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Tocotrienols activate diacylglycerol kinase a via 67 KDa laminin receptor

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ABSTRACT

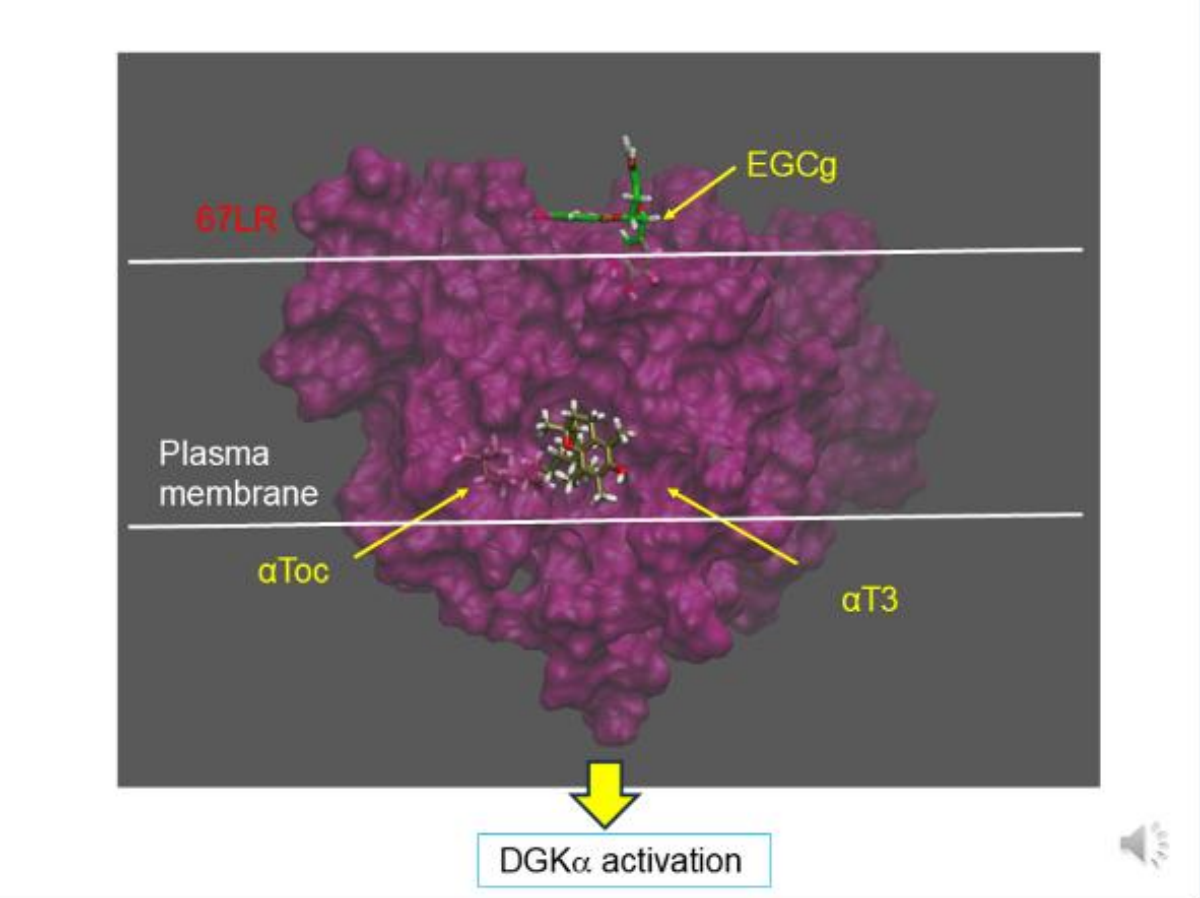
Background: Diabetes is a significant social issue. Controlling diabetic complications such as nephropathy is crucial for the quality of life (QOL) of diabetic patients. The abnormal activation of protein kinase C (PKC) through increased diacylglycerol levels (DG) due to hyperglycemia is a common mechanism that causes diabetic complications. Diacylglycerol kinase (DGK) can attenuate PKC activity by converting DG to phosphatidic acid. Thus far, d-α-tocopherol (αToc) treatment has been shown to prevent early changes of diabetic renal dysfunctions by activating DGKα via the 67KDa laminin receptor.
Vitamin E is classified into tocopherols (Toc) and tocotrienols (T3) with four derivatives: α, β, γα, β, γ, and δ. We have found that all Tocs induced the translocation of DGKα. However, it remains unclear if T3s also induced translocation of DGKα.

Objective: The objective of the study was to investigate the relationship between tocotrienols (T3s) and induced translocation of DGKα, while exploring the mechanism underlying T3s-induced translocation of DGKα.

Results: Similarly to a-tocopherol, α-T3 binds to 67LR, resulting in the induction of a translocation of DGKα. As well as α-T3, β, γ, and δ Tocs induced translocation of DGKα.

Conclusion: αT3, like αToc, activates DGKα by binding to Leu58 and Trp176 in 67LR. It means 67LR may be important for the functions of T3s, especially the non-oxidant effects of T3s.

