



Kinetics and mechanism of the reactions of Ni(II) oligopeptideamide-cyanide and Ni(II) oligopeptide-cyanide complexes with triethylene-tetramine
by William Anthony Marchese

A thesis submitted in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE
in Chemistry
Montana State University
© Copyright by William Anthony Marchese (1974)

Abstract:

The kinetics and mechanism of the reaction between - TRIEN and certain Nickel(II) oligopeptideamide cyanide and Nickel(II) oligopeptide cyanide complexes have been investigated. The individual rate constants for each reaction of the different substrates were evaluated. The contribution of the TRIEN's subspecies to the overall rate constant were, also, evaluated. The effects of steric hindrances in the substrate ring and alteration of the solvent's pH on the rate constants were investigated.

The rate constants for the Nickel(II) oligopeptideamide cyanide and Nickel(II) oligopeptide cyanide complexes are approximately 10,000 times slower than for the Nickel(II) triglycinate ion. Both the steric hindrances and pH effects have a direct effect on the rate constants;

STATEMENT OF PERMISSION TO COPY

In presenting this thesis in partial fulfillment of the requirements for an advanced degree at Montana State University, I agree that the Library shall make it freely available for inspection. I further agree that permission for extensive copying of this thesis for scholarly purposes may be granted by my major professor, or, in his absence, by the Director of Libraries. It is understood that any copying or publication on this thesis for financial gain shall not be allowed without my written permission.

Signature William A. Marchese

Date 8/7/74

KINETICS AND MECHANISM OF THE REACTIONS OF
Ni(II) OLIGOPEPTIDEAMIDE-CYANIDE AND
Ni(II) OLIGOPEPTIDE-CYANIDE COMPLEXES
WITH TRIETHYLENETETRAMINE

by

WILLIAM ANTHONY MARCHESE

A thesis submitted in partial fulfillment
of the requirements for the degree

of

MASTER OF SCIENCE

in

Chemistry

Approved:


Chairman, Examining Committee


Head, Major Department


Graduate Dean

MONTANA STATE UNIVERSITY
Bozeman, Montana

August, 1974

ACKNOWLEDGMENTS

I would like to thank the Department of Chemistry at Montana State University for their financial support in the way of teaching assistantships and tuition waivers. I would like to thank Dr. Pagenkopf for his continued help and interest throughout this project. Without his continued support, this thesis would not have been possible. I would also like to thank Dr. Jennings for all the support he has given me in the past two years, without which none of my graduate study would have been possible. I would also like to thank Dr. William Aziz Hanna, of the Electrical Engineering Department, and Dr. David Smith, of the Chemistry Department, whose help with the computer was priceless. I would also like to acknowledge the help of the staff members and graduate students of the department for their discussion, technical aid and friendship during the past two years.

TABLE OF CONTENTS

	<u>Page</u>
VITA	ii
ACKNOWLEDGMENTS	iii
TABLE OF CONTENTS	iv
LIST OF TABLES	vi
LIST OF FIGURES	viii
ABSTRACT	x
STATEMENT OF THE PROBLEM	1
INTRODUCTION	2
EXPERIMENTAL SECTION	6
Apparatus	6
Reagents	6
Kinetic Measurements	9
RESULTS	12
Determination of the General Rate Expression for the Reaction of TRIEN with $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$	12
pH Dependence of the $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$ System	15
Steric Effects on the Oligopeptideamide System	21
Determination of the General Rate Expression for the Reaction of $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ and TRIEN	26
pH Dependence of the $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System	29

	<u>Page</u>
Steric Effects of the Tripeptide System	37
Reaction of $\text{Ni}(\text{H}_2\text{SGG})\text{CN}^{2-}$ in Presence of Excess CN^- and Effect of Excess CN^- on k_d	41
DISCUSSION	50
REFERENCES	58

