Reviewer's report

Title: Metabolic and morphological alterations induced by proteolysis-inducing factor from Walker tumour in C2C12 myotubes.

Version: 2 Date: 29 November 2007

Reviewer: Kent KL Lundholm

Reviewer's report:

General

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

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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)

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Discretionary Revisions (which the author can choose to ignore)

This manuscript is improved following revision. It is more clearly described but is still unfocused in Introduction and Discussion. However, a main problem remains namely to evaluate whether WF is produced by tumour cells exclusively or by host cells predominantly or a combination of both. It may well be that WF is not tumour specific to any extent but present in all replicating or stressed non-malignant cells. The authors touch upon this dilemma by emphasizing that WF was not determined in tumour tissue. Despite this fact, the authors found it likely to be produced by the Walker-256 tumour cells without providing any information or data on this. Accordingly, it was interesting to know that the original PIF core-protein is also expressed in non-tumour cells obviously assessed in patients with oesophageal carcinoma.

There is no doubt that the myotubes are negatively affected by the biological material present in the ascites preparation. A matter of question is whether this is mainly unspecific reactions due to toxic material which may well be a glycosylated protein from either tumour cells or host cells. Therefore, some control experiments must be performed to exclude this possibility. The most appropriate approach would be to have non-malignant ascites secondary to some irrigating agents. An alternative could be to culture Walker-256 and non-transformed fibroblast for control preparations from the cell culture medium. Another concern to this reviewer is what high concentrations of ammoniumsulphate may mean concerning negative effects on subsequent cell cultures. Our own experience is that even small amounts of associated ions may introduce negative effects. Therefore, it is vital to the validity of this study to
perform some kind of relevant control preparation(s).

**What next?:** Reject because scientifically unsound

**Level of interest:** An article of limited interest

**Quality of written English:** Acceptable

**Statistical review:** No, the manuscript does not need to be seen by a statistician.