, láurico, linoleico y mirístico, y previene el desarrollo de la HP inducida con T en roedores así como los cambios histológicos y urodinámicos que caracterizan a la HP atípica inducida por fenilefírina, efectos que se relacionan con una inhibición de la 5α-reductasa y una inhibición de las respuestas mediadas por los adrenoreceptores (ADR)-1. Además presenta moderada acción anti-inflamatoria, asociada a la inhibición dual de las enzimas ciclooxigenasa (COX) y 5-lipooxigenasa (5-LOX). Estudios en voluntarios sanos mostró ser bien tolerado, sin producir las EA, mientras que en pacientes sanos y con HP mostró efectos antioxidantes. Por otra parte a dosis de 320 mg/día durante 4 y 6 meses fue tan efectivo como el SP (320 mg/día) en reducir los STBU evaluados mediante la reducción del puntaje IPSS, produciendo además un descenso modesto del volumen prostático. Una variante de este producto se encuentra registrado como suplemento nutricional en Cuba con el nombre de Palmex, que dado la información que lo sustenta pudiera ser un candidato importante a ser tomado en consideración como una alternativa natural en el manejo de la Hiperplasia Prostática Benigna en la población masculina china.

Key words: lipidic extract, Cuban Royal palm fruit, China, IPB
Helminth infections are among the most widespread infections in humans, especially in the tropical regions of the world. It causes enormous hazard to health and contributes to the prevalence of malnourishment, anemia and pneumonia among others. Mucuna pruriens and Canarium schweinfurthii used in traditional medicinal practices in Northern Nigeria, for the treatment of worms infestation were investigated. The in-vitro anthelmintic evaluation of the crude and solvent partitions extracts, on Pheretima pothuma and Ascaris suum (eggs and L2 stage) respectively. Showed that the hexane fraction (leaves and stem bark) of C. schweinfurthii and Mucuna pruriens (leaves) had ovicidal potency of (100.00 ±0.33) at 80mg/ml which was comparable to the positive control; Albendazole(100.00 ±0.33) at the same concentration. While the Chloroform fractions showed ovicidal activity of 98.20±0.12 and 98.20±0.12 80 mg/ml on the two plants respectively. The results of determination of larvicidal potency showed that the hexane fraction (0.06%) was the most potent, followed by the Chloroform fractions (0.1 and 0.2 % respectively). These results clearly justified the uses of these plants in the treatment of worms in ethno-medical practices.

**Key words:** anthelmintic activity, mucuna pruriens, canarium schweinfurthii, pheretima pothuma, a. suum,
The role and variety of chemical compounds in plants make them potential candidates for pest control. Such is the case of Adenophyllum aurantium (L.) Strother, whose root extract containing ethyl acetate reduced the mycelial growth and sporulation of phytopathogenic fungus Fusarium solani and Alternaria alternata, without harming the growth of the biofertilizer Bacillus licheniformis. Biotechnological techniques have been used to obtain natural products that are, for some reason, hard to obtain at large scale through chemical synthesis. Said techniques are useful to preserve germplasm; additionally, they help to prevent excessive predation of wild populations which maintains to a certain extent biodiversity and gene stability of species. Due to the above, strategies to obtain a large number of individuals for its use in phytochemical, structural and metabolic studies are important. Preliminary results show that we obtained 95% of establishment of aseptic cultures from seeds. Micro-cutting were sectioned from germinated seedlings, and the induction of multiple sprouts from axillary buds was achieved using MS medium (Murashige and Skoog, 1962) supplemented with BAP (2.5 milligrams per litre) and AIA (0.1 milligrams per litre). Root forming occurred spontaneously in 90% of the micro-cuttings.

Key words: adenophyllum aurantium, micropropagation, medicinal plant
Introducción: Desde 1988 el Laboratorio Central de Farmacología (LCF) apoyada por el Ministerio de Salud Pública (MINSAP), ha trabajado en las investigaciones los usos de plantas medicinales con el objetivo de incorporarlas en el Sistema Nacional de Salud. Además ha coordinado el Programa Cubano de Investigaciones de Plantas Medicinales (1988-2004). Objetivo: Exponer el trabajo realizado durante 25 años del LCF en la validación del uso tradicional de plantas medicinales. Métodos: se revisaron los principales resultados, trabajos de difusión y la docencia médica superior, así como los resultados publicados y los incluidos en la Farmacopea Vegetal Caribeña de TRAMIL desde 1988 hasta 2014. Resultados: se expone el trabajo realizado con plantas científicamente validadas durante 25 años, en el cual existe un elevado porcentaje de usos tradicionales respaldados por evaluaciones experimentales preclínicas, que ratifica la ausencia casi total de toxicidad en las formas de uso tradicional. Se presenta el trabajo de colaboración con la red TRAMIL para la difusión del uso tradicional, su eficacia y seguridad en Cuba y el Caribe. Conclusiones: Los datos indican la importancia de la validación del uso tradicional de plantas, la adecuada divulgación de los resultados y la necesidad de establecer una metodología adecuada para la validación.

**Key words:** plantas medicinales, farmacopea vegetal caribeña, fitoquímica, farmacología, microbiología,
SPN-P-001  Effects of policosanol pre-treatment on blood-brain barrier damage and inflammatory markers after ischemia-reperfusion brain injury in rats

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Blood-brain barrier (BBB) disruption following ischemia-reperfusion (I/R) is associated with poor outcomes in stroke patients. Policosanol, a mixture of sugarcane wax alcohols, has been shown to protect against cerebral functional and histological disturbances induced by I/R in gerbils. Nevertheless, the effects of policosanol on experimentally induced BBB disruption have not been explored. The objective of this study was to investigate the effects of policosanol on BBB disruption following the induction of cerebral I/R in rats. Rats were randomized into a negative vehicle control and five I/R groups: a positive vehicle control, three policosanol (50, 200 and 400 mg/kg), one aspirin (150 mg/kg). Treatments were given orally 1 h before ischemia induction. Brain ischemia was induced by 30 min bilateral occlusion of carotid arteries, followed by 24 h reperfusion. BBB permeability was evaluated according to Evans blue (EB) dye extravasation. Also, myeloperoxidase (MPO) activity in brain homogenates was measured. EB concentration and MPO values after 24 h of reperfusion were significantly increased in positive controls when compared with the negative controls. Oral pre-treatment with policosanol (50 - 400 mg/kg) significantly decreased EB extravasation (60.4% - 71.7%) as compared to the positive controls. Aspirin reduced significantly BBB leakage by 92.5%. Also, the I/R-induced increases of MPO activities were lowered significantly with policosanol (50 - 400 mg/kg) (46.3% - 65.3) and aspirin (68.4%). In conclusion, oral policosanol pre-treatment (50 - 400 mg/kg) protected against BBB damage induced by cerebral I/R in rats, and attenuated the increase of MPO, a marker of inflammation, in the brain tissue.

Key words: policosanol, blood-brain barrier damage, inflammatory markers, ischemia-reperfusion, brain injury, rat
SPN-P-002 Comparative study of the preventive effects of Policosanol and the omega-3 fatty acid of fish oil in models of cerebral ischemia

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Stroke is the third cause of death and the first of permanent adult disability. Pre-treatment with policosanol and omega-3 fish oil (FO) has been effective in experimental models of cerebral ischemia in rodents. This study compared the effects of policosanol and omega-3 fish oil in the global cerebral ischemia (GCI) model induced by ischemia-reperfusion (I/R) in Mongolian gerbils. Gerbils were randomized distributed in 7 groups: a negative control group and 6 with GCI: a positive control (vehicle), policosanol (100 and 200 mg/kg) and FO (1.25 and 2.5 g/kg) and aspirin (60 mg/kg). All treatments were administered by oral route during 4 weeks. Policosanol (100 and 200 mg/kg) and FO (1.25 and 2.5 g/kg) significantly reduced the clinical symptoms score (60 y 60%; 33.4 y 36.4%, respectively), the rate of clinical symptoms (63.6 and 73.3%; 32.7 and 58.8%, respectively), the hyper-locomotion (84.4 y 82.5%; 28.3 y 32.6%, respectively), the histological score of neurological damage (41.4 y 52.1%; 28.3 y 32.6%, respectively) and the plasmatic levels of malondialdehyde (76.7 y 83.3%; 59.9 y 72.7%, respectively) and sulphydrl groups (100 y 100%, respectively). In conclusion, Policosanol (100 and 200 mg/kg) and FO (1.25 and 2.5 g/kg) marked and significantly reduced the GCI in Mongolian gerbils, with a mayor potency and efficacy of Policosanol.

Key words: policosanol, omega-3 fish oil, cerebral ischemia, mongolian gerbils
El D002 es una mezcla de alcoholes grasos (C24 a C34) extraídos y purificados de la cera de abejas Apis mellifera, el cual constituye el ingrediente activo (IA) de las tabletas de Abexol® y BWA® las cuales presentan efectos antioxidantes y gastroprotectores. El presente estudio evaluó la estabilidad de 3 lotes almacenados bajo condiciones de degradación forzada, estabilidad acelerada (40ºC y 75% HR) y a largo plazo (3 años) en las zonas climáticas II y IV. Los parámetros medidos en todos los casos fueron: características organolépticas, contenido (%) total e individual de alcoholes mediante cromatografía de gases con columna capilar, aparición de productos de degradación así como las pérdidas por secado en los casos de la estabilidad acelerada y a largo plazo. Los resultados muestran que bajo condiciones de estrés (hidrólisis ácida y básica, oxidación, termólisis y fotólisis) no se formaron productos de degradación y se mantuvo el porcentaje de pureza superior al 87%. Por su parte, los estudios de estabilidad acelerada (12 meses) y a largo plazo demostraron la elevada estabilidad de estos alcoholes. Adicionalmente, no se observó, bajo ninguna de las condiciones ensayadas, cambios de coloración ni olores extraños, permaneciendo el IA como un polvo fino de consistencia cerosa y color blanco hueso. Las pérdidas por secado fueron inferiores al 1% lo cual se corresponde con las especificaciones de calidad para dicho producto. Los resultados del presente estudio confirman un tiempo de vida útil de 3 años, lo cual concuerda con los informados en estudios previos de estabilidad del D002 y sus formas terminadas.

**Key words:** d002, alcoholes de la cera de abejas, alcoholes grasos, estabilidad
**SPN-P-004  Long term stability study of 20 mg Policosanol tablets in blisters**

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El policosanol constituye una mezcla reproducible de ocho alcoholes grasos primarios de 24 a 34 átomos de carbono purificados de la cera de la caña de azúcar. Esta sustancia, con efectos hipocolesterolemizantes y antiagregante plaquetarios, es empleada en la elaboración de tabletas con 20 mg de alcoholes, forma farmacéutica terminada utilizada en los ensayos clínicos y en la práctica de rutina. El objetivo fue determinar la estabilidad de estas tabletas en blísteres de cloruro de polivinilo y aluminio. Muestras de tres lotes de tabletas (MedSol, Cuba) se pusieron en cajas de cartón y se mantuvieron en las condiciones de la zona climática IV (30 ± 2 °C, 70 ± 5 % de humedad relativa) durante cinco años. Los parámetros evaluados se mantuvieron dentro de sus especificaciones de calidad durante todo el estudio: apariencia (tabletas verdes con superficies enteras), peso promedio (inicial 10 %), contenido total de los seis alcoholes grasos (20 ± 2 mg), tiempo de desintegración (< 30 min), y contenido microbiológico (1000 bacterias/g y ? 100 hongos/g, ausencia de E. coli, S. aureus, Pseudomonas, Salmonella y C. albicans). Los resultados del presente estudio sustentan que las tabletas de policosanol con dosis de 20 mg, almacenadas en las condiciones de la zona climática IV, presentan un tiempo de vida útil de cinco años.

**Key words:** policosanol, cera de caña de azúcar, alcoholes grasos, estabilidad, tabletas
Los alcoholes de la cera de abejas constituyen una mezcla reproducible de seis alcoholes grasos primarios de 24 a 34 átomos de carbono purificados de la cera de Apis mellifera. Esta sustancia, con efectos antioxidantes y gastroprotectores, es empleada para la elaboración de las tabletas de Abexol® con dosis de 50mg de alcoholes, forma farmacéutica terminada utilizada en los ensayos clínicos y en la práctica de rutina. El objetivo fue determinar la estabilidad de las tabletas con 50 mg de alcoholes de cera de abejas (Abexol®) en blísteres y en frascos de polietileno de alta densidad. Muestras de tres lotes de tabletas de Abexol® con dosis de 50mg envasadas en blísteres de cloruro de polivinilo y aluminio y en frascos de polietileno de alta densidad (Rainbow & Nature, Sydney, Australia) se pusieron en cajas de cartón y se mantuvieron en las condiciones de la zona climática IV (30 ± 2 °C, 70 ± 5 % de humedad relativa) durante cinco años. En ambos envases, los parámetros evaluados se mantuvieron dentro de sus especificaciones de calidad durante todo el estudio: apariencia (tabletas blancas con superficies enteras), peso promedio (inicial 7,5 %), contenido total de los seis alcoholes grasos (50 ± 3,75 mg), tiempo de desintegración (< 15 min), dureza (> 3 kg/cm2) y contenido microbiológico (? 1000 bacterias/g y ? 100 hongos/g, ausencia de E. coli, S. aureus, Pseudomonas, Salmonella y C. albicans). Los resultados del presente estudio sustentan que las tabletas de Abexol® con dosis de 50mg, envasadas en ambos envases, almacenadas en las condiciones de la zona climática IV, presentan un tiempo de vida útil de cinco años.

Key words: abexol, alcoholes de la cera de abejas, alcoholes grasos, estabilidad, tabletas
D-002, a mixture of six higher aliphatic beeswax alcohols, has been shown to produce anti-inflammatory effects in experimental models and to exhibit gastroprotective, not gastrotoxic effects. The mechanism of the anti-inflammatory action of D-002, however, had not been explored yet. This study was aimed at investigating the effects of D-002 on cyclooxygenase (COX) and 5-lipoxygenase (5-LOX) activities in vitro. The addition of D-002 to preparations of the cytosolic fraction of rat blood polymorphonuclear leukocytes significantly (p<0.001) and dose-dependently (r=0.990) inhibited the in vitro activity of 5-LOX up to 81% at 500?g/mL concentration. In contrast, nordihydroguaiaretic acid (NDGA), the reference substance, inhibited 5-LOX activity by 55%. Also, D-002 produced a significant (p<0.001) and dose-dependent (r=0.984) inhibition of COX activity in preparations of rat liver microsomes up to 61% at 2500 ?g/mL, while indomethacin at 0.4 mg/ml produced a 90% inhibition. D-002 modified both kinetic parameters (Vmax and Km) of 5-LOX and COX activities through uncompetitive mode of inhibition. This study demonstrates that D-002 inhibits both COX and 5-LOX activities, with a highest affinity for 5-LOX, so that the dual inhibition of both enzyme activities may explain the observed anti-inflammatory effects of D-002 as well as the lack of D-002-related gastrotoxicity.

