

of this preparation exhibited the most potent inhibitory activity, with an  $IC_{50}$  value of 1.613  $\mu\text{g/ml}$ , followed by the ethanolic extract of *Ligusticum sinense* (Kot hua bua) ( $IC_{50}$ =3.769  $\mu\text{g/ml}$ ) and *Nigella sativa* (Thian dam) ( $IC_{50}$ =4.085  $\mu\text{g/ml}$ ). The water and ethanol-water extracts of all plants were apparently inactive ( $IC_{50} > 100 \mu\text{g/ml}$ ). These results can support using Prasaproyhai in Thai traditional medicine for antipyretic caused by inflammation. **References:** [1] Foundation of resuscitate and encourage Thai Traditional Medicine (2005) Thai Pharmaceutical Book Pikanate Printing Center Cooperation. [2] Tewtrakul, S. and Itharat, A. (2007). *J. Ethnopharmacol.* 109:412 – 416.

PJ56

#### Ethnopharmacological study of two plants of Northern Madagascar: bronchodilator activity of *Tetracera madagascariensis* and antispasmodic activity of *Mascarenhasia arborescens*

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Ethnobotanical investigations were conducted with population and traditional healers in several villages in the North of Madagascar. From this field research, two plants have been selected to justify their traditional use. *Tetracera madagascariensis* Willd. ex Schltld. (Dilleniaceae), a species endemic to Madagascar, is traditionally used to treat respiratory disorders. The bronchial asthma is a widespread disease in Madagascar. Bioassay-guided fractionation using isolated guinea pig trachea pre-contracted with histamine at  $2.10^{-5}\text{M}$  led to the identification of methylene chloride extract as the main active fraction. This extract induced a concentration-dependant relaxation with a median effective concentration ( $EC_{50}$ ) of  $53 \pm 0.5 \mu\text{g/ml}$  ( $n=6$ ). Subfractions are in analysis process. *Mascarenhasia arborescens* A. DC. (Apocynaceae), a tree growing in the East of Africa and Madagascar, is widely used in Northern of Madagascar to treat intestinal disorders and diarrhoea. It is on account of these data that we investigated this species for antispasmodic activity. Bioassay-guided fractionation using isolated guinea pig ileum pre-contracted with histamine at  $3.10^{-6}\text{M}$  to monitor the activity led to the isolation of davidigenin (dihydrochalcone) as the main active constituent from methylene chloride fraction. Effectively, it induced a concentration-dependant relaxation of the histamine pre-contracted guinea pig ileum with an  $EC_{50}$  of  $11.1 \pm 0.7 \mu\text{g/ml}$  ( $n=4$ ). This data is in accordance with the literature underlined an antispasmodic effect of davidigenin on mouse jejunum [1]. **Acknowledgements:** A. Rakotozafy (botanist, IMRA), J.P. Nicolas (association "Jardins du Monde"), population and healers surveyed. **Reference:** [1] Sato, Y. et al. (2007) *Biol. Pharm. Bull.* 30:145 – 149.

PJ57

#### Isoflavonoids in tropical and subtropical neglected leguminous species

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Isoflavonoids, bioactive compounds belonging to the phytoestrogens, are known to prevent some kinds of hormone-related cancers and may participate as alternatives to the conventional hormone replacement therapy [1]. Although they have already been identified in about 50 different plant families [2], the quantities were usually in trace amounts and therefore legumes remain the most important source of isoflavonoids. Since the tropical and subtropical regions offer wide range of neglected leguminous species, we decided to analyze 27 of them for

the presence of isoflavonoids. Aqueous/methanolic extracts obtained from dried seeds were pretreated on an immunoaffinity column (IAC) [3] and subsequently analyzed by reverse phase HPLC/UV-DAD. Immunosorbents for IAC are characterized by high molecular selectivity so that single group of structurally related compounds can be targeted. UV spectra and retention times were compared with set of standards. As a result, we have identified certain commonly known (e.g. genistein) and high levels of uncommon isoflavonoids in several samples. Genistein was present in *Spartium junceum* L., *Pachyrhizus tuberosus* Spreng. and *Trigonella foenum-graecum* L., in concentrations 13, 7 and 0.8  $\mu\text{g/g}$  of dry seeds, respectively. Interestingly, the extract of *P. tuberosus* contained significantly higher amounts of total isoflavonoids than soybean. Though presence of isoflavonoids in these species has never been published before, the fact itself is basically not striking, but the amounts are hint for their further investigation and possible use in dietary products **Acknowledgements:** This research was supported by project GACR 525/09/0994 and MSM 6046070901. **References:** [1] Adlercreutz, H. et al. (2004) *BioFactors* 22:229 – 236. [2] Mackova, Z. et al. (2006) *Phytochemistry* 67:849 – 855. [3] Delaunay, N. et al. (2000). *J. Chromatogr. B* 745:15 – 37.

PJ58

#### Antibacterial effects of leaves of *Vaccinium vitis-idaea* L

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This study was realized to investigate the antibacterial activity of extracts from leaves of *Vaccinium vitis-idaea* L. (Ericaceae) against eleven strains of *Escherichia coli*. Leaves of *Vaccinium vitis-idaea* L. collected on Klekovača Mountain (RS, Bosnia and Herzegovina, W. Balkans) were extracted by different solvents (water, ethanol and ethyl acetate). Cultures of bacteria were clinical isolates and standard strain of *E. coli* ATCC 25922. Minimal Inhibitory Concentrations (MICs) and Minimal Bactericidal Concentrations (MBCs) of extracts and antibiotic amoxicillin were determined by tube dilution method. The results revealed that water extract exhibited the highest activity against all strains of *E. coli* (MICs were 5 mg/ml). MICs of ethyl acetate extract were 20 mg/ml for all bacteria strains tested. Ethanol extract exhibited antibacterial activity with MIC values between 20 and 40 mg/ml. In conclusion, water extract from leaves of *Vaccinium vitis-idaea* L. showed high antibacterial activity against *Escherichia coli* with MBCs 5 mg/ml for nine strains and 10 mg/ml for two strains.

PJ59

#### In vitro antibacterial activity of cloves (*Syzygium aromaticum*) against MRSA

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The process of microbial resistance against antibiotics makes it essential to seek for novel drugs. There have been many studies over the last years, in which a lot of plant species have been checked for their antimicrobial activities. Cloves (*Syzygium aromaticum*), which is a commonly used spice worldwide, have antibacterial, antifungal, antiviral, antioxidant, antimutagenic, anaesthetic, insecticidal, anti-inflammatory, antithrombotic, antiparasitic and antiulcerogenic activities [1]. Staphylococci are a well known cause of both hospital and community acquired infections. Isolates that have acquired methicillin resistance pose serious problems for treatment and eradication. After the introduction of the drug, Methicillin Resistant *Staphylococcus aureus* (MRSA) strains were reported in the early 1960 s. Epidemics have occurred around the world and the clones have diversified since then. MRSA is still on the rise. Nowadays, the imminent threat of reduced susceptibility to vancomycin have emerged [2]. The purpose of this study was to evaluate antibacterial activity of cloves against MRSA. All 100 (one hundred) clinical MRSA isolates were screened by using agar dilution method. Both aqueous and ethanol extracts of cloves were obtained. All of the strains were found to