科技部補助

大專學生研究計畫研究成果報告

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＊ 計 畫 ＊
＊ : 探討下視丘神經?在糖尿病動物的食慾調控所扮演之角色 ＊
＊ 名 稱 ＊
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學生計畫編號： MOST 103-2815-C-040-040-B
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大專學生研究計畫研究成果報告

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執 行 單 位：中山醫學大學醫學系生理科

中華民國 104 年 03 月 09 日

探討下視丘神經肽在糖尿病動物的食慾調控所扮演之角色

中文摘要：

糖尿病目前是美國排名第四位的致死病因，而且在台灣自1983年以來已是台灣的前五大死因之一。常見的糖尿病分為先天和後天這兩種形式，並且糖尿病會引發許多的併發症包括多飲、多食，多尿還有視物模糊。目前已有許多針對糖尿病老鼠下視丘進行不同的研究，例如:下視丘的胰高糖血素(glucagon)抑制肝臟的葡萄糖生產。由先前的文獻探討得知食慾會受到下視丘的控制，因此想要了解糖尿病病患所產生的多飲還有多食的病徵是否和刺激下視丘的飲食中樞有所關連性。首先利用雌性去卵巢大鼠將其分為控制組以及糖尿病組，糖尿病組的大鼠是利用腹腔注射Streptozotocin藥物(STZ，65mg/kg)所誘導，接著在注射STZ 7天後犧牲並且取出大鼠的下視丘進行蛋白萃取。接著利用雄性大鼠，並且分為以下四種不同的處理分別是：控制組、糖尿病組、糖尿病接受胰島素治療組(insulin，8 IU/kg，每12小時給予一次並持續三天)以及糖尿病接受根皮苷治療組(phlorizin，400 mg/kg，每8小時給予一次並持續三天)。糖尿病組大鼠在注射STZ 7天後犧牲而接受胰島素治療組及接受根皮苷治療組在結束三天的治療後犧牲並取出大鼠的下視丘進行蛋白萃取。萃取出的蛋白利用Western blot進行蛋白質分析。根據實驗結果顯示NPY1R在雌性去卵巢糖尿病大鼠的下視丘表現量有顯著的增加，因此推論NPY1R對於雌性去卵巢糖尿病大鼠的食慾是具有影響力的。而相較於NPY1R蛋白表現量有明顯的差異，其他的接受器（NPY5R、MC3-R和MC4-R）還有神經胜肽（NPY、CART(55-102)和CART(61-102)）都沒有明顯的改變。然而，在雄性大鼠的實驗中，我們所進行的NPY、NPY1R、CAT(55-102)還有CART(61-102)的蛋白質表現量在雄性控制組大鼠、糖尿病組大鼠、注射胰島素治療的糖尿病組還有注射根皮苷治療的糖尿病組大鼠之間都沒有顯著的差異。根據實驗結果推論影響糖尿病大鼠食慾增加的主要因素可能不是因為下視丘神經胜肽蛋白表現量改變所造成，而是其他更複雜的因素所導致。

關鍵字：糖尿病、食慾、下視丘、神經胜肽

ABSTRACT

Current evidence revealed that diabetes mellitus (DM) is ranked fourth in the cause of the death in the United States. Moreover, DM is also among the top five causes of death in Taiwan since 1983. The onset of diabetes can be divided into nature and nurture. Diabetes also cause many complications including ploydipsia, polyphagia, polyuria, and blurred vision. There were many different researches focusing on hypothalamus of diabetic rats. For example, glucagon signal in the hypothalamus inhibits liver glucose production. Previous evidence shows that appetite is under the control of hypothalamus. Therefore, the present study investigated whether the diabetic symptoms in patients were connected with the changes in hypothalamus. Ovariectomized (OVX) female rats were divided into control and diabetic groups. Diabetic animals were induced by streptozotocin (STZ) (50 mg/kg; i.p.) injection. After 7 days of STZ injection, rats were scarified and their hypothalami were removed. The result showed that the expression of NPY1R was significantly increased in female diabetic rats. Therefore, NPY1R might affect the appetite of female diabetic rat. However, other receptors (NPY5R, MC3-R, and MC4-R) and their ligands (NPY, CART(55-102), and CART(61-102)) didn't reveal significant difference. Another experiment in diabetic male rats was carried out by dividing them into four groups: control, diabetic, insulin replacement diabetic, and phlorizin treated diabetic rats. Insulin replacement diabetic rats were given insulin subcutaneously in diabetic rats at the dose of 8 IU/kg every 12-hour for 3 days, while phlorizin was given subcutaneously in diabetic rats at the dose of 400 mg/kg every 8-hour for 3 days. Results revealed that NPY, NPY1R, CART(55-102), and CART(61-102) expression in four groups were similar. These results suggested the main reason for the hyperphagia in diabetic rats might not be due to the changes in hypothalamic neuropeptides and blood glucose, but might be caused by others complex factors.