LIST OF TABLES

<u>Table</u>	<u>Page</u>
I. Names of Compounds Used in This Study and Their Abbreviations	9
II. Equilibria and Equilibrium Constants	11
III. Concentrations of Species and k_{obs} for $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$ Reaction with TRIEN	14
IV. Correspondence of k_{obs} with pH for $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$ System	16
V. Calculated k_{obs} 's from pH Study	18
VI. Complexes and Their k_{t} 's at the $-\log[\text{H}^+]$ Evaluated	21
VII. $k_{\text{obs}}\{\text{sec}^{-1}\}$ and Corresponding $[\text{TRIEN}]_{\text{t}}$ for $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System	27
VIII. Relationship of $k_{\text{obs}}\{\text{sec}^{-1}\}$ and pH for $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$	29
IX. k_{obs} 's{experimental} and k_{obs} 's{calculated} for $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System	31
X. Steric Effects on k_{d} and k_{t} in Oligopeptide System	34
XI. k_{obs} 's and Reaction Conditions for Reaction of $\text{Ni}(\text{H}_{-2}\text{SGG})\text{CN}^{2-}$ with TRIEN	39

<u>Table</u>	<u>Page</u>
XII. k_{tt} and k_d with Excess Cyanide for Ni(H ₂ G ₂ GG)CN ²⁻ Reaction with TRIEN	44
XIII. Effect of Excess Cyanide on k_d for Tripeptide Systems	49

LIST OF FIGURES

<u>Figure</u>	<u>Page</u>
1. Pseudo First Order Kinetics for Reaction of TRIEN with $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$. $-\log[\text{H}^+] = 10.10$	13
2. Resolution of k_{trien} and $k_{\text{htrien}}^\ominus$ for the $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$ System	19
3. $k_{\text{obs}}\{\text{sec}^{-1}\}$ versus $-\log[\text{H}^+]$ for $\text{Ni}(\text{H}_{-2}\text{GGa})\text{CN}^{1-}$ from Calculated k_{trien} and $k_{\text{htrien}}^\ominus$	20
4. $k_{\text{obs}}\{\text{sec}^{-1}\}$ versus $[\text{TRIEN}]_t$ for $\text{Ni}(\text{H}_{-2}\text{AGa})\text{CN}^{1-}$ System. $-\log[\text{H}^+] = 10.63$	23
5. $k_{\text{obs}}\{\text{sec}^{-1}\}$ versus $[\text{TRIEN}]_t$ for $\text{Ni}(\text{H}_{-2}\text{GAa})\text{CN}^{1-}$. $-\log[\text{H}^+] = 10.74$	24
6. $k_{\text{obs}}\{\text{sec}^{-1}\}$ versus $[\text{TRIEN}]_t$ for $\text{Ni}(\text{H}_{-2}\text{AAa})\text{CN}^{1-}$. $-\log[\text{H}^+] = 10.65$	25
7. Rate Plot for $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System. $-\log[\text{H}^+] = 10.20$	28
8. Resolution of k_{trien} and $k_{\text{htrien}}^\ominus$ for the $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System	32
9. $\frac{k_{\text{obs}}\{\text{sec}^{-1}\}}{[\text{TRIEN}]_t}$ versus $-\log[\text{H}^+]$ for $\text{Ni}(\text{H}_{-2}\text{GGG})\text{CN}^{2-}$ System	33
10. $k_{\text{obs}}\{\text{sec}^{-1}\}$ versus $[\text{TRIEN}]_t$ for $\text{Ni}(\text{H}_{-2}\text{AAG})\text{CN}^{2-}$. $-\log[\text{H}^+] = 10.52$	35

