A147 Fractions of lowbush blueberry (*Vaccinium angustifolium*) polyphenols provide protection to cultured neurons against simulated stroke *in vitro*

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Recent feeding studies, conducted by our group, have shown that consumption of aqueous extract of lowbush blueberries (*Vaccinium angustifolium* Ait.) led to reduced neuronal damage after surgically-induced stroke in rats. The possible identity of the compounds responsible for this *in vivo* protection were investigated in this study using an *in vitro* assay which utilized neurons from the cerebellum of neonatal rats. Following the addition of the extract, cultured neurons were challenged with a six hour incubation of oxygen-glucose deprived medium (OGD), to simulate a stroke, or 100 µM hydrogen peroxide, to induce oxidative stress. Neuronal damage was determined by measuring extracellular lactate dehydrogenase (LDH) or cytosolic caspase-3 activity, indicators of necrosis and apoptosis respectively. A semi-purified extract of wild blueberries (30-1000 ng/ml) reduced both OGD- and hydrogen peroxide- induced necrotic cell death, which was apparent by a 36-38% decline in extracellular LDH. Protection against apoptotic neuronal death was also apparent by a 62-93% lower induction of caspase-3 activity. Using Sephadex LH-20 column chromatography, two fractions were identified that protected neurons against damage induced by OGD and hydrogen peroxide. The highly colored fraction, determined to largely contain anthocyanins, reduced LDH activity by 27-29% and caspase-3 activity by 61-69%. Neuroprotection against only OGD was observed for the second fraction that reduced LDH activity by 41% and caspase-3 activity by 56%. The second fraction was determined to contain predominantly proanthocyanidins. Individually these enriched fractions of anthocyanins and proanthocyanidins provided neuroprotection but they were less effective than the semi-purified blueberry extract. These results indicate that a semi-purified aqueous extract of lowbush blueberries can protect neurons against two distinct forms of cellular damage induced by a simulated stroke *in vitro*.

A148 Effects of the water extract of *Cyperus articulatus* and some anticonvulsant or antiepileptic compounds on the rat cortical wedge preparation

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Cyperus articulatus is a plant commonly used in traditional medicine in Africa and Latin America to treat many diseases (1,2). Extracts of rhizomes of *C. articulatus* have been shown to possess antiepileptic properties (3). In these studies, the water extract from rhizomes of *C. articulatus* concentration-dependently reduced spontaneous epileptiform discharges and NMDA-induced depolarisations in the rat cortical wedge preparation perfused with Mg^{2+} -free artificial cerebrospinal fluid (-Mg^{2+} aCSF). The epileptiform events were completely blocked at a concentration of 2.2 mg/ml. Concentration of 3 mg/ml inhibited 83.7% ± 5.8 of NMDA-induced depolarisations. At the same concentrations, AMPA-induced depolarisations were not affected. Phenobarbital, pentobarbital and phenythoin inhibited both AMPA and NMDA-induced depolarisations with a preference for AMPA rather than NMDA. For example, concentration of 2000 µM of phenobarbital inhibited 90% and 50% of AMPA- and NMDA-induced depolarisations respectively. Phenobarbital, pentobarbital and phenythoin completely blocked spontaneous epileptiform discharges at concentrations of 500, 200 and 200 µM respectively. A competitive NMDA antagonist DCPPene inhibited NMDA-induced depolarisations and epileptiform discharges and had no effect on AMPA-induced depolarisations. DCPPene completely blocked NMDA-induced depolarisations at a concentration of 1 µM. The two antiepileptic compound valproate and ethosuximide did not affect either epileptiform discharges or AMPA and NMDA-induced depolarisations. In conclusion, the mechanism actions of *C. articulatus* seem to be different from the one of phenobarbital, pentobarbital, phenythoin, valproate, and ethosuximide. With the same pattern of inhibiting NMDA-induced depolarisations, *C. articulatus* extract could contain components acting as NMDA antagonists.

References: 1. Burkill, H.M. (1985) The Useful Plants of Tropical Africa. Royal Botanic Gardens Kew, London. **2.** Schultes, R. E. and Raffauf, R.F. (1990) The Healing Forest: Medicinal plants of the Northwest Amazonia. Dioscorides Press, Portland. **3.** Ngo Bum et al. (2001), J. Ethnopharmacol. 76: 145-150.