Reviewer’s report

Title: Further Evidence for Increased Macrophage Migration Inhibitory Factor Expression in Prostate Cancer

Version: 4 Date: 1 June 2005

Reviewer: Robert A Mitchell

Reviewer’s report:

The manuscript by Meyer-Siegler et al. extends their prior observation that MIF serum levels are increased in patients with CaP while proposing an explanation as to why a recent manuscript contradicting their earlier work achieved conflicting results. The novelty of this manuscript lies in the fact that the discrepant results between the authors earlier studies and this conflicting report are due to the use of different reagents for an otherwise identical ELISA protocol. The authors present convincing data that the ELISA diluent used in the conflicting study interferes with MIF epitopes and ultimately with serum ELISA results from both control and CaP patient sera. Other studies are presented that are not necessarily novel but support the general conclusion that MIF mRNA and protein is overexpressed in prostate adenocarcinoma tissue and cell lines. Despite these plausible findings and a very good discussion of the results and implications, there are a couple of concerns that need to be addressed:

Major Compulsory Revisions

1. The data shown in Figure 3 do not support the stated conclusions that, 1) MIF exists in a high molecular weight complex that is not recognized by the MIF capture antibody or, 2) that the MIF ELISA detects less than 1% of total serum MIF. The authors need to demonstrate that the immuno-reactive bands observed in the non-sticking protein lanes are not simply due to non-specific cross-reactivity with the detection antibody (or secondary). Simply adding a blocking peptide or full length recombinant MIF to the detection antibody during the western blot should answer this question. Do the authors predict that a reducing gel would resolve this complex into the MIF monomer? Further, would the addition of a weak detergent (and/or reductant) to the ELISA diluent resolve this complex and result in values that are 100 fold higher?

2. In the first paragraph of the Discussion section, the authors suggest that chronic inflammation often precedes epithelial injury and subsequent prostate carcinogenesis. They go on to propose that MIF may be an important mediator of this process and that the present findings confirm our hypothesis. The findings presented in this manuscript do not support this statement and should be clarified to reflect this. There are no functional studies presented in this manuscript that suggest that MIF participates in any aspect of prostate carcinogenesis.

Minor Essential Revisions

3. Fig. 4 should read 40x and not 400x magnification

4. Despite their contention that BSA as a diluent may interfere with the MIF ELISA because of prior studies reporting MIF binds to human serum albumin (aka, sarcolectin), the authors should note that these same studies reported that bovine serum albumin does not bind to MIF even at high concentrations.

Discretionary Revisions (which the author can choose to ignore)
**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:** Acceptable

**Statistical review:** No

**Declaration of competing interests:**

I declare that I have no competing interests.