Key words: diabetes mellitus、appetite、hypothalamus、neuropeptides

目錄

（一） 前言

目前，糖尿病（diabetes mellitus, DM）在美國的致死病因排名第四位，並自1983 年以來一直位居台灣前五大死因之一，而常見的糖尿病可分為第一型糖尿病(幼年型)和第二型糖尿病(肥胖型)這兩種形式。糖尿病會引起許多的併發症，其中包括多飲、多食、多尿和視力模糊等。在正常動物，下視丘 (hypothalamus)是神經內分泌的中樞，它的主要功能是調節內臟活動、清醒及睡眠週期、內分泌活動、飲水行為、及攝食行為等。在攝食行為方面，下視丘弓形核 (arcuate nucleus)中會分泌神經胜肽，例如：neuropeptide Y (NPY)、pro-opiomelanocotin (POMC)、Agouti-related peptide (AgRP)、orexin、和 cocaine- and amphetamine-regulated transcript (CART)，這些神經胜肽會透過其相對應的接受器，對於攝食產生促進或是抑制的效果。目前已知，在正常動物，厭食劑(例如安非他命)之作用方式是透過：(1)減少下視丘 NPY，及(2)增加下視丘 POMC 及 CART 基因表現，去減少攝食行為及減輕體重。此外，許多訊息傳遞，例如 PKA、PKC、cFos/cJun、AP-1、CREB、及 NK-kB signaling 等，以及抗氧化酶，例如 superoxide dismutase (SOD)、peroxisome、及 catalase 等，都與這些神經胜肽一起被活化，來調控食慾。但是，此種調控方式發生在具貪食行為之糖尿病動物時，神經胜肽調控方式是否有改變？若有不同，是何種(或何類)神經胜肽發生改變？此外，究竟何種訊息傳遞或抗氧化酶會發生改變？因此，本研究將探討：下視丘的神經胜肽(NPY、POMC、及CART)、訊息傳遞(PKA-CREB signaling)、或抗氧化酶(SOD)，如何對第一型糖尿病動物的食慾進行控制。

（二） 研究目的

糖尿病 (*Diabetes mellitus,* DM)是一種代謝疾病，目前糖尿病主要分為兩種型式:(1)第一型糖尿病:主要為胰島素缺乏，因為產生胰島素的細胞，即胰腺 beta 細胞，無法產生足夠的胰島素，此為幼年型或先天型的糖尿病，因患者常伴有自體抗體(anti-insulin autoantibody)的產生。此外，患者需注射胰島素治療，故又稱胰島素依賴型糖尿病(insulin-dependent DM)；(2)第二型糖尿病:主要為胰島素接受器產生缺陷所導致，此為肥胖型或後天型的糖尿病。因患者胰島素分泌正常，治療時不需注射胰島素，故又稱故又稱非胰島素依賴型糖尿病(insulin-independent DM)。糖尿病早期的病徵包括：高血糖、糖尿、及三多(多飲、多食、多尿)等 *(Cooke DW et al., 2008)*。另外，糖尿病發後的併發症包括：血管疾病、周圍神經病變、腎臟疾病、視覺模糊、和對感染的易感性等。糖尿病透過針對飲食的控制、運動、注射胰島素、或口服降血糖藥物的治療，可以達到降低血糖水平的功能而控制病情，有效的血糖控制可以延遲或防止併發症。

下視丘 (hypothalamus)位於腦部的腦下腺上方，與腦下腺前葉以垂體門脈系統的血管相聯繫，控制身體多項功能 *(Melmed S et al., 2005)*。下視丘具有調節體溫、血糖、水平衡、脂肪代謝、攝食習慣、情緒、荷爾蒙的製作、以及晝夜節律等功能。在攝食調控方面，下視丘外側區 (lateral hypothalamic area)為進食中樞(feeding center)，主要功能是負責食物攝入量的控制，因此當此處受到刺激興奮時會導致食量增加；反之，若是這一區受到損害，則會導致減少食物的攝取量或厭食症。下視丘腹內側區(ventromedial hypothalamic)為飽覺中樞(satiety center)，主要功能是負責飽的感覺以減少食物攝入量，因此當此處受到刺激興奮時會導致食量減少；反之，若是這一區受到損害，則會導致增加食物的攝取量或肥胖。

