# Formation Constants of Metal Complexes of $N^3$-Benzyl-L-histidine and $N^{\alpha}N^3$-Dibenzyl-L-histidine and Stereoselectivity in the Ternary Complexes with D- and L-Histidine †

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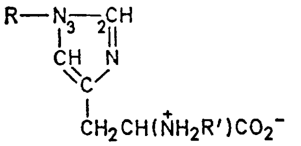
Formation constants for the complexes of $Ni^{II}$, $Cu^{II}$, and $Zn^{II}$ with $N^3$-benzyl-L-histidine and $N^{\alpha}N^3$-dibenzyl-L-histidine, and of the ternary complexes of these metals and ligands with D- and L-histidine, have been measured potentiometrically at 25 °C and $I$ = 0.10 mol $dm^{-3}$ (K[$NO_3$]). Stabilization of the ternary complexes is small, but there is significant stereoselectivity between the ternary complexes with $Ni^{II}$ and $Zn^{II}$, the *meso* complexes being preferred in all cases. With $Cu^{II}$, stereoselectivity is small or absent in the unprotonated ternary complexes, but significant in the monoprotonated ternary species {[Cu(D/L-His)(L-Ligand)H]}, the optically active complex being the more stable.

THE importance of metal–histidine interactions in biological systems has long been recognised. The imidazole group of histidine (His) serves many important biological functions, such as the catalytic site for hydrolytic enzymes, and contributes to the buffering of proteins in the physiological pH range. It is not surprising, therefore, that a great deal of work has been devoted to an understanding of the interactions between His and metal ions and the possible stereoselectivity in such interactions.[1] Much of the work has been restricted to $Cu^{II}$–His systems with little interest in substituted histidines. Many of the problems confronted in these studies are associated with the particular co-ordination centres in the intermediate pH region. A study of N-substituted histidines should, therefore, shed light on these problems as well as amplifying any stereoselective effects which may be present.

In some early work, Leberman and Rabin[2] studied the copper(II)–3-methylhistidine system potentiometrically in order to determine whether the acid dissociation of the mono complex of $HisO^-$, $[Cu(HisO)]^+$, which occurs at high pH, was from the pyrrole nitrogen ($N^3$) or a co-ordinated water molecule. Sigel and McCormick[3] studied the n.m.r. line-broadening effects associated with the copper(II) complexes of 3-methyl- and 1-methyl-histidine since the latter can only bond to $Cu^{II}$ in a glycine-like manner. They concluded that 3-methylhistidine bonds *via* both the imidazole and the amino-nitrogen atoms. Ritsma[4] studied the interactions of $Cu^{II}$, $Co^{II}$, $Ni^{II}$, and $Zn^{II}$ with *N*-methylhistidine, *NN*-dimethylhistidine, and *N*-benzylhistidine. He concluded that with $Cu^{II}$ the protonated metal complexes are somewhat less important than in the case of His, and that glycine-like behaviour is hindered by the substituents at the amino-nitrogen atom, imidazolepropionic acid-like behaviour becoming increasingly important with substitution.

Three possible bis complexes can be formed with a racemic mixture of the ligand, *e.g.* $[M(L\text{-}HisO)_2]$, $[M(D\text{-}HisO)_2]$, and *meso*-[M(L-HisO)(D-HisO)]. In the absence of stereoselectivity, the concentration ratios in an equilibrium would be expected to be 1 : 1 : 2. With $Co^{II}$, $Ni^{II}$, and $Zn^{II}$ and His the *meso* complex is formed preferentially. This has been confirmed by the measurement of both formation constants and enthalpy changes.[1] With $Cu^{II}$ there is no significant stereoselectivity in the formation constants of bis complexes, but the formation of the monoprotonated optically active complex $[Cu(L\text{-}HisO)_2H]^+$ appears to be favoured over its *meso* analogue, and formation of the optically active bis complex $[Cu(L\text{-}HisO)_2]$ has a more favourable enthalpy change.[5] In addition, stereoselectivity has been detected in the formation constants of a range of ternary complexes of $Cu^{II}$, His, and another amino-acid.[6] These examples of stereoselectivity are more likely to be significant with N-substituted histidines in which steric factors would be more important.

