Research Article

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Long-term Food Additive Monosodium Glutamate Induces the Differentiation of Pancreatic Adipose Tissue into newly Formed Islets of Langerhans

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Abstract

The aim of this study was to find the effect of long-term food additive monosodium glutamate (MSG) on the tissue of pancreas. For this purpose, two treated groups of adult wistar rats were administered 15 mg/kg body weight for 30 and 75 days respectively. Immediately after sacrificing, specimens of pancreas were collected and fixed. Routine histological processes were carried out to prepare the specimens for light microscopy. Hematoxylin and Eosin, and Gomori aldehyde fuchsin stains were used. The study confirmed that the pancreas had a histological compensatory defense mechanism. In terms of exocrine units, the size of islets of Langerhans were increased due to increase the size of alpha and beta cells and the engorgement of the surrounding blood vessels. The exocrine acini neighboring to islets of Langerhans were engorged with secretion. After 75 days, fusion of some preexisting islets among the exocrine units were recorded. The study firstly focused the light on the pancreatic adipose tissue differentiation to give rise a newly–formed islets of langerhans. The alpha and beta cells of the islets had a well cytological architecture with apparent network of functional blood vessels. Creation of a new generation of beta cells induced by appetizer, may regarded as an important approach in the treatment of type 1 diabetes mellitus. The study concluded that, in small animals, the endocrine mass of pancreatic islets proportionate with the need of insulin. Moreover, adipose tissue act as endocrine tissue and may secreting the hormone insulin. The study declared a positive relationship between exocrine and endocrine units, when the pancreas was subjected to external stressful factor.

Keywards: Rat, Pancreas, Adipose Tissue, Monosodium glutamate.

تحفيز النسيج الدهني للبنكرياس لإنتاج خلايا بيتا جديدة عن طريق الاستخدام طويل الأمد للملح الصيني على شكل جرعات

الخلاصة

إن الهدف من هذه الدراسة هو معرفة تأثير التجريع طويل الأمد للمضاف الغذائي أحادي صوديوم الكلوتاميت Monosodium glutamate على أنسجة البنكرياس. ولهذا الغرض تم تجريع مجموعتي المعاملة من الجرذان البالغة نوع wistar albino rat ب (15ملغم/كغم) من وزن الجسم (MSG) لمدة 30 و 75 يوما . أما مجموعة السيطرة فقد أعطيت ماء مقطر فقط. جمعت نماذج البنكرياس مباشرة بعد التضحية بحيوانات التجربة. ثبتت النماذج بالفورمالين الداريء المتعادل ثم أجريت عليها المعاملات النسيجية الروتينية لتهيئتها للفحص بالمجهر الضوئي. صبغت العينات بصبغتي الهيماتوكسلين والآيوسين وصبغة الكوموري الديهايد. أكدت الدراسة امتلاك أنسجة البنكرياس القابلية على الاستجابة الخلوية التعويضية. فعلى مستوى وحدات الإفراز الخارجي ازداد حجم جزر لنكرهانس الموجودة ضمن نسيج الإفراز الخارجي مع ازدياد أحجام خلايا ألفا وبيتا المكونة لها واحتقان الأوعية الدموية المرافقة لها. كذلك احتقنت وحدات الإفراز الخارجي المحاذية إلى جزر لنكرهانس بالإفرازات الأنزيمية. بعد مرور 75 يوما على تناول المضاف الغذائي تم تسجيل حالات اندماج بعض جزر لنكرهانس الموجودة أصلا ضمن وحدات الإفراز الخارجي فيما بينها. من ناحية ثانية على مستوى وحدات الإفراز الداخلي سجلت الدراسة لأول مرة نشوء جزر لنكرهانس حديثة التكوين فعالة ذات بناء خلوي متكامل محاطة بشبكة من الأوعية الدموية, نتيجة تخصص خلايا النسيج الدهني في الغالب إلى خلايا ألفا وبيتا. إن ذلك يزيد من دون شك مستوى إنتاج الأنسولين في الجسم ويساهم في النهاية في الاقتراب من علاج النوع الأول من السكري. تستنتج الدراسة أن الكتلة الخلوية الهرمونية الصماء في بنكرياس الحيوانات الصغيرة تتناسب مع الحاجة الى هرمون الأنسولين. أن نشاط النسيج الدهني في إفراز الأنسولين يضاف إلى مجموعة الهرمونات التي يسجلها الباحثون في السنوات العشر الأخيرة باتجاه ترشيح النسيج الدهني كغدة صماء. تستنتج الدراسة كذلك أن هناك علاقة إيجابية بين وحدات الإفراز الداخلي و الخارجي المحاذية لها فيما لو تعرض البنكرياس إلى عامل خارجي ضاغط.

