

## PHYSICAL BIOCHEMISTRY

PM-E3

I.M.R. AND E.S.R. STUDIES OF STAPHYLOCOCCAL PROTEASE. H. Du-  
ras, P. LeGuc and G. Drapeau, Department of Chemistry and  
Microbiology, University of Montreal, Montreal, Canada.  
(intr. by R.H. Marchessault, Department of Chemistry, Uni-  
versity of Montreal.)

The extracellular proteolytic enzyme of Staphylococcus aureus is composed of a single polypeptide chain of about 12,000 in molecular weight and the amino acid composition indicated and unusually high content of dicarboxylic amino acid (35%) and of proline residues (10%). The enzyme has a unique specificity for clearing carboxyl-terminal side of either aspartic acid or glutamic acid and exhibits maximum activity at pH 4.0 and 7.8. The N.M.R. technique was used to study the effect of pH and temperature of the protease. The major changes in the spectra are in the aromatic proton region and suggested a gradual exposure of these groups to the aqueous medium in going to higher pH values. Mono- and di-nitroxide analogs of DFP were used to label the enzyme at the active site. The spectra are comparable to labeled subtilisin BPN'. In conclusion, the staph protease has a rather "open" or "loose" structure in the native form.

FPM-E4

CONFORMATIONAL CHANGES OF POLYPEPTIDES IN INTENSE ELECTRIC FIELDS.  
A.J. Bennett\* and C.P. Bean, (intr. by D.C. Golibersuch), General  
Electric Research and Development Center, Schenectady, N.Y. 12305

Employing a simple "all or none" statistical theory, a calculation is given of the phase diagram in electric field-temperature space for the helix-coil transition of a polypeptide with nonpolar residues but charged end groups. The principal results are i) the transition field extrapolated to absolute zero is on the order of millions of volts per centimeter, ii) the normal transition temperature of large molecules is predicted to be significantly affected by fields as low as 30,000 v/cm, and iii) for temperatures just above the helix-coil transition temperature, the application of a field to a large molecule causes an initial transition to the helix state and with a further isothermal increase of field the coil state returns. The theory is extended to the case of the unfolding of a globular protein in an electric field. The fields are somewhat lower than those for the helix-coil transition and are always single valued at a given temperature. Lastly the effect of including the presence of charged residues is shown to decrease the estimated critical fields but keep them of the same order of magnitude as those given for the case of nonpolar residues.

## PHYSICAL BIOCHEMISTRY

FPM-E5

A DIRECT DETERMINATION OF THE  
SOLUTION. II: CONFORMATION OF  
NMR AND A SHIFT REAGENT. H. S.  
Biological and Medical Research  
Illinois, 60439 and F. A. Vict  
Hospital, Evanston, Illinois,

We have recently reported a  
conformation of molecules cont  
solution(1). The conformation  
was determined in acetone solu  
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between the crystalline and so  
this study in an attempt to el  
conformation of the flexible s  
variation and protonation of t  
mation. Computer methods for  
conformation changes will be d  
change on drug activity will a

(1) N. S. Angerman, S. S. Dan  
(1972)

Work supported in part by the  
This paper is contribution #10.

FPM-E6

CONFORMATIONAL ANALYSIS OF  
J.R. Weintraub and A.J. Hopling  
Western Reserve University, Cle

A set of empirical potential fun  
energy calculations of polymers,  
properties of a number of methyl-  
methylamine, phenyl-n-propylami  
conformational free energy was c  
four states: neutral charge-vacuo  
charge-vacuo (III), positive charge  
adopt one of two stable conformati  
perpendicular to the ring, and the  
conformation with the amine chain  
far from the ring. The folded con  
III, while the extended form is ad  
functions it was also possible to c  
with the minimum energy conform  
abilities to be determined. These  
density of each of the various low  
have a steric "bulge" below the pl  
studied which possess this "bulge"  
cases, also pharmacologically ina  
possess this "bulge".