**Key words:** ciclooxygenase and 5-lipoxygenase, enzymes, D-002, beeswax, alcohols
Effects of D-002 (beeswax alcohols), grape seed extract and their combined therapy on oxidative markers in rat plasma and liver

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D-002, a mixture of high molecular weight alcohols from beeswax (Apis mellifera), and grape seed extract (Vitis vinifera, L), rich in flavonoids, presents antioxidant effects, but no previous study had compared such effects. The aim of this study was to investigate the effects of D-002, grape seed extract and their combined therapy on rat plasma and liver oxidative markers. Two experiments were conducted. First experiment: rats were distributed into seven groups: a vehicle control, three D-002-treated, three grape seed extract-treated (25, 100 and 250 mg/kg/day, respectively). Second experiment: rats were randomized into four groups: one vehicle control and three treated with D-002, grape seed extract or their combined therapy (25 mg/kg/day). Treatments were given orally for 30 days. In the first experiment: D-002 and grape seed extract (25-250 mg/kg/day) reduced plasma malondialdehyde, total hydroperoxides, carbonyl groups and liver malondialdehyde formation with similar efficacy, but unchanged blood superoxide dismutase. Second experiment: each monotherapy produced significant and comparable reductions of plasma malondialdehyde, carbonyl groups and liver malondialdehyde formation. The combined therapy lowered plasma oxidative variables more effectively than each monotherapy, and reduced liver malondialdehyde more than grape seed extract, not than D-002. In conclusions D-002 and grape seed extract (25-250 mg/kg/day) significantly and similarly reduced plasma oxidative variables and liver malondialdehyde formation, without modifying blood superoxide dismutase. The combined therapy displayed benefits over each monotherapy regarding to plasma oxidative variables, whereas regarding to liver malondialdehyde was more effective than grape seed extract, not than D-002.

Key words: d-002, grape seed extract, lipid peroxidation, protein oxidation
SPN-P-008  Protective effect of D-002, a mixture of beeswax alcohols, against indomethacin-induced gastric ulcers and mechanism of action

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D-002, a mixture of higher aliphatic beeswax alcohols, produces gastroprotective and antioxidant effects. To investigate the gastroprotective effect of D-002 against indomethacin-induced ulcers, oxidative variables and myeloperoxidase (MPO) activity in the rat gastric mucosa were examined. Rats were randomized into six groups: a negative vehicle control and five indomethacin (50 mg/kg) treated groups, comprising a positive control, three groups treated orally with D-002 (5, 25 and 100 mg/kg) and one group with omeprazole 20 mg/kg intraperitoneally (ip). The contents of malondialdehyde (MDA), protein carbonyl groups (PCG), hydroxyl radical generation and catalase (CAT), glutathione peroxidase (GSH-PX), superoxide dismutase (SOD) and MPO enzyme activities in the rat gastric mucosa were assessed. Indomethacin increased the content of MDA and PCG, the generation of *OH radical and MPO enzyme activity, while it decreased the CAT, GSH-PX and SOD activities as compared to the negative controls. D-002 (5-100 mg/kg) significantly and dose-dependently reduced indomethacin-induced ulceration to 75%. Also, D-002 decreased the content of MDA and PCG, the generation of hydroxyl radicals and MPO activity as compared to the positive controls. The highest dose of D-002 (100 mg/kg) increased significantly GSH-PX and SOD activities, while all doses used increased CAT activities. Omeprazole 20 mg/kg, the reference drug, reduced significantly the ulcers (93%), MDA and PCG, the generation of hydroxyl radicals and MPO activity, and increased the CAT, GSH-PX and SOD activities. D-002 treatment produced gastroprotective effects against indomethacin-induced gastric ulceration, which can be related to the reduction of hydroxyl radical generation, lipid peroxidation, protein oxidation and MPO activity, and to the increase of the antioxidant enzymes activities in the rat gastric mucosa.

Key words: Protective effect, D-002, beeswax alcohols, indomethacin-induced, gastric ulcers, mechanism of action
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SPN-P-009  Effects of D-002 on models of acute inflammation

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Oral treatment with D-002, a mixture of 6 high molecular weight aliphatic primary alcohols purified from beeswax, has been shown to produce anti-inflammatory effects in the carrageenan-induced pleurisy and in the cotton pellet granuloma models, but its effects on other models of acute inflammation have not been investigated. This study was aimed to investigate the anti-inflammatory effects of single oral doses of D-002 on dextran-, histamine- and serotonin-induced rat paw oedema models, and on the xylene-induced ear oedema in mice. Oral pre-treatment with D-002 (200, 400 and 800 mg/kg) significantly inhibited dextran-induced paw oedema (p < 0.001) up to 71.4%. D-002 (800 mg/kg), not 200 or 400 mg/kg, inhibited significantly histamine - and serotonin-induced hind paw oedemas in rats by 57 (p < 0.01) and 36% (p < 0.001), respectively, while diphenhydramine (60 mg/kg) and serotonin (10 mg/kg) produced significant inhibitions of 60% (p < 0.05) and 79% (p < 0.01), respectively. Also, D-002 (200 and 400 mg/kg), not 50 mg/kg, significantly and dose-dependently (r= 0.951, p 0.05) reduced by 50 % a xylene- induced oedema formation in mice ear. Thus, this study reveals that oral treatment with single doses of D-002 exhibited significant anti-inflammatory activity in experimental models of acute inflammation.

Key words: D-002, acute inflammation
The gastric mucosa is susceptible to the effects of aggressive factors, which are counterbalanced by mucosal defensive factors. Acid peptic diseases result from the imbalance between these aggressive and defensive factors. Aspirin-induced ulcer is a model of NSAIDs-induced damage in which neutrophil infiltration play a key role. D-002, a mixture of 6 higher aliphatic alcohols, has been shown gastroprotective and anti-inflammatory effects. This work investigates the protective effect of D-002 against aspirin-induced ulcers and associated neutrophil infiltration in the gastric mucosa. Rats were randomized into six groups of 8 rats each. A negative vehicle control, and five aspirin (300 mg/kg)-treated groups: a positive control, orally treated with the vehicle, three with D-002 (25, 50 and 100 mg/kg, respectively) and other with 10 mg/kg Omeprazole. Five (5) hours after induced damage the rats were sacrificed. The stomachs were removed and opened, and lesions examined macroscopically and microscopically. Ulcer indexes and neutrophil infiltration per ulcer areas were measured. All positive, none negative, controls exhibited aspirin-induced ulcers. Oral treatment with D-002 (25-100 mg/kg) dose- dependently and significantly reduced aspirin-induced gastric lesions (37 to 75%), the mean number of microscopic ulcers (40 to 72%) and neutrophil infiltration (41.7 to 83.1%) in the rat gastric mucosa. In turn, omeprazole 10 mg/kg reduced significantly the ulcer indexes, the mean microscopic ulcer number and neutrophil infiltration per area of microscope field, respectively. Oral treatment with D-002 (25-100 mg/kg) effectively protects against aspirin-induced ulcers and in the gastric mucosa induced by aspirin ulceration.

**Key words:** D-002, aspirin induced ulcer, neutrophil infiltration, gastric mucosa
Osteoarthritis (OA), the most common age-related degenerative joint disease in adults, is characterized by a progressive cartilage loss which produces debilitating chronic pain in affecting individuals and tends to worsen over time as cartilage wears away. Non-pharmacologic interventions are the cornerstone of OA management. Nevertheless, optimal treatment should combine non-pharmacological and pharmacological modalities. Numerous studies have demonstrated that oxidative stress has been implicated in OA which support the evaluation of antioxidants for management of OA. D-002 is a mixture of six high molecular weight aliphatic alcohols purified from beeswax that has been shown to be effective in experimental models of acute and chronic inflammation and in experimental models of OA. The aim of this study was to compare effects of D-002 and Grape seed extract (GSE) in mono iodoacetate induce OA in rats. Male wistar rats were distributed into 6 groups: one negative control and five groups with MIA induce OA: a positive control group and four groups treated with D-002 (200 and 400 mg/kg) or GSE (200 and 400 mg/kg). OA was induced by MIA. Substances were administered during 10 days. Cartilage changes were measured by using histological and Mankin modified score. D-002 (200 and 400 mg/kg) and GSE (400 mg/kg) significantly prevented the increase on total histological score induced by the injection of MIA into the knee joint of the animals. Effects of D-002 were significantly higher than GSE ones in the reduction of total histological score. D-002 (200 and 400 mg/kg) significantly reduce histological parameters of the score except cellular abnormalities and osteoclasts presences. On its side, GSE was no effective on extension to damage, cellular abnormalities, bone loss and panus formation and osteoclast presence. D-002 was more effective to GSE in preventing cartilage damage in MIA-induced OA model in rats.

Key words: osteoarthritis, d-002, mono iodoacetate, rats
**SPN-P-012 Effects of D-002, a mixture of high molecular weight beeswax alcohols, on patients with non-alcoholic fatty liver disease (NAFLD)**

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**BACKGROUND.** Non-alcoholic fatty liver disease (NAFLD), characterized by excessive liver fat accumulation without habit of alcohol consumption, is intimately related to insulin resistance and ranges from a benign course to liver fibrosis and cirrhosis. NAFLD management mainly involves dietary modification and weight loss. Although no fully successful pharmacological intervention is available, alternative therapies to treat NAFLD have shown promising results. D-002, a mixture of beeswax alcohols with antioxidant effects, has been shown hepatoprotective effects experimentally. The effects of D-002 on NAFLD, however, had not been investigated. **AIM:** To investigate the efficacy and safety of D-002 on patients with NAFLD.

**METHODS:** This placebo-controlled study included 50 patients with NAFLD randomized to placebo (n = 25) or D-002 50 mg (n = 25) tablets twice a day for 24 weeks. Primary endpoint was to obtain a significant reduction of ultrasonography-detected liver fat infiltration as compared to placebo. Secondary endpoints were to obtain decreases of the Human Omeostatic Assessement (HOMA) index, insulin levels, serum liver enzymes, increase of plasma total antioxidant status (TAS) and unaffected the remaining parameters. The frequency of D-002 patients (12/25, 48.0%) referring symptom improvement was higher (p<0.001) than in placebo (1/25, 4.0%). Treatment was safe and well tolerated. Six patients, three of each group, withdrew from the study, only two (placebo) because of adverse experiences. **CONCLUSION:** D-002 (100 mg/day) was effective to improve ultrasonographic findings, indicators of insulin resistance (HOMA index, insulin levels), plasma TAS and clinical evolution on this population of patients with NAFLD. Further studies, however, are needed to confirm these results.

**Key words:** D-002, beeswax alcohols, patients non-alcoholic fatty liver disease, NAFLD
SPN-P-013 Effects of D-002 (beeswax alcohols) on gastrointestinal symptoms and oxidative markers in middle-aged and older patients

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This study investigated the persistence and coexistence of the effects of D-002, a mixture of higher aliphatic beeswax alcohols with antioxidant and gastroprotective effects, on symptoms and plasma oxidative markers in subjects with gastrointestinal complaints. Sixty subjects were double-blinded randomised to D-002 (50 mg) or placebo tablets twice daily for 24 weeks. Dose was titrated to three tablets daily if symptoms, assessed with the Gastrointestinal Symptom Rating Scale (GSRS), did not improve after 6 weeks. No significant changes occurred in placebo. D-002 persistently reduced (p < 0.001) GSRS overall score and several sub-scores versus placebo. Dose-titration (1/30) and antacids intake (7/30) with D-002 were less frequent (p < 0.001) than with placebo (29/30 and 28/30, respectively). D-002 persistently decreased (p < 0.00001) total hydroperoxides, (p < 0.001) plasma malondialdehyde and sulphhydril groups, while increased (p < 0.01) plasma total antioxidant status. Summarizing, D-002 (100 mg/day) administered for 24 weeks displayed persistent antioxidant and gastroprotective effects in subjects with gastrointestinal complaints. OBJECTIVES: In light of these issues, this study investigated the persistence and coexistence of the effects of D-002, given for 6 months, on the symptoms and plasma oxidative markers of middle-aged and older subjects with habitual gastrointestinal complaints. CONCLUSIONS This study demonstrates that oral treatment with D-002 (100 mg/day) for 24 weeks ameliorated symptoms and improved plasma oxidative markers in middle-aged and older patients with habitual gastrointestinal complaints. These results support the coexistence and persistence of symptom relief and antioxidant effects of D-002 and that its antioxidant effect could contribute, at least partially, to its gastroprotective effect in such population.

Key words: D-00, beeswax alcohols, gastrointestinal symptom, oxidative marker, middle-aged, older patient
El prevenox constituye una mezcla reproducible de ácidos grasos de 24 a 36 átomos de carbono purificados de la cera de la caña de azúcar. Esta sustancia, con efectos antioxidantes, es empleada en la elaboración de tabletas con 5 mg de ácidos, forma terminada utilizada en los ensayos clínicos y en la práctica de rutina. El objetivo fue determinar la estabilidad de estas tabletas en blísteres de cloruro de polivinilo y aluminio. Muestras de tres lotes de tabletas (MedSol, Cuba) se pusieron en cajas de cartón y se mantuvieron en las condiciones de la zona climática IV (30 ± 2 °C, 70 ± 5 % de humedad relativa) durante tres años. Los parámetros evaluados se mantuvieron dentro de sus especificaciones de calidad durante todo el estudio: apariencia (tabletas verdes con superficies enteras), peso promedio (inicial ± 10 %), contenido total de los ácidos grasos (5 ± 0,5 mg), tiempo de desintegración (< 30 min), y contenido microbiológico (≤ 1000 bacterias/g y ≤ 100 hongos/g, ausencia de E. coli, S. aureus, Pseudomonas, Salmonella y C. albicans). Los resultados del presente estudio sustentan que las tabletas de prevenox con dosis de 5mg, almacenadas en las condiciones de la zona climática IV, presentan un tiempo de vida útil de tres años.

