

Conference report: Second annual young researcher forum of the Japan Society of Medical and Pharmaceutical Sciences for Traditional Medicine

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Abstract

Five young researchers presented their studies in the field of traditional Japanese herbal medicine (Wakan-yaku) at the second annual young researcher forum entitled “Growing Research Buds by Young Investigators,” held during the annual meeting of the Japan Society of Medical and Pharmaceutical Sciences for Traditional Medicine 2021. Mr. Yoneda presented exploratory research on new pharmaceutical seed compounds from garlic (*Allium sativum*). Dr. Yang investigated whether diosgenin promoted proper axonal regeneration in the brain using a mouse model of Alzheimer’s disease. Her study clarified the molecular mechanisms of the diosgenin-driven accurate pathfinding of injured axons. Mr. Nishihara presented his research on promoting the effective utilization of *Phellodendron* trees in Nara Prefecture. His research team evaluated the possibility of short-term cultivation, screened for useful components in the leaves. Dr. Ogawa used the Japanese Adverse Drug Event Report (JADER) database, which contains reports of side effects associated with drug treatment, to demonstrate the effectiveness of *hokizai* in reducing side effects, particularly those of anticancer drugs. Dr. Kitamura reported that *Cnidium monnieri* (L.) Cusson fruit extract and the isolated active ingredients, osthol and imperatorin, strongly inhibited the activity of furin-like enzymes. Further screening indicated that root extracts of *Paeonia lactiflora* Pall. and *Scutellaria baicalensis* Georgi suppressed cell syncytial formation by the S protein. The event was held online, and we had 86 participants. We can improve this forum in terms of format, frequency, and publicity activities; however, we hope to hold this forum in 2022 and in the future.

KEYWORDS

Kampo medicine, traditional Japanese medicine, Wakan-yaku, young researchers

INTRODUCTION

The Japan Society of Medical and Pharmaceutical Sciences for Traditional Medicine was established in 1984 and has held an annual scientific meeting for more than three decades. Many young researchers are active at

the annual meeting, not only to present their research and study the latest topics, but also to network with researchers from other institutions. An annual symposium has also been organized by young researchers for several years. In addition to the annual meetings, since 2020 a virtual forum has been held by young

researchers in the society. At the virtual forum held on September 3, 2021, 86 participants, including students, attended. A plenary lecture was delivered by Prof. Kaoru Inokuchi from Toyama University, which focused on motivating young researchers, and a roundtable discussion was held after the meeting for interaction among researchers. Five young researchers presented their studies in the field of traditional Japanese herbal medicine (Wakan-yaku). Below is a summary of these presentations.

SYNTHESIS OF THIOPYRANS USING THIOACROLEIN, A GARLIC-DERIVED UNSTABLE COMPOUND, AND EVALUATION OF THEIR ACTIVITIES

Mr. Yoneda presented exploratory research on new pharmaceutical seed compounds from garlic (*Allium sativum* L.), which is used in Japanese and Chinese herbal medicine. He was the only student presenter at this forum, but was enthusiastic about his current research. Lively discussions took place, and discussions between the students were particularly impressive. It was a presentation suitable for virtual forums by young researchers in the society. Based on the presentation made, the forum conferred an award as an incentive and encouragement toward further work. Upon receiving the award, Mr. Yoneda commented, "I am honored to receive an extraordinary award. I am motivated to work harder on my research."

MOLECULAR MECHANISMS OF DIOSGENIN-INDUCED AXONAL REGENERATION AND MEMORY RECOVERY IN A MOUSE MODEL OF ALZHEIMER'S DISEASE

Alzheimer's disease (AD) is a neurodegenerative disease characterized by amyloid β ($A\beta$) deposition and subsequent neural network disruption in the brain. Dr. Yang and her group previously found that diosgenin, a constituent of *Dioscorea* Rhizome, restored $A\beta$ -induced neurite (axon and dendrite) atrophy in cultured neurons and improved memory function in a mouse model of AD, 5XFAD [1,2]. Importantly, intake of diosgenin-rich yam extract enhances cognitive function in healthy humans [3]. Although there are no studies having demonstrated that the degenerated axons regenerate toward their intrinsic target regions in adult brains, she investigated whether diosgenin promotes proper axonal regeneration in the brains of 5XFAD mice. Her study clarified the molecular mechanisms of the diosgenin-driven accurate pathfinding of injured axons.

SUSTAINABILITY-BASED LOGGING SURVEY AND STUDY OF EFFECTIVE USE OF PHELLODENDRON BARK

The herbal medicine *Phellodendron* bark consists of the bark of the Amur cork tree, *Phellodendron amurense* Rupr. or *Phellodendron chinense* C.K.Schneid. (Rutaceae), excluding the periderm. For a long time, it has been used solely in folk medicine as an internally ingested stomachic medicine (e.g., daranisuke), and used as a topical agent to alleviate bruises, sprains, and periodontal diseases. Furthermore, in Kampo medicine, it is a constituent of herbal formulas such as oren-gedokuto, shichimotsukokato, and chuohko.

More than 20 years are required to cultivate the *Phellodendron* tree, which is the raw material for the herbal medicine *Phellodendron* bark. Therefore, challenges such as cultivated land abandoned owing to the long cultivation period, shortage of successors, and aging of managers will create difficulties in securing *Phellodendron* bark in the future. Additionally, these limitations affect not only *Phellodendron* bark, but also other herbal medicines.

Therefore, six public testing and research institutes in Nara Prefecture collaborated to set up an interdisciplinary research headquarters. As one of their research themes, Mr. Nishihara and his team are researching the promotion and effective utilization of prefectural *Phellodendron* trees.

