No other amino acids were detected even as trace contaminants. No amide-NH₃ was detected in the peptide, and the isoelectric point was found to be 7.0 as determined by zone electrophoresis on starch.¹⁰ It may be noted that there is one additional residue of serine and one less residue of glutamic acid in the bovine MSH than in β -MSH.^{3,4,11} Hence, as would be expected, the isoelectric point^{4,5} (\not H 5.8) of β -MSH is lower than that of the bovine hormone.

By the paper-strip modification¹² of the phenvl isothiocyanate method.¹³ the amino acid sequence at the N-terminus was established as Asp.Ser.Gly.-Pro.Tyr.Lys.Met. A portion of this N-terminal sequence was confirmed by means of kinetic studies with leucine aminopeptidase.14 Aspartic acid was identified as the C-terminal amino acid by reaction with carboxypeptidase. Digestion of the peptide hormone with chymotrypsin gave rise to four peptides in accordance with the specificity of the enzvme:

 $- Ch-5B \longrightarrow Ch-3B \longrightarrow Ch-2B \leftarrow Ch-4B-$ H. Asp. Ser. Gly. Pro. Tyr. Lys. Met. Glu. His. Phe. Arg. Try. Gly. Ser. Pro. Lys. Asp. OH The F¹⁹ magnetic resonance spectra of

The separation and analysis of these peptide fragments in the chymotryptic hydrolysate were carried out as previously described.4,15 The amino acid sequence in peptide Ch-3B was also elucidated by the action of leucine aminopeptidase. These data permit the formulation of the structure shown above for the bovine MSH.

It will be seen that this structure differs from that proposed for the porcine hormone^{3,4} only at position 2, where a servi residue replaces a glutamyl. As far as we are aware, the replacement of an uncharged residue (serine) by a charged one (glutamic acid) has not previously been encountered in connection with species variations among biologically active peptides.

(10) I. D. Raacke and C. H. Li, J. Biol. Chem., 215, 277 (1955).

(11) P. Roos, Acta Chem. Scand., 10, 1061 (1956).

(12) H. Fraenkel-Conrat, THIS JOURNAL, 76, 3606 (1954).

(13) P. Edman, Acta Chem. Scand., 4, 283 (1950).

(14) D. H. Spackman, E. L. Smith and D. M. Brown, J. Biol. Chem., 212, 255 (1955). We are indebted to Dr. L. K. Ramachandran for a gift of this enzyme.

(15) I. I. Geschwind, C. H. Li and L. Barnafi, THIS JOURNAL, 79, 620 (1957).

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FLUOROTELLURATES

Sir:

We have studied the behavior of tellurium hexafluoride and of selenium hexafluoride toward various inorganic fluorides. Neither of these hexafluorides showed evidence of strong interaction or of fluorine exchange with covalent fluorides; however, the tellurium compound did react with certain ionic fluorides to form the new fluorotellurate salts. Selenium did not form fluoroselenates; apparently,

the requisite orbitals in selenium lie at too high an energy level.

The fluorotellurates, which have the limiting composition 2MF·TeF6, were prepared by heating a metal fluoride with an excess (100%) of tellurium hexafluoride in a pressure vessel to 250° and cooling this reaction mixture over a period of 24 hours to room temperature. After the excess gas was released, the solid was recovered. In the case of cesium the solid product analyzed closely for $2CsF \cdot TeF_6$. The X-ray pattern of the cesium compound consisted of many weak lines which were not indexed; no crystalline cesium fluoride was present. Rubidium and potassium fluorides gave products that had the empirical compositions $2RbF \cdot 0.52TeF_6$ and $2KF \cdot 0.32TeF_6$. X-Ray diffraction patterns of these solids showed the presence of the alkali metal fluoride and a second crystalline phase. The fluorides of sodium, lithium, and barium were inactive under the experimental conditions. Under comparable conditions,

selenium hexafluoride gave no sign of reaction with cesium and potassium fluorides.

binary systems of liquid tellurium (and selenium) hexafluoride with arsenic trifluoride and with boron trifluoride showed no evidence of compound formation or of a measurable fluorine exchange rate up to 60°.

The fluorotellurates dissociated at elevated temperatures to yield the starting materials. Crude measurements of the dissociation pressure of cesium fluorotellurate yielded an average value for the heat of dissociation of 0.4 kcal. over the temperature range 200-250°. Addition of the fluorotellurates to water resulted in evolution of tellurium hexafluoride and dissolution of the alkali metal fluoride. The fluorotellurates displayed no stability outside the solid state, and the stability of the lattice appeared to be inversely related to the polarizing power of the cation. The lattice could either be composed of M^+ and $TeF_8^=$ or of M^+ , F^- , and TeF_7^- aggregates. A unique solution of the crystal structure is being sought in analyses of the X-ray diffraction pattern and the F19 magnetic resonance spectrum of the cesium compound.

Contribution No. 405 CHEMICAL DEPARTMENT EXPERIMENTAL STATION E. L. MUETTERTIES E. I. DU PONT DE NEMOURS AND COMPANY WILMINGTON, DELAWARE

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FORMATION OF 6β -HYDROXY AND 6-KETO DERIVATIVES OF ESTRADIOL-16-C¹⁴ BY MOUSE LIVER MICROSOMES

Sir:

Further study of the protein-binding of estradiol by fortified liver preparations1 has revealed that mouse liver microsomes in the presence of reduced triphosphopyridine nucleotide and oxygen convert estradiol-16-C¹⁴ to at least five new metabolites² extractable into an acetone-benzene mixture (3:4). Preliminary chromatographic analysis on

(1) I. L. Riegel and G. C. Mueller, J. Biol. Chem., 210, 249 (1954). (2) G. Rumney, Fed. Proc., 15, 343 (1956).