Key words: catechins, diabetic nephropathy, diacylglycerol kinase, vitamin E



Graphical Abstract: Tocotrienols activate diacylglycerol kinase a via 67 KDa laminin receptor

INTRODUCTION

In recent years, diabetes has become a significant health concern with an estimated 400 million patients worldwide [1-2]. Prolonged diabetes may lead to complications such as diabetic retinopathy, diabetic neuropathy, and diabetic nephropathy (DN). One-third of diabetic patients suffer from DN, which is characterized by proteinuria, decreased renal glomerular filtration, hypertension, and edema. One of the causes of DN is an increase in diacylglycerol (DG), which activates protein kinase C (PKC), specifically the β subtype [3-7]. However, DG kinase phosphorylates DG to produce phosphatidic acid, decreasing DG [8-11]. Therefore, activation of DGK may improve diabetic nephropathy indirectly by suppressing the abnormal hyperactivity of PKC. Previous data have shown that oral administration of epigallocatechin gallate (EGCG), a type of catechin, and d-α-tocopherol (αToc), a type of vitamin E, increased DGKα activity in the renal glomeruli of diabetic mice, which improved DN [12-17]. The activation of DGKα was also achieved by direct binding of EGCG and αToc to the 67 kDa laminin receptor (67LR) [18-19]. However, the binding sites of EGCG and αToc were different. EGCG binds around the 166th lysine of the 67LR, whereas αToc

binds to the pocket created by the 67LR [18].

Vitamin E is the most abundant fat-soluble antioxidant in vivo and is classified into tocopherols (Toc) and tocotrienols (T3) with four derivatives, γ ,β ,α, and δ, respectively. The former has saturated phytyl groups and the latter has unsaturated isoprenoid side chains (Fig. 1) [20]. Toc is found in soybeans, rapeseeds, and sunflower seeds, while T3 is abundant in oil and cereals such as palm oil, wheat, and barley. αT3 has about 50 times higher antioxidant capacity than αToc, and its unique physiological functions, such as inhibition of atherosclerosis, cell proliferation, and nerve cell protection, have been reported [21-22]. However, the detailed mechanism of action is still unknown except for its antioxidant capacity.

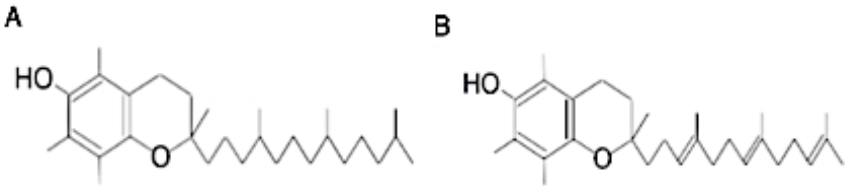


Figure 1. Basic structure of tocopherol (A) and tocotrienol (B)

Numerous functions of α-tocopherol have been studied, including ischemic protection by preventing LDL oxidation, vascular protection, prevention of diabetic complications, and prevention of skin blemishes. Most of these functions are thought to be due to anti-oxidation by oxidizing itself. The antioxidant alone cannot explain many reported functions, such as anticancer effects. Azzi et al. have evaluated these non-antioxidant functions since around 1990 [23]. Yet, this study found that αToc activates DGKα by direct binding to 67LR, which may account for the non-oxidative functions of αToc. T3 also has many functions, and its molecular structure is similar. This suggests that 67LR is also involved in some functions of T3s. Therefore, this study determined if T3 can bind to 67LR and activate DGKα.

MATERIALS & METHODS

Materials: Dulbecco's Modified Eagle medium (DMEM), trypsin-EDTA, fetal Bovine Serum (FBS), Opti-MEM, and penicillin-streptomycin were obtained from Gibco. Fugene HD Transfection Reagent was from Promega. Anti-FLAG mouse antibody was purchased from Santa Cruz Biotechnology. Peroxidase-conjugated affiniPure goat anti-Mouse IgG and peroxidase-conjugated affiniPure goat anti-Rabbit IgG were from Jackson Immuno Research Laboratories. Plasmids encoding FLAG-67LR, GFP-DGKα, and anti-GFP antibody were generated in our laboratory [12]. Anti-67LR antibody was from GeneTex. T3s and αToc were from Cayman Chemical and FUJIFILM Wako Pure Chemicals. EGCG was from LKT Laboratory.

Cell culture: DDT1-MF2 cell, a smooth muscle cell line derived from a leiomyosarcoma of the ductus deferens of a Syrian hamster, was cultured in DMEM-high glucose medium supplemented with penicillin (100 units/ml) and streptomycin (100 mg/ml), 5% FBS without heat treatment, and 20 mM L-glutamine. All cells were cultured at 37°C and 5% CO_2.

Lipofection: DDT1-MF2 cells (1.0×10^5) were seeded in a polylysine-coated glass-bottomed petri dish (MATSUNAMI, 3.5 cm diameter) and cultured for 24 hours. Three hundred µl of OPTI-MEM and 5 µl of Fugene HD transfection reagent (Promega) were mixed well for each glass-bottomed petri dish. Two µg each of plasmid encoding the cDNA of both GFP-DGKα and FLAG-67LR (or mutants) were added to this solution, mixed well, and allowed to stand at room temperature for 10 minutes.

The entire volume of the prepared gene transfer solution was slowly added to the center of a glass-bottomed petri dish seeded the previous day and incubated for 24 hours.

