Vascular Wall and Endothelium

Editores
J. Martins e Silva
Carlota Saldanha

2008
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Editors
J. Martins e Silva
Carlota Saldanha

Editorial Office
Institute of Chemical Biopathology,
Faculty of Medicine, University of Lisbon
Av. Prof. Egas Moniz
Lisboa – Portugal

Mailing Address
Actas de Bioquímica
Apartado 4098
1500-001 Lisboa – Portugal

Subscription Information
Subscription price is $25.00 (twenty five US dollars) or €25,00 (twenty five euros) per volume. An additional charge of €5,00 (five US dollars) per volume is requested for post delivery outside Portugal. Payment should accompany all orders. Correspondence concerning subscription should be addressed to the mailing address above.

ISBN: 972-590-076-6
VASCULAR WALL AND ENDOTHELIUM

LISBON (PORTUGAL),
SEPTEMBER 14, 2007

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Fundação Merck Sharp e Dohme
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IV
VASCULAR WALL AND ENDOTHELium

Proceedings of the Symposium on Vascular Wall and Endothelium
(7th Advanced Course on Applied Biochemistry)
Held in Lisbon, September 14, 2007.
Organized by the Institute of Biopatologia Química, Faculdade de Medicina,
and Unidade de Biopatologia Vascular do Instituto de Medicina Molecular,
Universidade de Lisboa

Editors
J. Martins e Silva
Carlota Saldanha

Published by
Instituto de Biopatologia Química
Faculdade de Medicina
Universidade de Lisboa
Agradecimentos à Fundação Merck Sharp e Dhome pelo apoio financeiro concedido à realização e publicação dos textos do 7.º Curso Avançado de Bioquímica Aplicada.

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Printed in Portugal
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HAEMORHEOLOGICAL CHANGES DURING RECOMBINANT HUMAN ERYTHROPOIETIN THERAPY IN A RAT MODEL OF RENAL FAILURE INDUCED BY PARTIAL NEPHRECTOMY

Elisio Costa1,2, Flávio Reis4, Petronila Rocha-Pereira4, Sofia Baptista4, André Dias4, Susana Rocha1,2, Elisabeth Castro1,2, Vasco Miranda6, Maria Sameiro Faria6, Alfredo Loureiro7, Edite Teixeira de Lemos4, Belmiro Parada8, Arnaldo Figueiredo9, Alexandre Quintanilha2,9, Frederico Teixeira4, Luis Belo1,2, Alice Santos-Silva1,2

1 Faculdade Farmácia, Serviço de Bioquímica, Universidade do Porto; 2 Instituto Biologia Molecular e Celular, Universidade do Porto; 3 Escola Superior de Saúde, Instituto Politécnico de Bragança; 4 Instituto de Farmacologia e Terapêutica Experimental, Faculdade Medicina, Universidade Coimbra; 5 Centro Investigação Ciências Saúde, Universidade Beira Interior, Covilhã; 6 FMC, Dinefro – Diálises e Nefrologia, SA.; 7 Uninefro – Sociedade Prestadora de cuidados Médicos e de Diálise, SA.; 8 Departamento de Urologia e Transplantação Renal, Hospitais Universidade de Coimbra; 9 Instituto Ciências Biomédicas Abel Salazar, Universidade do Porto

ABSTRACT

The aim of this work was to study the effect of recombinant human erythropoietin (rhEPO) therapy on haemorheological parameters, by using a rat model of chronic renal failure (CRF) induced by partial (3/4) nephrectomy.

The study used adult male Wistar rats and was performed in three groups: a control one (n=6) and in two groups with induced chronic renal failure (n=9), being one of them submitted to rhEPO therapy (n=4). Blood samples from the control group were collected at the beginning and at the end of the experimental procedure and from CRF rats at 3, 5, 8, 12 and 15 weeks after surgical partial nephrectomy. Haemorheology and renal function were evaluated.

Three weeks after the 3/4 nephrectomy, a statistically significant increase in serum urea and creatinine concentrations were found. This increase in renal function markers remained high along the 12 weeks of experimental procedure.

Comparing with controls, rhEPO treated rat have showed a statistically significant progressive increase in haemoglobin (Hb), haematocrit (Ht), red blood cells (RBC) count, mean cell volume
Partial nephrectomy seems to be a suitable methodology to induce CRF in rats and to study erythropoiesis biology. The rhEPO therapy is associated with an increased erythropoietic stimulation and a decrease in platelet count.