## PHYSICAL BIOCHEMISTRY

FPM-E3

I.M.R. AND E.S.R. STUDIES OF STAPHYLOCOCCAL PROTEASE. H. Dufras, P. Leduc and G. Drapeau, Department of Chemistry and Microbiology, University of Montreal, Montreal, Canada. (intr. by R.H. Marchessault, Department of Chemistry, University of Montreal.)

The extracellular proteolytic enzyme of Staphylococcus aureus is composed of a single polypeptide chain of about 12,000 in molecular weight and the amino acid composition indicated and unusually high content of dicarboxylic amino acid (35%) and of proline residues (10%). The enzyme has a unique specificity for clearing carboxyl-terminal side of either aspartic acid or glutamic acid and exhibits maximum activity at pH 4.0 and 7.8. The N.M.R. technique was used to study the effect of pH and temperature of the protease. The major changes in the spectra are in the aromatic proton region and suggested a gradual exposure of these groups to the aqueous medium in going to higher pH values. Mono- and dinitroxide analogs of DFP were used to label the enzyme at the active site. The spectra are comparable to labeled subtilisin BPN'. In conclusion, the staph protease has a rather "open" or "loose" structure in the native form.

FPM-E4

CONFORMATIONAL CHANGES OF POLYPEPTIDES IN INTENSE ELECTRIC FIELDS. A.J. Bennett\* and C.P. Bean, (intr. by D.C. Golibersuch), General Electric Research and Development Center, Schenectady, N.Y. 12305

Employing a simple "all or none" statistical theory, a calculation is given of the phase diagram in electric field-temperature space for the helix-coil transition of a polypeptide with nonpolar residues but charged end groups. The principal results are i) the transition field extrapolated to absolute zero is on the order of millions of volts per centimeter, ii) the normal transition temperature of large molecules is predicted to be significantly affected by fields as low as 30,000 v/cm, and iii) for temperatures just above the helix-coil transition temperature, the application of a field to a large molecule causes an initial transition to the helix state and with a further isothermal increase of field the coil state returns. The theory is extended to the case of the unfolding of a globular protein in an electric field. The fields are somewhat lower than those for the helix-coil transition and are always single valued at a given temperature. Lastly the effect of including the presence of charged residues is shown to decrease the estimated critical fields but keep them of the same order of magnitude as those given for the case of nonpolar residues.

## PHYSICAL BIOCHEMISTRY

FPM-E5

A DIRECT DETERMINATION OF THE SOLUTION. II. CONFORMATION OF NMR AND A SHIFT REAGENT. N. S. Biological and Medical Research Illinois, 60439 and T. A. Vic Hospital, Evanston, Illinois,

We have recently reported a conformation of molecules in solution (1). The conformation was determined in acetone solution and now be presented for CQ in methanol between the crystalline and solution. This study in an attempt to elucidate the conformation of the flexible side chain variation and protonation of the amine group. Computer methods for determining conformation changes will be discussed. Change on drug activity will be discussed.

(1) N. S. Angerman, S. S. Danforth (1972)

Work supported in part by the National Science Foundation. This paper is contribution #10.

FPM-E6

CONFORMATIONAL ANALYSIS OF POLYMER CHAINS. J.R. Weintraub and A.J. Hopfinger, Western Reserve University, Cleveland, Ohio

A set of empirical potential functions for the energy calculations of polymers, the properties of a number of methyl-substituted methylamine, phenyl-n-propylamine, and the conformational free energy was calculated for four states: neutral charge-vacuum (I), positive charge-vacuum (II), positive charge-vacuum (III), and positive charge-vacuum (IV). The conformation with the amine chain perpendicular to the ring, and the conformation with the amine chain parallel to the ring, far from the ring. The folded conformation (III), while the extended form is adopted. In addition, functions it was also possible to calculate the conformational free energies with the minimum energy conformations to be determined. These conformational free energies have a steric "bulge" below the plane of the ring. The studied which possess this "bulge" conformation, also pharmacologically inactive, possess this "bulge".