Detection of DGKα activation: Twenty-four hours after the lipofection, cells were stimulated with several concentrations of T3s or αToc for 15 minutes and fixed with 4% paraformaldehyde. Localization of GFP-DGKα was then observed based on the green fluorescent under confocal laser microscopy. DGKα translocation rate (%), which is the number of cells showing membrane localization of GFP-DGKα/total cells observed expressing GFP-DGKα, was used for the activation because DGKα is translocated from the cytoplasm to the plasma membrane when activated.

Molecular docking: Since the αToc binding site is closed in the crystal structure of 67LR (PDB ID: 3BCH) [24], the 3D structure of αToc-bound form of 67LR in the previously performed MD simulation [18] was employed as the receptor. The receptor was prepared using the Protein Preparation Wizard [25, 26]. αToc and EGCG were prepared using the LigPrep function [27, 28]. The box center for the docking calculations was defined using the centroid of selected residues that were found to constitute the EGCG and αToc binding site [29, 18]. The grid required for the docking procedure was generated using a scaling factor of 1.0 and a partial charge cutoff of 0.25, while the X, Y, Z dimensions of the inner box were set to cover the whole binding site. For the ligand docking, a scaling factor of 0.8 and a partial charge cutoff of 0.15 were used to allow complete flexibility of the structures. The poses were selected according to the binding mode and the XP GScore. The Glide Extra-Precision (XP) scoring function was used for the calculations [30]. The binding free energy was calculated by the MM-GBSA method using Prime-MM-GBSA [31].

RESULTS & DISCUSSION

To investigate whether αT3 activates DGKα, we first stimulated DDT1-MF1 cells with 200 μM αT3, which significantly increased the rate of DGKα translocation compared to untreated cells. (Fig. 2A、B).

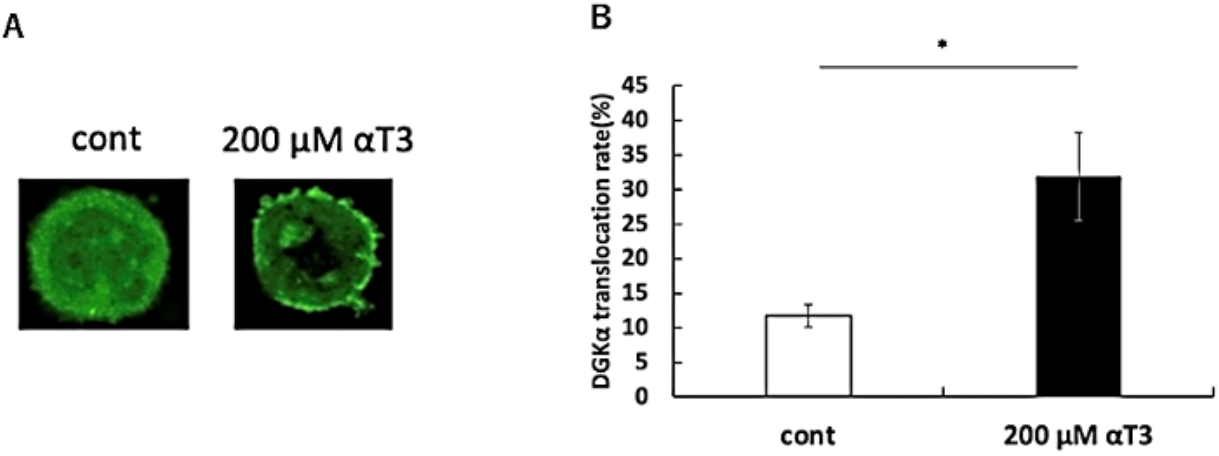


Figure 2. Effect of aT3 on DGKα activation (A) Typical image of GFP-DGKα before (control) and after DDT-MF2 cells expressing both FLAG-67LR and GFP-DGKα by 200 mM αT3 (experiments). (B) The translocation rate was calculated as described in Materials and methods, * shows $p<0.05$ by student's T-test.

Next, the translocation rate of DGKα was examined by varying the concentration of αT3. The results showed that αT3 above 50 μM increased the translocation rate of DGKα, which is a percentage of the cells showing membrane localization of GFP-DGKα in a concentration-dependent manner (Fig. 3A, B).

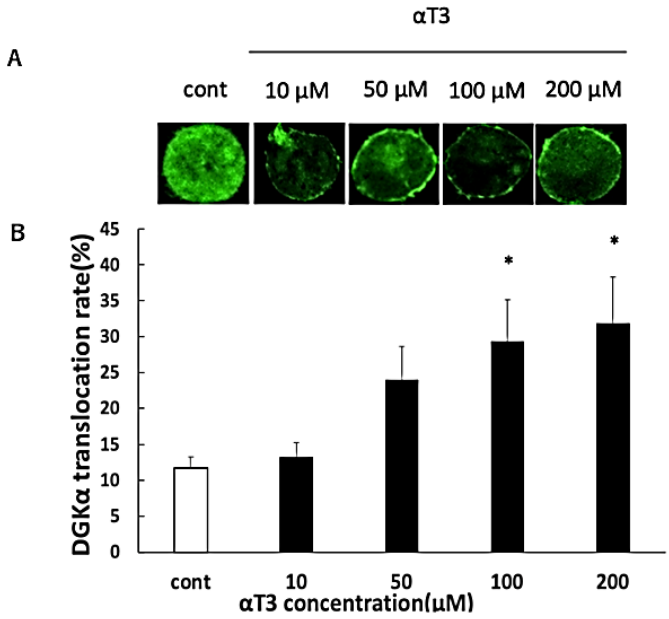


Figure 3. Dose-dependent effect of aT3 on DGKα activation. DDT-MF2 cells expressing both FLAG-67LR and GFP-DGKα were stimulated by 10, 50, 100 or 200 mM αT3. We performed the experiments more than 3 times and observed more than 30 cells in each experiment. Pictures above the graph are typical images of the cell showing the membrane localization of GFP-DGKα, so-called translocation. The percentage of the cells showing the membrane localization of GFP-DGKα (=translocation rate) gradually increased dependent concentration of αT3. * Shows p<0.05 by Dunnett test.

