Biochemistry

SOME CHARACTERISTICS OF A CELL-FREE DNAase SENSITIVE SYSTEM INCORPORATING AMINO ACIDS INTO PROTEIN. J. Heinrich Matthaei* and Marshall W. Nirenberg*. National Institutes of Health, Bethesda, Md.

Extracts of E. coli W3100 prepared by grinding cells with alumina actively incorporate C^{14}-valine into protein. The extracts were centrifuged at 20,000 x g for 20 minutes. Remaining intact cells and debris were removed from the supernatant suspension by centrifuging at 30,000 x g for 60 minutes. Ribosomes were obtained by centrifuging the supernatant suspension for 2 hours at 105,000 x g. For maximum incorporation of C^{14}-valine into protein reaction mixtures require ATP, Mg^{++}, an ATP generating system, a complete amino acid mixture, ribosomes and a 105,000 x g supernatant solution. Incorporation of C^{14}-valine into protein proceeds at a rapid rate for 15 minutes at 37°. Incorporation is markedly inhibited by 50 μg. chloramphenicol per ml. 10 μg. per ml. DNAase inhibited approximately 70% of the incorporation whereas an equivalent amount of RNAase was completely inhibitory. Inhibition by DNAase cannot be reversed by addition of polyanions. Addition of a DNAase digest of salmon sperm DNA had no effect upon incorporation of C^{14}-valine into protein. Although DNAase markedly inhibits amino acid incorporation into protein, it is not known whether intact DNA is necessary for this process.