Reviewer's report

Title: Antiglucocorticoid RU38486 inhibits reduction of protein synthesis due to stress, but not uremia-specific net protein catabolism in experimental acute renal failure

Version: 1 Date: 12 October 2004

Reviewer: Alan Bevington

Reviewer's report:

General
This study describes experiments on the role of glucocorticoid in the protein wasting illness that occurs during acute renal failure. This is an important subject for research, but I have a number of serious reservations about the way that this study has been designed and performed.

Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

1) The animal model. In the untreated BNX group, the animals were subjected to total bilateral nephrectomy followed by 48h of starvation and then a period of water restriction (to limit pulmonary oedema). The author should justify this model and explain in what clinical situation such extreme conditions would occur in humans. The author should also explain why it was necessary to apply two catabolic stimuli (starvation and uraemia) simultaneously.

2) The title of the paper refers to protein synthesis and uremia-specific net protein catabolism but, as far as I can see, protein synthesis rate was not measured directly in this study, nor is it clear how the author has distinguished between the uremia-specific and starvation-induced or stress-induced contributions to the measured net protein catabolism. The basis of these distinctions needs to be explained more clearly.

3) Most of the measurements in this study concern the release of amino acids into the extracellular compartment of the perfusion medium. In the case of 3-methyl-histidine release, this can be interpreted, as this amino acid is only released from myofibrillar protein and is not re-utilised, so it is an accurate index of myofibrillar protein degradation. The release of the other amino acids however is more difficult to interpret because they can be re-utilised in cell metabolism and, in the case of the non-essential amino acids, can be synthesised in the cells. Consequently their rate of appearance in the extracellular compartment will show a complex dependence on protein synthesis rate, protein degradation rate, amino acid synthesis and catabolism rates, and also on the rate of transport across the plasma membrane. Several of these factors (including amino acid transport rate) can be affected by glucocorticoid in the hind limb perfusion model (Hundal HS. Babij P. Taylor PM. Watt PW. Rennie MJ. Effects of corticosteroid on the transport and metabolism of glutamine in rat skeletal muscle. Biochimica et Biophysica Acta. 1092(3):376-83, 1991). The author should therefore provide evidence or cite references to show how such amino acid data are to be interpreted. For example, was protein synthesis blocked with cycloheximide during the perfusion to eliminate the contribution from protein synthesis?

4) To maintain efficient oxygenation during the perfusion, calf erythrocytes were used in the perfusion medium. The author should state (or estimate from data in the literature) the contribution of erythrocyte amino acid efflux to the extracellular amino acid accumulation during the 60 min recirculation perfusions. Has it been shown previously to be negligible?
5) The dose of RU38486 used in this study is unclear. In the abstract it is stated to be 25 mg/kg body weight/day, whereas elsewhere in the paper it is stated to be 50 mg/kg body weight/day. This is important because RU38486 is difficult to dissolve in water and consequently it is difficult to administer sufficient to rats to achieve efficient blockade of the glucocorticoid receptors. The author should present or cite evidence that the receptors are effectively blocked under the conditions used in this study. For example, even at a dose of 50 mg/kg body weight/day given to rats by gavage, only 80% of the receptors were blocked (Pickering WP, Baker FE, Brown J, Butler HL, Govindji S, Parsons JM, Pawluczyk IZ, Walls J, Bevington A. Glucocorticoid antagonist RU38486 fails to block acid-induced muscle wasting in vivo or in vitro. Nephrology Dialysis Transplantation. 18(8):1475-84, 2003).

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Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

6) Even though many interesting and relevant references are cited, the reference list is out of date, with nothing cited after 2001, and most of the references are before 1992. More recent work in this field should also be cited.

7) The components of the perfusion diagram in Fig 1 should be labelled in more detail.

8) Error bars and statistical analysis should be shown on Fig 2.

9) The Conclusion refers to "non-operated animals" which were not included in the study. This should be removed.

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Discretionary Revisions (which the author can choose to ignore)

10) I suggest that the second sentence of the Abstract (Background) should be altered to ".....the question to what extent does corticoid action specific to uremia cause the observed muscle degradation, and does inhibition of glucocorticoid action reduce the protein wasting?"

What next?: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

Level of interest: An article whose findings are important to those with closely related research interests

Quality of written English: Needs some language corrections before being published

Statistical review: No

Declaration of competing interests: None