**Key words:** prevenox, cera de caña de azúcar, ácidos grasos, estabilidad, tabletas
Osteoporosis is a chronic degenerative systemic disease characterized by microarchitectural disrepair of bone tissue accompanied by the progressive reduction of bone mass. D-003 is a mixture of very long-chain aliphatic acids purified from sugarcane which has shown cholesterol-lowering effects in experimental and clinical studies. Previous studies in rats have shown that D-003 prevent bone loss and decreased bone resorption in ovariectomized rats. This study was undertaken to investigate the effects of D-003 in a model of osteoporosis induced by ovariectomy and glucocorticoids administration in female rabbits. Female New Zealand white rabbits skeletally matured were used in 4 groups. A sham group of false operated rats and three groups of ovariectomized and treated with prednisolone animals; a positive control group administered with the vehicle and two groups treated with D-003. Treatment with D-003 (50 and 200 mg/kg) significantly prevented decreased trabecular number and thickness while decreased trabecular separation, number of osteoclast and osteoclast surface increased by the induced damage. In line with these results, D-003 significantly prevented the increase in Total cholesterol and TG at both doses improving the lipid profile of animals treated with D-003, allowing for the first time linking lipid lowering effect-antiresorptive effect.

Key words: d-003, glucocorticoids, osteoporosis, rabbit
Effect of D-003 and soy standardized extract rich in daidzein on osteoporosis induced in ovariectomized rats

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D-003 is a mixture of higher fatty acids purified from sugarcane wax with antiosteoporotic effects in ovariectomized (ovx) rats. Soy standardized extract rich in daidzein (SSE-D) have shown bone-protective effects. This study compared the effect of D-003 and SSE-D on bones of ovx rats. Rats were randomized into 6 groups: one false-operated and five ovx groups: a positive control treated orally with the vehicle, two with D-003 (50 and 200 mg/kg) and two with SSE-D (15 and 30 mg/kg) for 3 months. Ovx decreased trabecular volume, number and thickness of trabeculae in the fifth vertebrae and femoral neck, and increased trabecular separation, osteoclast number and surface versus the false-operated group. D-003 and SSE-D prevented all changes induced by ovx, but the effects of D-003 were significantly greater than those of SSE-D. Concluding, D-003 (50 and 200 mg/kg) and SSE-D (15 and 30 mg/kg) prevented osteoporotic changes in ovx rats, but D-003 was more effective than the SSE-D preparation assayed.

Key words: D-003, soy standardized extract, daidzein, osteoporosis, ovariectomized rat
SPN-P-017  Effects of D-003 on quality of life in postmenopausal women at moderate to high risk of osteoporosis.

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Osteoporosis, a disease characterized by reduction of bone mineral density (BMD) and alteration of bone microarchitecture that increases bone fragility and fracture risk, affects the quality of life of the sufferers. Pharmacological management of osteoporosis includes the use of drugs that reduce bone resorption and/or increase bone synthesis. D-003 is a mixture of high molecular weight aliphatic acids purified from sugar cane wax with antiresorptive effects demonstrated in experimental and clinical studies. This randomized, double-blinded, placebo-controlled study investigated the effects of D-003 on the quality of life of postmenopausal women at moderate to high risk of OP. Forty postmenopausal women were randomized to receive placebo or D-003 (10 mg/d) for three months. Significant reductions on Qualeffo total score and serum low-density lipoprotein-cholesterol (LDL-C) levels versus placebo group were considered as the primary and secondary efficacy variables, respectively. Evaluation of safety and tolerability included the analyses of the effects on physical and blood indicators, and the adverse events (AE) reports, as well. Data were analyzed as per the Intention-to-Treat method. Both groups displayed similar baseline characteristics. After complete three months on therapy, D-003 significantly decreased Qualeffo total scores (p<0.0001 vs baseline, p<0.05 vs placebo), as well as LDL-C values (17.1 %). The treatment was safe and well tolerated. Only one patient (D-003-treated) withdrew from the study, and it was due to protocol violation. Four patients, all from placebo group, reported some AE. In conclusion, D-003 (10 mg/d) administered during three months improved quality of life in postmenopausal women at moderate to high risk of OP and produced additional benefit on serum LDL-C values.

Key words: D-003, postmenopausal women, osteoporosis
Osteoarthritis (OA) has been linked with bone marrow edema (BME) pattern, identified by magnetic resonance imaging and histopathology, which involves a mix of different pathologies, so that BME term has been progressively replaced by that of bone marrow lesions (BML). BML is the first sign of OA after experimental ligament damage, precedes cartilage erosion and degeneration in experimentally-induced OA, strongly correlate with knee pain incidence and progressing in humans, and the reduction of BML size is accompanied by pain improvement. Some studies have revealed positive findings with bisphosphonates and their impact on the progression of cartilage defects. Oral administration of D-003, a mixture of high molecular weight sugar cane wax acids, has been shown to produce bone antiresorptive effects in experimental and clinical studies. This study investigated the effects of D-003 on BML, subchondral bone and cartilage by sodium monoiodoacetate (MIA)-induced OA in rats. Animals were distributed into 5 groups: a negative control and four injected with MIA: one positive control and three with D-003 (100, 200 and 400 mg/kg). The damage was induced by a single injection of MIA into the synovial cavity of the knee, after which rats were treated by gastric gavage for 10 days. Knee joint were removed and processed for light microscopy. Slides were stained with hematoxylin and eosin and toluidine blue to stain proteoglycan for analyze the cartilage. A Mankin score was used to assess the depth and extent of cartilage damage. No negative, but all positive control animals, exhibited BMLs, meanwhile D-003 treatment decreased significantly the frequency of animals with BMLs. Negative controls did not show tibiia plate extent of damage, bone loss or presence of osteoclasts, all these variables were significantly increased in the positive control group. Oral D-003 ameliorated the damage to the subchondral bone, lowering the extent of tibiia damage, bone loss, and the presence of osteoclasts in the knee joint, as compared with positive controls. Oral treatment with D-003 was effective for reducing BMLs, subchondral bone and cartilage damage, in MIA-induced OA in the rat knee, which suggests that it may benefit more components of osteoarthritic joint.

**Key words:** D-003, sugarcane wax acids, bone marrow lesions, bone and cartilage damage, osteoarthritis
El D-004 es un ingrediente farmacéutico activo obtenido de los frutos de la palma real cubana (Roystonea regia), compuesto fundamentalmente por una mezcla de ácidos grasos libres entre 8 y 18 átomos de carbono, con potencialidades en el tratamiento de la hiperplasia prostática benigna. Para determinar la vida útil del D-004 en frascos de vidrio ámbar y de polietileno de alta densidad, en las zonas climáticas IV y II, muestras de tres lotes pilotos se colocaron a 30 °C/70 % HR y 25 °C/ 60 % HR, se determinaron las características organolépticas y los contenidos de material insaponificable, ácidos grasos y microbiológicos desde el tiempo inicial hasta los 51 meses, y se calcularon los tiempos de vida útil mediante análisis estadísticos. En vidrio ámbar, en ambas zonas el D-004 mantuvo sus especificaciones de calidad por más de 24 meses. La disminución del contenido de ácidos, ocurrida primero en la zona IV, permitió establecer los nuevos tiempos de vida útil para ambas zonas. En polietileno, en ambas zonas, el D-004 cumplió con los parámetros evaluados durante los primeros 30 meses, posteriormente el contenido de ácidos disminuyó hasta resultar inferior al límite establecido en varias de las muestras. Se demostró que el ingrediente activo D004, obtenido a escala piloto y envasado en vidrio ámbar, presenta un tiempo de vida útil superior a los 24 meses (33 y 48 meses en las zonas climáticas IV y II, respectivamente), mientras al ser envasado en frascos de polietileno de alta densidad presenta tiempos de vida útil de 30 y 33 meses en las zonas climáticas IV y II, respectivamente.

**Key words:** d-004, estabilidad, vidrio ámbar, polietileno de alta densidad, roystonea regia
El desarrollo de cápsulas blandas de D004, ingrediente activo con potencialidades en la prevención y tratamiento de la hiperplasia prostática benigna, conllevó al estudio de estabilidad de esta forma farmacéutica. Se emplearon tres lotes piloto de cápsulas con 320 mg de ácidos totales (LIPA PHARMACEUTICALS, Australia) envasadas en frascos ámbar de tereftalato de polietileno (40 cápsulas/frasco). El estudio de estabilidad acelerada se realizó a 40 °C y 75 % HR y los estudios a largo plazo se desarrollaron en condiciones de las zonas climáticas II (25 °C y 60 % HR) y IV (30 °C y 70 % HR). En el tiempo inicial y en cada muestreo se determinaron las características organolépticas, que coincidieron con las iniciales durante los seis primeros meses del estudio acelerado y en todos los demás muestreos de los estudios a largo plazo; la variación del peso, que se mantuvo in 90-110 % del peso promedio inicial, el tiempo de Desintegración, que fue 30 min y el Contenido microbiológico (una vez al año), que se mantuvo estable. También se determinó el contenido de ácidos grasos por Cromatografía de Gases, el cual se mantuvo en 320 mg ± 32 mg (10%) por cápsula y durante todo el estudio en condiciones aceleradas y en ambas zonas climáticas. Según los resultados del estudio de estabilidad acelerada, se prevé que estas cápsulas pudieran resistir hasta 6 meses en condiciones drásticas y alcanzar un tiempo de vida útil de 3 años en las condiciones de la zona climática IV, los resultados obtenidos hasta el momento a largo plazo avalan un tiempo de vida útil de al menos 2 años en las condiciones de ambas zonas climáticas.

**Key words:** d-004, estabilidad, cápsulas blandas, tereftalato de polietileno, roystonea regia
La determinación de los productos de degradación en ingredientes activos (IA) y formas terminadas es esencial como parte de su conocimiento químico-farmacéutico y un aspecto exigido por los organismos reguladores. Teniendo en cuenta lo anterior, se implementaron métodos analíticos para identificar y cuantificar los productos de degradación en el D004. A muestras sometidas a oxidación (peróxido de hidrógeno), fotólisis (luz UV) y termólisis (80 °C y 185 °C), se les determinaron las características organolépticas, índice de peróxido (IP), índice de ácido tiobarbitúrico (IATB), porcentaje de dienos conjugados (%DC) y compuestos volátiles (CVol), y sus resultados se compararon con el de una muestra de D004 sin degradar; mientras que a muestras sometidas a 110 °C solo se les determinaron los dos últimos parámetros. Se determinó que la oxidación fue la que provocó la mayor degradación de los AG insaturados, así como los mayores IP, mientras que la termólisis fue la que conllevó a mayores contenidos de DC, IATB y CVol. En el caso de la termólisis a 110 °C se pudo observar que el % DC aumentó hasta las 7 h y disminuyó a partir de este tiempo, se determinó por primera vez la presencia de 2-hexenal, 2-heptenal, 2-hexanona, 2-heptanona, 2-hepteno, tolueno, octano, 1-pentanol y pentilfurano en el D004 degradado.

**Key words:** d004, productos de degradación, ácidos grasos, estabilidad
D-004, a lipid extract of royal palm (Roystonea regia) fruits that contains a reproducible mixture of fatty acids, has been shown to prevent testosterone and phenylephrine-induced prostate hyperplasia in rodents. This study investigated the long-term oral toxicity of D-004 in rats. Rats from both sexes were randomized into four groups (20 rats sex/group): a control and three treated with D-004 (800, 1500 or 2000 mg/kg/day, respectively). At study completion, rats were sacrificed under anesthesia. Determinations of blood biochemical and hematological parameters and organ weight were done. Also, necropsy and histopathological studies were performed. Four of 160 rats died before study completion. No clinical signs of toxicity were observed throughout the study. Food and water consumption, bodyweight, blood biochemical and hematological parameters, organ weight ratios and histopathological findings were similar in control and treated groups. The histological lesions found in treated animals are commonly present in this specie and strain according to literature and our historical data. In conclusion, long-term (12 months) oral treatment of rats with D-004 (800-2000 mg/kg/day) did not show evidences of D-004-related toxicity under our conditions. The highest dose tested (2000 mg/kg) was a no-observed adverse effect level in this study.

**Key words:** toxicity, D-004, lipid extract, Roystonea regia fruit, Sprague Dawley rat
D-004 is a lipid extract of royal palm (Roystonea regia) fruits that consists in a mixture of free fatty acids, wherein oleic, lauric, palmitic, and myristic are the most abundant; that prevents prostate hyperplasia induced with testosterone in rodents. D-004 has shown to inhibit the enzyme activity of prostate 5- reductase in vitro. Then, D-004-impairment of reproductive toxicity could be expected. This study investigated the possible reproductive toxicity of D-004 administered to fertile Sprague Dawley rats of both sexes. Rats were randomised into four groups: a control group, treated with the vehicle and three with D-004 at 500, 750 and 1 000 mg/kg/day, respectively. Female rats were treated for 15 days prior to mating, throughout mating and from pregnancy to day 21 of lactation. Male rats were administered 10 weeks before, and during mating. The effects on growth, development, reproductive performance, and fertility of the F1 generation were also assessed. No evidence of D-004-related toxicity was observed. Thus, maternal body weight and food consumption, litter size, survival through the weaning period and weights of male and female pups did not show differences between control and treated groups. Drug-treated and control groups’ offspring were comparable in both physical and sensorial development and in reproductive performance. There were no significant differences among F2 generation groups in the number of born pups, mean pup’s weight and postnatal pups survival. In conclusion, D-004 orally administered to rats of both sexes did not affect the fertility and general reproductive function, which indicates that it does not alter the lactation, growth, development and sexual maturation of the progeny.

Key words: toxicity, D-004, Roystonea regia, Sprague Dawley rat
SPN-P-024  Teratogenesis of D-004 in rabbits

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El D-004 es un extracto lipídico de los frutos de la palma real (Roystonea regia) que contiene una mezcla reproducible de ácidos grasos, fundamentalmente oleico, palmítico, láurico y mirístico. El tratamiento oral con D-004 previene significativamente la hiperplasia prostática inducida en roedores. El objetivo del estudio consistió en evaluar el potencial teratogénico del D-004 en conejos. Las hembras preñadas se distribuyeron aleatoriamente en tres grupos: un control, tratado con vehículo (tween 65/agua) y dos tratados con D-004 (500 y 1000 mg/kg). Se administró el producto desde el día 6 hasta el día 18 de la gestación por vía oral. Se sacrificaron las hembras el día 29 de la gestación y sus fetos se examinaron en busca de anormalidades externas, viscerales y esqueléticas. No hubo diferencias significativas entre los animales tratados y los controles en cuanto al incremento del peso de las madres y el consumo de alimentos durante la gestación. Los grupos tratados y el control fueron comparables en el número de cuerpos lúteos, sitios de implantación, reabsorciones, fetos vivos, peso de los fetos y proporción de los sexos. Se encontraron tres fetos con malformaciones viscerales: uno del grupo control y dos tratados con 1000 mg/kg. No se observaron malformaciones externas o esqueléticas en ninguno de los grupos. Las variaciones esqueléticas observadas no tuvieron una incidencia estadísticamente significativa. Los resultados del estudio permiten concluir que la administración por vía oral de D-004 en dosis de 500 y 1000 mg/kg durante el periodo de organogénesis no es teratogénica o embriotóxica en conejos.