Next, the translocation rate of DGKα was examined by pretreatment with 67LR antibody prior to αT3 stimulation, to determine whether 67LR, as well as αToc, mediates DGKα activation by αT3. The results showed that stimulation of αT3 after pretreatment with 67LR antibody was significantly reduced, compared to the groups that were not treated with the antibody or treated with the control (Fig. 4). This indicates that the activation of DGKα by αT3 was mediated by 67LR.

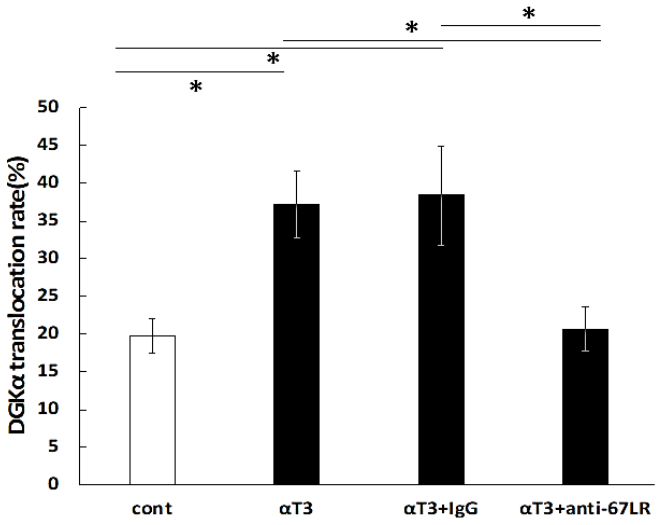


Figure 4. Involvement of 67LR in the aT3-induced activation of DGKα. After preincubation without or with anti-67LR or control antibody (IgG), DDT-MF2 cells expressing both FLAG-67LR and GFP-DGKα were stimulated by 200 mM αT3. We performed the experiments more than 3 times and observed more than 30 cells in each experiment. The translocation rate was calculated as described in Materials and methods, * shows p<0.05 by student's T-test.

To examine the binding sites of αT3 and 67LR, we overexpressed FLAG-67LR with mutations on Leu58 and Trp176, which are the αToc binding sites, or on Lys166, which is the EGCG binding site, and examined their effects on the translocation of DGKα. Figure 5 showed that the translocation rate of DGKα was not significantly changed with the use of the Lys166 mutant. However, the translocation rate significantly decreased in the group mutated in the αToc binding site.

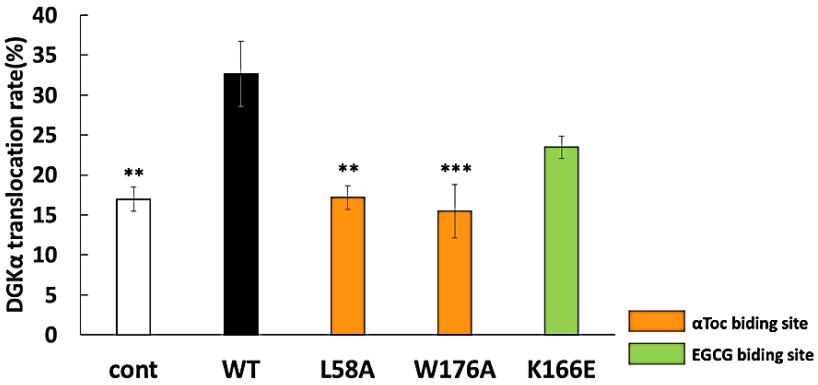
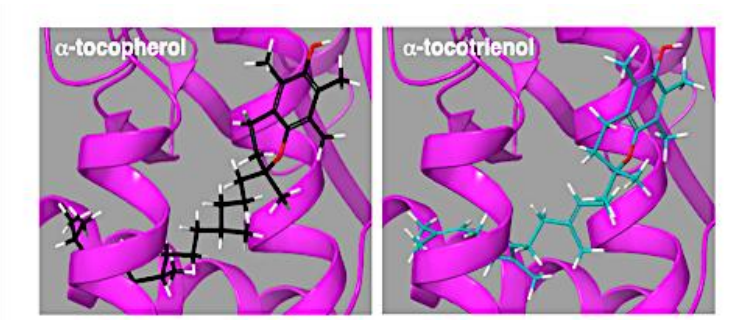


Figure 5. The binding site of αT3 in 67LR. To determine the binding site of αT3, DDT-MF2 cells expressing both GFP-DGKα and FLAG-67LR (WT) or its mutant were stimulated by 200 mM αT3. We performed the experiments more than 4 times and observed more than 30 cells in each experiment. * shows p<0.05 by Student's T-test vs WT.

