A Model for the Cobalamin-Dependent Methionine Synthase

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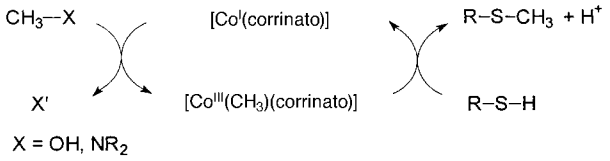
The acid-catalyzed transfer of a Me group from *N*,*N*-dimethylaniline (**6**) to vitamin-B_{12}-derived Co^{I} complexes **2a**,**b** was realized (*Scheme 3*). Hexane-1-thiol (**8**) was methylated by the methylcobalt complexes **4a**,**b** in the presence of pyridine. Conditions for the complete cycle, *i.e.*, Me transfer from **6** to **8** with Co^{I} complexes acting as a nucleophile and a nucleofuge have been established. The importance of Zn^{2+} as activating agent and of the basicity of tertiary amines for the Me transfer has been investigated.

Introduction. – Vitamin-B_{12}-dependent catalysis is observed in prokaryotes as well as in eukaryotes with methyl transferases and mutases playing important rôles [1–3]. Methylmalonyl-succinyl mutase and methionine synthase catalyze the two vitamin-B_{12}-dependent reactions essential in mammalians. In the methionine synthase reaction, a Me group from tetrahydro-N^5-methylfolate (= *N*-{4-{[(2-amino-1,4,5,6,7,8-hexahydro-5-methyl-4-oxopteridin-6-yl)methyl]amino}benzoyl}-L-glutamic acid) is used for the synthesis of methionine with a methylcobalt complex as an intermediate [4][5]. *Matthews*, *Ludwig* and coworkers have recently investigated the vitamin-B_{12}-dependent methionine synthase from *Escherichia coli* and provided solid information about the structure and reactivity of the holoenzyme [1]. The catalytic cycle consists of two half-reactions involving first the transfer of the Me group from tetrahydro-N^5-methylfolate to cob(I)alamin. The methylcob(III)alamin reacts further with homocysteine to give methionine in the second half-reaction (*Scheme 1*).

Scheme 1



The mechanism of activation of the tetrahydro-N^5-methylfolate for transfer of the Me group to cob(I)alamin is still speculative [4]. Protonation at N(5), one-electron oxidation, and two-electron oxidation have been proposed for this activation. The protonation at N(5) seems to be more reasonable than an oxidative pathway, because an oxidative activation would require an enzymatic electron acceptor with a high positive redox potential[1]). For the second half-reaction, a nucleophilic substitution of

[1]) Model reactions for Me transfer from tetrahydro-N^5-methylfolate *via* oxidation have been unsuccessful [6], while Me transfer from $RNMe_3^+X^-$ or $R_2NMe_2^+X^-$ compounds was feasible [6–8].

the thiolate with the methylcob(III)alamin and a radical-induced transfer reaction are discussed [9]. Since the Me transfer occurs with overall retention of configuration, the two half-reactions should each proceed with inversion of configuration compatible with two S_N2 reactions [10][11]. Under *in vivo* conditions, the cob(I)alamin is occasionally oxidized to cob(II)alamin, which itself is catalytically inactive and must be recycled to the active state by reduction of Co^I and methylation. This is achieved by reaction with reduced flavodoxin and adenosyl-methionine [1]. Recently *Matthews* and coworkers provided compelling evidence for involvement of Zn^{2+} in the activation of homocysteine [12–14].

The first half-reaction of methionine synthase has an analogy in methyl transferase, where MeN- as well as MeO-containing substrates are used for the transfer of the Me group to a [Co^I(corrinato)] complex. Typical Me sources used by microorganisms are MeOH or methyl aryl ethers [10][15–18]. In these cases, the corrinato(methyl)cobalt formed as an intermediate is further used to generate CH_4, MeCOOH, or 5-methylguanine, or to transfer the Me group to 2-mercaptoethanesulfonic acid (coenzyme M) [3][17]. *Thauer* and coworkers pointed out that Zn^{2+} ions are an essential cofactor for the Me transfer from MeOH to coenzyme M [17]. The importance of Zn^{2+} as a cofactor in biotransformations has been recognized by other authors as well [13][18–22].

The discovery of transmethylation and the elucidation of the biosynthetic pathway by which methionine is formed, has led to numerous model studies and mechanistic propositions for this intriguing corrinatocobalt-dependent reactions [23][24]. The transfer of a Me group from a substrate Me–X (X = R_2N, OH, or RO) to a thiolate, with a corrinatocobalt(I) or another Co complex serving as shuttle, has been dissected into the two half-reactions (*Scheme 1*).

While the early experiments for inducing transfer of a Me-group from an N-atom, *i.e.*, tetrahydro-N^5-methylfolate, tertiary amines or quaternary ammonium salts, to vitamin B_{12s} or B_{12r} were unsuccessful [25], *Pandit* and coworkers showed that Cob(I)aloxime and Cob(I)alamin (= vitamin B_{12s}) can be methylated by an N^5-methylpterinium salt, bearing a quaternary Me group [7]. Subsequently, *Pratt* and coworkers reported the successful methylation of cob(I)alamin by a trimethylanilinium cation [8]. Transfer of Me was also observed from 1-methyl-1-alkylpiperidinium cations to an arenethiolato-cobaloxime by *Tada* and coworkers [27]. Thus, we are not aware of any examples where secondary or tertiary amines were successfully used for Me transfer to corrinatocobalt complexes.