Key words: teratogenesis, D-004, rabbit
The aim of this study was to investigate the effects of D-004, antioxidant and/or anti-inflammatory substances on prostate hyperplasia (PH) induced by testosterone (T) and phenylephrine (PHE) in rats. Two experiments were conducted. First experiment: Rats were randomized into seven groups: a negative control and six groups injected with T; a positive control and five were treated with D-004 (400 mg/kg), grape seed extract (GSE) (250 mg/kg), vitamin E (VE) (250 mg/kg), ibuprofen (200 mg/kg) or celecoxib (50 mg/kg). Second experiment: Rats were randomized into eight groups: a negative vehicle control and seven groups injected with PHE: a positive control, three treated with D-004 (200, 400 and 800 mg/kg) and three others with tamsulosin (0.4 mg/kg), GSE (250 mg/kg) and VE (250 mg/kg), respectively. In the first experiment: Oral administration of D-004, not of any other treatment, significantly reduced prostate weight and prostate weight/body weight ratio as compared to the positive control. D-004, GSE and VE, not ibuprofen or celecoxib, lowered significantly prostate MDA values. In contrast, significant inhibitions of prostate MPO activity were achieved with D-004, GSE, ibuprofen and celecoxib, not with VE. Second experiment: D-004 (200-800 mg/kg) increased significantly volume voided per micturition (VM) and urinary total volume (UTV), lowered significantly MDA in prostate and bladder homogenates, and reduced carbonyl groups (GC) levels only in the prostate. Tamsulosin increased significantly VM and UTV, but unchanged oxidative variables. GSE and VE unchanged the UTV, whereas VE, not GSE, modestly but significantly attenuated the PHE-induced decrease of VM. In conclusions, oral treatment with D-004 (400 mg/kg), but not with the antioxidant and anti-inflammatory substances tested, prevented T-induced PH in rats, so that the preventive effect of D-004 on this model does not seem to be associated to its antioxidant or anti-inflammatory effects. The single oral administration of D-004 (200-800 mg/kg) was the only treatment that ameliorated the urodynamic changes and reduced increased oxidative variables in the prostate of rats with PHE-induced PH.

Key words: anti-inflammatory; antioxidant; d-004; prostate hyperplasia; urodynamic changes
Short-term (14 days) treatment with D-004, a lipid extract from Roystonea regia fruits, has been shown to prevent testosterone (T)-induced prostate hyperplasia (PH) and increased oxidative stress in rat prostate, but no study has documented if such effects persist after longer treatment. This study investigated the persistence of the effects of D-004 orally given for 60 days for preventing prostate enlargement and increase of oxidative markers in rats with T-induced PH. Rats were randomized into three groups: a negative control and two T-injected that received orally vehicle (positive control) or D-004 (400 mg/kg/day), respectively. Prostate weights of positive controls significantly and markedly increased over the time, while persistent and significant reductions of such increases were seen in D-004-treated rats. Also, D-004 significantly reduced the T-induced increase of prostate conjugated diene generation and sulphydril groups concentrations, achieving a complete reduction from the day 30 after starting the treatment. Concluding, the effects of oral treatment with D-004 (400 mg/kg/day) on T-induced prostate enlargement and increased prostate oxidative markers persisted over 60 days of treatment.

**Key words:** d-004, prostatic hyperplasia, oxidative markers, roystonea regia, testosterone
El D005, nuevo ingrediente activo antiinflamatorio, se obtiene a partir del aceite de los frutos maduros de Acrocomia crispa. Al no existir una metodología para el procesamiento de estos frutos y no encontrarse reportes sobre las características físico-químicas o la composición del material vegetal o del aceite nos propusimos como objetivo establecer el método de tratamiento de los frutos y caracterizar el material vegetal y el aceite. Se encontró que la mejor forma de secado era a temperatura ambiente a la sombra durante 25 días, y que la molienda debía realizarse con molino de martillos. Ocho lotes de material vegetal se caracterizaron en cuanto a humedad residual (< 8%), contenido de cenizas (totales: 2,30-3,87%; insolubles en HCl: 0,70-0,97%; solubles en agua: 0,44-1,59%), contenido de aceite (13-18%) y contenido de ácidos grasos (totales: 92-96% y libres: 1-3%) en el aceite. Además, se obtuvieron ocho lotes de aceite a partir de cada lote de material vegetal con un rendimiento superior al 11%, los cuales fueron caracterizados encontrándose que todos fueron líquidos oleosos translúcidos color amarillo-naranja y olor dulce, con pérdidas por secado < 5%, densidad relativa (0,88-0,91 g/ml) e índice de refracción (1,449-1,458). El índice de saponificación estuvo entre 211-239, el contenido de material no saponificable < 1%, el contenido de ácidos grasos totales 90% y elevados índices de acidez (48-345) lo que concuerda con un alto contenido de ácidos grasos libres (9-60%). Se realizó un extracto metanolico a un lote de material vegetal encontrándose un 0,007% de polifenoles expresado como pirogalol.

**Key words:** acrocomia crispa, caracterización de aceite, material vegetal
SPN-P-028 Characterization of D005 a lipidic extract from Acrocomia crispa fruits

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Los fitofármacos, representan una parte importante del comercio de medicamentos, requiriendo no solo de procesos de obtención reproducibles sino también de controles de calidad que permitan verificar el cumplimiento de los atributos deseados. El D005, es un nuevo ingrediente activo compuesto por una mezcla de ácidos grasos libres extraídos y purificados a partir de los frutos de A. crispa con potente efecto antiinflamatorio. Tomando en consideración lo dicho, el trabajo tuvo como objetivo caracterizar los lotes de D005, para establecer los intervalos en los que se encuentran sus principales características. Se encontró que todos los lotes eran líquidos oleosos traslúcidos de color naranja-rojizo y olor dulce agradable. Los rendimientos de obtención a partir del aceite estuvieron por encima del 70 %, las pérdidas por secado <1%, la densidad relativa entre 0,88-0,91 g/mL, el índice de refracción osciló entre 1,446 y 1,453, el contenido de material no saponificable < 2 %, el índice de acidez por encima de 350, el contenido de ácidos grasos libres superior al 90 %. La composición individual de ácidos grasos, mostró que el D005 presenta como componentes mayoritarios los ácidos láurico (C12:0= 34,75 %) y mirístico (C14:0=13,22 %) y oleico (C18:1= 30,41 %) seguido de los ácidos palmitico (C16:0= 8,94 %), esteárico (C18:0= 3,62 %), cáprilico (C8:0=1,04 %), cáprico (C10:0=2,29 %), palmitoleico (C16:1= 0,07 %). La determinación de los intervalos en los que se encuentran estos parámetros permitirá establecer las especificaciones de calidad para este ingrediente activo.

**Key words:** acrocomia crispa, d005, extracto lipídico
Aunque se encuentran ampliamente distribuidas en Cuba y hay varios reportes de sus usos etnomédicos, las palmas cubanas han sido poco estudiadas; sin embargo, teniendo en cuenta la gran variedad de palmas existentes en Cuba y lo factible que pudiera resultar el empleo de sus frutos para la obtención de productos naturales con usos nutracéuticos y/o farmacéuticos; resulta interesante realizar estudios fitoquímicos preliminares a algunas de las especies nativas o introducidas del país. Para la realización de este trabajo se recolectaron frutos maduros frescos de S. repens, Roystonea regia, Sabal palmetto, Sabal maritima, Colpothrinax whitii, Thrinax radiata, Veichia merrillii, Chrysalidocarpus lutescen, Wotyetia bifurcata, Caryota urens y Phytosperma elegans. Los frutos se secaron a temperatura ambiente a la sombra durante 20 días, se molieron y se sometieron a extracción con hexano en soxhlet. Los extractos lipídicos se secaron y se les determinaron el rendimiento de extracción y sus contenidos de ácidos grasos como ésteres metílicos por cromatografía de gases. Estos extractos presentaron rendimientos de extracción entre 0,6 y 25,5 %; contenido de ácidos grasos totales desde 58,4 hasta 92,5 % y contenidos de ácidos libres entre 1,9 y 70,2 %. La composición de ácidos grasos presentó como ácidos mayoritarios en las todas las especíes estudiadas el palmitico (9,2-52,2 %) y el oleico (9,6-42,7); mientras que los ácidos linoleico (0,1 - 3,4 %) y esteárico (1,7 -6,1 %) fueron los minoritarios. Los ácidos láurico, mirístico y linoleico fueron mayoritarios en unas especies y minoritarios para otras. El presente trabajo permitió caracterizar las fracciones lipídicas de once especies de arecaceas presentes en la isla, lo cual contribuyó al conocimiento de algunas de las principales características físicas y químicas, con vistas a su posible empleo en la industria farmacéutica.

**Key words:** palmas cubanas, aceite, ácidos grasos, frutos de arecaceae
The Lamiaceae family has been characterized by the occurrence of unsaturated acids in their seed oils. The importance of such acids as chemotaxonomic markers, for the cosmetic, nutritional and medicinal industries has also been demonstrated. *Salvia coccinea* L., known as banderilla and *Leonotis nepetaefolia* (L.) R.Br., known as Bastón de San Francisco are two members of this family and are mainly cultivated in Cuba as ornamental plants. The purpose of this work was to determine the fatty acid composition of seed oil of these species. Seeds were collected, milled and extracted with hexane. The extracts were filtered and the solvent was evaporated under reduced pressure in a rotary evaporator. Organoleptic characteristics and extraction yields of the oils were determined. The content of fatty acids, analysed as methyl esters, were determined by GC-FID and GC/MS. The main fatty acids found in the oil of *S. coccinea* were linolenic (33.1%), linoleic (25.2%) and oleic (13.3%) acids; whereas in *L. nepetaefolia* oil were found oleic (43.2%), laballenic (18.4%), palmitic (15.0%), and linolenic (13.4%) acids. Other fatty acids were also found but in minor proportions. The results of this study are a contribution to the chemical composition knowledge of both species and suggest that such oils could be used in several industries.

**Key words:** salvia coccinea, leonotis nepetaefolia, lamiaceae, fatty acid, gas chromatography
Varias de las especies del género Terminalia (Combretaceae) han demostrado internacionalmente propiedades farmacológicas como cardioprotectoras, hipolipemiantes, vasodilatadoras, diuréticas, antioxidantes, gastroprotectores, hepatoprotectores, antiinflamatorios y analgésicos. Estas propiedades han sido atribuidas fundamentalmente a los polifenoles y sus glicósidos, encontrados en las hojas y la corteza. Teniendo en cuenta además, que en Cuba la especie Terminalia Catappa es catalogada como una planta invasora y que existen pocos trabajos relacionados con su composición química y estudios farmacológicos, se llevó a cabo un estudio preliminar para determinar ácidos polifenólicos en el extracto metanólico de sus hojas. Las hojas fueron recolectadas, secadas, molidas, desgrasadas con hexano y posteriormente extraídas con metanol en un baño ultrasónico. El extracto se filtró y el disolvente se eliminó al vacío. El extracto seco se hidrolizó y se extrajo con acetato de etilo. Se determinó el rendimiento de extracción, las características organolépticas y los polifenoles totales mediante el método de Follin-Ciocalteu, así como la composición química por CG/EM previa formación de derivados TMS. Se obtuvo un líquido de color pardo oscuro de olor característico y con un rendimiento de extracción de 10,8%. El contenido total de polifenoles fue 184,6 (mg Pirogalol/100g Extracto) y como ácidos polifenólicos mayoritarios se encontraron el ácido gálico (3,4,5-trihidroxi-benzoico), ácido vanílico (3- metoxi-4-hidroxibenzoico), ácido 3,4-dihidroxi-benzoico, ácido 2,5-dihidroxi-benzoico, ácido 4- hidroxibenzoico y ácido benzoico. También se detectaron otras estructuras relacionadas con polifenoles. La presencia de estos compuestos pudiera justificar las propiedades medicinales atribuidas a esta especie, a la vez que servirían de base para continuar con futuras pruebas farmacológicas que avalen sus usos etnomédicos.

Key words: terminalia catappa, combretaceae, ácido gálico, polifenoles, cg/em, tms
The national and international regulator organisms demand, for the use of natural products like nutritional supplements or drugs, to know with accuracy the content of some toxic elements for men, such as the Arsenic (As). The Policosanol, Prevenox and Abexol are active pharmaceuticals ingredients extracted of the wax of the sugar cane and bees and they are constituted by mixtures of alcohols or aliphatic acids, both with high molecular weight, showing cholesterol lowering, antioxidant and gastroprotective effects. In the present study, a microwave-assisted digestion technique (HNO3- H2O2, EPA 3052) was employ for the determination of total arsenic in the three matrices of natural products by Hydride Generation - Atomic Absorption Spectrometry. The accuracy of the determination was evaluated by the standard additions method. The recoveries were satisfactory by three samples between 95% and 105. The detection limit was 0.002 µg.g-1. The content of As in the three analysed natural products were under the method detection limit as well as the established limit of the United States Pharmacopeia 33 Ed. for similar products.

**Key words:** arsenic, antioxidants, hydride generation, atomic absorption spectrometry
SPN-P-033 Characterization of by-product from the obtaining process of propolis for its possible use as nutritional supplement

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El propóleo es un producto natural que las abejas elaboran a partir de exudados de diversas plantas. Es usado por las abejas para recubrir las paredes de la colmena y mantenerla libre de bacterias. Está compuesto básicamente por sustancias resinosas procedentes de distintas especies vegetales con un 50% de bálsamos y resinas, 35% de cera, 10% de aceites esenciales y un 5% de polen. Este material resinoso es sometido a una extracción hidroalcohólica para la obtención de tintura de propóleos y extracto blando. Como subproducto de este proceso se obtiene un residuo ceroso que se saponificó, se extrajo con acetona y se purificó empleando seis combinaciones de hexano-acetona. Los mejores resultados se encontraron después de dos purificaciones con hexano y tres con acetona, obteniéndose una concentración de 94% de una mezcla de alcoholes de alto peso molecular, donde predominan los alcoholes C30 y C32. El objetivo de este trabajo fue caracterizar este residuo ceroso y evaluar su posible uso como suplemento alimentario, teniendo en cuenta las propiedades conocidas de los alcoholes presentes en la cera de abejas y la cantidad de subproducto generado en la obtención de tintura de propóleos y extracto blando.