The results indicated that αT3, like αToc, activates DGKα via binding to Leu58 and Trp176 in 67LR. Since αToc and αT3 have very similar structures (Fig. 1), this finding aligned with expectations. Molecular docking simulation supports the hypothesis that both bind to the same pocket with similar strength; binding free energy of αToc and αT3 was -94.3 and -75.3, respectively (Fig. 6). However, binding free energy of EGCG to the site was -58.01, which was very low compared with αToc and αT3 (data not shown). These results indicate that αToc and αT3 bind to the same site in 67LR, activating DGKα. According to the analysis, 67LR is most often expressed in cancer cells [32]. However, both αToc and αT3 have anti-cancer effects. So, the binding of both αToc and αT3 may be involved in this phenomenon.



	α-tocopherol	α-tocotrienol
Binding free energy *ΔG* (kcal/mol)	-94.3	-75.3

Figure 6. Comparison of the binding free energy of αT3 and αToc in 67LR by docking simulation. The docking simulations were performed using 3D structures of 67LR as a receptor and αToc and αT3 as ligands. The docking poses that gave the best binding score were shown, and the calculated binding free energy was calculated by the MM-GBSA method.

T3 has four isoforms (α-, β-, γ-, and δ-), characterized by a Croman ring attached to the isoprenoid side chain at the C2 position. T3 also has anticancer properties, inducing growth arrest and cell death in several cancer cells derived from breast, gastrointestinal, liver, prostate, lung, and bone cancer [33-36]. However, anticancer effects vary among the T3 isoforms. It was hypothesized that 67LR would show a different affinity to the T3 isoforms. Thus, we examined whether there were differences in DGKα translocation rate among the T3 isoforms. All T3 isoforms significantly activated DGKα, and the strengths to activate DGKα were β-, δ-, α-, and γ-T3, respectively (Fig. 7). However, no significant differences were observed (Fig. 7).

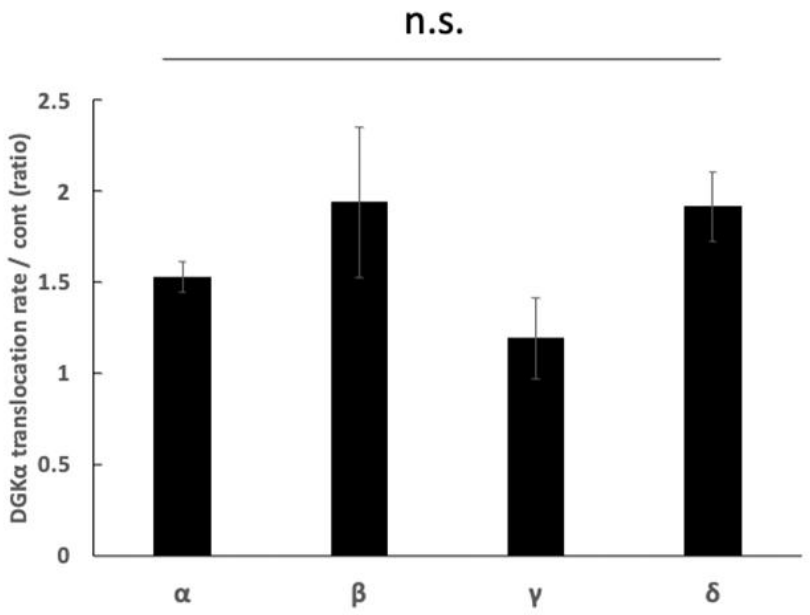
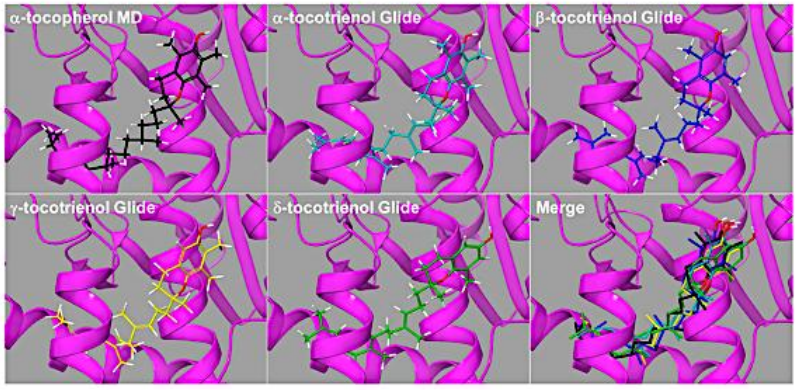


Figure 7. Comparison of the ability of α, β, γ, and dT3 to activate DGKα. To compare the activity to induce DGKα translocation (activation), DDT-MF2 cells expressing both GFP-DGKα and FLAG-67LR (WT) were stimulated by 100 mM α, β, γ, or δT3. We performed the experiments 4 times and observed more than 30 cells in each experiment to calculate the translocation ratio. Each column shows a ratio to control (no stimulation). * Shows $p<0.05$ by the Turky-Kramer test.