The first model for the Me transfer from MeOH was recently described by us [28]. We reported that heptamethyl cob(II)yrinate **2a** can be methylated by MeOH in the presence of $NaBH_4$ used for the reduction of Co^{II} complex **2a** to Co^I complex **3a**, and in the presence of Zn^{2+} ions used for activation of the leaving Me group. Further investigations showed that the Me transfer from MeOH or *N*,*N*-dimethylaniline can be achieved electrochemically under acidic conditions [29].

While the first half-reaction is best described as a nucleophilic substitution with the substrate bearing the activated Me group and the supernucleophilic [Co^I(corrinato)] complex; it was not clear whether the Me transfer from the Co-atom to the thiol involves a nucleophilic attack by a thiolate anion, or a reactive R–S radical and homolysis of the Co–C bond [9][30]. In 1963, *Johnson* and coworkers [31] suggested a

homolytic cleavage of the C–Co bond because methionine was found when methylcobalamin was photolyzed in the presence of homocysteine. *Schrauzer et al.* postulated in 1968 a nucleophilic attack of a thiolate anion on the [Co(Me)] complex [32–34]. Results at variance with this proposal [35][36] led *Hogenkamp et al.* to reinvestigate this reaction in 1985 [37]. They proposed a mechanism which involves the nucleophilic reaction of a thiolate with the methylcobalt complex, leading to methionine and cob(I)alamin.

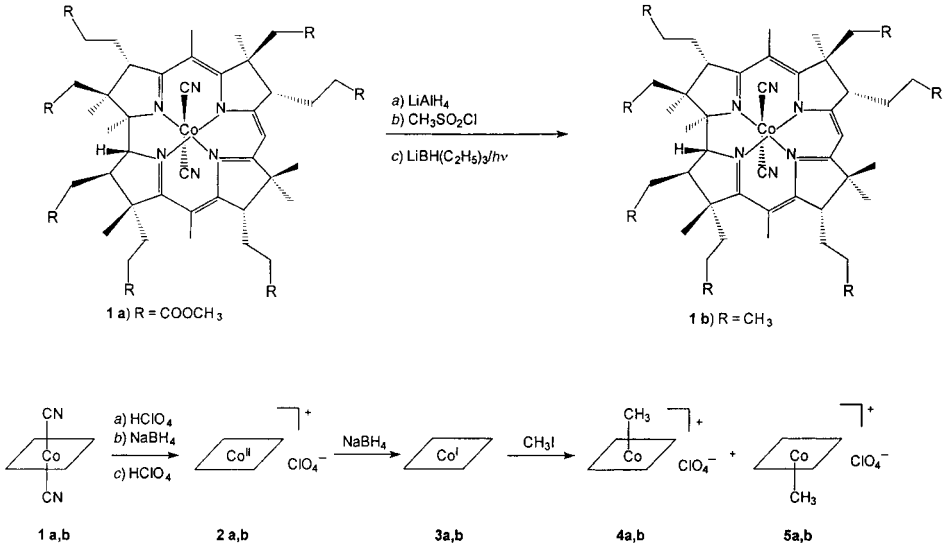
We developed a model system for the two half-reactions of the methionine synthase and explored reaction conditions under which the complete cycle involving the transfer of a Me group from a tertiary amine to thiol with [Co^{I}(corrinato)] complexes as mediators could be established.

Results and Discussion. – *Preamble.* For the first half-reaction, we formulated a S_N2 reaction between an amine bearing the Me group and the supernucleophilic [Co^{I}(corrinato)] complex (see **3a**,**b** in *Scheme 2*). Our major concern was the activation of the substrate for the Me transfer under conditions compatible with the fast decay of the [Co^{I}(corrinato)] complex under protic and acidic conditions. We considered Zn^{2+} to be a sufficiently strong *Lewis* acid for complexation with a tertiary methylamine, in the presence of $NaBH_4$ or Zn^0 used as reducing agents, for the [Co^{II}(corrinato)] **2a**,**b**. Ideally the reducing agent should not lead to reductive cleavage of the [Co(Me)-(corrinato)] formed (see below). The leaving-group ability of the Me-group-bearing amine must be adjusted in such a way, that the Me transfer competes efficiently with the oxidative pathways available for the Co^{I} complex.

For the second half-reaction, we again assumed a S_N2 reaction by which the Me transfer from the [Co(Me)(corrinato)] complex to the thiol should proceed. This implies that the [Co^{I}(corrinato)] behaves not only as a supernucleophile – required for the first half-reaction – but also as an efficient leaving group, analoguous to the well known reactivity of I^- in substitution reactions [38]. In addition, the nucleophilicity of the thiolate – related to the pK_a of the thiols – must be considered. An appropriate solvent must be selected for both half-reactions. Although a variety of Co complexes were developed for modelling the [Co(corrinato)] moiety of vitamin B_{12}, we chose the vitamin-B_{12}-derived [Co(corrinato)] complexes **1a**,**b**, rather than synthetic Co complexes, for this study [39–47]. We surmised that the different ligand field and charge in cobaloximes and salen-derived Co complexes might lead to a decrease in the catalytic efficiency to such an extent that one or both half-reactions might not occur at all. In addition, the solubility of **2a**,**b** in a variety of organic solvents should allow the efficient separation and identification of the [Co(corrinato)]-derived products. Finally, it was hoped that the judicious choice of the parameters for both half-reactions might lead to the complete cycle. This was indeed achieved, though the yield was rather modest. The Co complexes **2a**,**b** and the reference complexes used in this study were prepared as shown in *Scheme 2*.

