Use of natural agent from Giant knoweeds for supplementary treatment of peri-implantitis – in vitro study

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**Background:** Prevention of peri-implantitis is essential for the success of implant rehabilitation. Anaerobic gram-negative organisms are most commonly found in peri-implantitis affected sites. Giant knotweeds originating from East Asia – F. japonica, F. sachalinensis and hybrid F. bohemica are invasive plants in Europe and North America, and a traditional East Asian drug used in Korean folk medicine to improve oral hygiene. Our earlier studies have shown that these plants contain high levels of polyphenols.

**Aim/Hypothesis:** The aim was to obtain polyphenol enriched natural extracts and fractions and to evaluate their antibacterial activity against gram-negative anaerobic bacteria as well as inhibition activity of one of the most important proteases in development of peri-implantitis - elastase.

**Material and Methods:** Rhizomes of studied plants were extracted using ultrasound-assisted extraction and fractionated between solvents of increasing polarity. All extracts and fractions were screened for total polyphenols and tannins as well inhibition of elastase. Extracts were phytochemically characterized by methods of liquid chromatography coupled to mass spectrometry using the electrospray ionization interface - HPLC-DAD-ESI-MS - assay. Than their antibacterial activity was measured, minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC) were evaluated. Peri-implantitis is caused by gram-negative and anaerobic bacteria and as a model for gram-negative bacteria Escherichia coli was used. Human gingival fibroblasts were obtained from healthy volunteers and cytotoxicity of all the extracts was evaluated by MTT assay. The MTT assay was used to estimate the mitochondrial metabolic function through the spectroscopic measurement of mitochondrial dehydrogenase.

**Results:** Butanol and water fractions have shown the highest inhibition of elastase, where fractions in concentration of 80 μg/ml, inhibited up to 50% of elastase. HPLC-DAD-ESI-MS analysis has shown that most active fractions contain highly polymerized proanthocyanidins (up to procyanidin heptamer). Among evaluated plants the strongest antibacterial activity against anaerobic gram-negative organisms had F.japonica, its extracts revealed minimal bactericidal concentration (MBC) 6 mg/ml. The MTT assay revealed that F.japonica has low cytotoxicity.

**Conclusions and Clinical Implications:** Since the predictable treatment protocol of peri-implantitis is not yet known, new substances that express activity against pathogenic bacteria should be evaluated for their use in peri-implantitis treatment. Extracts obtained from Giant knoweeds plants, mainly from F.japonica expresses high activity against gram-negative anaerobic bacteria. With the limitation of this study the results suggests that these plants should be considered as a potential natural substance peri-implant biofilm control.