The team investigated the growth and logging of trees in Nara Prefecture. They also measured the content of berberine-type alkaloids in the bark according to a previous report [4] to determine whether it meets the standards described in the Japanese Pharmacopoeia for *Phellodendron* bark, which are typically difficult to meet. To evaluate the possibility of short-term cultivation, they measured the alkaloids in the bark not only from trees cultivated for more than 20 years, but also from young trees cultivated for three and six years.

In addition, during logging of the trees, only the bark is obtained as a crude drug and the other parts (leaves, fruits, and heartwood) are discarded at the logging site. The team hypothesized that by utilizing these unused parts, further cultivation and maintenance of trees would be possible. Therefore, their focus was first on the leaves, that are obtained in high yields, screening them for useful components, and further analyzing these components for commercialization.

ELUCIDATING THE EFFECTIVENESS OF HOKIZAI BASED ON ANALYSIS USING THE JAPANESE ADVERSE DRUG EVENT REPORT (JADER) DATABASE

Hochuekkito and other qi deficiency treatment formulas (*hokizai*) increase physical strength and alleviate

fatigue and are therefore used to reduce the side effects of anticancer agents. However, it is difficult to explain the effect of qi deficiency treatment scientifically. There have been various reports analyzing data on the adverse events of Kampo medicine [5,6], but not many studies have focused on their effectiveness. Therefore, using the Japanese Adverse Drug Event Report (JADER) database, which contains reports of side effects associated with drug treatment, Dr. Ogawa examined whether *hokizai* is effective in reducing side effects, particularly those of anticancer drugs.

Dr. Ogawa focused on the following *hokizais*, “hochuekkito,” “juzendaihoto,” and “rikkunshito,” which are frequently reported in JADER. Typical adverse events of anticancer drugs, such as nausea, vomiting, diarrhea, malaise, erythropenia, leukopenia, thrombocytopenia, and decreased appetite, were selected as the target adverse events. For analysis, cases of anticancer drugs were extracted and further divided into two groups according to the use or non-use of *hokizai*. To examine the effectiveness of *hokizai*, the reported odds ratio (ROR) was calculated for the use and non-use of *hokizai*. In addition, the time to the onset of adverse events was calculated and the results were compared.

Of the total data used in the analysis, 123,610 adverse events were reported for anticancer drugs and 479 cases were reported for the combination of anticancer drugs and *hokizai*. In the analysis using the ROR, most of the target adverse events showed an ROR of approximately 1, suggesting that the adverse events caused by anticancer drugs were not suppressed by *hokizais*. Next, the median time to the onset of target adverse events was compared between the use and non-use of *hokizais*. In cases without *hokizais*, erythrocytopenia developed at a median of 14 days after the administration of anticancer drugs but was prolonged to 73 days with the use of *hokizais*. The time of onset of adverse events was also extended for other target adverse events. Therefore, these results indicated that although *hokizai* did not completely suppress the onset of adverse effects caused by anticancer drugs, it extended the time to onset.

This study demonstrated the effectiveness of *hokizai* by using adverse event reports. As a future study, the authors would like to investigate whether Kampo medicine can suppress adverse events for drugs other than anticancer drugs. In addition, the contribution of each crude drug to the effectiveness of Kampo medicine will be studied.

SCREENING FOR HERBAL MEDICINAL EXTRACTS THAT CONTROL THE CLEAVAGE OF THE SARS-COV-2 SPIKE PROTEIN

Dr. Kitamura presented “Screening for inhibitory effects of crude drugs on the activity of the severe acute

respiratory syndrome coronavirus-2 spike protein cleavage (SARS-CoV-2 S protein).” SARS-CoV-2, the causative virus of coronavirus disease 2019 (COVID-19), has spread worldwide, and the pandemic shows no signs of abating. The first step in the intracellular entry of SARS-CoV-2 involves processing of S proteins [7]. After processing and activating the S protein, the viral membrane fuses with the host cell. The major difference between SARS-CoV-2 and other coronaviruses is the presence of a proprotein convertase motif at the interface of the S1 and S2 domains of the S protein, which is cleaved by the serine protease furin. Inhibition of S protein processing by furin suppresses SARS-CoV-2 infection [8]. The herbal extracts that inhibited S protein cleavage were investigated; 124 herbal medicinal extracts that inhibited furin-like enzyme activity were screened. *Cnidium monnieri* (L.) Cusson fruit extract was observed to strongly inhibit enzyme activity. Furthermore, the isolated active ingredients, osthol and imperatorin, present in *Cnidium monnieri* fruits, showed the ability to inhibit the activity of furin-like enzymes [9]. Next, through observation of S protein transduction into HEK293 cells, crude drug extracts were screened by evaluating their inhibitory activity on syncytial formation (a phenomenon in which cells fuse and multiple nuclei aggregate). As syncytial formation is suppressed by inhibiting the cleavage of the S protein, it is possible to search for crude drug extracts that are, directly and indirectly, active toward the cleavage of the S protein. The results of this screening indicate that the root extract of *Paeonia lactiflora* Pall. and *Scutellaria baicalensis* Georgi suppressed cell syncytial formation by the S protein. In the future, Dr. Kitamura plans to determine the molecular mechanisms of this process using various approaches, including proteome analysis.

POSTSCRIPT

Although the event was held online, as in the previous year, we had more participants attending. We can improve this forum in terms of format, frequency, and publicity activities, and we hope to continue to hold this forum in the coming years. We are planning to hold the next forum in August 2022, and are hoping for the readers' participation.

CONFLICT OF INTEREST

The authors declare that there are no conflict of interests regarding the research, authorship, and publication of this article.

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