<u>Figure</u>	<u>Page</u>
11. $k_{\text{obs}} \{\text{sec}^{-1}\}$ versus $[\text{TRIE N}]_t$ for Ni(H ₋₂ GAG)CN ²⁻ . $-\log[\text{H}^+] = 10.65$	36
12. $k_{\text{obs}} \{\text{sec}^{-1}\}$ versus $[\text{TRIE N}]_t$ for Ni(H ₋₂ GGG Bnz-E)CN ¹⁻ . $-\log[\text{H}^+] = 10.53$	37
13. $k_{\text{obs}} \{\text{sec}^{-1}\}$ versus $[\text{TRIE N}]_t$ for Ni(H ₋₂ SGG)CN ²⁻ . $-\log[\text{H}^+] = 10.73$	40
14. $k_{\text{obs}} \{\text{sec}^{-1}\}$ versus $[\text{TRIE N}]_t$ for Ni(H ₋₂ GGG)CN ²⁻ System with $[\text{CN}^-]_t = 4.0 \times 10^{-5} \text{ M}$. $-\log[\text{H}^+] = 10.27$	42
15. $k_{\text{obs}} \{\text{sec}^{-1}\}$ versus $[\text{TRIE N}]_t$ for Ni(H ₋₂ GGG)CN ²⁻ System with $[\text{CN}^-]_t = 6.0 \times 10^{-5} \text{ M}$. $-\log[\text{H}^+] = 10.18$	43
16. $k_d \{\text{sec}^{-1}\}$ versus $[\text{CN}^-]_t$	44
17. A Stereochemical Drawing of L-alanine	48

ABSTRACT

The kinetics and mechanism of the reaction between TRIEN and certain Nickel(II) oligopeptideamide cyanide and Nickel(II) oligopeptide cyanide complexes have been investigated. The individual rate constants for each reaction of the different substrates were evaluated. The contribution of the TRIEN's subspecies to the overall rate constant were, also, evaluated. The effects of steric hindrances in the substrate ring and alteration of the solvent's pH on the rate constants were investigated.

The rate constants for the Nickel(II) oligopeptideamide cyanide and Nickel(II) oligopeptide cyanide complexes are approximately 10,000 times slower than for the Nickel(II) triglycinate ion. Both the steric hindrances and pH effects have a direct effect on the rate constants.

STATEMENT OF THE PROBLEM

The investigation and elucidation of the kinetics and mechanism for the reactions of some Ni(II) Oligopeptide cyanide complexes with triethylenetetramine are the major problems presented in this work.

The studies involve the determination of the general rate expression, pH effects and steric effects introduced by variation in substrate complex.

INTRODUCTION

Oligopeptide-metal complexes have generated much interest because of their possible biologically enzymatic properties (1).

Two major areas that have received much attention are the structural and kinetic aspects of these complexes.

Originally, the question of how Ni(II) and Cu(II) oligopeptide complexes formed was pursued hotly. After some initial confusion, it became evident that the formation of these complexes is assisted by the ionization of the peptide's imide protons and the peptide's nitrogen atoms are coordinated to the metal (2,3)

The kinetics of both Cu(II) and Ni(II) complexes of oligopeptides have been investigated quite thoroughly. The kinetics of these complexes involving proton transfer, ligand exchange, steric and pH effects have been studied.

The proton transfer reactions of these complexes are of both the general acid catalysis (4,5,6) and specific hydrogen ion catalysis (7,8).

The ligand exchange reactions of these Cu(II) and Ni(II) oligopeptide complexes most often involve the substitution of a multidentate ligand for the oligopeptide.

Triethylenetetramine and ethylenediamine tetraacetate ion are used most frequently (4,5,9).

The Cu(II)-Triglycine, $\text{Cu}(\text{H}_{-2}\text{GGG})^{-}$, reaction with multidentate ligands proceeds by two general mechanisms (9). In a general acid catalysis mechanism, a proton is transferred to a peptide nitrogen to assist the dissociation of the complex. The nucleophilic mechanism involves coordination of the displacing ligand to the metal, and this speeds the breaking of the metal-peptide nitrogen bonds. Steric effects are very important and steric hindrance in the substituting ligand can block the reaction.

Ni(II)-Triglycinate, $\text{Ni}(\text{H}_{-2}\text{GGG})^{1-}$, can react by a nucleophilic mechanism similar to that proposed for the analogous copper complex, $\text{Cu}(\text{H}_{-2}\text{GGG})^{1-}$ (4). Ethylenediamine and polyamines have second-order rate constants of $(1.2 - 1.7) \times 10^4 \text{M}^{-1} \text{sec}^{-1}$ with these complexes (4). Steric effects, chelation and donor ability are important factors in the substituting ability of the nucleophile (4).