Key words: propolis, nutritional supplement, wax
Introducción: El aceite de girasol ozonizado OLEOZON® Oral es un medicamento que presenta excelentes propiedades en la parasitosis por Giardia Lamblia. En este trabajo se presenta el estudio de estabilidad acelerada de tres lotes de producción de OLEOZON® Oral a partir de la puesta en marcha de la planta de producción de aceite ozonizado CNIC y teniendo en cuenta las indicaciones del CECMED. Materiales y Métodos: Se utilizaron frascos plástico de baja densidad y de vidrio. Los lotes en estudio se mantuvieron con las condiciones requeridas a una Temperatura de (25 ±2°C) Y 60% de Humedad. Se analizaron durante tiempo 0, 1, 2, 3 y 6 meses las características de calidad: índice de peróxido, índice de acidez, concentración de aldehídos, viscosidad, características organolépticas y límite microbiano. Se utilizó computadora y el programa estadístico Origin para el análisis de los resultados. Resultados y Discusión: Durante los seis meses de mantener la muestra en estudio acelerada en frascos de plástico y de vidrio se ha podido apreciar que el índice de peróxido a medida que transcurre el tiempo el valor de concentración en el tiempo cero disminuyó, el índice de acidez aumentó para cada uno de los lotes, la viscosidad y la concentración en aldehídos volátiles se mantuvo cercana al límite inferior de especificación, no hubo crecimiento microbiano para ninguno de los lotes estudiados y el comportamiento en los diferentes envases es similar. Conclusiones: El producto en condiciones de Temperatura de (25 ±2)0C y 60% de Humedad mantiene sus especificaciones de calidad hasta los tres meses de elaborado en las nuevas condiciones de producción.

Key words: oleozon, estabilidad acelerada
La edición digital Guía Interactiva “Plantas Medicinales” representa la obra de dos médicos y un ingeniero agrónomo, cubanos, interesados en poner al servicio de los profesionales de la salud, y de los lectores en general, una herramienta práctica para el empleo racional de las plantas medicinales. La temática abordada se presenta con un lenguaje claro y sencillo, al alcance de todas aquellas personas interesadas en los beneficios que su uso reporta sobre la salud humana. De una manera resumida y didáctica, se refleja en esta multimedia información actualizada sobre cuarenta y seis de las plantas medicinales reconocidas por las autoridades reguladoras de productos naturales y aprobadas por el Programa Nacional para el Desarrollo y la Generalización de la Medicina Natural y Tradicional. Gran parte de las plantas incluidas, son conocidas y empleadas en el mundo con muy buenos resultados en el alivio de ciertas dolencias y malestares comunes; pueden ser encontradas en cualquier latitud del planeta, solo con la distinción de que en muchas localidades y países, el nombre común cambia por la influencia de culturas y costumbres, no obstante mantienen sin variación su nombre científico; tampoco sufren modificación significativa los principios activos y componentes de las mismas, por lo cual se conserva su carácter terapéutico; en este material se reflejan además los principales usos por aparatos y sistemas, las formas naturales o farmacéuticas de presentación, su modo de empleo, la dosificación, las precauciones que deben tenerse en cuenta para su elaboración y administración; entre otros aspectos de interés. Dispone además, de una galería de imágenes que refleja las plantas de interés, un índice de fórmulas farmacéuticas y un breve glosario para facilitarle al lector no especializado la comprensión de algunos términos. Surge como resultado de la necesidad de disponer de un material integrador en la docencia de pregrado y postgrado para la formación de profesionales de las ciencias médicas en la Atención Primaria de Salud ya que los existentes hasta el momento en nuestro país, muestran los contenidos de una forma muy dispersa y fraccionada. Hasta la actualidad se han realizado diversos estudios científicos en el ámbito de la medicina natural y tradicional especialmente relacionados con plantas medicinales, sin embargo, no se dispone de una publicación que oriente adecuadamente su manejo, objetivo que nos trazamos con nuestra guía encaminada a dotar a profesionales y promotores de salud, de un instrumento práctico y eficiente en su capacitación.

**Key words:** Medicinal Plants, Interactive Guide, Guía interactiva, Plantas Medicinales
Introduction: the main objective of the inflammatory reaction is delete or dilute the injurious agent with a series of humoral and cellular events that preferably heal and rebuild to the injured tissue (Howard, 1996). Most commonly used nonsteroidal anti-inflammatory agents are the steroids and NSAIDs, both with very serious adverse effects (Rang, 2008) objective: to evaluate the anti-inflammatory activity of the aqueous extract of Sedum praealtum Methodology: extract is prepared by infusion, took dry sprinkler, 6 groups of rats each with 6 were distributed as (1) witness without trading), (3) treated with (50, 200 and 400 mg/kg of the extract), (5) with diclofenac (4 mg/kg), (6) positive control, an inflammation was induced by granuloma and the Administration was orally for seven days, at the end is dissected granulomas, stomach and blood samples. Results: phytochemical: flavonoids, alkaloids, saponins and tannins; pharmacological, you notice an anti-inflammatory process dose-dependent to exist significant differences with the positive control and with diclofenac, the histologically showed a more severe irritation of the mucosa in stomach with diclofenac, Melo and cols isolated four flavonoids with anti-inflammatory activity of this plant, which traps radicals and antioxidants are considered free (Marcano et al., 2002). The aqueous extract of Sedum praealtum DC, presented anti-inflammatory effect.

Key words: anti-inflammatory activity, Sedum praealtum, always live, San Martín de las pirámides, México
Human babesiosis is an emerging zoonotic disease caused by Babesia spp. Have importance to public health and veterinary since it has economic impact by affecting the production of cattle. In Central America and Mexico, human cases have been confirmed as in South America and Brazil. The infection of humans and animals occurs through the bite of infected ticks inoculated sporozoites that become trophozoites and reproduce, the erythrocyte rupture, releasing merozoites that invade other erythrocytes, another mechanism of transmission is through transfusion. Treatment regimes, depending on the degree of B. microti infection is used: azithromycin with clindamycin and atovaquone with quinine. These drugs cause in patient side effects so do not complete their treatment, resistant strains causing. An alternative is to search for plants with antiparasitic activity. In this paper the antiparasitic activity of the methanol extracts of Ibervillea sonorae, Krameria sonorae and Phoradendrom californicum in mice infected with Babesia microti was evaluated. Parasitemia curve was determined and the effect of the extracts was evaluated for 33 days. The results were subjected to statistical analysis and Zigma Zigma Stat Plot. Observed that two extracts showed activity in different degree. So we can conclude that plants are a potential source of chemical structures, which could be an alternative in controlling parasites.

Key words: antiparasitic activity, Babesia microti, plant species
The textile industry is one of the activities that generate more pollution due to the large volumes of water used in their different processes. Among the contaminants in effluents are textile dyes, which do not exhibit high toxicity to living organisms, however, the strong coloring that they may come to abolish the photosynthetic process. The adsorption by activated carbon is one of the most used processes in the purification of textile effluents; however the high cost of this material has led to research into new adsorbent materials, including agro-industrial waste. In this work, the peanut shell and parchment coffee were characterized as adsorbents for Orange II (NII), Acid Blue 113 (AA 113) and Basic Green 4 (VB4). The wastes were ground, sieved and subjected to different pre-treatments (acidic, basic and microwave), and then the kinetic adsorption studies were performed. Of the three dyes tested, only AA 113 and VB4 were adsorbed by both materials during the first 6 hours of treatment. Optimization of dye adsorption was conducted using a central composite design by analyzing the effect of exposure time (2 and 10 min) and power (10 to 50%) of the microwaves pretreatment on the adsorption capacity. It was determined that the removal of dye by the residue increased when the power and the microwave exposure time had the higher value.

**Key words:** adsorption, waste, coffee, peanuts, textile dyes, microwave
SPN-P-039 In Vitro Activity of some fractions from extract of Ruta graveolens against Trypanosoma cruzi NINOA strain

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Trypanosoma cruzi causes Chagas disease. It is estimated that 11–18 million people in South America are infected by this disease and 100,000 people are at risk of contracting it each year. The chemotherapy used to treat this illness has serious side effects and its efficacy on the chronic phase of disease is still a matter of debate. Previous studies have shown trypanocidal effect of Ruta graveolens. The objective of this work was to find the active fractions, against T. cruzi strain NINOA, in vitro evaluation, after making a chromatographic separation of the chloroform extract. Bioassay-guided phytochemical study of the chloroform extract of R. graveolens was carried out using as toxicity bioassay Artemia salina. The in vitro trypanocidal assays were carried out according to the protocol reported elsewhere, with slight modifications. Bloodstream trypomastigotes were quantified by the method of Pizzi. The chloroform extract showed a 67% mortality Artemia salina, was separated by column chromatography. Fractions 232, 234, 306, 311, 328 and 397 which presented the lowest LC50 for biotoxicity test were selected. Fractions 306 and 328 for a second chromatographic separation were chosen, obtaining fractions 292-318 and 340-390 which presented an LC50 less. Of fractions tested against Trypanosoma cruzi was found that the fraction from 340 to 390 at a concentration of 100 ug / ml showed higher percent lysis against Nifurtimox reference drug concentrations 50, 25 and 100 ug / mL.

Key words: trypanosoma cruzi, trypanocidal activity, ruta graveolens
Evaluation of the in vitro effect of plant extracts of Ruta graveolens and Euphorbia serpyllifolia on Trypanosoma cruzi strains NINOA and INC-5

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Chagas disease caused by the protozoan Trypanosoma cruzi is life-threatening, treatment with Benznidazole and Nifurtimox base ensures no cure for the disease, causing side effects causing patients to discontinue treatment. In this work, in vitro trypanocidal activity of methanol extracts of Ruta graveolens and Euphorbia serpyllifolia on Trypanosoma cruzi strains Ninoa and INC - 5 was determined. The strains were maintained by serial passage in CD1 mice, inoculated intraperitoneally with infected blood. Determining the trypanocidal effect in vitro was performed by placing blood trypomastigotes with plant extracts and reference drugs to a final concentration of 10, 25, 50 and 100 mg / mL. Percent lysis was determined by the method of Pizzi, parasite numbers that survived the treatment, comparing with the negative control. With Ninoa strain showed Ruta graveolens a high percentage of lysis, while Euphorbia serpyllifolia had no effect, Benznidazole obtained a percentage of 55,25,20 and 5 %, Nifurtimox 47,29,10 and 3%. The INC- 5 strain, Ruta graveolens obtained a low percentage of lysis, while Euphorbia serpyllifolia showed little activity, Benznidazole was 28, 18, 13 and 2 %, Nifurtimox was 81, 72, 62 and 52%. The methanol extract Ruta graveolens trypanocidal has greater activity than Benznidazole and Nifurtimox on Trypanosoma cruzi strain Ninoa concentrations tested.

Key words: ruta graveolens, euphorbia serpyllifolia, trypanosoma cruzi
Nutraceutical quality components of microencapsulated noni juice (Morinda citrifolia L.) by spray drying

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Noni is a fruit with nutraceutical potential, but its unpleasant odor and flavor limit its consumption. Encapsulation is the process to entrap active agents into particles; it is a promissory technology that may allow mask bad taste and bad smell components. The aim was to assess the nutraceutical potential in noni juice microencapsulated by spray drying. The drying temperatures were 180 and 85 °C of inlet and outlet, respectively. The carrier materials (maltodextrine DE-10, arabic gum, and soy lecithin) were mixed in different proportions, searching maximum bad-taste and bad-smell masking and minimum active compounds alteration. Noni juice was obtained from ripe, cleaned, and scalded fruits (92 °C 5 min). A pulp-removing machine was used and then the juice was obtained by centrifugation of the pulp (11000 G, 5 °C 30 min). Phenols, flavonoids, vitamin C and antioxidant activity (by the DPPH inhibition technique), drying efficiency and residual humidity were measured. There was some important alteration of active compounds in different levels (upon the compound) by high temperatures, however, the best treatment of microencapsulated noni juice was with 70 % maltodextrin + 29.9 % arabic gum + 0.1 % soy lecithin (treatment 3) to keep the major quantity of total phenols, antioxidant activity, this treatment showed the highest drying efficiency and caused the lowest residual humidity. A high positive correlation between phenols and antioxidant activity was observed. The sensory evaluation showed that the microencapsulated noni juice with treatment 3 was significantly less rejected (22 %) than the natural juice (46 %).

Key words: phytochemicals, microencapsulated, spray drying, antioxidant activity, sensory evaluation
Noni is a bush of Asiatic origin which produces fruits that when ripe have an unpleasant and penetrating smell. The noni has a lot of popularity because of its nutraceutical and healing properties, however, its unpleasant smell disfavors its consumption. The microencapsulation by multiple emulsions is an alternative for masking smells and minimize the factors that interfere in secondary metabolites stability. The nutraceutical compounds of noni juice are sensible to high temperatures, for which the objective of this study is to microencapsulate noni juice by multiple emulsions disregarding the use of heat. To achieve this procedure noni juice was trapped in multiple emulsions, using as wall material soy protein isolate (SPI) and Arabic gum (GA). Phenolic, flavonoid and ascorbic acid content of three emulsions W/O/W with polymer concentration (SPI-GA) of 6, 7.5 and 9 % were measured. The results showed that did not exist significant difference ($P < 0.05$) in phenolic and ascorbic acid content among treatments. The levels of flavonoids were higher in the treatments than the ones that existed in the noni juice control. As conclusion the polymer content in the multiple emulsions did not interfered in the phytochemical content among treatments.