糖尿病動物具貪食現象，因此，下視丘的調控機制可能發生病變，我們將探討下視丘神經胜肽類(neuropeptides)之改變方式。透過下視丘的兩種神經元群體：一種為增進食慾的神經元: NPY/AgRP 神經元，這種神經元受到刺激會使得攝食量增加，而另一種為抑制食慾的神經元: POMC/ CART 神經元，此種神經元若是受到刺激則會使得攝食量減少。與正常動物相比較，糖尿病動物下視丘之此兩類神經胜肽必有某些程度的改變。

（三） 文獻討論

下視丘的主要功能是調節內臟活動和內分泌活動，因此它在自主神經系統以及內分泌系統中扮演著重要的角色，而下視丘所能控制的多項功能中，包括可以調控攝食行為。從早期的實驗可以知道，下視丘外側區 (lateral hypothalamic area)是「飢餓中樞」而下視丘腹內側核 (ventromedial hypothalamic nucleus)則是「飽食中樞」*(Mayer J et al., 1967)*。此外，下視丘的弓形核 (arcuate nucleus)的主要產物有兩種神經元群體：一種為增進食慾的神經元: NPY/Agouti related peptide (AgRP)神經元，這種神經元受到刺激會使得攝食量增加 *(Kalar SP et al., 1999; Morton GJ et al.,2001; Ellacott KL et al.,2004)*，而另一種為抑制食慾的神經元: POMC/ CART 神經元，此種神經元若是受到刺激則會使得攝食量減少。這兩種神經元群體的作用雖然是相互拮抗的 *(Zigman JM et al., 2003; Schwartz MW et al., 2005)*，但是確能藉由整合荷爾蒙(如:瘦素)和營養訊號共同調控攝食量和體重 *(Cone RD, 2006; Elmquist JK et al., 1999; Schwartz MW et al., 2000; Spieglman BM et al., 2001)*。

NPY 在下視丘中是一種重要的促進食慾胜肽 *(Clark JT et al., 1984; Levine AS et al.,1984; Stanley BG et al., 1984)*，NPY 在弓形核中可以利用促進攝食量控制身體能量的平衡。下視丘弓形核能夠分泌 NPY，NPY 可以藉由和 NPY Y1 receptor（Y1R) 接受器或 NPY Y5 receptor（Y5R) 接受器的結合，促進食物的攝取量 *(Roseberry AG et al., 2004)*，其作用分別為 NPY 透過刺激 Y5R 接受器可以增加對食物的攝取量，另外 NPY 透過對 Y1R 接受器作用影響能夠對食物的囤積作用 *(Keen-Rhinehart E et al., 2007)*。

POMC 神經元製造 POMC 後須進行修飾作用，以產生功能性蛋白質。而 POMC 進行乙醯化修飾後所製造出的 α-melanocyte-stimulating hormone（α-MSH) 可以調節食物的攝取量和身體能量的支出 *(Takahashi A et al., 2013; Monutjoy KG, 2010)*。α-MSH 可以和存在於下視丘室旁核 (paraventricular nucleus) 的 melanocortin-4 receptor (MC-4R)或 melanocortin-3 receptor (MC-3R)接受器結合，產生抑制攝食量的效果 *(Shwartz MW et al., 2000)*。

CART 在下視丘中和能量的動態平衡還有神經內分泌的調控有所關聯，另外 CART 在弓形核中作用的位置與 POMC 相同，能夠與 MC-4R 結合產生抑制攝食量的效果 *(Kristensen P et al.,1998)*。

在正常動物，我們實驗室發現，作用在神經中樞的厭食劑(例如安非他命或 phynylpropanolamine)，可透過減少下視丘 NPY，及增加下視丘 POMC 來調控攝食行為*(Hsieh et al., 2011; Kuo et al., 2011)*，此外，抗活性氧族群 (reactive oxygen species) 及 free radicles 之酵素，例如 SOD and nitric oxide synthesis (NOS)，也參與厭食的調控 *(Kuo et al., 2009; Kuo et al., 2012)*。許多訊息傳遞，例如：protein kinase A（PKA) *(Hsieh et al., 2007)*、protein kinase C (PKC) *(Kuo et al., 2009)*、cAMP response element binding protein (CREB) *(Hsieh et al., 2008)*、nuclear protein kappa B (NF-kB) *(Kuo et al., 2012)* signaling，參與 POMC 及食慾的調控。此外，Y1R receptor 及 MC3R 也參與厭食劑之調控機制 *(Hsieh et al., 2011; Hsieh et al., 2013)*。但是，這些神經胜肽、訊息傳遞、或神經胜肽接受器的調控方式，若發生在具貪食行為之糖尿病動物時，是否有類似現象？因此，本研究將探討下視丘的神經胜肽及訊息傳遞如何對第一型糖尿病動物的食慾進行控制。