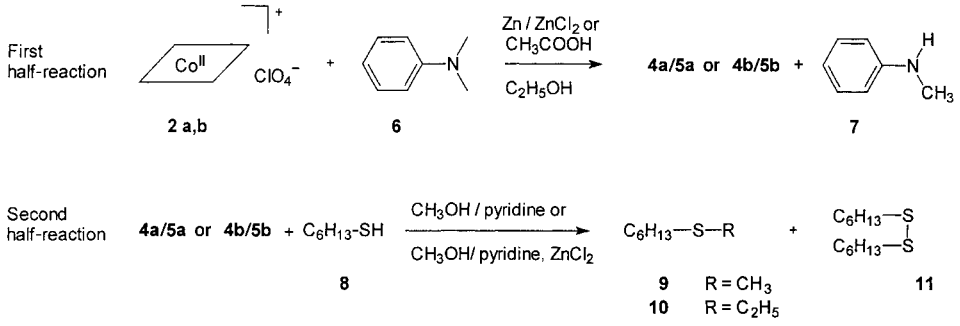
First Half-Reaction. When a solution of *N*,*N*-dimethylaniline (**6**) and heptamethyl cob(II)yrinate **2a** in EtOH/AcOH was stirred with Zn powder for several hours at room temperature, the methylcobalt complexes **4a**/**5a** and *N*-methylaniline (**7**) were formed (*Scheme 3*, *Table 1*). Reduction of **2a** by other reducing agents such as titanium(III) citrate or $NaBH_4$ did not lead to methylation of the [Co^{I}(corrinato)]

Scheme 2



complex. Zn/AcOH as reducing agent gave the best results, **4a** being formed in 10% yield. Other light-sensitive compounds were also formed, but were not identified. An inter- or intramolecular Me transfer from one of the methoxycarbonyl groups of the heptaester **2a** could be excluded, because *N*-methylaniline (**7**) was identified in 0.5% yield[2]). It is noteworthy that no aniline was detected under these conditions.

Scheme 3



Furthermore, the (heptaalkylcorrinato)cobalt **2b** gave **4b** and **5b** in 29% yield (**4b**/**5b** 21:7) and 12.5% of **7**[3]) under the same conditions. The modest yields of the methylcobalt complexes **4a**/**5a** and **4b**/**5b** might be due to their limited stabilities under

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[2]) The GC yield of **7** is based on the 100-fold excess of **6** used in this reaction; based on the amount of **2a** used, it corresponds to a yield of 50% and a turnover number of 0.5.

[3]) For this reaction, a 50-fold excess of **6** was used. Based on the amount of the corrinatocobalt **2b** used, this yield corresponds to 625% or 6 turnovers.

Table 1. *Methyl Transfer from* N,N-*Dimethylaniline* (**6**) *to the Co*^{I} *Complex* **3a**, *Obtained from* **2a** *with Zn as Reducing Agent*, EtOH/AcOH 5 : 1

Entry	Solvent	Ratio **2a**/**6**	Additive	Time [h]	Yield of **4a** [%][a])
1	EtOH/AcOH	1 : 5	–	6	5
2	EtOH/AcOH[b])	1 : 5	–	4	[c])
3	EtOH/AcOH	1 : 5	–	2	8
4	EtOH/AcOH	1 : 100	–	2	10
5	EtOH	1 : 5	$ZnCl_2$	2	6
6	EtOH	1 : 5	AIE[d])	2	[c])
7	EtOH	1 : 5	NH_4Cl	2	2

[a]) Yield calculated from ^{1}H-NMR integration of the methylcobalt complex **4a** formed. [b]) Reaction under reflux. [c]) <2% of product. [d]) AIE = anion-exchange resin.

the reaction conditons. Indeed, when the mixture **4a**/**5a** and Zn in the presence of $ZnCl_2$ or AcOH was kept in MeOH at room temperature, it decomposed rather rapidly[4]).

Second Half-Reaction: Methyl Transfer from the Co-Atom to Thiol. When the mixture **4a**/**5a** was treated with hexane-1-thiol (**8**) in MeOH in the presence of pyridine for 24 h at room temperature, no Me transfer was observed. At 50°, the reaction proceeded slowly and gave the hexyl methyl sulfide (**9**) in 4% yield (*Fig. 1*). Increasing the temperature to reflux yielded 37% of **9**. Upon addition of $ZnCl_2$, Me transfer occurred with 69% yield. With *N*,*N*-dimethylaniline (**6**) instead of pyridine, the yield of **9** decreased to 23%, even in the presence of $ZnCl_2$. In view of the rather rapid decomposition of **4a**/**5a** in the presence of Zn, the decrease for Me transfer was not surprising when Zn and $ZnCl_2$ were used as additives (*Table 2*). Me Transfer from MeOH (used as the solvent) *via* **4a**/**5a** could be excluded, because no methyl sulfide **9** could be detected when a mixture of the Co^{II} complex **2a**, $ZnCl_2$, Zn, thiol **8**, and pyridine in MeOH was refluxed for 72 h.

