（学位第３号様式）

学 位 論 文 要 旨

氏　名	Natsuko Tsurudome
題　目	Analysis of the triggering mechanism of vascular abnormal contraction and its preventive mechanism by mulberry leaves （血管異常収縮の発症機構と桑葉由来成分による予防機序に関する研究）

Vascular diseases such as cardiovascular and cerebrovascular diseases are directly related to blood vessels and account for fatal diseases around the world. Their causes can be classified into three categories: vascular abnormal contraction, arteriosclerosis, and hemorrhage. Vascular abnormal contraction has afflicted many people for years without any treatment or preventive methods. Sphingosylphosphorylcholine (SPC) is a causative factor, but its mechanism still remains unknown, making it difficult to establish treatment and preventive methods. However, prevention by food has attracted considerable attention in recent years. It has been reported that vegetables and fruits have various physiological functions. Mulberry (Morus australis Poir.) is known to have some physiological effects, but no studies on abnormal contraction have been conducted. Therefore, this study confirmed their preventive effect against SPC-induced vascular abnormal contraction and identified their active compounds and mechanisms.

The preventive effect against SPC-induced abnormal contraction was evaluated using cell imaging of cultured human coronary artery smooth muscle cells (HCASMCs). Mulberry leaves were classified by their ages or processing method, their preventive effect was verified by co-culturing HCASMCs with their extract, and it was found that mulberry leaves over 20-years-old trees were effective. In addition, the active compound was identified as fisetin, which is more abundant in mulberry leaves than in strawberries and apples, and their hydroxyl group at the C-3 was considered the active site through structure-activity relationships.

To reveal the preventive mechanism of fisetin and the pathogenesis of SPC, the mechanism around the plasma membrane, which is upstream of the reaction field in the vicinity of the cell, were the focus of attention. Consequently, through molecular interaction analysis, fisetin was found to act directly on HCASMCs and not inhibit SPC activity. It was also confirmed that nitrobenzoxadiazole-SPC, the fluorescent-labeled SPC, is incorporated into cells via endocytosis and that exocytosis is occurring specifically in abnormally contracted cells. However, such occurrences were also observed in fisetin-treated cells. Therefore, it was concluded that these phenomena are not related to the preventive mechanism of fisetin but to the pathogenesis of SPC-induced vascular abnormal contraction.