Introduction

Adipocytes arise from mesenchymal stem cells (MSCs) by a sequential pathway of differentiation. MSCs develop either from ectoderm or mesoderm and commit into different undifferentiated precursors, which upon the expression of key transcription factors enter a differentiation program to acquire their specific functions. In mammals, the adipose tissue is composed of white adipocytes (primary site in energy storage) and of brown adipocytes (specialized in thermogenesis) (1).

Adipose tissue is formed at stereotypic times and locations in a diverse array of organisms. Once formed, the tissue is dynamic, responding to homeostatic and external cues. The formation and maintenance of adipose tissue is essential to many biological processes and when perturbed leads to significant diseases. Despite this basic and clinical significance, understanding of the developmental biology of adipose tissue has languished (2).

It appears that fat tissues evolved primarily as a safe harbor to store energy in times of plenty and to provide fuel when food sources become insufficient. However, in addition to serving as purely a storage depot, adipose tissue is now recognized as the body's largest endocrine organ, controlling many aspects of systemic physiology by secreting hormones (adipokines), lipids, cytokines and other factors (3, 4). MSG used extensively in the food preparation as flavor enhancer. A previous studies have been reported the effects of MSG when it given at a high dosages (5, 6), therefore, the present study was done. In 1959, the FDA classified the MSG as a "generally recognized as safe" substance. But in 1995, the FDA reported that many people may respond to MSG and progress the MSG signs complex, this condition is characterized by many symptoms includes: headache, vomiting, bronchospasm, rapid heartbeat, weakness, chest pain, drowsiness and sweating (7). In large animals, as well as humans, a proportionate increase in the pancreas size, islet number, and total islet mass, proportionate with an increased demand for insulin (8, 9).

Materials and method

Animals:

Twenty-four adult wistar male rats aged between (6-8) weeks old and weight between (190-250 g). Animals were housed in an individual cages during autumn months (September, October and November) and evaluated clinically by physical examination before initiation of experiment. Animals were provided with food and water ad libitium and maintained in the animal house of veterinary medicine collage\university of Baghdad. Animals were divided into three groups. First group (A) included eight (8) mature male rats served as a control group and supplied only by water. Second group (B) included eight (8) mature male rats that were given a daily oral dose of MSG (15mg/ kg /BW) for 30 days and then they were scarified. Third group (C) included eight (8) mature male rats that were given a daily oral dose of MSG (15mg/ kg /BW) for 75 days and then they were scarified (10).

Histological study:

Samples from pancreas were preserved in 10% Neutral buffered formalin for 72 hrs then the specimen were processed by routine histological processing method (11). H & E stain for routine tissue details, Gomori stain to differentiate between alpha and beta cells of pancreas (12). A Histomorphometric measurements Were done by aid of optica view7 image analysis software, Which is a professional image analysis software that perform a series of processing or measurements and incorporates with optica camera (13). The histomorphometric measurements of the pancreas include, diameter of the islet of Langerhans and nuclei of alpha and beta cells, Number of alpha & beta cells per islet of Langerhans, and Percentage of small and

large diameter of Langerhans. All data presented as mean ± standard error. The comparisons of the data were done between groups at the same age. The significance of the differences between means was estimated with one way ANOVA by using SPSS version 20 at level ($P<0.05$).