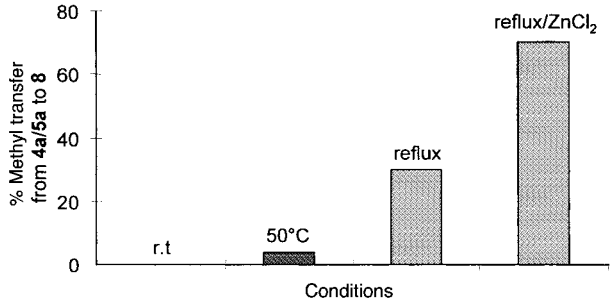


Fig. 1. *Second half-reaction: Methyl Transfer from the Co-Atom of* **4a**/**5a** *to thiol* **8**. Conditions: **4a**/**5a** as catalyst, 10-fold excess of **8**, 5-fold excess of pyridine, MeOH, 24 h, in the dark.

With the peralkylated methylcobalt complexes **4b**/**5b**, Me transfer to give hexyl methyl sulfide (**9**) was achieved in 58% yield in the presence of $ZnCl_2$. Direct methylation of the thiol **8** by MeOH, used as solvent, in the presence of $ZnCl_2$ could be

4) Decomposition: 50% after 1 h, 65% after 3 h, and 95% after 7 h; according to the ^{1}H-NMR data, the corrin ring of **2a** as well as of **2b** was modified under the reaction conditions and lost their transfer activity.

Table 2. *Methyl Transfer from* **4a**/**5a** *to Hexanethiol* **8** *in the Presence of 5 mol-equiv. of Pyridine* (24 h reflux in MeOH)

Entry	Ratio **4a** + **5a**/**8**	Additives	*T* [°C]	Yield of **9** [%][a])
1	1:10	–	r.t.	0
2	1:10	–	50	4
3	1:10	–	reflux	37
4	1:10	Zn, $ZnCl_2$	reflux	28
5[b])	1:10	$ZnCl_2$	reflux	33
6	1[c]):10	$ZnCl_2$	reflux	58
7	0:10	$ZnCl_2$	reflux	0
8	1:10[d])	$ZnCl_2$	reflux	0
9	1:10	$ZnCl_2$	reflux	69
10	0:10	Zn, $ZnCl_2$	reflux	0

[a]) GC Yield. [b]) In EtOH. [c]) **4b**/**5b** instead of **4a**/**5a** was used. [d]) **11** instead of **8** was used as S-compound.

excluded because no methyl sulfide **9** was formed in the absence of any methylcobalt complex. In all of these experiments, we observed the formation of dihexyl disulfide (**11**) as a by-product. To exclude the possibility that **11** reacted with the methylcobalt complex **4a/5a**, we ran the reaction with dihexyl disulfide (**11**) instead of thiol **8** as the only S-compound. However, under the latter conditions, no hexyl methyl sulfide (**9**) was formed. A reversible reaction also did not take place, since on treatment of **2a** with **9** under the reaction conditions used for the complete catalytic cycle (see below), neither the methylcobalt complexes **4a**/**5a** nor dihexyl disulfide (**11**) could be detected. However, methylation of the Co^I-complex **3a**, formed by reduction of **2a**, with dimethylhexylsulfonium *p*-toluenesulfonate could be achieved in high yield at room temperature. Furthermore, it was of interest to establish whether $ZnCl_2$ would affect the lifetime of the Co^I complex formed from **2a**. When the Co^I complex **3a**, prepared by electrochemical reduction of **2a** in EtOH, was kept at room temperature in the dark, the UV/VIS spectrum of **2a** was reconstituted within 20 min in the absence of $ZnCl_2$, whereas it took 40 min in the presence of this *Lewis* acid.

As mentioned above, *Johnson* and coworkers suggested a homolytic cleavage of the intermediate methylcobalt complex for the Me transfer to homocysteine [31]. Therefore, we photolyzed a mixture **4a**/**5a** with an excess of pyridine and hexanethiol **8** with a 150-W sun lamp at room temperature or under reflux. However, no Me transfer to the thiol could be detected under these conditions. Also, **9** was not formed when the disulfide **11** was used instead of **8**.

Complete Cycle. For the complete cycle, we considered the possibility of using the methylcobalt complexes **4a**/**5a** as catalyst: In a first reaction step, the Me group would be transferred to the thiol forming the methyl sulfide **9** and a Co^I complex **3a**. The Co^I complex in turn would react as a supernucleophile with the activated methylamine leading to **4a**/**5a** and completion of the catalytic cycle (*Scheme 4*).

Since these experiments did not lead to the demethylation of *N*,*N*-dimethylaniline (**6**), we considered an *in situ* formation of the Co^I complex as the starting reaction. Indeed, when the perchloratocobalt(II) complex **2a** was reduced with Zn in the presence of $ZnCl_2$, the demethylation of *N*,*N*-dimethylaniline (**6**) was observed with the concomitant formation of hexyl methyl sulfide (**9**) in 2–6% yield (*Table 3*).