Figure 8 shows no difference between binding free energies of T3 isoforms in αToc binding sites of 67LRs, which supports the results in Fig. 7. This finding suggests that all T3s activate DGKα via 67LR. A difference in translocation of DGKα in each T3 may be observed when there is a concentration dependence. However, a significant correlation between increased 67LR expression and tumor progression has been recently reported. The overexpression of 67LR has been observed in many cancers such as chronic lymphocytic leukemia, acute myeloid leukemia, multiple myeloma, and prostate cancer [37-40]. Yet, it has been reported that EGCG binding to 67LR may induce apoptosis via protein kinase B, nitric oxide, and cyclic AMP34, 35. Therefore, the mechanism of 67LR-mediated DGKα activation by T3 revealed in this study may explain the anticancer effects of various T3, but further studies are needed to conclude.

As described above, DGKα activation via 67LR is crucial to ameliorating DN by EGCG and VtE. Therefore, T3 may be effective on DN. Similarly, Naigenin may activate DGKα via 67LR [41]. Yet, we cannot rule out the possible involvement of an anti-oxidative effect in the VtE or EGCG-mediated amelioration of DN because anti-oxidative impact is vital for the amelioration of DN [15, 42].



	α-tocopherol	α-tocotrienol	β-tocotrienol	γ-tocotrienol	δ-tocotrienol
Binding free energy ΔG (kcal/mol)	-94.3	-75.3	-81.1	-85.4	-79.6

Figure 8. Comparison of binding free energy of α, β, γ, δT3, and αToc to 67LR by docking simulation. The docking simulations were performed using 3D structures of 67LR as a receptor and αToc, α, β, γ, and δT3 as ligands. The docking poses that gave the best binding score were shown, and the binding free energy was calculated using the MM-GBSA method.

CONCLUSION

αT3, like αToc, activates DGKα by binding to Leu58 and Trp176 in 67LR. Therefore, 67LR may be important for the functions of T3s, especially the non-oxidant effects of T3s, such as the anti-cancer function.

Abbreviations: d-α-tocopherol (αToc), diacylglycerol (DG), diacylglycerol kinase (DGK), Dulbecco's Modified Eagle medium (DMEM), diabetic nephropathy (DN), ethylenediamine tetraacetic acid (EDTA), epigallocatechin gallate (EGCG), fetal bovine serum (FBS), green fluorescent protein (GFP), knock out (KO), protein kinase C (PKC). tocopherol (Toc), tocotrienol (T3), 67 KDa laminin receptor (67LR)

Competing interests: The Authors have no conflict of interest.

Author's contribution: Tomoko Namba performed almost all the experiments, Dr. Daiki Hayashi performed the Glide simulation, Dr. Yasuhito Shirai designed and conducted the experiments, and Drs. Itsuko Fukuda and Shuji Ueda helped design and financially supported a series of experiments.

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REFERENCE

1. Gupta S., Sharma N., Arora S., Verma S. Diabetes: a review of its pathophysiology, and advanced methods of mitigation. *Curr Med Res Opin.*2024; 40, 773-780. DOI: https://doi.org/10.1080/03007995.2024.2333440.
2. Goyal R., SinghalM., Jialal, S. Type 2 Diabetes. StatPearls; 2023. DOI: https://www.ncbi.nlm.nih.gov/books/NBK513253/
3. Banerjee S., Lu S., Jain A., Wang I., Tao H., Srinivasan S., et al. Targeting PKCalpha alleviates iron overload in diabetes and hemochromatosis through the inhibition of ferroportin. *Blood.* 2024; 144, 1433-1444. DOI: https://doi.org/10.1182/blood.2024023829.
4. Zhang R., Qin C., Zhang J., Ren H., Wang Y., Wu Y., et al. DNA hypomethylation of Syk induces oxidative stress and apoptosis via the PKCbeta/P66shc signaling pathway in diabetic kidney disease. *FASEB J.* 2024; 38, e23564. DOI: https://doi.org/10.1096/fj.202301579R.
5. Nadendla R.R., Khairunnisa K., Aziz N., Pyne C., Panigrahy

P.U., Wal P., et al. An updated review on diabetic nephropathy: potential mechanisms, biomarkers, therapeutic targets and interventional therapies. *Curr Diabetes Rev*. 2024; 21, e240624231266. DOI: https://doi.org/10.2174/0115733998291920240611063402