We now report the results of a potentiometric study of the complexes of $Ni^{II}$, $Cu^{II}$, and $Zn^{II}$ with $N^3$-benzyl-L-histidine (bzh) and $N^{\alpha}N^3$-dibenzyl-L-histidine (dbzh), and of the ternary complexes of these metal ions and ligands with D- and L-histidine.



His R = R' = H
bzh R = $PhCH_2$, R' = H
dbzh R = R' = $PhCH_2$

## EXPERIMENTAL

Amino-acids were obtained from the Sigma Chemical Co. (SIGMA grade).

Complex formation constants were calculated from potentiometric-titration curves obtained as described previously.[7] The glass electrode was calibrated in terms of hydrogen-ion concentrations and titrations were carried out at 25 °C with an ionic background of 0.10 mol $dm^{-3}$ K[$NO_3$]. Calculations were made with the aid of the MINIQUAD computer program.[8]

## RESULTS AND DISCUSSION

*Binary Complexes.*—Complex-formation constants are expressed in terms of the recognised convention, usually in the form of overall formation constants, β. For

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† No reprints available.

[1] L. D. Pettit and J. L. M. Swash, *J.C.S. Dalton*, 1976, 588.

[2] R. Leberman and B. R. Rabin, *Trans. Faraday Soc.*, 1959, **55**, 1660.

[3] H. Sigel and D. B. McCormick, *J. Amer. Chem. Soc.*, 1971, **93**, 2041.

[4] J. H. Ritsma, Thesis, Groningen, 1973.

[5] D. S. Barnes and L. D. Pettit, *J. Inorg. Nuclear Chem.*, 1971, **33**, 2177.

[6] G. Brookes and L. D. Pettit, *J.C.S. Chem. Comm.*, 1974, 813; 1975, 385.

[7] G. Brookes and L. D. Pettit, *J.C.S. Dalton*, 1975, 2106.

[8] A. Sabatini, A. Vacca, and P. Gans, *Talanta*, 1974, **21**, 53.

example, in reaction (1) the formation constant is $\beta_{xyz}$.

$$M_x + L_y + H_z \longrightarrow [M_xL_yH_z] \qquad (1)$$

If the reaction includes formation of a ternary complex, *e.g.* $M_w + His_x + L_y + H_z \longrightarrow [M_w(His)_xL_yH_z]$, the formation constant would be $\beta_{wxyz}$.

Protonation constants are given in Table 1 together with those for His for comparison. Replacing the hydrogen at $N^3$ with a benzyl group results in a lowering of the basicity of the imidazole nitrogen. Likewise the introduction of a benzyl group at the α-amino-nitrogen also results in a significant lowering of the basicity of that nitrogen. These effects result from the electron-attracting inductive effect of the benzyl group. The values of the protonation constants of the carboxyl groups are of low precision because of their very low basicities. The calculated standard deviations take account only of random errors and, while not a direct measure of the accuracy of the constants calculated, are useful guides to the importance of the particular species and the probable precision of the constant.

TABLE 1

Protonation constants at 25 °C and $I = 0.10$ mol dm$^{-3}$ ($K[NO_3]$)

| | bzh | dbzh | L-His |
|---|---|---|---|
| log *K* (amine N) | 9.206(1) | 8.471(4) | 9.128(1) |
| log *K* (imidazole N) | 5.525(2) | 5.504(8) | 6.052(3) |
| log *K* (carboxyl O) | 1.94(2) | 1.97(2) | 1.58(2) |

Overall metal complex-formation constants are given in Table 2. L-Histidine was also studied and the results calculated are very close to those already published.[1] These results are important when studying the formation of ternary complexes. The pH values studied were limited to those before the onset of formation of hydroxo-complexes.[1]

