ENDOGENOUS BENZODIAZEPINES AND INTESTINAL BACTERIAL FLORA IN NORMAL AND PATHOLOGICAL CONDITIONS

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Evidence has been provided that benzodiazepine (BZD)-like substances are increased in the brains of animals with experimental HE, as well as in serum and in cerebrospinal fluid of patients with HE due to liver cirrhosis and fulminant hepatic failure. Thus, BDZ-like compounds can be regarded as precipitating factors of hepatic encephalopathy requiring the presence of a cerebral supersensitivity to these compounds. The endogenous biosynthetic pathway for the production of natural benzodiazepines has not yet been found while their presence in food has been already described. Furthermore it has been demonstrated by *in vitro* experiments that microorganims can synthesise molecules with BDZ structures and by *in vivo* studies that intestinal bacterial flora contributed by 40% to level of total natural benzodiazepines in serum of cirrhotic patients. The aim of the present study was to: 1) confirm the partial contribution of intestine to the level of circulating natural BDZ in commercial BDZ free patients, 2) to study the influence of the chronic administration of a symbiotic compound (Zyrfos-Alfa Wassermann) on the serum concentration of BDZ-like compounds in rats.
The serum samples obtained by acid precipitation was chromatographed at 0.8 ml/min on a LiChrospher 100 RP-18 column (250x4.0 mm; 5 μm) equilibrated with 80% water/0.1% TFA and 20% acetonitrile. The sample was analysed using a water/0.1% TFA and acetonitrile gradient at 0.5% from 20 to 58% acetonitrile. 75 fraction were collected, lyophilised and tested for their ability to inhibit [^{3}H]RO 15-1788 specific binding to central BDZ binding site.
The concentration of BDZ-like substances in portal vein of cirrhotic patients (n=10) reach levels of 1024.24 ± 22.78 ng diazepam equivalent (DE)/mL in comparison with the circulating value of 162.44 ± 10.89 ng DE/mL. These data further demonstrate that there is a intestinal source of the above mentioned compounds. As far as regard the influence of the symbiotic which is composed by probiotic, prebiotic and B vitamins, herein we report that after 14 days of chronic administration the level of BDZ-like compounds decreased from level of 173.5 ± 43.76 ng DE/mL present in controls to level under the detection limit (0.5 ng DE/mL).
The reported experiments and data seem to further demonstrate that the intestinal bacterial flora contribute to the amount of circulating natural BDZs both in normal and pathological conditions. Moreover it might be possible to modulate the level of these compounds by the administration of dietary supplements.