Results and discussion

The histological sections of pancreas of control group showing normal architectures of exocrine and endocrine portions of pancreas (Figure 1). The islet of Langerhans were positively respond to the MSG, Their size were increased as a result of increase diameter of alpha and beta cells (Table 1 and 2). Some Islets of Langerhans were fused with each other especially after 75 days in order to increase its efficiency (Figure 2). The exocrine acini were engorged with enzymatic secretions especially that surrounded the Islet of Langerhans with congestion of associated blood vessels. This phenomena indicates a positive relationship between exocrine and endocrine portion of pancreas (Figure 3).The study focused on the interstitial adipose tissue present between the exocrine pancreatic lobules and the incidents of cellular specialization that occurred in it. The interstitial adipose tissue of pancreas showed a process of differentiation into newly formed islet of Langerhans. The architecture of the beta cells were integrated building and highly vascularized. Each islet consists mainly of alpha and beta cells.

On the other hand, the study recorded firstly that the hormone insulin was excreted from the adipose tissue. This result was added a new hormone to the hormones previously reported by researchers in last decade (Figures 4, 5, 6, and 7).

The present results showed a significant increase in the diameter of islet of Langerhans in MSG treated groups. The increase in size of islet of Langerhans may be due to presence of MSG receptors on endocrine tissues (14). The Increased size of islets of Langerhans and hyperinsulinemia in hypothalamic obesity are well known (15). Additionally, the MSG treated group is characterized by central obesity and insulin resistance and these may lead to diabetes (16). Also, (17) showed that rats received 4 mg/kg body weight MSG via SC administration exhibited glucose intolerance, insulin resistance, and hypertriglyceridemia. The clarity of the cell membrane of the endocrine cells of the islet of the Langerhans were very weak, so that it was difficult to recognize the boundaries between alpha and beta cells and also difficult to detect the size of these cells. However, we could calculate the number of alpha and beta cells by calculate the number of their nuclei.

The current results showed a significant increase in the number of Alpha and Beta cells in MSG treated groups in comparison with control group. These results were in agreement with results of (18) who showed an increasing in number of beta cells. On the other hands, it has been reported that the number of alpha cells were increased due to MSG (19).

The present result referred to the process of differentiation of newly-formed pancreatic islets of Langerhans from the adipose tissue. The presence of different sizes of newly-formed islets indicates the steps of formation of these islets. Each islet consists mainly of alpha and beta cells. The study regarded these islets as functionally active as integrated in cytoarchitecture and function. Moreover, the presence of a network of blood vessels around these pancreatic islets confirms their functional and viability .

In large animals, including humans, a proportionate increase in the pancreas size, islet number, and total islet mass, proportionate with an increased demand for insulin (8, 9). The present result declared that small animals were included with this fact.

Conclusion

The study concluded that the newly formed Islet of Langerhans and their fusion with each other is

an advanced adaptive condition and undoubtedly increases the total amount of insulin that excreted and finally aid in the treatment of the type 1 diabetes. The current study concluded that there was an interaction between exocrine and endocrine units of the pancreas due to differences of the acini that surrounding the islet of Langerhans compared to the farther ones.

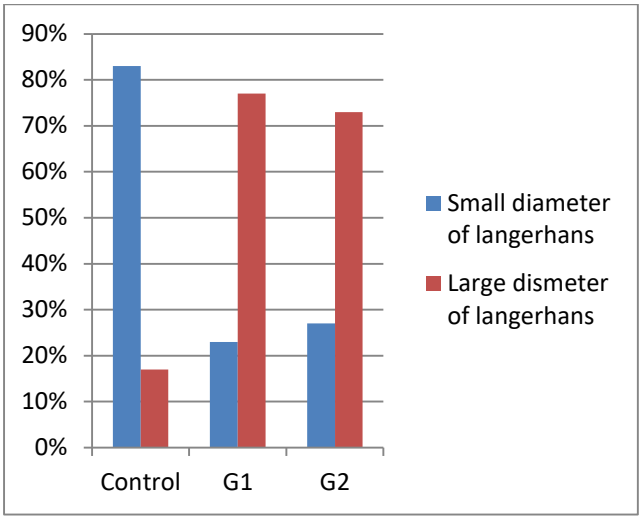

Diagram (1) showing the percentage of small and large diameter of Langerhans in control and MSG treated groups.