Scheme 4

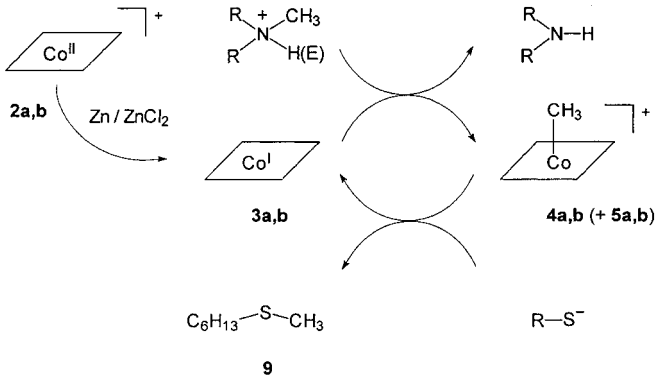


Table 3. *Complete Cycle with* **2a**, N,N-*Dimethylaniline* (**6**), *Hexanethiol* **8**, *and Zn as Reducing Agent and* $ZnCl_2$ *as* Lewis *Acid in EtOH at Reflux*

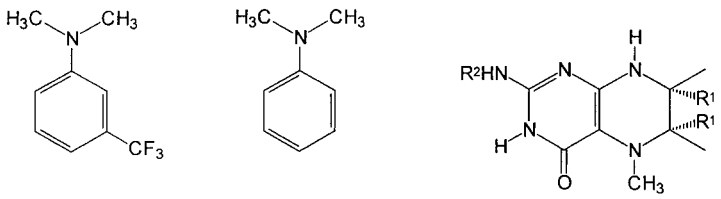
Entry	Solvent	Ratio **2a**/**8**/**6**	Time [h]	Yield [%][a])	
				9	**10**
1	EtOH	1 : 40 : 45	24	5.9	0.8
2	EtOH[b])	– : 40 : 45	165	0	0
3	EtOH	1 : 40 : 135	24	0.7	0.4
4	EtOH[c])	1 : 48 : 43	24	0	0
5	EtOH	1 : 40 : 22.5	48	2.2	0.9
6[d])	EtOH	1 : 40 : 45	24	0.7	0.6
7	EtOH	1 : 40 : –	165	0	0
9	EtOH	1 : 40[e]) : 45	72	0	0
10	i-PrOH	1 : 40 : 45	24	1.3	0
11	$CHONEt_2$	1 : 40 : 45	24	1.1	0
12	THF	1 : 40 : 45	24	0.7	0

[a]) GC yield with internal standard. [b]) Reaction without **2a**. [c]) No Zn added. [d]) **2b** instead of **2a** was used. [e]) Thiophenol instead of **8** was used.

Similarly, the corrinatocobalt(II) complex **2b** acted to shuttle the Me group from *N*,*N*-dimethylaniline (**6**) to hexanethiol **8**. However, under the same reaction conditions (24 h reflux in EtOH) only 3.8% of **7** and 0.7% of **9** were detectable. In view of the efficiency of the first half-reaction with **2b**, this rather low yield in the complete cycle seemed surprising. However, the corrinatocobalt complexes **2b** and **4b**/**5b** were rather labile compounds, and the reaction conditions under which the complete cycle was performed were different from those of the two separate half-reactions.

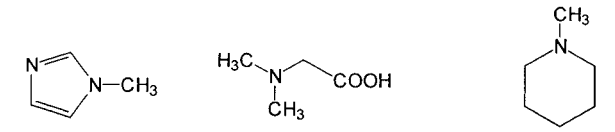
From the solvents tested (EtOH, i-PrOH, THF, and DMF), EtOH gave the highest yield, but it also led to the formation of ethyl hexyl sulfide (**10**) as a by-product. A control experiment showed that, in the absence of *N*,*N*-dimethylaniline (**6**), no methyl sulfide **9** was formed.

After the successful realization of the complete catalytic cycle, the question of the transfer ability of various methylamines was addressed. For this investigation, the methylamines **12**–**16** with different pK_a values were chosen and their Me-transfer activity compared with that of *N*,*N*-dimethylaniline (**6**). The yield of hexyl methyl



pK_a 2.1 [48] pK_a 5.1 [49] pK_a (**13a**) 5.7 [50]

12 **6** **13a** $R^1 = CH_3$, $R^2 = H$
b $R^1 = H$, $R^2 = C(O)C(CH_3)_3$



pK_a 7.2 [51] pK_a 9.8 [52] pK_a 10.1 [53]

14 **15** **16**

sulfide (**9**) was highest (*ca.* 6%) when **6** with a pK_a of *ca.* 5 was used, which is comparable to that of N^5-methyltetrahydrofolate (pK_a 4.8 [54]).

A further aspect of the model system concerns the methylation of different thiols. We had assumed that the thiol group in homocysteine and hexanethiol have comparable nucleophilic and redox properties and expected appropriate activity in our model system. As shown above, this assumption has been realistic. Thus, when hexanethiol **8** (p$K_a \approx 10.7$ [55]) was replaced by thiophenol (pK_a 6.8 [55]), no thioanisol was formed under the reaction conditions of the complete catalytic cycle. If the nucleophilicity of the thiolates in the Me-transfer reaction from the methylcobalt complex is related to their basicity, thiophenol should react more slowly than hexanethiol **8**, while the decomposition of **4a**/**5a** still proceeds at a constant rate.