6. Sanapalli V., Sigalapalli D.K., Shaik A.B., Bhandare R.R., Sanapalli B.H.R. Computational elucidation of human beta-defensin-2 as a dual inhibitor of MMP-9 and PKC-betaII for diabetic wound management. *ACS Omega*. 2025; 10, 3575-3584. DOI: https://doi.org/10.1021/acsomega.4c08292.
7. Li W., Zhu K., Liu Y., Liu M., Chen Q. Recent advances in PKC inhibitor development: Structural design strategies and therapeutic applications. *J Med Chem*. 2025; 2287, 17290. DOI: https://doi.org/10.1016/j.ejmech.2025.117290.
8. Barbernitz X. & Raben DM. Phosphorylation of DGK. *Adv Biol Regul*. 2023; 88, 100941. DOI: https://doi.org/10.1016/j.jbior.2022.100941.
9. Martin-Salgado M., Ochoa-Echeverría A., Mérida I. Diacylglycerol kinases: A look into the future of immunotherapy. *Adv Biol Regul*. 2024; 91:100999. DOI: https://doi.org/10.1016/j.jbior.2023.100999.
10. Liu Y., Yang Z., Zhou X., Li. Z., Nakanishi H. Diacylglycerol kinases and its role in lipid metabolism and related diseases..*Int J Mol Sci.* 2024; 25, 13207. DOI: https://doi.org/10.3390/ijms252313207.
11. Sakane F., Murakami C., Sakai H. Upstream and downstream pathways of diacylglycerol kinase: Novel phosphatidylinositol turnover-independent signal transduction pathways. *Adv Biol Regul.* 2025; 95, 101054. DOI: https://doi.org/10.1016/j.jbior.2024.101054
12. Hayashi D., Ueda S., Yamanoue M., Saito N., Ashida H., Shirai Y. Epigallocatechin-3-gallate activates diacylglycerol kinase alpha via a 67 kDa laminin receptor: A possibility of galloylated catechins as functional food to prevent and/or improve diabetic renal dysfunctions. *J Funct. Foods* 2015; 15, 561-569. DOI: https://doi.org/10.1016/j.jff.2015.04.005.
13. Hayashi D., Yagi K., Song C., Ueda S., Yamanoue M., Topham M., et al. Diacylglycerol kinase alpha is involved in the vitamin E-induced amelioration of diabetic nephropathy in mice. *Sci. Rep*. 2017; 7, 2597. DOI: https://doi.org/10.1038/s41598-017-02354-3.
14. Kakehi T., Yagi K., Saito N., Shirai Y. Effects of vitamin E and its derivatives on diabetic nephropathy in Rats and identification of diacylglycerol kinase subtype involved in the improvement of diabetic nephropathy. *Functional Foods in Health and Disease.* 2017; 7, 816-832. DOI: https://doi.org/10.31989/ffhd.v7i10.386.
15. Hayashi D., Ueda S., Yamanoue M., Ashida H., Shirai Y. Amelioration of diabetic nephropathy by oral administration of d-α-Tocopherol and its mechanisms. *Biosci. Biotechnol. Biochem.* 2018; 82, 65–73. DOI: https://doi.org/10.1080/09168451.2017.1411184.
16. Hayashi D., Wang L., Ueda S., Yamanoue M., Ashida H., Shirai Y. The mechanisms of ameliorating effect of a green tea polyphenol on diabetic nephropathy based on diacylglycerol kinase α. *Sci. Rep*. 2020; 10, 11790. DOI: https://doi.org/10.1038/s41598-020-68716-6.
17. Fukunaga-Takenaka R., ShiraiY., Yagi K. Adachi N., Sakai N., Merino E., et al. Importance of chroman ring and tyrosine phosphorylation in the subtype-specific translocation and activation of diacylglycerol kinase alpha by D-alpha-tocopherol. *Genes Cells*. 2005;10, 311-319. DOI: https://doi.org/10.1111/j.1365-2443.2005.00842.x
18. Hayashi D., Mouchlis D.V., Okamoto S., Namba T., Wang L., Li S., et al. Vitamin E functions by association with a novel binding site on the 67 kDa laminin receptor activating diacylglycerol kinase. *J Nutr Biochem*. 2022; 110, 109129. DOI: https://doi.org/10.1016/j.jnutbio.2022.109129.
19. Hayashi D. & Shirai Y. The role of diacylglycerol kinase in the amelioration of diabetic nephropathy. *Molecules* 2022; 27, 6784. DOI: https://doi.org/10.3390/molecules27206784.
20. Tian H., Li Y.F., Jiao G.L., Sun W.Y., He R.R. Unveiling the antioxidant superiority of alpha-tocopherol: Implications for vitamin E nomenclature and classification. *Free Radic Biol Med*. 2024; 216, 46-49. DOI: https://doi.org/10.1016/j.freeradbiomed.2024.03.003.
21. Jiang, Q. Different roles of tocopherols and tocotrienols in chemoprevention and treatment of prostate cancer. *Adv Nutr*. 2024 ;15, 100240. DOI: https://doi.org/10.1016/j.advnut.2024.100240.
22. Jang Y. and Kim C.Y. The role of vitamin E isoforms and metabolites in cancer prevention: mechanistic insights into sphingolipid metabolism modulation. *Nutrients,* 2024; 16, 4115. DOI: https://doi.org/10.3390/nu16234115.
23. Zingg J.M. and Azzi A. Non-antioxidant activities of vitamin E. *Curr Med Chem*. 2004; 11; 1113-1133. DOI: https://doi.org/10.2174/0929867043365332.
24. Nelson J., McFerran N.V., Pivato G., Chambers E., Doherty C., Steele D., et al. The 67 kDa laminin receptor: structure, function and role in disease. *Biosci Rep*. 2008; 28, :33-48. DOI: https://doi.org/10.1042/bsr20070004.
25. Afanamol M.S., Dinesh A.D., Ali K.S., Vengamthodi A., Rasheed, A Drug repurposing by in silico prediction of cyclizine derivatives as antihyperlipemic agents. *In Silico Pharmacol*. 2023; 11, 27. DOI: https://doi.org/10.1007/s40203-023-00164-2.