**Key words:** multiple emulsions, secondary metabolite, nutraceutical, noni juice
SPN-P-043  Antifungal activity of the root of Adenophyllum aurantium (L.) Strother

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Medicinal plants research has made available substances for the treatment of different human and agricultural pests. The natural products represent an alternative to synthetic antifungal. México is the country with more species of Asteraceae in the world, accounting for 12-15% of México's vascular flora. It has been reported that the Asteraceae family is an important source of pesticides. Specie that belongs to this family is Adenophyllum aurantium (L.) Strother known as 'ruda montes' endemic of México. The presence of bioactive compounds has been demonstrated recently against root nematode Nacobbus aberrans and fungal pathogens Fusarium solani and Alternaria alternata. Hexane and the ethyl acetate extract of root were prepared to determine inhibition of growth mycelial in Fusarium oxysporum and Rhizoctonia solani; the agar diffusion assay, Petri dishes with potato dextrose agar, thiabendazole (+) and ethanol (-) controls was carried out. Mycelial growth rate of the second, third and fourth day of incubation was determined. Like preliminary result was found that fungi were sensitive to extracts, and it would be interesting to identify the substance responsible for this activity.

Key words: adenophyllum aurantium, antifungal activity, growth rate
Wax cochineal is an agrowaste product which contains considerable amount of coccera. This has a good potential for obtaining policosanol. The objective of this study was to examine the aliphatic alcohols content of wax cochineal (Dactylopiauscoccus) extracts from nymphs and adult stages. Extracts were obtained by applying alkaline hydrolysis reaction. All the obtained extracts were characterized chemically by Fourier Transform Infrared Spectroscopy (FTIR) in the attenuated total reflectance (ATR) sampling mode. For other side, the aliphatic alcohols were identified and quantified by Gas Chromatography coupled to Mass Spectrometric techniques (GC-MS). ATR-FTIR analysis indicate presence of aliphatic alcohols in all the extracts, which was confirmed by GC-MS. The composition of aliphatic alcohols of the waxy extracts derived from cochineal was very similar, being 1-triacontanol the most abundant alcohol. The highest content of 1-triacontanol was found by the method 1 (5.24±0.36 g/kg), followed of method 2 (1.575±0.43 g/kg) and method 3 (0.520±0.03 g/kg) of wax cochineal from adult stage. In wax cochineal extracts from nymphs stage, was found likewise by method 1 the highest content (1.66±0.41 g/kg), followed of method 2 (0.515±0.05 g/kg) y method 3 (0.173±0.07 g/kg) (p<0.05). High yield of triacontanol was recuperated of cochineal wax in both insect stages. This component is important in the farmaceutical industry and in the agriculture, by the which wax cochineal could be an important source of triacontanol.

**Key words:** policosanol, triacontanol, cochineal, wax, insect
Introducción. La hipertensión arterial es una enfermedad crónica, de etiología multifactorial y que corresponde a la elevación persistente de la presión arterial sobre los límites referenciales. Entre los problemas de salud a los que puede verse enfrentado el paciente hipertenso, destacan las alzas de presión arterial. Aquí, el paciente puede optar por acudir a algún sistema público de urgencia o realizar medidas de autoatención. A nivel popular y en diferentes fuentes de información, se menciona que el zumo de Citrus limon L. Burm. f. (Rutaceae) es útil para disminuir la presión arterial. Sin embargo, este uso no tiene un respaldo científico del todo claro. Objetivo. El objetivo del trabajo fue analizar el uso del zumo de C. limon, en el manejo del alza de presión arterial. Metodología: Para esto se analizó la composición química de frutos de C. limón disponibles en la ciudad de Concepción, Chile, a través de métodos espectroscópicos y cromatográficos. Para caracterizar el uso del zumo, se aplicó una encuesta a 506 pacientes hipertensos del CESFAM Dr. Víctor Manuel Fernández de la ciudad de Concepción, y se realizó un estudio piloto con 5 pacientes hipertensos, para evaluar efecto del zumo frente un alza de presión arterial. Resultados: Como resultados se identificaron polifenoles presentes en el zumo de C. limon como los flavonoides eriocitrina, hesperidina y diosmina. Con respecto a los datos compilados de la encuesta, el 95.2% de la población encuestada ha usado alguna vez zumo de C. limon ante un alza de presión arterial, con una mejoría de los síntomas aproximadamente en media hora. En el estudio piloto se observó una disminución de la presión arterial en hipertensos en situación de alza de presión, que tiende a ser sostenida en el tiempo, no así en pacientes normotensos. Discusión y Conclusión. El uso del zumo de C. limon frente un alza de presión arterial, está ampliamente arraigado en la cultura popular y se concluyó que disminuye la presión arterial por un mecanismo de acción no establecido, probablemente relacionado con el contenido en polifenoles y vitamina C, compuestos a los cuales se les han atribuido efectos antihipertensivos.

Key words: Citrus lemon L., treatment, arterial pressure
Introduction. Diabetes mellitus is a chronic disease that has become increasingly prevalent in recent decades. The objective of treatment is to reduce cardiovascular risk. The use of medicinal plants in the treatment of chronic diseases has attracted scientific interest in recent years. In Chile, a common form of alternative medicine for diabetes is the leaf of the Bauhinia forficata ("Pata de Vaca") plant. Objective. To evaluate the hypoglycemic effect of B. forficata leaves, administered as complementary therapy for patients with poorly controlled type-2 diabetes mellitus. Methodology. A semi-experimental study was conducted on Chilean patients in a primary healthcare setting who have poorly controlled type-2 diabetes mellitus (HbA1C >7%). These patients were administered a 0.4% infusion of B. forficata leaves, twice daily for a period of three months, and then a period of one month without infusion. Clinical parameters such as body mass index (BMI), blood pressure and blood glucose were measured every month. Total cholesterol, triglycerides and glycated hemoglobin (HbA1C) were measured every three months. Results. For patients receiving the infusion of B. forficata leaves, a decrease in HbA1C levels of 0.5% was observed. No significant changes in weight, blood pressure, total cholesterol, triglyceride and glucose levels were observed. Discussion and Conclusion. The infusion of B. forficata leaves, when used as an adjunct to pharmacological treatment along with a healthy lifestyle, may decrease the levels of HbA1C in the adult diabetic population.

Key words: hypoglycemic effect, Bauhinia forficata, patient, diabetes
Heterakis gallinarum impedes the productivity of village chickens and hence their socio-economic contribution to rural livelihoods. Smallholder farmers are endowed with vast indigenous knowledge for controlling H. gallinarum and they predominantly use Aloe ferox, Agave sisalana and Gunnera perpensa. However, their anti-helminthic efficacy is unknown. Therefore, the study objective was to determine the anthelmintic efficacy of A. ferox, A. sisalana and G. perpensa against H. gallinarum in vitro. Ten H. gallinarum worms were randomly introduced into each of the 42 petri-dishes. In triplicate, treatments were positive control (distilled water), negative control (mebendazole) and A. ferox, A. sisalana and G. perpensa aqueous leaf extracts with doses 7.25, 14.5, 29 and 58 mg/mL. Worm motility inhibition and worm recovery was noted over 60 h. Data was analysed using general linear model of Statistical Analyses System. Agave sisalana (14.5 mg/mL) had highest (80%) worm motility inhibition at 12-h interval (p<0.05) and highest worm mortality index (80%) showing that the plant has anthelmintic properties. Worms (70%) were recovered after 48 h following treatment with A. ferox (58 mg/mL) and 50% for G. perpensa all dose levels. This signifies that G. perpensa has moderate anthelmintic activities while those of A. ferox are primarily ascribed to its purgative effects. Agave sisalana (14.5 mg/mL) was the most efficacious in reducing motility and causing mortality of H. gallinarum in vitro. Findings are useful to resource-constrained farmers for selecting the best plant and pharmaceutics for manufacturing anthelmintic drugs.

Key words: chickens, heterakis gallinarum, plant leaf extracts, worm mortality, worm motility
SPN-P-048  Immune modulatory effect of Korean native chicken meat

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Introduction: Chicken essence has been well known for its health-promoting and enhancement of body immunity. However, the mechanism underlying its effect remains unclear. This study aims to examine the immune-modulatory effect of four different Korean native chickens (Yeonsan Ogye, Hyunin Black, Hwangbong, and Hoengseong Yakdak) on macrophages. White Leghorn was used as a control. Materials and Methods: Breast and leg meat were grounded and extracted with 70% ethanol at 60 oC for 6 hours. The proliferation of macrophage like RAW264.7 cells was assayed by a colorimetric immunoassay, which is based on quantititing bromodeoxyuridine (BrdU) incorporation into the newly synthesized DNA. The uptake of fluorescein-labeled Escherichia coli was measured to determine whether chicken extract stimulates the phagocytic activity of RAW264.7 cells. The quantity of nitric oxide (NO) was determined by Griess assay in LPS-stimulated RAW 264.7 cells. Results: All chicken strains of breast meat extracts (50-200 ug/mL) increased cell proliferations while leg meats fail to affect cell proliferations. Breast meat extracts (200 ug/mL) of two strains of Korean native chicken (Hyunin Black and Hoengseong Yakdak) enhanced the phagocytic ability. Furthermore, LPS-stimulated release of nitric oxide was significantly decreased by breast meats of four Korean native chicken strains in dose-dependent manner while breast meat of White Leghorn fail to affect nitric oxide release. Nitric oxide production was not affected by leg meats. Conclusion: These results suggest that Korean native chicken meat, especially breast meat, stimulates immune functions of macrophages.

Key words: Korean native chicken, raw 264.7 cells, nitric oxide, phagocyte activity, immune function
Concentrations of Se as an essential metal in human milk of Venezuelan women

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Human milk is a biological fluid, which provides all the nutrients, including essential metals that are required for the development and growth of newborn children during the first months of life; also, it contains enzymes, specific proteins, nucleotides, etc. Se concentration in milk is directly affected by levels in the food chain, also is a component of two enzymes, glutioneperoxidase that protects against oxidative damage, and of type I iodothyronine 5-de-iodinase, this antioxidative cellular Se might have a role in preventing Cancer. As essential nutrients, many chemicals can be transferred from body storage sites and the blood of nursing mothers to breast milk. Nowadays, researches on the importance of essential trace metals in human milk, its nutritional quality and its relationship with environmental factors are growing. In this work, it is presented the total concentrations of Se in samples in human milk using ICP-MS. The samples were collected from breastfeeding women between 2 and 8 post-partum months in the Hospital of Children of Veritas in Maracaibo city. For destruction of organic material, digestion of the real samples with HNO3 / H2O2, both concentrated, was applied. The total metal concentration was 0.011 mg L−1 of Se. The limit of detection (LD=3σ/m) obtained was 0.003 for Se. The precision (expressed as RDS) was < 5%. Accuracy was assessed by recovery studies, obtaining mean recoveries percentages of 100 ± 5%. Developed methods for the determination of total concentration of the studied metal was accuracy, precise and free from interferences.

Key words: human milk, essential metal, icp-ms
Chemical composition of Salvia aratocensis extract obtained by supercritical fluid extraction and its antigenotoxicity against ultraviolet radiation-induced DNA damage

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Introduction: Salvia is a cosmopolitan and important genus, widely used in flavoring and traditional medicine for treatments of respiratory and gastrointestinal diseases. The present work evaluated the chemical composition of Salvia aratocensis extract obtained by CO2 supercritical fluid extraction, as well as its DNA protective effect against ultraviolet radiation (UVR)-induced genotoxicity. Material and Methods: S. aratocensis extract composition was determined by gas chromatography-mass spectrometry (GC-MS). The antigenotoxic effect of S. aratocensis extract was assayed in co-incubation procedures using the SOS chromat test in Escherichia coli PQ37 cells. Results and Discussion: The major compounds of the plant extract were \( \beta \)-cadinol (31.8%), 1,10-epi-cubenol (12.9%), trans-\( \beta \)-caryophyllene (7.7%), \( \beta \)-cadinene (6.6%), and squalene (6.5%). S. aratocensis extract protected bacterial cells against UVR-induced genotoxicity at doses between 0.063 and 1 mg/mL. The results were discussed in relation to the DNA protective potential of the S. aratocensis extract and its major constituents.

Key words: salvia aratocensis, supercritical fluid extraction, antigenotoxicity, sos chromat test, ultraviolet
Lippia origanoides extract obtained by supercritical fluid extraction and its major constituent pinocembrin diminish ultraviolet radiation-induced DNA damage and modulate cell division in Escherichia coli

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Introduction: Lippia origanoides is an aromatic shrub native to Central and South America. The plant is used as seasoning and in traditional medicine for treatment of gastrointestinal, genitourinary and respiratory diseases as well as anti-inflammatory and analgesic remedy. The present work evaluated the chemical composition of L. origanoides extract obtained by CO2 supercritical fluid extraction, and its antigenotoxic activity against UVR-induced genotoxicity. Major constituent of this extract, pinocembrin, was also studied.

Material and Methods: L. origanoides extract composition was determined by gas chromatography-mass spectrometry (GC-MS). The antigenotoxic effect of L. origanoides extract was assayed in co-incubation procedures using the SOS chromotest in Escherichia coli PQ37 cells. Fluorescence microscopy analysis was used in cell treatments in order to determine interferences with cell division; different nucleoid shapes categories were measured.

Results and Discussion: The principal constituents of the plant extract were pinocembrin (54.9%), trans-? caryophyllene (10.9%), carvacrol (6.3%), p-cimene (6.2%), thymol (4.6%), and 1,8-cineole (3.5%). L. origanoides extract protected bacterial cells against UVR-induced genotoxicity at doses between 0.016 and 1 mg/mL; this effect was related to its major constituent pinocembrin. Fluorescence microscopy analysis showed that pinocembrin interfered with E. coli cell division, what provide first evidence on bioantimutagenic potential of the pinocembrin. The results were discussed in relation to the DNA protective potential of the L. origanoides extract and its major constituents.

Key words: antigenotoxicity, sos chromotest, ultraviolet radiation, lippiaoriganoides, supercritical fluid
SPN-P-052 Chemical composition of essential oil and extract obtained by supercritical fluid extraction from Turnera diffusa and its antigenotoxicity against UVR-induced DNA damage

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Introduction: Turnera diffusa, commonly known as damiana, is a shrub largely used as aphrodisiacs in American traditional medicine, but also for treatments of skin, respiratory and gastrointestinal diseases. The present work evaluated the chemical composition of T. diffusa essential oil (EO) and extract obtained by CO2 supercritical fluid extraction (SFE), as well as, its DNA protective effect against ultraviolet radiation (UVR)-induced genotoxicity. Material and Methods: T. diffusa extracts compositions were determined by gas chromatography-mass spectrometry (GC-MS). The antigenotoxic effect of T. diffusa EO and extract were assayed in co-incubation procedures using the SOS chromotest in Escherichia coli PQ37 cells. Results and Discussion: The major constituents in T. diffusa EO were drima-7,9(11)-diene (28%) and dihydro-karanone (25%); in the extract obtained by SFE mostly 3-deoxy-isopetasol (64%) and 4,5-di-epi-aristolochene (22%), were detected. Both T. diffusa EO and extract obtained by SFE protected bacterial cells against UVR-induced genotoxicity in a dose-dependent manner. These results were discussed in relation to the DNA protective potential of the T. diffusa EO and extract obtained by SFE and their major constituents.