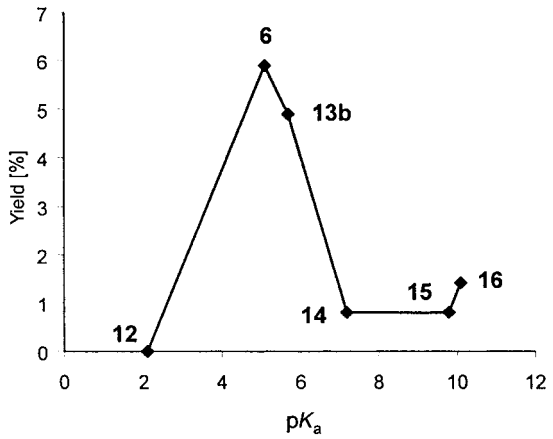


Fig. 2. *Complete cycle with* **2a**: *yield of* **9** *in relationship to the* pK_a *of the amines* **6** *and* **12**–**16**. Standard reaction conditions.

Conclusion. – Based on the mechanistic assumption that the Me-transfer from tetrahydro-N^5-methylfolate to homocysteine *via* methylcob(III)alamin proceeds by two S_N2 reactions, with cob(I)alamin as a supernucleophile and as a 'super'-nucleofuge, and an activation of both half-reactions by Zn^{2+} ions, we developed a model reaction for the complete catalytic cycle. Under carefully controlled conditions, the two half-reactions, as well as the complete catalytic cycle, were realized. Although a temperature much higher than that of the enzymatic system was required, and the yield is still rather modest, we consider our results an important contribution to the understanding of possible reaction pathways for the methionine synthase. The importance of Zn^{2+} ions as activating agent and the effect of the basicity of the tertiary methylamine on the reactivity have been demonstrated.

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Experimental Part

General. The reactions were carried out with reagents and solvents of *puriss.* grade under Ar. Hexane-1-thiol (**8**; *Fluka*), dihexyl disulfide (*Lancaster*), and dibutyl sulfide (*Aldrich*) were purified by distillation and checked for purity by GC and NMR. *N,N*-Dimethylaniline (**6**; >99.5%), *N*-methylaniline (**7**; >98%), *N,N*-dimethylglycine (**15**; >99%), thiophenol (>99%), 1-methyl-1*H*-imidazol (**14**; >99%, *Fluka*), and 1-methylpiperidine (**16**; Aldrich, 99%) were checked for purity by GC and NMR and used as received. The solns. were degassed by sonicating under reduced pressure. Column (CC) and flash chromatography (FC): distilled commercial-grade solvents, CH_2Cl_2/THF/Et_2O 2:1:3 for CC, silica gel (40–63 μm) from *Fluka* (analyzed reagents). TLC: *Merck-F-254* precoated sheets, CH_2Cl_2/THF/Et_2O 2:1:3, visualization by 5% phosphomolybdic acid hydrate/EtOH or by UV. GC: *Hewlett-Packard-HP-5890* instrument; *HP-5-Ultra* capillary column (length 10 m, i.d. 0.2 mm), 5% phenylpolysiloxane; temp. program 40–270° (6°/min). UV/VIS: *Hewlett-Packard-8451-A* diode-array spectrophotometer in MeOH; λ_{max} (log ε) in nm. IR: *Perkin-Elmer-PE-782* spectrometer; $CHCl_3$ soln. in 0.2-mm path NaCl cells; in cm^{-1}. NMR: *Bruker-AC-300* (^1H, 300 MHz; ^{13}C, 75 MHz) and *Bruker-AC-500* (^1H, 500 MHz; ^{13}C, 125 MHz) instruments: δ in ppm (internal lock, $CDCl_3$ (δ(H) 7.24, δ(C) 77.00)), *J* in Hz; ^{13}C multiplicities from DEPT spectra. MS: *Varian MAT-CH-7A*, 70 eV; in *m/z* (%). FB-MS: *Fision Autospec-Q*, acceleration voltage 8 kV, ionization Cs^+ (32 keV); matrix: dithiothreitol (DTT)/dithioerythriol(DTE); in *m/z* (%). ESI-MS: *Fisions Instrument VG Platform II*, positive-ion measurements (3.5 kV); negative-ion measurements (2.5); in *m/z* (%) in the solvents given.

*Heptamethyl- [*Coα, Coβ-*Di(cyano-*κC)*]cob(III)yrinate* (**1a**) [56]. This compound was prepared from 20.0 g (14.76 mmol) of vitamin B_{12} (commercial cyanocob(III)alamin): 11.4 g (71%) of **1a**.

*Heptamethyl (*Co-*Perchlorato)-cob(II)yrinate* (**2a**) [57]. This compound was prepared from 500 mg (0.46 mmol) of **1a** as reported: 453 mg (87%) of **2a**.

a,b,c,d,e,f,g-*Heptamethyl (*Coβ-*Methyl)(*Coα-*perchlorato)- and (*Coα-*Methyl)(*Coβ-*perchlorato)cob(III)yrinate* (**4a**/**5a**). The mixture of stereoisomeric methylcobalt complexes **4a**/**5a** (β/α) 6:1 was prepared as described in [57].

