REVISTA ESPAÑOLA DE FISIOLOGIA, 44 (1), 31-34, 1988

Serum Androgen Levels after Streptozotocin Administration in the Male Rat

I. Centol, F. López-Blanco, A. Deniz, L. Benítez, I. Marrero-Arencibia, L. Fanjul and C. M. Ruiz de Galarreta*

Departamento de Bioquímica y Biología Molecular
Universidad de La Laguna
Colegio Universitario de Las Palmas
35016 Las Palmas de Gran Canaria (Spain)

(Received on June 8, 1987)

I. CENTOL, F. LOPEZ-BLANCO, A. DENIZ, L. BENITEZ, I. MARRERO-ARENCIBIA, L. FANJUL and C. M. RUIZ DE GALARRETA. *Serum Androgen Levels after Streptozotocin Administration in the Male Rat.* Rev. esp. Fisiol., 44 (1), 31-34, 1988.

Serum levels of testosterone and dihydrotestosterone were measured in control and diabetic animals 5, 10 and 15 days after streptozotocin administration. The diabetic state produced a marked reduction in serum androgen levels 10 and 15 days after streptozotocin administration. Insulin treatment partially restored the circulating androgen levels when administered to diabetic rats.

Key words: Streptozotocin, Testosterone, Dihydrotestosterone.

The diabetic state is accompanied by reproductive disturbances in the male rat (2, 5, 10). Some of the effects of experimental diabetes include reduced testicular weight (6, 8), accessory sex gland atrophy (8) and degenerative changes in seminiferous tubules (7, 10). It has been postulated that diabetes results in low gondotrophin levels as well as altered endocrine function of the testes (8, 9, 12, 13). Insulin has been reported to partially reverse the gonadal alterations of diabetic rats (3, 12, 16). The present investigation aimed to determine whether the experimental period might influence serum androgen levels in streptozotocin diabetic rats and whether insulin treatment could restore them to control levels.

Materials and Methods

Male Sprague-Dawley rats from our breeding colony (250-300 g BW) were housed in a temperature (24 ± 1° C) and light cycle controlled room (12 h light/12 h dark) with tap water and standard pelleted diet available *ad libitum*. Diabetes was induced by a single i.p. dose (40 mg/kg BW) of streptozotocin (Sigma) freshly prepared in citrate buffered saline, pH = 4.5, with control rats receiving vehicle alone. The following experimental

* To whom correspondence should be addressed.

groups (n = 5 animals per group) were studied: 1) Controls, 2) Diabetics, and 3) Diabetics treated daily with insulin (Novo Lente), 2 IU/day s.c. Treatment was begun 1 day after the induction of diabetes, with the other groups receiving similar volumes of saline/day. Blood samples were obtained by orbital sinus puncture under light ether anesthesia just before streptozotocin or vehicle administration day on days 5th, 10th and 15th thereafter. Blood was allowed to clot at room temperature, and stored as serum at −20° C until assayed for hormones. Serum glucose was assayed by a standard GOD method (Boehringer Mannheim). The criteria used to assess the severity of the diabetic state included the daily measurement of glucosuria, polydipsia, polyphagia and ketonuria. Testosterone (T) and dihydrotestosterone (DHT) were determined in triplicate by radioimmunoassay using a commercial kit (The Radiochemical Centre). T and DHT were separated by column chromatography (1).

Results were compared by analysis of variance (ANOVA) and the least significant difference (LSD) method (15).

Results and Discussion

The diabetic state was characterized by polydipsia, polyuria, glucosuria and ketonuria (results not shown), high serum glucose levels and body weight loss (fig. 1), despite the marked polyphagia observed (results not shown). Insulin treatment partially prevented weight loss and reduced serum glucose levels (fig. 1). Serum androgen levels showed a time dependent decline after streptozotocin administration while insulin treatment significantly increased T and DHT levels in diabetic animals (fig. 2).