TABLE 2

Formation constants for binary complexes at 25 °C and $I = 0.10$ mol dm$^{-3}$ ($K[NO_3]$)

| | | log $\beta_{xyz}$ | | |
|---|---|---|---|---|
| Metal ion | *xyz* | bzh | dbzh | L-His |
| $Ni^{II}$ | 110 | 8.823(1) | 8.014(4) | 8.669(1) |
| | 120 | 16.111(4) | 15.135(4) | 15.555(2) |
| | 111 | 11.53(7) | ? | 12.06(6) |
| | 121 | 20.18(9) | 19.32(6) | 20.32(4) |
| $Cu^{II}$ | 110 | 10.194(1) | 8.958(3) | 10.111(1) |
| | 120 | 18.519(1) | 16.76(1) | 18.078(3) |
| | 111 | 13.740(3) | 12.46(4) | 14.089(5) |
| | 121 | 23.623(1) | 21.02(6) | 23.883(2) |
| | 122 | 27.05(1) | 25.34(6) | 27.56(1) |
| $Zn^{II}$ | 110 | 6.580(1) | 5.744(3) | 6.453(2) |
| | 120 | 12.17(1) | 11.488(2) | 12.007(3) |
| | 111 | ? | ? | 11.30(1) |
| | 121 | ? | 16.92(1) | 17.80(2) |

The results for the complexes with $Cu^{II}$ suggest that bonding in the complexes of bzh is similar to that in the complexes of His itself. Complexes with dbzh are significantly weaker, protonated species in particular being considerably less important. A comparison of the equilibrium constants for reaction (2), where HL is histidine

$$Cu^{2+} + H_2L^+ \longrightarrow [Cu(HL)]^{2+} + H^+ \qquad (2)$$

or substituted histidine, with complexes of glycine, imidazolepropionic acid, and histamine suggests glycine-like bonding in the monoprotonated complex (111 species) of bzh (as with His itself [1]), but results for dbzh are less conclusive. Glycine-like bonding is also supported by a comparison of the constants for reaction (3) where L = His, bzh, or 2,4-diaminobutyric acid.[9]

$$Cu^{2+} + H_3L^{2+} \longrightarrow [Cu(HL)]^{2+} + 2H^+ \qquad (3)$$

In the mono complex, $[CuL]^+$ (110), the bonding of bzh is again probably the same as with His (*i.e.* tridentate). However, the complex with dbzh is significantly weaker, although it is more stable than that with glycine (log $\beta_{110}$ 8.38, log $\beta_{011}$ 9.85) or histamine (log $\beta_{110}$ 9.55, log $\beta_{011}$ 9.80), when the markedly lower basicity of dbzh (log $\beta_{011}$ 8.47) is taken into account. The bulky substituent on the α-amino-nitrogen atom must make it more difficult for the carboxyl group to interact with $Cu^{II}$ at the apical position. Similar arguments would apply to the formation of the bis complexes, $[CuL_2]$. The high values for the stepwise constant, log $K_{121}{}^{110}$, suggest that the mode of bonding of $L^-$ to the complex $[Cu(HL)]^{2+}$ is tridentate. However, again, the value for dbzh is smaller than with the other ligands, suggesting significant steric interference.

With His, protonated complexes are far less important with $Ni^{II}$ than they are with $Cu^{II}$. Their importance with the substituted histidines and $Ni^{II}$ is even less, and inclusion of the 111 species makes only a marginal improvement in the statistics of the fit of trial models to the experimental data. In a titration of $Ni^{II}$ with bzh in the ratio 1 : 3, the protonated complex 121 $[Ni(HL)L]^+$ involved a maximum of 6% of the total metal at *ca.* pH 5.5.

In the mono and bis complexes (110 and 120 species) all the ligands appear to bond in a tridentate manner. Steric hindrance is significant in the complexes with dbzh (*cf.* with $Cu^{II}$) making them significantly less stable than those of the other ligands. However, the complexes of bzh are more stable than those of His itself. This is somewhat unexpected but may be partly the result of greater contributions of the α-amino-nitrogen and carboxyl-oxygen atoms to the overall stability of the species $[NiL]^+$. If this is the case the greater basicities of these centres in bzh compared to His would encourage higher values for $\beta_{110}$ and $\beta_{120}$. This trend is also found with $Cu^{II}$ and $Zn^{II}$.