In this work, both oligopeptideamides and Oligopeptides were complexed with Nickel(II). Oligopeptideamides are small peptides composed of two amino acids, and the small peptide's carboxylic acid end, its C end, has

been converted into an amide. Glycyl-glycine amide is an example of an oligopeptideamide. Oligopeptide refers to a small peptide composed of three amino acids, and the carboxylic acid of this small peptide is untouched.

Glycyl-glycyl-glycine is an example of an oligopeptide.

In this paper, a cyanide ion, CN^- , is added to enhance the stabilization of the complex. The CN^- occupies the fourth position of the square planar coordination sphere of Ni(II). Such complexes of Ni(II) Oligopeptide or oligopeptideamide and cyanide shall be referred to in this work as "mixed complexes".

The mixed complexes are square planar, yellow and diamagnetic. $\text{Ni}(\text{H}_2\text{GGa})\text{CN}^{1-}$ has $\lambda_{\text{max}} = 405 \text{ nm}$, $\epsilon = 170 \text{ M}^{-1}\text{cm}^{-1}$, Cf. $\lambda_{\text{max}} = 452 \text{ nm}$, $\epsilon = 136 \text{ M}^{-1}\text{cm}^{-1}$, for the aquo-complex $\text{Ni}(\text{gga})\text{H}_2\text{O}$ (7). The $\text{Ni}(\text{H}_2\text{GGG})\text{CN}^{2-}$ absorption maxima and molar absorptivities are $\lambda_{\text{max}} = 410 \text{ nm}$ and $\epsilon = 174 \text{ M}^{-1}\text{cm}^{-1}$ (7). The mixed complexes behave as weak acids, with pK_a 's Ca 9. Previous attempts at determining the stability constants of these complexes have been made. Because of the complete formation of these complexes, the stability constants must be greater than 10^7 (7).

Previous studies with the Ni(II) mixed complexes have been limited to proton transfer studies (7,8). It was found with these proton transfer studies that the system responded to specific hydrogen catalysis only, and the cyanide ion had a stabilization effect on the substrate that reduced the rate of proton transfer 38,000 times (7).

EXPERIMENTAL SECTION

Apparatus

For the gathering of all spectral data, either absorption spectra or absorbance measurements at a fixed wavelength, a Cary Model 14 spectrophotometer was used. The instrument is equipped with thermostated cell compartments capable of maintaining a constant temperature to within $\pm 0.1^{\circ}\text{C}$.

A Radiometer Model PHM26c pH meter was used to measure pH. The instrument was standardized using standard buffer solutions.

To record the passage of time, a A.R. & J.E. Meylan type 208A stopwatch was used.

Reagents

Nickel (II) perchlorate stock solution, 0.00916M , was prepared by diluting to volume a pre-determined volume of $0.0916\text{M Ni}(\text{ClO}_4)_2$. The $0.0916\text{M Ni}(\text{ClO}_4)_2$ was prepared from twice recrystallized $\text{Ni}(\text{ClO}_4)_2$ and standardized by EDTA titration (10).

Sodium cyanide solution (0.095M) was standardized by the argentimetric method (11), then diluted to 0.0095M .

Anhydrous sodium perchlorate was prepared by dissolving a weight of hydrated sodium perchlorate in boiling, double distilled water, filtering the solution using a millipore filter, evaporating to dryness and heating in an oven overnight. To prepare a 2.00M solution of sodium perchlorate, the required weight of sodium perchlorate was dissolved in the required volume of double distilled water.

A stock solution of borate buffer was prepared by dissolving enough sodium borate in double distilled water to provide total boron concentration equal to 0.2973M .

