



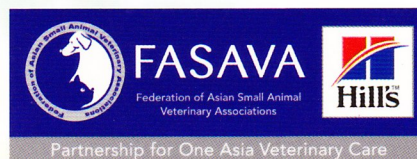
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CATEGORY: TRADITIONAL VETERINARY MEDICINE

TOPIC:

Development of novel earthworm powder and investigation of its therapeutic effects in animals

S. Akazawa¹, Y. Machida¹, H. Okawa², T. Watanabe³, and S. Wakimoto³

1. Department of Materials Engineering, National Institute of Technology, Nagaoka College, Niigata, Japan; 2. Scarecrow Inc., Tokyo, Japan; 3. Waki Pharmaceutical Co., Ltd., Nara, Japan.

Background and Objectives:

Earthworms are well-known soil decomposers. Because they are polyphagous animals and their cast contains a large amount of nutrients, application of earthworms in composting has been studied extensively. Earthworms have been also studied for their therapeutic effects against human diseases. In particular, dried earthworm powder, called "Earth Dragon" or "Jiryu" (in Japanese), has been used as an antipyretic and diuretic treatment since ancient times. Furthermore, earthworms have fibrinolytic enzymes, and dried earthworm powder has been found to have health benefits for humans. However, the powder manufactured by conventional methods does not have high fibrinolytic activity because of the heat treatment involved in the final step. Thus, we have developed a novel production system that involves pressure treatment. Furthermore, we characterized the powder and investigated its therapeutic effects after oral administration in animals.

Materials and Methods: The earthworms, *Eisenia fetida* Waki, and its lyophilized powder were kindly provided by Waki Pharmaceutical Co., Ltd., Nara, Japan. The conditions for the high-pressure treatments were as follows: 100 MPa at 60 °C for 16 h by using SHP100-50A (Shinada Co., LTD., Niigata, Japan). Lumbrokinase (LR) activity (same as fibrinolytic activity) was measured at 37 °C by measuring the formation of *p*-nitroaniline (*p*NA) at 405 nm with a Corona Absorbance Microplate Reader MTP-310Lab (Corona Electric Co., Ltd., Ibaraki, Japan). S-2251 (Sekisui Medical Co., Ltd., Tokyo, Japan) and S-2288 (Sekisui Medical) were used as substrates for plasmin and tissue-plasminogen activator (t-PA), respectively. A fibrin plate assay was performed as follows: Hundred milligrams of fibrinogen was mixed with 10 mL of FP buffer (50 mM Tris-HCl, 93 mM NaCl, 1.66 mM CaCl₂·2H₂O, 0.96 mM MgCl₂·6H₂O). The mixed solution was centrifuged and the supernatant was transferred into a plastic plate. Then, 50 µL of thrombin (20 NIH units/ml) was added into the plate. Therapeutic effects in

animals were investigated by determining reactive oxygen metabolites-derived compounds (d-ROMs) and biological antioxidant potential (BAP) test.