[Coα,Coβ-*Di(cyano-*κC)*](2,7,18-triethyl-1,2,5,7,12,12,15,17-octamethyl-3,8,13,17-tetrapropylcorrinato-*κN^{21}, κN^{22},κN^{23},κN^{24})cobalt* (**1b**). Reduction of 2.0 g (1.83 mmol) of **1a** with $LiAlH_4$ according to [58] gave 1.58 g (91.5%) of heptol. Reaction of this heptol with mesyl chloride according to [59] gave 1.03 g (39.5%) of heptamethanesulfonate. Reaction of the latter (1.03 g, 0.72 mmol) in the presence of 62 ml of 0.1M $LiHB(C_2H_5)_3$ in THF according to [58] gave 0.213 g (39%) of **1b**.

Perchlorato(2,7,18-triethyl-1,2,5,7,12,12,15,17-octamethyl-3,8,13,17-tetraproylcorrinato-κN^{21},κN^{22},κN^{23},κN^{24})-*cobalt* (**2b**) was prepared from **1b** (0.16 g, 0.2 mmol) as described above for **2a**: 0.156 g (92%) of **2b**: TLC (CH_2Cl_2/MeOH 99:0.5; impregnated with $NaClO_4$): R_f 0.17. IR: 3028v*s*, 3014*s*, 2400*m*, 1520*w*, 1460*w*, 1424*m*, 1264v*s*, 1232*m*, 1208v*s*. UV ($c = 1 \cdot 10^{-5}$ M): 268 (16283), 318 (15284), 466 (6881), 486 (6542), 490 (6196), 578

(992), 582 (970), 586 (920), 594 (853). ESI-MS: 787.5(8), 773.6(15), 754.5(30), 728.5(100, $[M-OClO_3]^+$). FAB-MS: 881.34(2), 760.10(5), 728(38, $[M-OClO_3]^+$), 700.10(9), 656.30(8).

(Coβ-Methyl)(Coα-perchlorate)- and (Coα-Methyl)(Coβ-perchlorato)(2,7,18-triethyl-1,2,5,7,12,12,15,17-octamethyl-3,8,13,17-tetrapropylcorrinato-κN²¹,κN²²,κN²³,κN²⁴)cobalt (**4b**/**5b**). The methylations were carried out under Ar and under green light. After reduction of **2b** (20 mg, 0.024 mmol) with $NaBH_4$ (32 mg) in MeOH (10 ml) and addition of MeI (0.32 g, 1.62 mmol), the mixture was stirred for 25 min at r.t. For workup, phosphate buffer (pH 6; 20 ml) and $NaClO_4$ (20 mg) were added. The mixture (pH 6) was extracted with CH_2Cl_2 (3 × 10 ml), the org. phase dried ($MgSO_4$), and evaporated, and the residue chromatographed (silica gel (impregnated with $NaClO_4$), CH_2Cl_2/MeOH 98 : 2): 14.7 mg (72%) of **4b**/**5b** (β/α) 4.5 : 1. TLC (CH_2Cl_2/MeOH 98 : 2; impregnated with $NaClO_4$): R_f 0.55. IR: 3016*vs*, 2932*s*, 2254*m*, 1520*m*, 1488*m*, 1382*w*, 1348*w*, 1262*vs*, 1224*vs*. UV ($c = 10^{-5}$ M): 264 (18161), 304 (17008), 460 (7487), 586 (528), 592 (537). ^{1}H-NMR: 6.76 (*s*, 0.18 H (α)); 6.62 (*s*, 0.82 H (β)); 4.56 (*d*, 0.18 H (α)); 4.40 (*d*, 0.18 H (β)); 3.27 (*m*, 2.46 H (β)); 3.17 (*m*, 0.54 H (α)); 3.20–3.15 (*m*, 1 H); 2.44 (*s*, 3 H); 2.40–1.18 (*m*, overlapped by 2.33 (*s*, 3 H), 1.66 (*s*, 3 H), 1.62 (*s*, 3 H), 1.43 (*s*, 3 H), 1.31 (*s*, 3 H), 1.27 (*s*, 3 H), 25 H); 1.15–0.82 (*m*, 21 H); −0.20 (*s*, 0.54 H, α-Me); −0.27 (*s*, 2.46 H, β-Me). ^{13}C-NMR ($CDCl_3$): 0.71 (*q*); 8.85 (*q*); 9.60 (*q*); 13.76 (*q*); 14.24 (*q*); 14.40 (*q*); 14.48 (*q*); 15.47 (*q*); 16.09 (*t*); 16.26 (*q*); 18.28 (*t*); 18.54 (*q*); 20.03 (*q*); 20.16 (*q*); 20.57 (*t*); 20.72 (*t*); 21.01 (*t*); 24.00 (*t*); 24.81 (*q*); 29.67 (*t*); 30.03 (*t*); 32.18 (*q*); 32.76 (*t*); 32.86 (*t*); 33.60 (*t*); 34.48 (*t*); 44.76 (*d*); 46.46 (*s*); 46.90 (*s*); 51.23 (*s*); 53.93 (*d*); 56.16 (*d*); 56.41 (*d*); 59.39 (*s*); 76.21 (*d*); 86.46 (*s*); 96.28 (*d*); 99.98 (*s*); 106.58 (*s*); 107.18 (*s*); 163.75 (*s*); 165.59 (*s*); 173.24 (*s*); 175.03 (*s*); 176.93 (*s*); 177.20 (*s*). NOE ($CDCl_3$): irradiation at −0.27 → increase at 4.4 (*d*, H–C(19) (β)). ESI-MS: 777.45(5), 754.54(6), 743.50 (100 $[M-OClO_3^-]^+$), 728.61(6). FAB-MS: 881.34(2), 760.35(4), 745.10(30), 728.29 (100 $[M-Me-OClO_3]^+$), 700.10(7), 656.30(6).

