# Relevance of Protein Binding of CEM-102 (Fusidic acid) and its pH-dependent Effect on in vitro Activity.

*T.P. DEGENHARDT[1], P.R. RHOMBERG[2], R.N. JONES[2], P. FERNANDES[1] D. JOHNSON[3]*
*[1]Cempra Pharmaceuticals, Chapel Hill, NC, [2]JMI Lab., North Liberty, IA*
*[3]MicroConstants, San Diego, CA*

## Background:

The effect of drug concentration on protein binding and the pH-dependent effect of protein binding on the in vitro activity of CEM-102 (fusidic acid), an oral antibiotic in development for the treatment of complicated skin and skin structure infections, was examined.

## Methods:

Mouse and human plasma, human serum albumin (HSA, 50 mg/mL) and α1-acid glycoprotein (AAG, 1 mg/mL) were spiked with CEM-102 at 75, 750, and 7500 ng/mL and ultrafiltrates were analyzed for CEM-102 concentrations by LC/MS/MS. Susceptibility tests against *Staphylococcus aureus* were performed with and without 10% human serum at various pH ranges in Mueller Hinton (MH) broth, using a modification of standard CLSI methods.

## Results:

CEM-102 binds to mouse and human plasma with approximately the same degree of affinity (97.0%-97.9%). In human blood, CEM-102 binds primarily to HSA (binding ratios 96.3%-96.8%), but does not bind significantly to AAG (0.5%-9.8%). Binding was not concentration dependent (75-7500 ng/mL), indicating that there is no saturation of binding sites at the concentrations studied. MIC values (total of 7 MSSA, MRSA) increased 4 to 8-fold in the presence of 10% serum at pH 7.2-7.4. At pH conditions of 6.0 the effect of serum was negated and the MICs were restored 0.12 -0.25 µg/mL, identical to the MICs in MH broth without serum. This would be expected based on the low pKa of 5.7 of CEM-102, suggesting that the extent of protein binding to CEM-102 is less in acidic environments, such as those commonly encountered in sites of infection.

## Conclusions:

The CEM-102 MICs for *S. aureus* (7 strains) increased 4 to 8-fold in the presence of serum. However, this effect was mitigated in a more acidic milieu, at pH ranges below 7.2. These results indicate that although CEM-102 binds highly to plasma protein, this binding appears to be weak, reversible, and pH dependent.