Table (1) Islet of Langerhans diameter in control and MSG treated groups

Groups	**Islet of Langerhans diameter Mean ± SE**
Control	90.4±1.14 B
G1	136.8±0.14 A
G2	152.3±1.57 A

Different letters denote significant ($p<0.05$) differences between groups

G1= MSG treated group after 30 days , G2= MSG treated group after 75 days

Table (2) number of alpha and beta cells and diameter of their nuclei

Groups	**No. of alpha cell Mean ± SE**	**Diameter of alpha cells Mean ± SE**	**No. of beta cells Mean ± SE**	**Diameter of beta cells Mean ± SE**
Control	5.46±0.96 B	1.18±0.004 AB	13.53±2.73 B	1.19±0.03 B
G1	11.53±0.49 A	1.21±0.054 A	25.63±1.91 A	1.28±0.02 AB
G2	12.36± 1.12 A	1.08±0.021 B	24.53±2.81 A	1.33±0.02 A

Different letters denote significant ($p<0.05$) differences between groups
G1= MSG treated group after 30 days , G2= MSG treated group after 75 day

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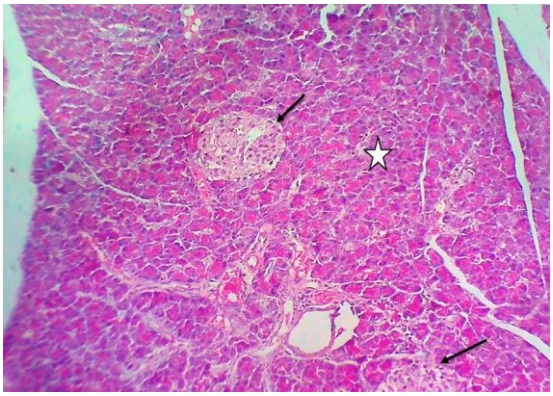

Fig.1 Pancreas of wistar rat (control). Arrows refer to two endocrine islets of Langerhans embedded among the pancreatic exocrine acini (star). Hematoxylin and Eosin. X200.

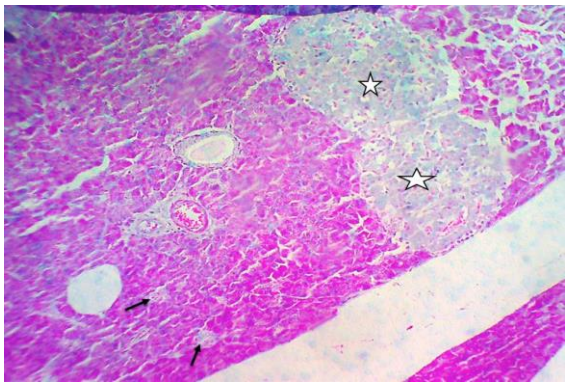

Fig.2. Histological section of pancreas in MSG treated group after 75 days showing hyperplasia of cells in two fused islets of Langerhans (stars). The area was congested with blood vessels. Note the newly-formed blood vessels (angiogenesis) (arrows) near the large pre-existing blood vessel. H&E stain (100x).

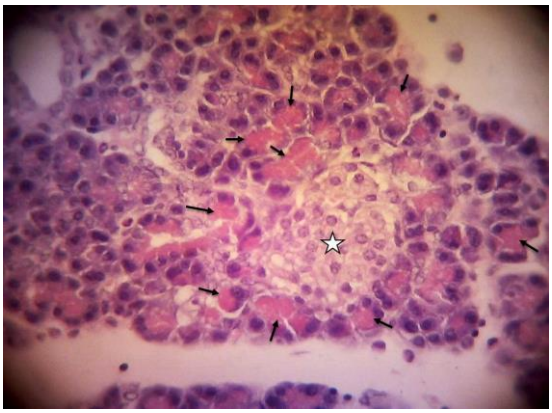

Fig.3. Pancreatic islet of langerhans after 75 days of MSG-treated group (star) surrounded by activated acini engorged with secretion (white arrows). Hematoxylin and Eosin stain. X 400.