Key-Words: rhEPO, Erythropoietin, renal failure, rat model, Haemorheology.

INTRODUCTION

Erythropoietin (EPO) is a glycoprotein hormone produced by kidneys, which regulates the proliferation, differentiation and maturation of erythroid cells. Chronic renal failure (CRF) patients develop anaemia due, mainly, to the low production of EPO by kidneys. To treat this anaemia, recombinant human EPO (rhEPO) therapy is currently used in these patients1-3.

The aim of this work was to study the effect of rhEPO therapy on haemorheological parameters, by using a rat model of CRF induced by partial (3/4) nephrectomy.

MATERIALS AND METHODS

Animals

Male Wistar rats (Charles River Lab. Inc.), 380-400g, were maintained in an air conditioned room, subjected to 12-h dark/light cycles and given standard laboratory rat chow (IPM-R20, Letcia) and free access to tap water.

Blood samples and Assays

Blood samples from the control group were collected at the beginning and at the end of the experimental procedure, and from CRF rats at 3, 5, 8, 12 and 15 weeks after surgical partial nephrectomy.

In all animals (controls and CRF) renal function was evaluated by determination of serum levels of urea and creatinine. Red blood cells (RBC) count, haema-
tocrit (Ht), haemoglobin concentration (Hb), haematimetric indices [mean cell volume (MCV) and mean cell Hb (MCH)], red cell distribution width (RDW), white blood cells (WBC) and reticulocyte counts were also evaluated by using a blood cell counter.

**Data Analysis**

For statistical analysis, we used the Statistical Package for Social Sciences, version 14.0. Results are presented as means ± SEM. Comparisons between groups at different times were performed using one-way ANOVA and Fisher’s tests. Significance was accepted at p less than 0.05.

**RESULTS**

Three weeks after the 3/4 nephrectomy, a statistically significant increase in serum urea (71.00 ± 2.66 vs 41.00 ± 0.68 mg/dL, p<0.05) and creatinine (0.828 ± 0.036 vs 0.412 ± 0.019 mg/dL, p<0.05) concentrations were found. This increase in renal function markers remained high along the 12 weeks of experimental procedure (Fig. 1).

Comparing with controls, rhEPO treated rats showed a statistically significant progressive increase in Hb, Ht, RBC count, MCV, MCH and RDW, showing at 12 weeks an inverse change, though still presenting significantly higher values; a decrease in platelet count, during the first 9 weeks of rhEPO therapy. When comparing haemorheological data from non-treated CRF and controls, we found only a trend to increased MCV and MCH values and a decrease in percentage of reticulocyte count. Comparing the two groups of CRF rats, we found that rhEPO treated rats presented significantly higher values in RBC count, Hb, Ht and RDW. In both groups of CRF rats, at five weeks, there was a decrease in their values, showing at the end a significantly lower value when compared to controls. No consistent alterations were found in white blood cells in CRF rats, with or without rhEPO therapy.

**CONCLUSION**

Partial nephrectomy seems to be a suitable methodology to induce CRF in rats and to study erythropoiesis biology. The rhEPO therapy is associated with an increased erythropoietic stimulation (increase Hb, Ht, RBC count and RDW) and a decrease in platelet count. This method might be useful used to study the cellular and molecular underlying EPO resistance.

![Fig. 1](image-url) - Renal function markers (serum urea and creatinine levels) along the 12 weeks of experimental procedure.

- **Controls**
- **Non-treated CRF rats**
- **rhEPO treated**

| a | p < 0.05, aa: p<0.01, aaa: p<0.001, non-treated CRF rats vs controls; b: p < 0.05, bb: p<0.01, bbb: p<0.001, T1 vs T0; T2 vs T1, etc.; e: p < 0.05, ee: p<0.01, eee: p<0.001, non-treated CRF rats vs rhEPO treated rats. |
ACKNOWLEDGEMENTS

We are very grateful to Roche Pharmaceuticals to provide the rhEPO used in this work. This study was supported by a PhD grant (SFRH/BD/27688/2006) attributed to E. Costa by FCT and FSE.

BIBLIOGRAPHY