本實驗的主要目的是利用第一型糖尿病大鼠模式，觀察其攝食量的變化和下視丘神經胜肽(以 NPY、POMC 和 CART 為主)含量變化間的關聯性。此外，下視丘訊息傳遞（PKA-CREB signaling)，神經胜肽接受器（Y1R, MC3R)，及抗氧化酶(SOD) 含量變化也將一併探討。

（四） 研究方法

本實驗旨在探討下視丘的神經胜肽如何對糖尿病動物的食慾進行控制。

1. 實驗分組

Male Wistar rats (200~300 g, Animal Center of National Cheng Kung University Medical College) were housed individually in a cage, maintained at 22±2°C according to a 12:12-h light-dark cycle (lights on at 6:00 AM), and habituated to frequent handling. Drug administration and food intake assessment (LabDiet, PMI Nu- trition International, Brentwood, MO) were performed daily at the start of dark phase (6:00 PM). This study has been carried out in accordance with the

Guide for the Care and Use of Laboratory Animals as adopted by the National Institutes of Health. Rats were divided into 4 groups:

Group 1: Control (normal rats)

Group 2: Diabetes rats (50 mg streptozotocin in 0.05 M citrate buffer)

Group 3: Diabetes + insulin treatment (8 IU/kg every 12-hour for 3 days)

Group 4: Diabetes + phlorizin treatment (400 mg/kg every 8-hour for 3 days)

2. 誘導第一型糖尿病動物及血糖測試

(1) Induction of diabetic condition,

Rats were fasted for 24 h and then given with a single injection of STZ (65 mg/kg, i.v.) into the femoral vein under the anesthesia of pentobarbital (Nembutal, 40 mg/kg, i.p.). A fasting blood glucose level greater than 350 mg/dL at 72 h after STZ injection confirmed the presence of diabetes. Hyperglycemia was confirmed once for every 3 days during this stage and sustained for at least 3 weeks. Clinical features of the disease (polyuria, polydipsia, polyphagia, weight loss and malaise) were also observed during this stage. Blood glucose levels were measured using glucose oxidase method as described in the purchased Glucose Kits. One week after establishing basic hyperglycemia, the animals were divided into several groups for further studies

(2) Determination of plasma glucose level (PGL)

To examine the change of PGL, blood samples were taken from femoral vein for 4 days in both normal and diabetic rats. Blood were centrifuged immediately at 15000 rpm for 3 min and PGL was measured using glucose oxidase method.

(3) Normalization of PGL by phlorizin treatment

To examine the effect of restored euglycemia on NPY, POMC, and CART expression in diabetic rats, diabetic rats was given with phlorizin (0.4 g/kg, s.c.) twice a day for 4 days (Day -3 ~ Day 0). The drug phlorizin was used to normalize the PGL. Phlorizin normalizes PGL by blocking reabsorption of glucose from kidney tubule and increasing glycosuria *(Rossetti et al., 1987)*. In addition to phlorizin, insulin was also used to normalize the PGL as described below.

(4) Insulin replacement

The preliminary study revealed that repeated treatments of insulin (1 unit/ml, i.p.) twice a day for at least 7 days were necessary for the restoration of feeding behavior, PGL and NPY content in diabetic rats. Therefore, insulin was

administered to diabetic rats twice daily for 7 days (Day –6 ~ Day 0).

3. 取下視丘組織，抽取及分析 mRNA

（1） RNA extraction

Hypothalamic NPY, POMC and CART mRNA levels were measured in a block of mediobasal hypothalamic tissue as described previously *(Morris, 1989)*. The block of mediobasal hypothalamic tissue was dissected rostral-caudally from the optic chiasma to the mammillary body, and extended laterally from the midline of hypothalamus to the perihypothalamic nucleus and superiorly to the anterior commissure. Total RNA was isolated from tissues using a modified guanidinium thiocyanate-phenol-chloroform method *(Chomczynski & Sacchi, 1987)*. Each hypothalamic block was homogenized in 1 ml of TRIZOL reagent using an Ultrasonic Processor (Vibra Cell, Model CV17; Sonics & Materials Inc., Danbury, Connecticut, USA). After an incubation at 22℃ for 5 mins, each sample was added with 0.2 ml of chloroform, shaken vigorously for 15 secs, incubated at 22℃ for 3 mins, and then centrifuged at 12,000 g for 15 mins under 4℃. After removal of aqueous phase and precipitation with 0.5 ml isopropanol, samples were incubated at 22℃ for 10 mins and centrifuged at 12,000 g for 15 mins under 4℃. The gel-like RNA pellets were washed with 75% ethanol by vortexing and centrifugation at 7,500 g for 5 mins under 4℃. Thereafter, RNA pellets were dried briefly, dissolved in RNase-free water, and stored at -80℃. The RNA was determined spectrophotometrically at 260 nm (Hitachi U-3210, Japan).

