氏　　名	Rahul Sharma
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学位論文題目	Octacosanol prevents high fat diet-induced obesity by activating energy expenditure and thermogenesis in brown and beige fats （オクタコサノールは褐色脂肪細胞およびベージュ細胞のエネルギー消費および熱産生を活性化させることにより高脂肪食誘導性の肥満を抑制する）

	（職名）	（学位）	（氏名）
主　　査	筑波大学　教授	薬学博士	熊谷　嘉人
副　　査	筑波大学　助教	博士（医学）	加藤　広介
副　　査	筑波大学　助教	博士（神経科学）	小金澤　禎史
副　　査	筑波大学　教授	博士（医学）	柳沢　裕美
副　　査	筑波大学　教授	医学博士	島野　仁

論文の要旨
Abstract of thesis

Background and purpose: Obesity develops when energy intake exceeds energy expenditure, resulting in the accumulation of white adipose tissue (WAT) in the body. Over the past 10 years, nutritional supplements, or nutraceuticals, have become increasingly popular among the general public. One such supplement, policosanol, has been the subject of numerous studies. Policosanol is a mixture of very-long-chain saturated fatty alcohols purified from natural sources such as rice bran, wheat, sugarcane and beeswax, whose main component is octacosanol, a high molecular weight primary aliphatic alcohol. Octacosanol has a number of indications for its use, many of which are currently being researched. Previous studies have demonstrated that octacosanol lowers blood cholesterol, suppresses platelet aggregation, reduces inflammation, increases athletic performance, protects cells, alleviates stress and restores stress-affected sleep. The β-oxidation of FAs is critical for the function of BAT and beige fat and hence thermogenesis. Recent studies using experimental models and humans suggest that the long chain saturated FAs, mainly stearic acid (C18:0), regulate mitochondrial function. Moreover, the ELOVL family member 6 (Elovl6), a microsomal enzyme responsible for converting C16 FAs into C18 species, has been suggested to regulate BAT

thermogenic capacity. Considering that the longer fatty acids could be preferred substrates for thermogenesis and the abovementioned effects of octacosanol or policosanol on lipid metabolism, their role in the prevention or treatment of obesity and in the thermogenic function of brown and beige adipocytes has not yet been established. In the present study, the author investigated anti-obesity and thermogenic function of octacosanol and policosanol in mice.

Methods: The author used 10 weeks old C57BL/6 male mice in this study. The mice were randomly divided into the following four groups: chow group (fed on standard chow with vehicle (10 mg/ml Acacia-gum water), HFD group (fed with vehicle treatment), the HFD + octacosanol group (60 mg/kg/day; Sigma-Aldrich, Tokyo, Japan), and HFD + policosanol group (60 mg/kg/day). Octacosanol, policosanol, or the same volume of vehicle was administered via oral gavage once every four weeks. At the end of four weeks, all mice were sacrificed in the early light phase in a non-fasting state. Blood samples and tissue [BAT, inguinal WAT (iWAT), epidedrmal WAT (eWAT) and Liver] samples were collected to analyse plasma TG and glucose and to identify specific genes/proteins that regulate brown/beige adipocyte function.

Results and discussion: This study aimed to determine whether purified octacosanol and policosanol have beneficial effects on HFD-induced obesity in mice. The author showed that both octacosanol and policosanol significantly ameliorated HFD-induced fat gain and fatty liver. Furthermore, octacosanol and policosanol activated signalling pathways that regulate thermogenesis and enhance energy expenditure in BAT and iWAT. Octacosanol or policosanol treatment completely suppressed HFD-induced BW gain. Policosanol is a fatty alcohol, which is converted to saturated and monounsaturated FAs by fatty aldehyde dehydrogenase (FALDH, alternatively known as Aldh3a2). In this study, the author also observed higher expression of *Aldh3a2* in BAT and iWAT of chow- or HFD-fed mice treated with octacosanol or policosanol, suggesting the conversion of octacosanol and policosanol into FAs. The findings shown by the author suggest that shortened saturated and unsaturated FAs metabolized from octacosanol and policosanol perform cellular functions important for thermogenesis and energy expenditure in BAT and WAT. In this study, the author showed that the treatment of HFD with octacosanol or policosanol induced Free fatty acid receptor-4 (*Ffar4)* expression in BAT and iWAT of mice and decreased lipid content in BAT and WAT, suggesting that the treatment of HFD with octacosanol or policosanol triggers brown/beige fat activation by upregulating *Ffar4* expression in BAT and WAT of HFD-induced mice. Since adipose tissue in obese individuals is characterized by adipocyte hypertrophy and chronic inflammation, the author also investigated the effects of octacosanol and policosanol on the inflammatory state of WAT. Treatment of HFD with octacosanol or policosanol lowered the expression levels of pro-inflammatory genes including F4/80, *CD68*, *Tnfα* and *Il1b* in eWAT. In conclusion, the author showed that octacosanol and policosanol exert beneficial metabolic effects by activating thermogenic changes in HFD-induced obesity in mice. Although precise molecular mechanisms underlying the involvement of octacosanol and policosanol in the thermogenic activity of BAT and browning of WAT are unknown. The author's data showed hat octacosanol is a potent dietary anti-obesity molecule, which increases the thermogenic activity of BAT and iWAT, thereby increasing energy expenditure and reducing fat mass.

審査の要旨
Abstract of assessment result

【批評 Review】

In this study, the author showed that both octacosanol and policosanol significantly ameliorated HFD-induced fat gain and fatty liver. Furthermore, octacosanol and policosanol activated signalling pathways that regulate thermogenesis and enhance energy expenditure in BAT and iWAT. One of the major advantages is the natural occurrence of this compound, which makes it to be used as a drug or food supplement without any side effects. The author also found that octacosanol and policosanol exert beneficial metabolic effects by activating thermogenic changes in HFD-induced obesity in mice. However, the precise molecular mechanism is still unknown. Studying the effect of octacosanol (policosanol) on human subjects would be required to further reveal the beneficial effect of octacosanol and policosanol on BAT and WAT of obese and diabetic individuals.

【最終試験の結果 Result】

The final examination committee conducted a meeting as a final examination on 7 May, 2019. The applicant provided an overview of dissertation, addressed questions and comments raised during Q&A session. All of the committee members reached a final decision that the applicant has passed the final examination.

【結論 Conclusion】

Therefore, the final examination committee approved that the applicant is qualified to be awarded a Doctor of Philosophy in Human Biology.