Although the significance of the findings obtained after streptozotocin administration are difficult to apply to spontaneous diabetes mellitus, KÜHN-VELTEN *et al.* (11) recently reported that high streptozotocin concentrations *in vitro* had no effect on basal and LH stimulated steroidogenesis of Leydig cells from normoglycemic rats. These data suggest that alterations in the testicular function of the

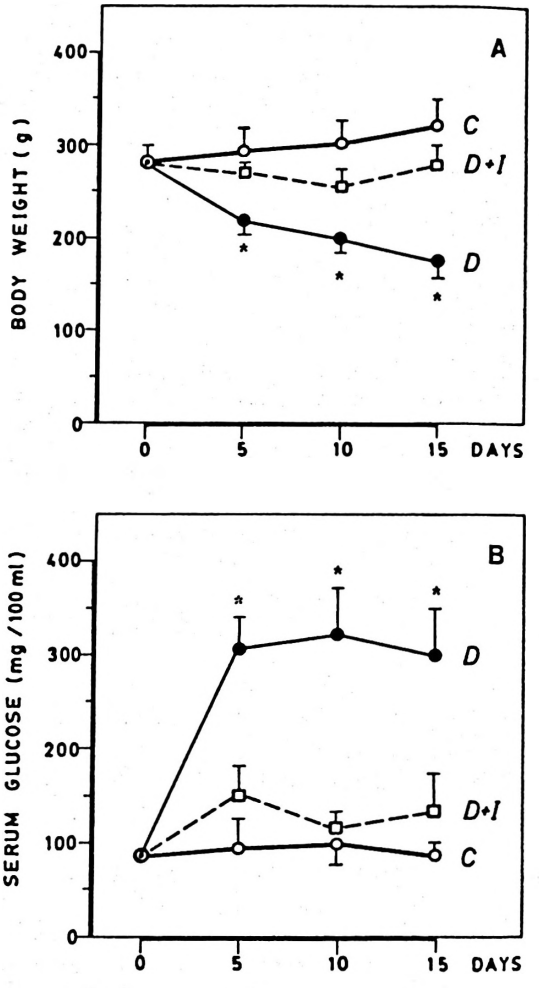

Fig. 1. *Changes in body weight (A) and serum glucose (B) of control (—○—), diabetic (—●—) and diabetic treated with insulin (—□—) animals over the experimental period.*
Diabetes was induced on day O, immediately after the first blood sampling. Values given represent the mean ± S.E.M. (vertical bars), (n = 5 animals per group). * $p < 0.05$ versus controls.

streptozotocin diabetic rat are not due to a toxic effect of the drug.

Alterations in the testicular function of diabetic rats have been described (3, 8, 10) and related with low circulating levels of gonadotrophic hormones (8, 9). These alterations included a reduced Leydig cell population (13, 16), a reduced hCG/LH receptor number (4) as well as reduced activity of different stroidogenic (12, 16) and NADPH generating enzymes in Leydig cells from diabetic rats (3). Most of these alterations, however, have been described at three or more weeks after streptozotocin administration, and therefore little is known about the development of the gonadal failure after the induction of the diabetic state. In human patients the age as well as the duration of the disease seems to influence the reproductive failure of the male (14).

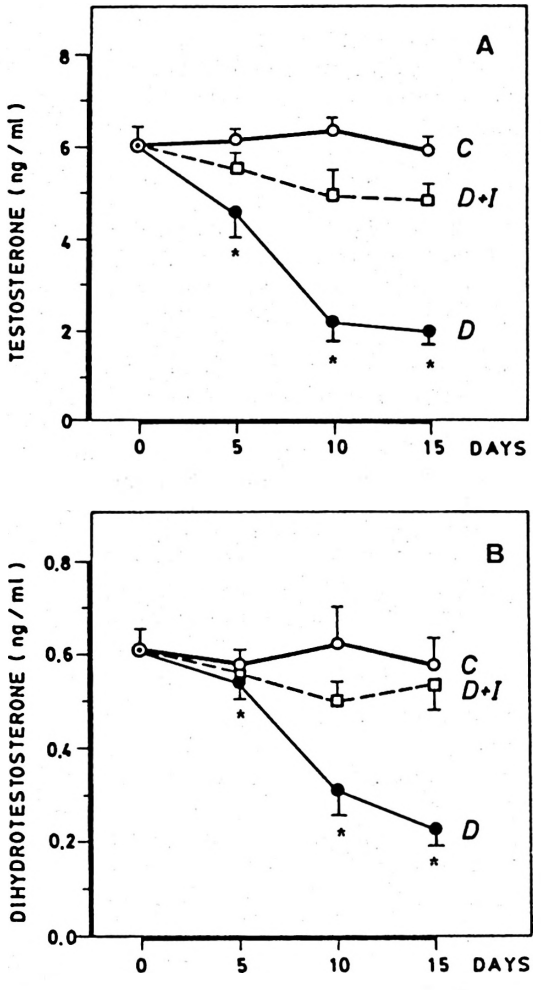

Fig. 2. *Changes in serum testosterone (A) and dihydrotestosterone (B) of control (—O—), diabetic (—●—) and diabetic treated with insulin (—□—) animals over the experimental period.*
Diabetes was induced on day O, immediately after the first blood sampling. Values given represent the mean ± S.E.M. (verticals bars), (n = 5 animals per group). * $p < 0.05$ versus controls.