(2) RT-PCR

With the use of the First-Strand cDNA Synthesis Kit (Boehringer Mannheim), RNA was reverse transcribed into single- stranded cDNA. For each sample, 8 μl of sterile diethyl pyrocarbonate (DEPC)-water containing 2 μg of RNA were added to oligo-p(dT)15 primer (0.8 μg/μl) followed by a heating at 65°C for 15 min, a cooling at 25°C for 10 min, and then addition to a reaction mixture consisting of 10X reaction buffer (100 mM Tris, 500 mM KCl; pH 8.3), deoxynucleotide mix (10 mM each), MgCl2 (25 mM), RNase inhibitor (40 units/μl), and AMV reverse transcriptase (25 units/μl). Reaction mixtures were incubated at 42°C for 2 h and then brought to 95°C for 5 min to terminate the reaction, followed by soaking at 16°C. PCR was subsequently carried out by mixing 3 μl of cDNA product with mastermix solution consisting of DEPC-water, 10X reaction buffer, MgCl2 (25 mM), deoxynucleotide mix (10 mM each), P1 and P2 primers (1 μg/μl each), and *Taq* polymerase (5 units/μl). GAPDH was

used as the internal standard calibrator. PCR reactions for NPY were carried out on a PCR thermocycler (Perkin-Elmer GeneAmp 2400) for 28 cycles with the following steps: 91°C for 1 min (denaturing), 60°C for 1 min (annealing), and 72°C for 30 s (extension), followed by a final elongation step at 72°C for 7 min; and finally the PCR products were soaked at 16°C. PCR reactions for the other molecules analyzed were carried out in steps similar to those de- scribed above except for changes in two steps (annealing and cycles), described as follows: NPY (60°C, 28 cycles); POMC (60°C, 28 cycles); CART (60°C, 28 cycles).

(3) Gel electrophoresis

After RT-PCR, 8 μl of each PCR product was subsequently separated by flat-bed gel electrophoresis on a 3% agarose gel. Gels stained by ethidium bromide (0.5 μg/ml) were visualized under UV light, photographed, and then scanned densitometrically (Hoefer, San Francisco, CA, USA). Ratios of NPY and GAPDH mRNA for each treatment day were calculated to determine relative NPY mRNA levels. Contents of NPY mRNA in AMPH-treated group were indicated as the percentage of control group. Similar steps were used to determine hypothalamic NPY, POMC and CART mRNA levels.

4. 取下視丘組織，抽取蛋白質（Western blot)

Protein samples extracted from hypothalamus tissue were separated in a 12.5% polyacrylamide gel, transferred onto a nitrocellulose membrane, and then incubated separately with specific NPY antibody (Santa Cruz Biotechnology, Santa Cruz, CA) or POMC antibody or CART antibody (Phoenix Pharmaceuticals) and β-actin (Sigma-Aldrich, St Louis, MO). After incubation with horseradish peroxidase goat anti-rabbit IgG and donkey anti-goat IgG, the color signal was develop by 4-chloro-1-napthol-3,3’ -diaminobezidine and 0.9% (wt/vol) NaCl in Tris-HCl (Sigma Chemical). Relative photographic density was quantified by scanning the photographic negiative film on a Gel Documentation and Analysis System (Alphalmager 2000; Alpha Innotech, San Leandro, CA).