Key words: turneradiffusa, essential oil, supercritical fluid extraction, antigenotoxicity, sos chromotest,
The majority presence of polyunsaturated fatty acids and phenolic compounds in seed oil extract of Carapa guianensis Aublet has great potential as antioxidant. But the extract effect on DNA it is ignored. The aim of this work was evaluated the mutagenic and genotoxic potential of the extract. The studies were performed in vitro (Ames test) with and without addition of the microsomal hepatic fraction, in vivo assays that included on peripheral blood leukocytes of Sprague-Dawley rats using the comet assay, the micronucleus assays and chromosomal aberrations in BALB/c mice of both sexes administered during 14 days orally in repeated doses (400, 1000 and 2000 mg/kg) and testing of the sperm head morphology of male BALB/c mice administered during 35 days with doses of 400, 1000 and 2000 mg/kg. In bacteria, the extract did not cytotoxic and mutagenic. In the in vivo assays, there was not increase the formation of alkali labile sites in DNA of SD rats leukocytes and there was not increase the frequency of bone marrow cells with micronucleus and cells with chromosomal aberrations (structural and numerical). It did not increase in the spontaneous frequency of anomalies in the sperm head morphology; however, the results of the sperm concentration in the treated group with 2000 mg/kg of the extract were different with the negative and positive control groups. The results indicate that this extract is not mutagenic or genotoxic according to the performed assays.

**Key words:** carapa guianensis, genotoxicity assay, antioxidant
The seed oil extract of Carapa guianensis Aublet is a natural product constituted by a complex mixture among whose components were the polyunsaturated fatty acids and terpenoids. The aim of this work was to evaluate the potential of the extract like antioxidant nutritional supplement in Sprague-Dawley rats with future potential as antimutagenic supplements useful in support of antineoplastic therapy. Different dose of the extract 100, 200 and 400 mg/kg were administered in co-treatment with Cyclophosphamide (CP) a potent inductor of oxidative stress and it was also administered the maximum dose of the product 400 mg/kg. In all the cases were determined the influence of these treatments in the water and foods intake, corporal weight, haematology and biochemistry sanguine test and the plasmatic antioxidant state. The obtained results demonstrated that the consumption of the extract in co-treatment with the CP did not affect the water and foods intake neither the increase the corporal weight when 200 mg/kg doses were used. The haematological and biochemical parameters analyzed were not affected for none of the evaluated doses, being able to correct the hepatotoxicity caused by the drug and the disorders in the lipid metabolism. The seed oleaginous extract of Carapa guianensis Aublet was a good antioxidant that which demonstrated in the reported values of enzymatic activity, of the main biomarker of oxidative stress used. With these results, we recommend the beginning of the security studies (preclinical toxicology studies) and the preclinical antimutagenic studies of the extract useful during the antineoplastic therapy.

**Key words:** carapa guianensis aublet, antioxidant, in vivo, antimutagenesis, cyclophosphamide
The extracts hydro-alcoholic obtained starting from the leaves of Schinus terbinthifolius Raddi (false copal), are broadly used by their anti-inflammatory and antimicrobial properties. Our objective was to develop a farmacognóstico study of Schinus terebinthifolius Raddi (false copal). The botanical species was classified and it evaluated the morphology of the leaves visually. The drying study was developed under different conditions being measured humidity, drying temperature and loss of weight. The vegetable dry material was stored in flask of glass amber, bags of paper kraft and polyethylene bags being evaluated humidity, characteristic organoleptic and microbial load. Starting from an extract hydro-alcoholic to 80% obtained by maceration secondary metabolites were identified using the technique of Rondina and Coussio (1969) modified by Durand (1986). To this extract they were determined physical-chemical parameters: characteristic organoleptic, pH, relative density, and refraction index, alcoholic content and total solids. Metabolites of interest were identified by chromatographic in thin layer. Total phenols were quantified by visible UV to 765 nm. The results threw that it was about the species Schinus terebinthifolius Raddi in correspondence with the consulted bibliography. The solar dryer and the nylon bags were the most efficient methods for the obtaining and storage of the vegetable dry material. Secondary several metabolites was identified in the extract hydro-alcoholic (pH: 5.30, density: 0.8902, total solids: 8.4%, refraction index: 1.3786, alcoholic content: 72%), steroids, resins, alkaloids, phenols and tannins being obtained, a 7.88% of total phenols agreeing with that obtained by other authors.

Key words: schinus terebinthifolius raddi, farmacognóstico, physical-chemical parameters
SPN-P-056  Extraction of flavonoids from leaves of Annona muricata L.

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The soursop (Annona muricata L) is used in the ethno-medicine for the treatment of different diseases. The presence of flavonoids in the leaves have been determined which have antidiabetic, antioxidant and anti-inflammatory properties. The study aims to evaluate the influence of operational parameters in obtaining extraction of flavonoids from the leaves of Annona muricata L. Materials and Methods: The evaluation of the operating variables was performed using an experimental design of response surface rotational comprised central $2^3$ with star points, being selected the intervals between 50 and 70 mL/g for the relation vegetal-volume material of solvent, between 40 and 80% the ethanol concentration and between 2 and 3 h the extraction time. The quantification of total flavonoids was performed by a colorimetric method at 430 nm, expressed as quercetin. Results and Discussion: the presence of flavonoids and quercetin in the extracts was determined. The significant variables resulted the quadratic effects of plant-material relationship solvent volume and the concentration of ethanol. The optimum extraction condition in total flavonoids was obtained at a concentration of ethanol of 96%, a ratio of 70 mL/g and a time of 1.6 h, to yield 87% process. To the best experimental condition triplicate experiments were performed and compared with the value predicted by the design, a good correlation was found between them. Conclusions: The best extraction conditions were selected in obtaining an extract from the leaves of soursop with presence of flavonoids.

**Key words:** annona muricata l, flavonoids, solid-liquid extraction
Study in silico of the DNA protective effects of the 2,6-disecbutilphenol extracted from the plant Phyllanthus orbicularis K

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The appearance of chronical degenerative deseases has a close relationship with the DNA damage in somatic cells. The damage can be originated by different genotoxic agents, as for example the aromatic amines. At the present it’s considered as possible way to prevent and treat these affections the use of chemoprotective fitocompounds. It has been studied that the total aqueous extract of the cuban specie Phyllanthus Orbicularis K reduce significantly the damage induced by the aromatic amines: m-PDA), 4-aminobiphenyl (4-ABP) y 2-amino- 1-methyl-6-phenylimidazo[4,5-b]pyridine (PhIP) and from the total aqueous fractioned extract was isolated and identified the 2,6 – disecbutilphenol (DSBP). However, it is not known how the DSBP protects at a molecular level. Thank’s to the advances in computational chemistry and modelations tools is posible to study the direct chemical interaction between the DSBP and the aromatic amines as possible genoprotective mechanism. The differentes association complexes between the phenol and the aromatic amines were generated by Multiple Minimum Hypersurface (MMH) and optimized with the semiempirical methods AM1 and PM6 – d and PM7. The association energies for each complex were obtaining with the program Q3. The results allowed demonstrated that the association between the phenol and the amines was thermodynamically favored and it also can be predicted a genoprotective order for the phenol against each amine. This research work allow us to propose the chemical direct interaction between the phenol and the amines as possible molecular mechanism for the DNA protection.

Key words: dna, in silico, phyllanthus orbicularis k, dsb, m – pda, 4 - abp, phip
SPN-P-058 Malpighia glaba L. (Acerola): analytical studies

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Introduction: Malpighia glaba L. (Acerola, Miguel variety) has beneficial health virtues, is used in traditional medicine as an immunostimulant, detoxifying, remineralizing, among others. Methods: agricultural studies were conducted at the Experimental Station of Medicinal Plants Dr. Juan Tomas Roig of the Drugs Research and Development Center (CIDEM), determining the optimal harvest time. While technological and analytical studies were conducted in the research department belonging to the same center, demonstrating through a chromatographic method by High Performance Liquid Chromatography (HPLC), applicable to the quality control and stability study the high content of Vitamin C, which is attributed to their therapeutic effects. Simultaneously phytochemical studies for identification by Thin Layer Chromatography (CCD) of different components in the plant material is performed in addition specifications established quality finished product using the method of K tabulated. Results: agricultural studies identified the best time of year to harvest the plant material. The method for controlling quality and stability study was specific, accurate, linear, accurate in the range of concentrations studied. The phytochemical studies proved the presence of saponins and flavonoids in plant material. Conclusions: a raw material of natural origin that meets quality specifications required for the development of pharmaceutical products was obtained.

Key words: Malpighia glaba L., Acerola, analytical study
This study was conducted to determine the effect of dietary supplementation with sanguinarine on growth performance, serum metabolites and antibodies in growing pigs. A total of 120 [Yorkshire×(Duroc×Landrace)] pigs, at 6 weeks of age (3 weeks postweaning) were randomly allotted to 3 dietary treatments. The dietary treatments were: the basal diet (BD)-fed group, the sanguinarine (sangrovit®)-fed group (BD+0.04 g/kg), and the antibiotic-fed group (BD+0.2 g/kg colistin) (n=40 per group). On days 7, 14 and 28 after initiation of treatment, serum levels of amino acids (AA), antibodies and some biochemical parameters were measured. Sanguinarine used as supplemental increased (P<0.05) the average daily feed intake (ADFI), average daily gain (ADG), nutrient intakes and final body weight (BW), improved the feed/gain ratio (F/G) (P<0.05), and reduced (P<0.05) the incidence of diarrhea in growing pigs compared with the non-additive and antibiotic groups. Results indicated that the sanguinarine increased (P<0.05) the serum contents of AA, such as Gly, Ile, Lys, Met, Arg, Ala and Thr in comparison with the antibiotic-supplemented group and/or BD on days of trial. In contrast to colistin, sanguinarine-fed group increased (P<0.05) serum concentration of triglycerides and increased (P<0.05) concentration of glucose, as well as this medicinal extract enhanced (P<0.05) serum concentration of IgG, but only on day 7 in relation to the other 2 groups. These findings indicate a beneficial effect of the sanguinarine on growth performance, serum concentration of some essential and non-essential AA, humoral immune response and diarrhea incidence, which may offer an effective alternative to antibiotics for growing pigs.

Key words: sanguinarine, pig, amino acid, serum antibody, serum metabolite, growth performance
Purslane (Portulaca oleracea L.) is a widely extended plant in Cuban fields and its use for alimentary and medicinal purposes is well known. Traditionally it has been employed as diuretic, anti-helmintic, anti-inflammatory, etc. It is reported that Portulaca oleracea L. can reduce atherosclerosis after prolonged hypercholesterolemia due to the presence of phytosterols which are able to inhibit the intestinal absorption of cholesterol. In this work, a method by High Performance Liquid Chromatography (HPLC) for qualitative and quantitative analysis of stigmasterol and ß-sitosterol in Portulaca oleracea L. powder was developed. A Lichrospher RP-18 (5 µm) 125 X 4 mm chromatographic column (reversed phase); isocratic operation mode and UV detection at a wave length of 208 nm were set. The developed method was validated for the quality control of the mentioned plant powders by means of the following parameters: linearity, precision, accuracy, specificity and detection / quantification limits, as described in international standards. The developed and validated technique allowed to simultaneously determining stigmasterol and ß-sitosterol in Portulaca oleracea L. powders. A 0,243 % (dry base) of stigmasterol and 0,815 % (dry base) of ß-sitosterol were obtained from quantitative analysis. The HPLC method was reliable and could be employed as a quality control method.

Key words: phytosterols, hypercholesterolemia, hplc, validation, portulaca oleracea l
SPN-P-061  Toxicity studies of Curcuma longa extracts

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The CUMIN EXTRACT is a pharmaceutical form prepared from the Curcuma longa L. The Curcuma longa L. is harvested from the north-east of Pinar del Río province, Cuba. From 1995 was isolated an antirretroviral protein (Tyrphostines) of 18-19D molecular weight (Garcia Martin J. et al 2003.NPU). Toxicity in Curcuma longa extracts was our principal objective in this case. In first place biological assays were realized using 40 female mice from Switzerland Stock of-1 (Barcelona), with maintenance diet using Aqueous Extract of Curcuma longa L. Twenty mice received extract and twenty were the control group. This group received a standard diet. Experimental studies with 4mg/kg mouse per day = 0.4 mg/Kg mouse per day with curcumin in human doses. The evaluation made to the mice was about in the Thomas media measure, weight sequence, muscular coordination method, String-Test of Miguel Blasco (Don't fall in a row), Spontaneous explorer (Labyrinth test in T: explore the first arm of the labyrinth in 60 seconds). Passing four weeks the mice were sacrificed and we take blood from the neck, a piece of liver (Lipid peroxidation) looking for Malonaldehyde concentration by thiobarbituryc method and proteins concentrated by Lowry method. As a result of this research we obtained that Curcuma longa extracts didn't cause any toxicity in mice, besides decreased lipids peroxides in blood and finally the 25 higher doses of Curcuma are not toxics.

Key words: toxicity studies, Curcuma longa, cumin
The search of new molecules from natural sources to treat infectious and parasitic diseases has pointed antimicrobial peptides as a promising alternative. Besides, the important role of these peptides in innate immunity, especially in invertebrates, makes this group of animals a secure source for obtaining antimicrobial peptides. Due to the different characteristics and versatility in functions of these molecules and the continuous development of resistance to conventional antibiotics in many microbial strains; the aim of this work is to improve the antimicrobial activity of Cm-p1, a peptide isolated from the sea mollusk Cenchritis muricatus. It was performed from synthetic derivative peptides from Cm-p1 taking into account properties such as Boman index, cationicity, and mass weight of the original fragment of Cm-p1, parameters currently related to antimicrobial activity. In order to accomplish our objective, the antibacterial and antifungal activities of the three synthetic peptides Cm-p3, Cm-p4, Cm-p5 were evaluated and compared with Cm-p1. They showed a little and null antibacterial activity in contrast to antifungal activity which was increased mostly Cm-p5. This peptide resulted, conveniently, non-toxic for animal cells in vitro, at relatively high concentrations.