Hexyl Methyl Sulfide (**9**). Hexane-1-thiol (**8**; 3.36 g, 28.42 mmol) was methylated according to [60]; 2.86 g (76%) of **9**. Colorless liquid.

Ethyl Hexyl Sulfide (**10**). As described for **9**, **8** (1 g, 8.46 mmol) was treated with EtI (1.7 ml, 16.92 mmol); 1.01 g (82%) of **10**. Colorless liquid.

N,N-*Dimethyl-3-(trifluoromethyl)aniline* (**12**). According to [48], 3-(trifluoromethyl)aniline (20 g, 124 mmol) was methylated: 14.2 g (60%) of **12**. Colorless liquid.

N-*(*cis*-3,4,5,6,7,8-Hexahydro-5,6,7-trimethyl-4-oxopteridin-2-yl)pivalamide* (**13b**). The 2,5,6-triaminopyrimidin-4-ol sulfate (12.86 g, 50 mmol) was transformed into 2-amino-6,7-dimethylpteridin-4-ol (9.48 g, 99%) according to [61]. Acylation with pivaloyl chloride yielded *N*-(4-hydroxy-6,7-dimethylpteridin-2-yl)pivalamide (11.39 g, 84%) [62], and hydrogenation of this compound over 10% Pd/C according to [63] gave 9.83 g (85%) of *N*-(*cis*-5,6,7,8-tetrahydro-4-hydroxy-6,7-dimethylpteridin-2-yl)pivalamide. Methylation [63] (2.0 g, 7.16 mmol) with MeI gave **13b** (1.42 g, 68%). White powder.

Methylation of **2a** *with* N,N-*Dimethylaniline* (**6**): *General Method*. All methylating reactions were carried out under Ar and green light. After reduction of **2a** with Zn, the additive was added and the mixture stirred at the specified temp. and for the time as given in *Table 1*. The pH was adjusted to 8 by addition of 0.1M NaOH and the mixture extracted with CH_2Cl_2 (3 × 10 ml). The org. phase was dried ($MgSO_4$) and evaporated and the residue chromatographed with CH_2Cl_2 for the amines and with MeOH/0.5% $NaClO_4$ for the corrinato complexes. The yield of **4a**/**5a** was determined from the ratio in the ^{1}H-NMR signals for the β- and α-methylcob(III)yrinate at −0.13 and −0.19 ppm and the sum of all H–C(10) signals at 5–7.2 ppm. *N*,*N*-Dimethylaniline (**6**) and *N*-methylaniline (**7**) were analyzed by GC.

Methylation of **2b** *with* **6**. As described for **2a**, **2b** (20 mg, 0.024 mmol) was reduced with Zn (0.1 g, 1.53 mmol) in EtOH/AcOH 5 : 1 (12 ml) for 20 min. Then, **6** (153 µl, 1.21 mmol) was added and the mixture stirred for 4.5 h at r.t. Workup and CC gave a yield of 28% (by ^{1}H-NMR) of **4b**/**5b** 3 : 1.

Methylation of Hexane-1-thiol (**8**) *with* **4a**/**5a** *or* **4b**/**5b**: *General Method*. a) *Reaction of the Dark:* A soln. of **4a**/**5a** (10 mg, 0.0087 mmol) or **4b**/**5b**, 5 mol-equiv. of pyridine, and 10 mol-equiv. of **8** in degassed solvent (10 ml) was heated after addition of the additives at the specified temp. for 24 h (*cf. Table 2*). The yield of **9** was determined by GC with dibutyl sulfide as internal standard.

b) *Reaction under Irradiation:* A soln. of **4a**/**5a** (23.5 mg, 0.0204 mmol), pyridine, and **8** (ratio of mol-equiv. 1 : 5 : 10 for the reaction at r.t., 1 : 10 : 10 for reflux) in degassed MeOH (10 ml) was irradiated with a 150-W sun lamp for 7 h at r.t. and 5 h at reflux until all **8** had reacted to **11**. No hexyl methyl sulfide (**9**) was found under these conditions.

Complete Catalytic Cycle: General Method. To a soln. of **2a** (10 mg, 8.8 µmol), Zn (50 mg), and $ZnCl_2$ (40 mg) in degassed solvent (40 ml), **6** and **8** were added in the ratio of mol-equiv. given in *Table 3*. The mixture was heated at reflux for the specified time. The amounts of hexyl methyl sulfide (**9**), excess **8**, ethyl hexyl sulfide

(**10**), and dihexyl disulfide (**11**) were determined by GC with dibutyl sulfide as internal standard. *N*-Methylaniline (**7**) was also determined by GC.

The reactions with the methylamines **12**–**16** were performed analogously.

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