5. Statistical analysis

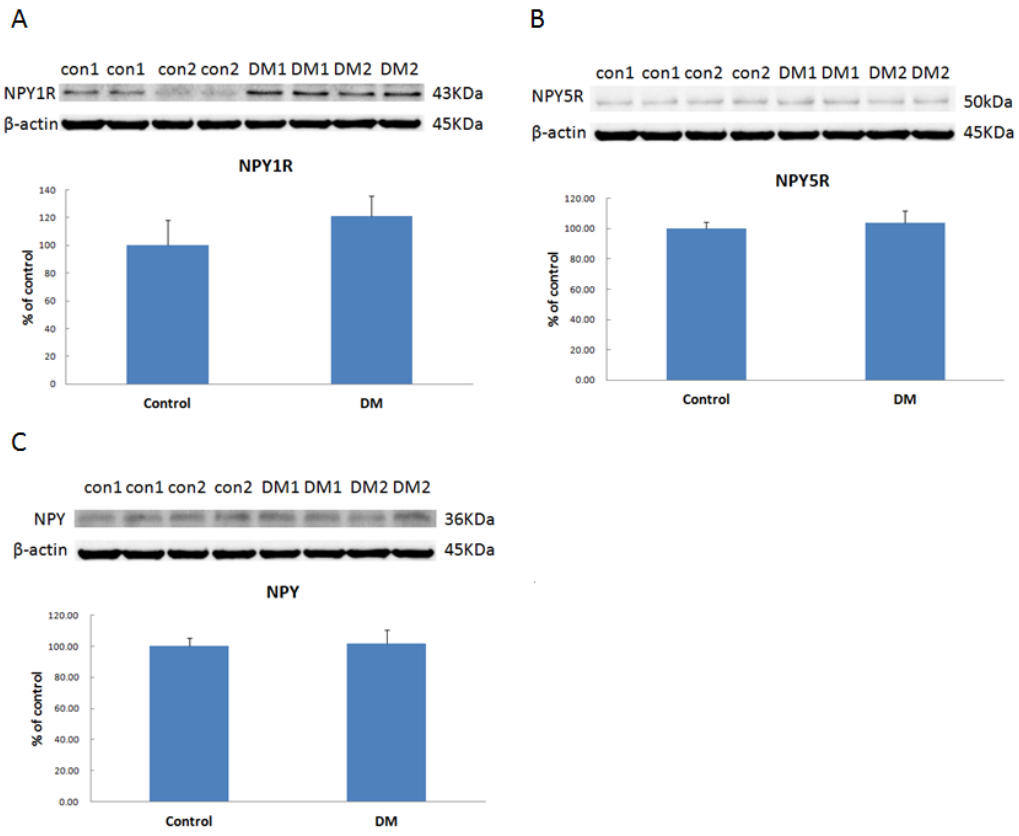
Statistical data were assessed by *t*-test, one-way or two-way ANOVA followed by post-hoc Dunnett’s test. $P<0.05$ was considered to be statistically significant.
Values are represented as mean±standard error (S.E.M.).

（五） 結果與討論

根據實驗結果分析刺激食慾的 NPY 神經胜肽及其接受器在控制組以及糖尿病組大鼠之間下視丘的蛋白的表現量差異發現：接受器 NPY1R 的表現量在雌性去卵巢糖尿病大鼠下視丘中相對於控制組的大鼠有明顯的增加，然而，NPY 以及其另一接受器 NPY5R 在雌性去卵巢的控制組及糖尿病組的大鼠之間的下視丘蛋白表現量並沒有顯著的差異（圖一）。此外，不管是 NPY 或是 NPY1R 在雄性的控制組大鼠、糖尿病組、糖尿病並接受胰島素(insulin)治療組和糖尿病並接受根皮苷(phlorizin)治療組的大鼠的下視丘蛋白表現量，四組之間都沒有顯著的差異（圖二）。另一方面，在抑制食慾的 CART 神經胜肽及其接受器在控制組以及糖尿病組大鼠之間下視丘的蛋白的表現量差異的實驗結果顯示：不論是 CART(55-102)、CART(61-102)、MC3-R 或是 MC4-R 在雌性去卵巢的控制組及糖尿病組的大鼠之間的下視丘蛋白表現量並沒有顯著的差異（圖三）。而在雄性的控制組大鼠、糖尿病組、糖尿病並接受胰島素治療組和糖尿病並接受根皮苷治療組的大鼠下視丘 CART(55-102)與 CART(61-102)的蛋白表現量，在四組間也是沒有顯著的差異（圖四）。

