

## **PROJECT REVIEW**

### ***"Investigation of in vitro/ex vivo TB-500 metabolism, synthesis of relevant metabolites and detection limits in urine and plasma"***

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In the recent years, the use of performance-enhancing peptides has become a growing issue in human as well as in equine sports drug testing. A product called TB-500, claimed to increase muscle growth and tissue repair in horses and other mammals, is available on the Internet and officially distributed. Anecdotal use not only in horse racing, but also in human sport has been documented, reaching also worldwide attention from the media. The active content has been identified as the N-terminal acetylated 17-23 fragment of thymosin beta4 (Ac-LKKTETQ) by our group.

Detection of the intact TB-500 and several metabolites has been achieved in horse urine and plasma, but no data are available for detectability in human. It has already been demonstrated that in vitro/ex vivo studies can be used to obtain reliable indication on the metabolism of small peptides.

The aims of the project are therefore to 1) characterize human metabolism of TB-500 by using human liver microsomes and human S9 fraction (for in vitro studies) and human plasma/serum (ex vivo studies); 2) synthesize appropriate amounts of certified reference standards of TB-500 and its most representative metabolites by solid-phase synthesis; 3) determination of the detection limits of the relevant metabolites and implementation of the metabolites into peptide-screening methods. The synthesized reference standards will be distributed to other human doping control laboratories