26. Wang K., Zhang P., Sun H., Cui S., Ao L., Cui M., et al. Dual-function natural products: Farnesoid X receptor agonist/inflammation inhibitor for metabolic dysfunction-associated steatotic liver disease therapy. Chin J Nat Med. 2023; 22, 965-976. DOI: https://doi.org/10.1016/S1875-5364(24)60706-5.
27. Halder D., Das S., Joseph A., Jeyaprakash R.S Molecular docking and dynamics approach to *in silico* drug repurposing for inflammatory bowels disease by targeting TNF alpha. *J Biomol Struct Dyn*. 2023; 41, 3462-3475. DOI: https://doi.org/10.1080/07391102.2022.2050948.
28. Zhou C., Yan L., Xu J., Hamezah H.S., Wang T., Du F., et al. Phillyrin: an adipose triglyceride lipase inhibitor supported by molecular docking, dynamics simulation, and pharmacological validation. *J Mol Model*. 2024; 30, 68. DOI: https://doi.org/10.1007/s00894-024-05875-7.
29. Tachibana H., Koga K., Fujimura Y., Yamada K. A receptor for green tea polyphenol EGCG. *Nat. Struct. Mol. Biol.* 2004; 11, 380–381. DOI: https://doi.org/10.1038/nsmb743.
30. Abdel-Kader M.S., Almutib F.S., Aldosari A.F., Soliman G.A., Elzorba H.Y., Alqarni M.H., et al. *In vitro* and *in silico*anti-picornavirus triterpene alkanoic acid ester from saudi collection of *Rhazya strict* decne. *Metabolites.* 2023; 13, 750. DOI: https://doi.org/10.3390/metabo13060750.
31. Police A., Shankar V.K., Pandey P., Rangappa S., Doerksen S.J., Murthy S.N. Novel topical anandamide formulation for alleviating peripheral neuropathic pain. *Int J Pharm.* 2023; 641, 123085. DOI: https://doi.org/10.1016/j.ijpharm.2023.123085.
32. Huang Y., Kumazoe M., Bae J., Yamada S., Takai M., Hidaka S., et al. Green tea polyphenol epigallocatechin-O-gallate induces cell death by acid sphingomyelinase activation in chronic myeloid leukemia cells. *Oncology Reports.* 2015; 34, 1162-1168. DOI: https://doi.org/10.3892/or.2015.4086.
33. Ranasinghe R., Mathai M. Zulli A. Revisiting the therapeutic potential of tocotrienol. *Bio Factors*. 2022; 48, 813-856. DOI: https://doi.org/10.1002/biof.1873.
34. Pang K.L., Foong L.C., Abd Ghafar N., Soelaiman I.N., Law J.X., Leong L.M., et al. Transcriptomic analysis of the anticancer effects of annatto tocotrienol, delta-tocotrienol and gamma-tocotrienol on chondrosarcoma cells. *Nutrients* 2022; 14, 4277. DOI: https://doi.org/10.3390/nu14204277.
35. Younes M., Loubnane G., Sleiman C., Rizk S. Tocotrienol isoforms: The molecular mechanisms underlying their effects in cancer therapy and their implementation in clinical trials. .*J Integr Med*. 2024; 22, 1-11. DOI: https://doi.org/10.1016/j.joim.2024.01.002.
36. Montagnani M.M., Macchi C., Ruscica M., Sartori P., Moretti R.M. Anticancer activity of delta-tocotrienol in human hepatocarcinoma: involvement of autophagy induction. *Cancers.* 2024; 16, 2654. DOI: https://doi.org/10.3390/cancers16152654.
37. Bae J., Kumazoe M., Lee K.W., Fujimura Y., Tachibana H. 67-kDa laminin receptor mediates oolonghomobisflavan B-induced cell growth inhibition in melanoma. *Phytomedicine*. 2023; 118, 154970. DOI: https://doi.org/10.1016/j.phymed.2023.154970.
38. Weiss S.F.T. & Ferreira E. Downregulation of LRP/LR with siRNA inhibits several cancer hallmarks in lung cancer cells. *FEBS Open Bio*. 2023; 13, 323-340. DOI: https://doi.org/10.1002/2211-5463.13544.
39. Cloutier G. and Beaulieu J.F. Reconsideration of the laminin receptor 67LR in colorectal cancer cells. *Biomol Biomed*. 2024; 24, 1117-1132. DOI: https://doi.org/10.17305/bb.2024.10323
40. Bae J. and Park S.J. The combination of oolonghomobisflavan B and diallyl disulfide induces apoptotic cell death via 67-kDa laminin receptor/cyclic guanosine monophosphate in acute myeloid leukemia cells. Curr Issues *Mol Biol.* 2024; 46, 2444-2455. DOI: https://doi.org/10.3390/cimb46030154.
41. Chinthammit B., Okamoto S., Shirai Y. Almond skin polyphenol extracts stimulate the activation of diacylglycerol kinase alpha via a 67 kDa laminin receptor. Functional Foods in Health and Disease 2022; 12,151-160. DOI: https://www.doi.org/10.31989/ffhd.v12i3.894.
42. Ramadan O.I., Taha R.S., Awad M.M., Abdelhay O.M., Ereba R.E., Younes A.M., et al., Combination of empagliflozin and curcumin ameliorates the induced diabetic nephropathy in male albino rats, through their synergistic anti-diabetic and antioxidative effects. Bioactive Compounds in Health and Disease 2025; 8, 90 - 103. DOI: https://www.doi.org/10.31989/bchd.8i3.1588