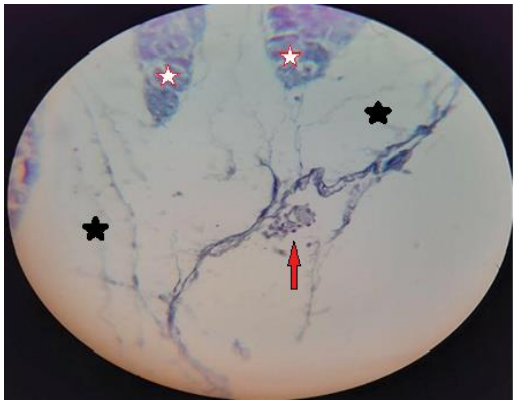

Fig.4. The onset of differentiation of pancreatic adipose tissue into endocrine islet of Langerhans (red arrow). Black stars refer to adipose tissue. white stars indicate two exocrine pancreatic acini. Gomori stain. X 200.

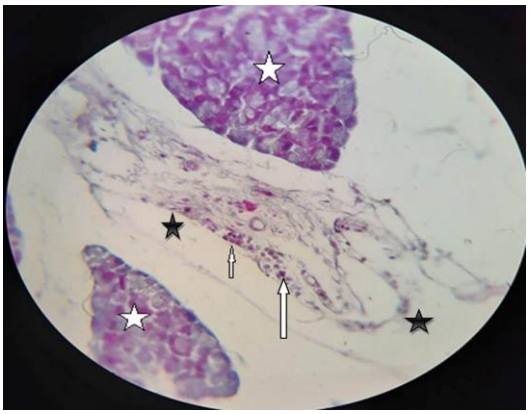

Fig.5. Advanced stage of differentiation of pancreatic adipose tissue (black star). White stars refer to the exocrine pancreatic acini. Note the red alpha cells (small arrow) and the blue beta cells (large arrow). Alpha and beta cells were surrounded by small size blood vessels. Gomori stain. X200.

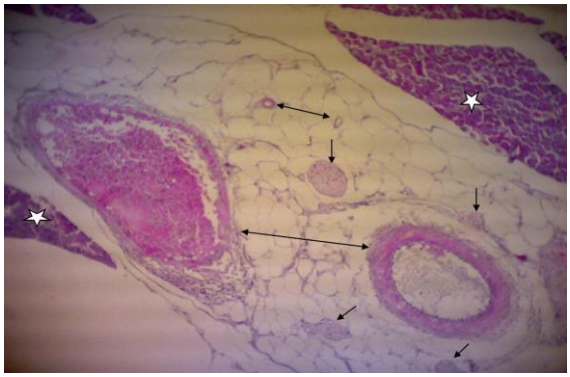

Fig.6. One-head Arrows refer to large and small newly-formed endocrine islets of Langerhans embedded within adipose tissue. Note the different sizes of blood vessels near the islets (two-headed arrows). White stars refer to exocrine acini of pancreas. Hematoxylin and Eosin stain.X200.

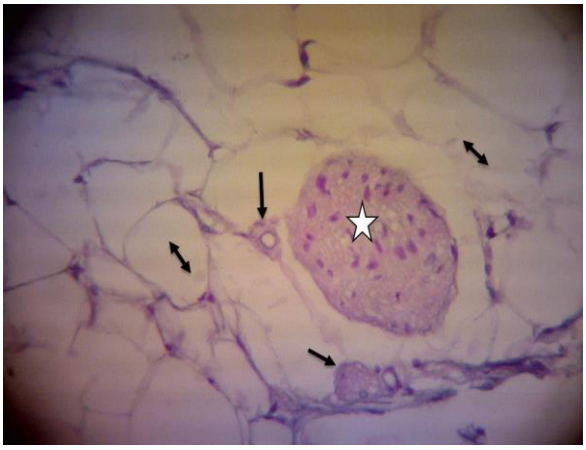

Fig.7. White star refers to Large functional islet of Langerhans packed mostly with alpha and beta cells. Arrow refer to smaller newly-formed two islets. All islets were embedded in adipose tissue (two-headed arrows). Hematoxylin and Eosin stain. X400.

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