Triethylenetetramine was converted to its disulfate salt by adding dropwise 2.0 moles of concentrated sulfuric acid to 1.0 mole of triethylenetetramine dissolved in ice cold toluene. The temperature of the triethylenetetramine-toluene solution was maintained at 0°C by using an ice bath. After the addition of the sulfuric acid, the resulting mixture was stirred for one hour. The precipitated disulfate salt was twice recrystallized from double distilled water.

To dissolve the disulfate salt in double distilled water for the preparation of the standard solution, a small amount of dilute NaOH solution was required.

Standardization was achieved by mole ratio at 550 nm using $0.077M$ $Cu(ClO_4)_2$ and acetate buffer.

Prior to each kinetic run, the required volume of the standard triethylenetetramine was adjusted to the desired pH using strong NaOH solution and then diluted to the desired volume.

The mixed complexes were prepared by mixing aliquots of $0.00916M$ $Ni(ClO_4)_2$ and the particular oligopeptide, $0.001M$, to be studied so that a 100% molar excess of the oligopeptide was present. The ionic strength was adjusted to $0.10M$ $NaClO_4$. The pH of the resulting solution was raised to 9.5 with dilute NaOH, borate buffer and a stoichiometric amount of NaCN, equivalent to total Nickel(II), added and the pH adjusted to the desired value with either dilute NaOH or $HClO_4$.

Solutions of the complexes were always prepared, within three hours, prior to any kinetic run.

After preparation, the Ni(II) oligopeptide cyanide solution was placed in a water bath at $25^\circ C$, for two hours, to insure temperature homogeneity during the reaction.

Names and their abbreviations of all the chemicals used in this study are summarized in Table I. Also, the suppliers and compounds' purities are supplied.

Table I. Names of Compounds Used in This Study and Their Abbreviations.

Name	Abbreviation
Glycyl-Glycine amide	GGa ^{a, g}
Alanyl-Glycine amide	AGa ^{a, g}
Glycyl-Alanine amide	GAa ^{b, g}
Alanyl-Alanine amide	AAa ^{a, g}
Glycyl-Glycyl-Glycine	GGG ^{a, g}
Glycyl-Alanyl-Glycine	GAG ^{a, g}
Alanyl-Alanyl-Glycine	AAG ^{a, g}
Glycyl-Glycyl-Glycine-Benzyl/Ester	GGG-BnzE ^{a, g}
Sarcosyl-Glycyl-Glycine	SGG ^{c, g}
Nickel(II) perchlorate	Ni(ClO ₄) ₂ ^{d, h}
Sodium cyanide	NaCN ^{e, h}
Sodium perchlorate	NaClO ₄ ^{e, h}
Triethylenetetramine	TRIEN ^{f, h}
Sodium borate	Na ₂ B ₄ O ₇ · 10H ₂ O ^{e, h}

- a) Fox Chemical Company
 b) Cyclo Chemical Company
 c) Nutritional Biochemicals Corporation
 d) G. Frederick Smith Chemical Company
 e) J. T. Baker Chemical Company
 f) E. H. Sargent & Company
 g) Chromatographically Pure
 h) Reagent Grade

Kinetic Measurements

The substrate molecule exhibits a characteristic maximum absorption peak at 245 nm. The course of each

reaction was followed by monitoring the disappearance of this peak, using the previously described Cary 14 spectrophotometer, with time after mixing of the reactants.

The reaction was initiated by mixing 50 ml of substrate solution and from 0.5 ml to 4.0 ml of the TRIEN solution. After the addition of the TRIEN, the reactants were stirred for an unspecified amount of time to insure mixing.

Ionic strength was maintained at 0.10M NaClO_4 . The temperature throughout the reaction time was maintained at 25°C.

The reaction rates were measured over a range of TRIEN concentrations and except where specified constant substrate concentration.

A_t will be defined as a substrate absorption at anytime, t , during the reaction. And A_∞ is the absorption that does not decrease with further time, i.e., the end of the reaction. $A_t - A_\infty$ is a function of the substrate concentration. Therefore, by calculating the slope of $-\ln(A_t - A_\infty)$ plotted versus time, one obtains the individual rate constants for each reaction, designated as k_{obs} .