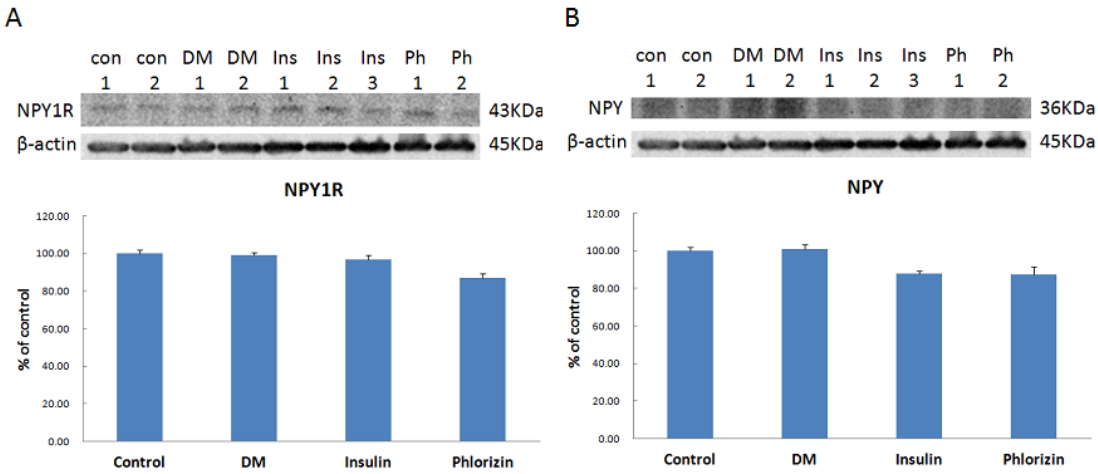
綜合以上結果，實驗透過 STZ 藥物誘導的糖尿病大鼠，在其食慾以及飲水量有顯著增加的情況之下，分析糖尿病大鼠下視丘與攝食相關的神經胜肽的蛋白表現量與控制組大鼠之間的差異發現只有在雌性去卵巢的糖尿病組大鼠的下視丘接受器 NPY1R 的蛋白表現量有顯著的增加情況，而在文獻中顯示，NPY 能夠透過與其接受器 NPY1R 的結合調控並刺激食慾*(Elbers et al., 2009)*。然而，其他的神經胜肽的蛋白表現量在控制組以及糖尿病組大鼠之間並沒有顯著的差異，因此推測影響糖尿病大鼠食慾增加的主要因素可能並不是因為食慾中樞下視丘影響攝食的神經胜肽蛋白表現量改變而造成的，而可能是其他更複雜的原因所導致。

（圖一）雌性去卵巢糖尿病大鼠的下視丘對 NPY 及其接受器的蛋白質表現量。



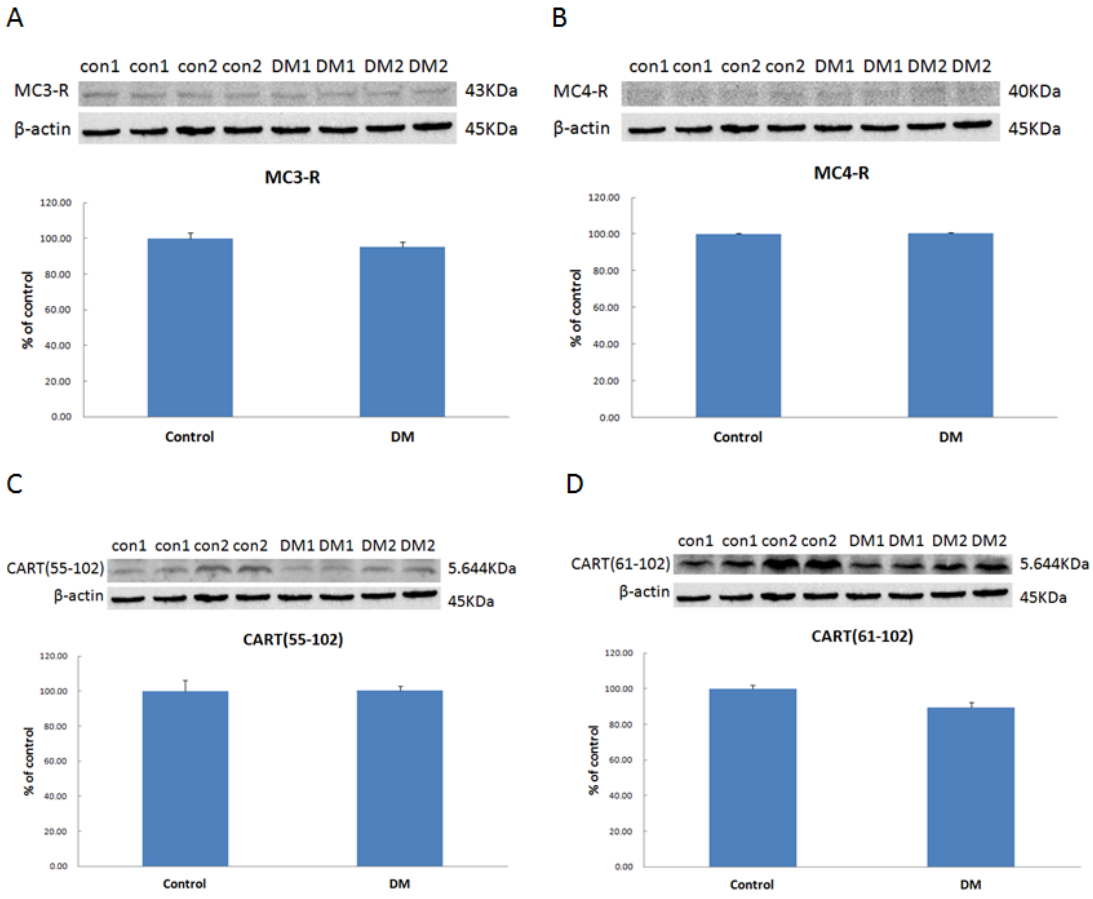
由圖表顯示，NPY1R 在雌性去卵巢糖尿病大鼠下視丘中的表現量相對於控制組的大鼠有明顯的增加，然而在 NPY 和 NPY5R 的表現量都沒有顯著的增加趨勢。(A)控制組與糖尿病組大鼠下視丘 NPY1R 蛋白表現量，(B)控制組與糖尿病組大鼠下視丘 NPY5R 蛋白表現量，(C)控制組與糖尿病組大鼠下視丘 NPY 蛋白表現量。（圖表顯示 mean±SEM）