Complexes formed between $Zn^{II}$ and His are less stable than those with $Co^{II}$, opposite to the order found with other amino-acids.[10] This, together with other evidence suggests that the readiness of $Zn^{II}$ to adopt tetrahedral co-ordination may result in the His being effectively bidentate with the carboxyl oxygen unco-ordinated. This is supported by an *X*-ray study of solid $Zn^{II}$–His complexes which shows the bonding to be, essentially, histamine-like with a distorted tetrahedral co-ordination shell.[11]

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[9] G. Brookes and L. D. Pettit, *J.C.S. Dalton*, 1976, 42.

[10] D. R. Williams, *J. Chem. Soc.* (*A*), 1970, 1550.

[11] K. A. Fraser and M. M. Harding, *J. Chem. Soc.* (*A*), 1967, 415.

Hydroxo-complexes were unimportant under the experimental conditions used. The monoprotonated species $[Zn(HL)]^{2+}$ was important with His, but not with the substituted histidines, while the monoprotonated bis complex $[Zn(HL)L]^{+}$ (121 species) was also important with dbzh. The ligand dbzh is also interesting because, with $Zn^{II}$, the stepwise constants, log $K_{110}$ and log $K_{120}$, are effectively the same (5.74). Abnormalities in the stepwise constants for zinc complexes are known and probably result from changes in co-ordination geometry. With this ligand it is possible that steric effects, caused by the benzyl substituent groups, could promote a change from essentially tetrahedral to essentially octahedral co-ordination when the second ligand is bonded to $Zn^{II}$.

*Ternary Complexes.*—Calculated complex-formation constants for ternary complexes of the two substituted histidines with D- and L-histidine and $Ni^{II}$, $Cu^{II}$, and $Zn^{II}$ are given in Table 3 together with some derived quantities. The only protonated ternary complexes of any significance were these of $Cu^{II}$.

TABLE 3

Formation constants for ternary complexes of formula $[M_w(D/L\text{-His})_xL_yH_z]$ at 25 °C and $I = 0.10$ mol dm$^{-3}$ ($K[NO_3]$)

| | | bzh | | dbzh | |
|---|---|---|---|---|---|
| Metal ion | | D-His | L-His | D-His | L-His |
| $Ni^{II}$ | log $\beta_{1110}$ | 16.523(3) | 16.060(8) | 16.106(7) | 15.36(1) |
| | $\Delta$ log $K$ | −0.97 | −1.43 | −0.58 | −1.32 |
| | log $X$ | 1.38 | 0.45 | 1.63 | 0.03 |
| | $\Delta$ log $\beta_{1110}$ | −0.46 | | −0.74 | |
| $Cu^{II}$ | log $\beta_{1111}$ | 23.993(1) | 24.111(2) | 22.73(1) | 22.797(8) |
| | log $\beta_{1110}$ | 18.587(1) | 18.586(2) | 17.41(2) | 17.24(2) |
| | $\Delta$ log $K$ | −1.72 | −1.72 | −1.66 | −1.83 |
| | log $X$ | 0.051 | 0.051 | −0.02 | −0.36 |
| | $\Delta$ log $\beta_{1111}$ | 0.12 | | 0.25 | |
| | $\Delta$ log $\beta_{1110}$ | 0 | | −0.17? | |
| $Zn^{II}$ | log $\beta_{1110}$ | 12.583(9) | 12.27(1) | 12.282(6) | 12.037(8) |
| | $\Delta$ log $K$ | −0.45 | −0.76 | +0.09 | −0.16 |
| | log $X$ | 0.99 | 0.36 | 1.07 | 0.58 |
| | $\Delta$ log $\beta_{1110}$ | −0.31 | | −0.25 | |

The apparent stabilization of some ternary complexes has been considered in detail by Sigel.[12] Following his convention, the quantitative stabilization of ternary complexes may be expressed in two ways. One considers the disproportionation constant, $X$, expressed as in equation (4); hence log $X = 2 \log \beta_{MAB} - \log \beta_{MA_2} - \log$

$$MA_2 + MB_2 \longrightarrow 2MAB, \qquad X = [MAB]^2/[MA_2][MB_2] \quad (4)$$

$\beta_{MB_2}$. For statistical reasons the value expected for $X$ is 4, *i.e.* log $X = 0.60$. The second method compares the differences in stability ($\Delta\log K$) for the reactions of MA + B and M + B. It therefore corresponds to reaction (5); hence $\Delta\log K = \log \beta_{MAB} - \log \beta_{MA} - \log$

$$MA + MB \longrightarrow MAB + M \quad (5)$$

$\beta_{MB}$. Since there are more available co-ordination sites on the hydrated metal ion than on the mono complex, it would normally be expected that $\Delta\log K$ would be negative, generally between −0.5 and −2.0.[12] A positive value suggests significant stabilization of the ternary complex.