The present investigation was undertaken in order to elucidate if there is a causal relationship between the gonadal alterations observed in the diabetic rats and the experimental period. Our data show a slight reduction in circulating levels of T as early as 5 days after streptozotocin administration followed by a clear cut reduction in serum levels of T and DHT in rats rendered diabetic 10 and 15 days. In addition, insulin treatment increased serum androgen levels of diabetic rats, 5,10 and 15 days after streptozotocin administration. Similar effects of Insulin on serum T have been reported in rats with 8-15 day long diabetes (3) but not in longer experimental periods (13).

Our data, suggests that at least 10 days after the induction of the diabetic state, one or more of the alterations in the pituitary-testicular axis of the rat (3, 8, 10, 12) could be detected. Whether early alterations are primarily due to a reduced gonadotrophin secretion (8, 9) or to a direct consequence of insulin defficiency at the Leydig cell level needs further investigation.

Acknowledgements

These studies were supported in part by grant n.° 67/85 from the «Dirección General de Universidades e Investigaciones del Gobierno Autónomo de Canarias» (Spain).

Resumen

Se miden los niveles séricos de testosterona y de dihidrotestosterona en ratas control y diabéticas 5, 10 y 15 días después de la administración de estreptozotocina. Los resultados muestran una clara disminución de los niveles circulantes de andrógenos 10 y 15 días después de la administración de estreptozotocina. El tratamiento con insulina a ratas diabéticas incrementa los niveles circulantes de andrógenos.

Palabras clave: Estreptozotocina, Testosterona, Dihidrotestosterona.

References

1. Abraham, G. E., Baster, J. E., Lucas, A., Corrales, P. C. and Telle, R. C.: *Anal. Lett.*, 5, 509-514, 1977.
2. Babbot, D., Rubin, A. and Grinsburg, S. J.: *Diabetes*, 7, 33-39, 1958.
3. Calvo, J. C., Biella de Souza Valle, L., Barañao, J. L. and Charreau, E. H.: *Horm. Metab. Res.*, 10, 161-164, 1975.
4. Charreau, E. H., Calvo, J. C., Tesone, M., Biella de Souza Valle, L. and Barañao, J. L.: *J. Biol. Chem.*, 253, 2504-2506, 1977.
5. Federlin, K., Schöfling, K., Neubronner, P. and Pfeiffer, G. F.: *Diabetologia*, 1, 85-89, 1965.
6. Foglia, V. G., Borghelli, R. F., Chierri, R. A., Fernández Collazo, E. L., Spindler, I. and Wesley, O.: *Diabetes*, 12, 231-233, 1963.
7. Foglia, V. G., Rossner, J. M., Cattaneo de Peralta Ramos, M. and Lema, B.: *Horm. Metab. Res.*, 1, 72-77, 1969.
8. Howland, B. E. and Zebrowski, E. J.: *Horm. Metab. Res.*, 8, 465-469, 1976.
9. Howland, B. E. and Zebrowski, E. J.: *Experientia*, 36, 610-611, 1981.
10. Irisawa, S., Shirai, M., Matsushita, S., Kagayamu, M. and Ichigo, S.: *Tohoku J. Exp. Med.*, 88, 311-317, 1966.
11. Kühn-Velten, N., Waldenburger, D. and Staib, W.: *Diabetologia*, 23, 529-533, 1982.
12. López-Blanco, F., Fanjul, L. and Ruiz de Galarreta, C. M.: *Endocrinology*, 109, 1248-1253, 1981.
13. Paz, G. and Hommonai, Z. T.: *Experientia*, 35, 1412-1414, 1979.
14. Rubin, A. and Babbott, D.: *J. A. M. A.*, 4, 498-499, 1958.
15. Sachs, L.: «Statistishe Auswertungsmethoden», Springer-Verlag, Berlin, 1978.
16. Tesone, M., Biella de Souza Valle, L., Foglia, V. G. and Charreau, E. H.: *Acta Physiol. Latinoam.*, 26, 387-394, 1976.