（圖二）雄性大鼠的下視丘對 NPY 及其接受器 NPY1R 的蛋白質表現量。



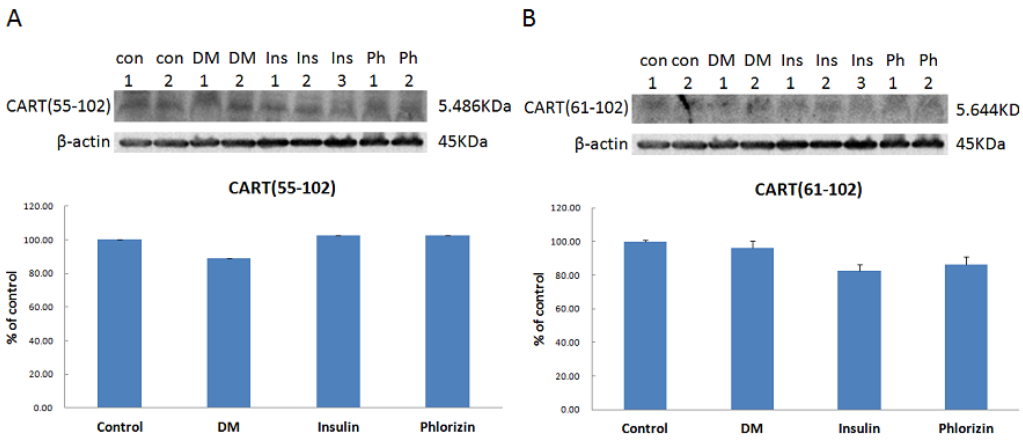
由圖表顯示，不管是在控制組、糖尿病組、糖尿病並接受胰島素(insulin)治療組以及糖尿病並接受根皮苷(phlorizin)治療組的大鼠，其下視丘 NPY 與其接受器 NPY1R 的蛋白表現量之間都沒有顯著的差異。(A)控制組、糖尿病組、糖尿病並接受胰島素治療組以及糖尿病並接受根皮苷治療組的大鼠大鼠下視丘 NPY1R 蛋白表現量，(B)控制組、糖尿病組、糖尿病並接受胰島素治療組以及糖尿病並接受根皮苷治療組大鼠下視丘 NPY 蛋白表現量。（圖表顯示 mean±SEM）

（圖三)雌性去卵巢糖尿病大鼠的下視丘對 CART 和其接受器的蛋白質表現量。



根據圖表顯示，不管是在控制組或是糖尿病組大鼠，其下視丘 CART(55-102)、CART(61-102)、MC3-R 和 MC4-R 的蛋白表現量之間都沒有顯著的差異。(A)控制組與糖尿病組大鼠下視丘 MC3-R 蛋白表現量，(B)控制組與糖尿病組大鼠下視丘 MC4-R 蛋白表現量，(C)控制組與糖尿病組大鼠下視丘 CART(55-102)蛋白表現量，(D)控制組與糖尿病組大鼠下視丘 CART(61-102)蛋白表現量。（圖表顯示 mean±SEM）

（圖四）雄性糖尿病大鼠的下視丘對 CART 的蛋白質表現量。



根據圖表顯示，不管是在控制組、糖尿病組、糖尿病並接受胰島素治療組以及糖尿病並接受根皮苷治療組的大鼠，其下視丘 CART(55-102)和 CART(61-102)的蛋白表現量之間都沒有顯著的差異。(A)控制組、糖尿病組、糖尿病並接受胰島素治療組以及糖尿病並接受根皮苷治療組的大鼠大鼠下視丘 CART(55-102)蛋白表現量，(B)控制組、糖尿病組、糖尿病並接受胰島素治療組以及糖尿病並接受根皮苷治療組大鼠下視丘 CART(61-102)蛋白表現量。（圖表顯示 mean±SEM）

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（七）　研究成果發表資料

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