Values for log $X$ and $\Delta\log K$ are given in Table 3 and demonstrate that none of the ternary complexes shows significant stabilization as measured by $\Delta\log K$, with the possible exception of the $Zn^{II}$–dbzh–His system. This behaviour parallels the finding in the binary zinc(II) complex of the substituted histidine when the second stepwise constant was as large as the first. The absence of stabilization in the complexes with $Cu^{II}$ is compatible with the explanations given by Sigel[12] for possible stabilization of ternary complexes.

Stereoselectivity was found to be common in these ternary complexes. Stereoselectivity may be expressed quantitatively as in equation (6). With $Ni^{II}$ the *meso*-ternary complexes were the more stable, stabilization of

$$\Delta\log \beta_{1110} = \log \beta(L\text{-}L)_{1110} - \log \beta(L\text{-}D)_{1110} \quad (6)$$

the species with bzh being $\Delta\log \beta_{1110} = -0.46$ compared to a value of −0.48 in the bis(His) complexes.[1] With dbzh the stabilization is greater (−0.74). There is now extensive evidence for stereoselectivity in bis(histidinato)nickel(II) complexes. Apart from potentiometric[1,4] and calorimetric data,[5] there is kinetic evidence,[13] and in all cases the *meso* complex is preferred. This suggests a common reason for the stereoselectivity, and the fact that stereoselectivity is greatest with the most heavily substituted ligand (dbzh) reinforces an explanation in terms of steric hindrance.

With $Cu^{II}$, stereoselectivity between the unprotonated species ($\Delta\log \beta_{1110}$) is zero with bzh, just as it is zero in the binary His complexes.[1] The value for dbzh is small, favouring the *meso* complex, but it is associated with a comparatively large error [$\Delta\log \beta_{1110}$ $-0.17 \pm 0.08$ (*i.e.* $2\sigma$)]. Stereoselectivity between the monoprotonated complexes (1111 species) is significant and, in both cases, in favour of the optically active complex. This is identical to the trend found with His itself[1,4] and with other ternary complexes of $[Cu(HisO)]^{+}$ with ligands containing positively charged substituents.[6] A similar stabilization has also been reported in complexes of $Cu^{II}$ with N-substituted histidines[4] when the following values for

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[12] H. Sigel, *Angew. Chem. Internat. Edn.*, 1975, **14**, 394.

[13] J. E. Hix and M. M. Jones, *J. Amer. Chem. Soc.*, 1968, **90**, 1723; J. R. Blackburn and M. M. Jones, *J. Inorg. Nuclear Chem.*, 1973, **35**, 1605.

$\Delta\log\beta_{121}$ were reported: *N*-methylhistidine, 0.072; *N*-benzylhistidine, 0.133. The *N*-benzyl group is responsible for stereoselectivity in the ternary complex [Cu(benzylproline)(valine)] [14] and in [Cu(benzylproline)$_2$].[15] The stereoselectivity found in the ternary 1110 complex with dbzh may well, therefore, result from a similar steric effect.

With Zn$^{II}$ the stereoselectivity is in favour of the *meso* complex and is therefore comparable to that found in [Zn(HisO)$_2$] complexes.[1] Increased substitution of the ternary ligand does not appear to increase the stereoselectivity as it did with Ni$^{II}$. This is presumably a result of the changes in co-ordination geometry, which appear to cause the stabilization of the mixed ternary complex incorporating the heavily substituted dbzh.

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[14] V. A. Davankov and P. R. Mitchell, *J.C.S. Dalton*, 1972, 1012.

[15] G. G. Aleksandrov, Tu. T. Struchov, A. A. Kurganov, and V. A. Davankav, *J.C.S. Chem. Comm.*, 1972, 1328.