Key words: antibacterial activity, antifungal activity, antifungals, minimum inhibitory concentration.
SPN-P-063  Applications of essential oil clove as fungicide on environments of archives and libraries

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The preservation of historical memory has been a priority of all civilization. Archives and libraries hoard much of the documentary heritage on paper. Though, sometimes there are problems of biodeterioration caused by fungi, conditioned mainly by fluctuations of environmental parameters which promoted the development thereof. For this in the biodeterioration control, chemicals have been used, although it has not been a definitive solution, as these may affect the health of staff, the document and the environment, therefore is continued in the search for safer products. At present, scientific studies are conducted in order to find alternatives for controlling biodeterioration by using plant products as biocides. The present study aims to evaluate the antifungal activity of the essential oil of clove against three strains from environments of the repositories of the National Archives of the Republic of Cuba and its effect on paper. The following procedures were used: the wells method and the tube dilution method, biofilm inhibition test and biocidal activity test on paper colonized by fungi. On paper was measured the copper number, the pH value and the appearance of stains before and after accelerated again. The results showed biocidal activity to values of 3% of the oil, affectation in the conidiogenesis, the formation of biofilms was inhibited as well as the fungal colonization. Also, there was no significant changes in the paper pH, no structural changes in the paper, or stains on them.

Key words: essential oils, vegetable biocide, fungicidal activity, biodeterioration, documentary heritage
Numerous population studies have linked elevated concentration of total cholesterol in plasma with increased incidence of atherosclerotic events. The leaves of Moringa oleifera Lam (Moringaceae) are used in many cultures as a hypocholesterolemic agent in obese patients. Taking into consideration that B6.129P2-Apoetm1Unc/JKO mice exhibit a marked increase in plasma cholesterol level and develop atherosclerotic lesions resembling those seen in humans; the main objective of our study was to evaluate the hypocholesterolemic and anti-atherosclerotic effects of crude extract of leaf of Moringa oleifera in ApoE KO mice. Males and females B6.129P2-Apoetm1Unc/JKO with 4 - 8 weeks of age were fed ad libitum with normal diet. The half of the animals was fed with crude leaf extract of Moringa oleifera during 90 days and the other served as control. C57Bl/6 mice were used as control. Body weight, food consumption, haematological and biochemical parameters, relative weight of the organs and aortic atherosclerotic lesions were measured. It was found that administration of the crude leaf extract of Moringa oleifera decreased the cholesterol increases in ApoE KO mice serum in comparison with control group, while no changes were found in wild type mice (C57Bl/6). Atherosclerotic plaque formation in aorta was also reduced approximately 35 % in treated ApoE KO mice. It was concluded that the leaves of Moringa oleifera have hypocholesterolemic and anti-atherosclerotic activity in B6.129P2-Apoetm1Unc/JKO mice and that there is valid pharmacological basis for employing them for this purpose in humans.

**Key words:** atherosclerosis, apoie mice, hypocholesterolemic, moringa oleifera
Introduction: Chicken essence is regarded as a health food in Chinese communities and South East Asia. In western countries, chicken soup has also been traditionally used for recovery from cold. Present study investigated the anti-inflammatory effects of four different Korean native chickens (YeonsanOgye, Hyunin Black, Hwangbong, and HoengseongYakdak). White Leghorn was used as a control.

Methods: Breast meats were grounded and extracted with 70% ethanol at 60 °C for 6 hours. The concentration of released histamine was determined in compound 48/80 stimulated HMC-1 cell supernatants by spectrofluorometric procedure. Quantitative real-time polymerase chain reaction (RT-PCR) was used to quantify the expression of mRNAs for IL-4, IL-10, IFN-γ, and TNF-α in LPS-stimulated RAW 264.7 cells.

Results: Histamine release was not affected by all strains of breast or leg meat extracts. However, breast meat extracts of four strains of Korean native chicken significantly reduced the gene expressions of IL-4 and IFN-γ while White Leghorn fail to affect these parameters. IL-10 mRNA expressions were decreased by Hwangbong and HoengseongYakdak breast meat extracts, and TNF-α gene expressions were not changed by chicken meat extracts. Conclusion: Present study suggests that breast meat of Korean native chicken manifest its anti-inflammatory effect, at least in part, by regulating the pro-inflammatory cytokine gene expressions.

Key words: korean native chicken, raw 264.7 cells, pro-inflammatory cytokine, gene expression, il-4, il-10
SPN-P-066  Evaluation of the use of fibre dietary in ice cream

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It was evaluated the use of microcrystalline cellulose in the milk ice cream formulation by means of the global evaluation of the product and its sensorial quality. For this purpose there were rehearsed different proportions of the dietary fibre and in parallel was elaborated the control ice cream of milk. It was evaluated the viscosity of the mixtures and the melting, yield and sensorial quality of the ice creams. To the selected variant was evaluated the composition and general characteristics. With the level of 4.5 % of microcrystalline cellulose was achieved a product of very good sensorial quality and satisfactory melting and nutritional characteristics.

Key words: microcrystalline cellulose, ice cream
SPN-P-067 Development of fermented milks from buffalo’s semiskimmilk and culture of lactobacillus acidophilus

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Pitching rate influence of industrial culture of Lactobacillus acidophilus on incubation time, acidity and pH of fermented milk from buffalo’s semi skim milk and 8 % of sugar, were studied. The best formula, with inoculation rate and emulsion, was used for producing batches of fermented milk at pilot scale to conduct physicochemical and microbiological analysis, and a sensory acceptance test. Shelf life was determined trough the graphic method for incomplete failuer data using Weibull’s distribution. It was concluded that the product showed good phisico - chemical, sensory and microbiologic characteristics. Shelf life at 4ºC was 15 days during witch it maintains a cellular viability of was 10^8 UFC/g. In the mass acceptance test this fermented milk rated within the "I like very much" and "I like extremaly intervals".

Key words: fermented milk, buffalo's semi skim milk, lactobacillus acidophilus
The present work had as objective to obtain a frozen and functional dairy dessert, of appropriate nutritional value, that benefits the consumers and especially to the populations clinically needy as the diabetics. In this ice cream is present the soy because of its low glucémic index, their composition with a high level of dietary fiber, low-level of fat and carbohydrates where the sugar is not present. There were carefully selected the raw materials to use, keeping in mind the intention a developing a low Calorie ice cream. Different formulations were evaluated where sorbitol was present, in order to increase the total solids, viscosity and sweetness, together fruit pulps that imparted freshness and good flavour. It is concluded that is it possible the obtaining an ice cream of satisfactory sensorial and nutritional characteristic, with low contain of fat and a caloric value 35% lower than the traditional ice cream, for feeding of diabetics and other needful clinically sectors. In the massive acceptance evaluation of the product, 80% of the consumers evaluated the ice cream as I like it very much.

Key words: diabetics, soy milk, sorbitol, hipocaloric sweeters, ice cream
SPN-P-069  Use of cheese whey in ice cream

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It was evaluated the use of whey from cheddar cheese as substitute of non fatty solids of milk in ice cream formulation, by means of the global evaluation of the product and its sensorial quality. Different substitution levels (15, 25 and 35%) and ice cream control were elaborated at laboratory scale. The viscosity of de mixtures, melting, yield and sensorial quality of the ice cream were evaluated. To the selected variant, the composition and general characteristics were determined at pilot plant scale. With the lever 25% of substitution of non fatty solids of milk for solids of whey, a product of very good sensorial quality, excellent melting, yield, and satisfactory nutritional characteristics was achieved.

Key words: cheese whey, ice cream
SPN-P-070  Potentialities of leaves from Clusia minor L. species

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Los estudios fitoquímicos realizados al género Clusia (Guttiferae) han demostrado que es una fuente rica de metabolitos secundarios. En este estudio nos proponemos determinar la capacidad secuestradora del radical DPPH y antiinflamatoria de cuatro extractos obtenidos a partir de hojas de la especie endémica Clusia minor L. Los extractos se prepararon por maceración en orden creciente de polaridad (hexano (CMH-A), acetato de etilo (CMH-B) y metanol (CMH-C)) y por extracción directa con etanol (CMH-E). Los estudios por cromatografía en capa delgada revelaron las diferencias cualitativas de estos extractos. El tamizaje fitoquímico corroboró estas diferencias cualitativas y se detectaron principalmente compuestos como: flavonoides, triterpenos, compuestos fenólicos. La actividad secuestradora del radical, mediante el ensayo de DPPH, mostró que las fracciones CMH-B y CMH-E presentaron un efecto significativo, mientras que la actividad antiinflamatoria de los extractos en el modelo de edema en la oreja de ratón inducido por aceite de crotón mostraron un alto por ciento de actividad en todos los extractos con excepción del extracto polar CMH-C.

Key words: género clusia, actividad antioxidante, actividad antiinflamatoria, tamizaje fitoquímico,
Almonds Intake as a tidbit snack, or as an ingredient of numerous kind of gourmet dishes either in stews or in desserts is a common practice by population all over the world, however, nutritional value is not taken into consideration fatty acids con...
Enzymatic hydrolysis of whole bovine plasma protein: optimization and enzyme activity of peptide fractions obtained by ultrafiltration

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In this study, the reaction of enzymatic hydrolysis of proteins from whole bovine plasma was optimized in a batch reactor, using the methodology of response surface (MSR), the antioxidant activity of peptide fractions were analyzed using analytical methods such as ABTS and FRAP and system ultrafiltration fractionation of antioxidant peptides interest was designed. Alcalase 2.4L was used for the hydrolysis, where three experimental factors: pH, temperature and enzyme-substrate (E /S) ratio, were analyzed in order to maximize the degree of hydrolysis (DH) reaction for two hours reaction. The GH was measured using the method of valuation of the proton (pH-stat). The effect of GH on experimental factors was adjusted to a quadratic equation which has coefficient of determination of 0.931. Optimal reaction conditions for the enzymatic hydrolysis of bovine plasma were: Temperature 60.4 °C, pH 9 and E / S 10% (w / w). The predicted value for GH was 23%. An analysis of interaction of experimental factors evidences the dual effect of temperature and pH, on the other hand the direct relationship of the E / S in the assessed levels was noted. The antioxidant activity of bovine plasma enzymatic hydrolysates with Alcalase 2.4 L increases significantly with the GH. Through an ultrafiltration system is possible to concentrate peptide fractions of molecular <8kDa weight, presenting significant antioxidant activity relative to the native protein and the complete hydrolyzate.

Key words: Enzymatic hydrolysis, bovine plasma, protein, optimization, enzyme activity, peptide, ultrafiltration
Probiotic fermented milk with prebiotic syrup

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Among dairy products with functional properties, the symbiotic fermented milks are commercially available worldwide. Numerous studies refer that the incorporation of probiotic bacteria and prebiotic product in the fermented milk are beneficial for health. Syrup prebiotic was used at 0.3 % and Bioyogur starter (Lactobacillus acidophilus + Streptococcus thermophilus) was inoculated at 2 % v/v. Lactic acid formation and pH values were determined for samples collected at the recently inoculated milk and every 0.5 h to the coagulation. The sugar contents at 0 y 3 h and cellular viability of L. acidophilus at 24 h were determined. The global quality was evaluated. The shelf life parked in polyethylene plastics containers and stored at 4 - 6 oC was determined. The acidification capacity and growth of starters were not affected of the used prebiotic syrup. The syrup for growth meanwhile the fructooligosaccharides fraction was intact. The products had good sensorial, physics-chemistry and microbiological quality. The viability of L. acidophilus reached counts in order of 10^10 ucf/mL. The shelf life was 21 days.

Key words: probiotic, prebiotic, fermented milk, fructooligosaccharides, symbiotic
Mangiferin an important bioactive constituent of mango with numerous pharmacological properties. The solubility behavior is important in order to obtain complete information about physicochemical data. Otherwise, temperature-solubility dependence allows perform the respective thermodynamic analysis. In the present work, we have measurement the solubility of mangiferin in water and six organic compounds commonly used in the pharmaceutical industry in the range of 5 to 60 °C of temperature. Materials and methods: The mass of mangiferin was chosen to be in excess in the solvents. This solution was put in a glass vessel inside an incubator under constant agitation for about 3 h at work temperature. The solution was centrifuged and the samples were filtered through a 0.45 µm membrane. Concentrations were determined by measuring absorbance in a UV spectrophotometer at 254 nm. Results and discussion: The solubility data were observed increased in the solubility of mangiferin in water and six organic compounds with an increased in the temperature in each solvent. Mangiferin is sparingly soluble in ethanol, slightly soluble in methanol and water and practically insoluble in diethyl ether, acetone and n hexane. The solubility were correlated according to the Van’t Hoff equation, the standards heat of solution was obtained from the slope of ln Cs vs 1/T plot. Conclusions: Solubility of mangiferin was determined experimentally in six solvents at different temperatures. For all cases was observed increased in the solubility of mangiferin respect to increase of temperature. The experimental solubility data were correlated with Van’t Hoff equation and the standards heats of solution for the different solvents were calculated.

**Key words:** mangiferin solubility, solvents, pharmaceutical industry
SPN-P-076 Assessment of genotoxic and cytotoxic effects of Phyllanthus plants aqueous extracts in Caulobacter crescentus

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Plants of the genus Phyllanthus have been widely used by traditional medicine all over the world. In Cuba, there is a plentiful variety and endemism of them, and various species have proven antiviral, antioxidant and antimutagenic properties against chemical and physical mutagens. In order to extend the search for natural compounds presenting genoprotective properties to other species of this genus grown in our country, previous studies about their possible genotoxic and cytotoxic properties must be done. In the present work, the aqueous extracts of four Phyllanthus species were assessed: P. williamioides, P. chamaecristoides, P. microdictyus, and P. epiphyllanthus, at concentrations ranging from 0.1 to 2 mg/ml and using Caulobacter crescentus cells as the in vitro experimental model. Genotoxicity was assessed through evaluating primary DNA damage by means of the SOS Colorimetric assay. Cytotoxicity was measured through colony formation assay. As a result, the genotoxic and cytotoxic effects were only statistically significant by way of a Dunnett Test (p<0.05), for the highest concentration tested and only in case of the P. williamioides plant extract. In all cases the LD50 values were higher than the maximum concentration assayed and P. chamaecristoides and P. microdictyus extracts showed a survival rate over 90%. These outcomes are in correspondence with preceding studies about the genotoxicity and cytotoxicity of several Phyllanthus species, with different experimental models, in vitro and in vivo, and they validate future researches about the possible antimutagenic properties of these plants aqueous extracts.

Key words: phyllanthus, caulobacter crescentus, genotoxicity, cytotoxicity, plant aqueous extract