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Bernhard Kräutler: A brief curriculum vitae is found in Chimia 41 (1987) 277. Areas of research: preparative and mechanistic electro-, photo-, radical-, and organometallic chemistry; chemistry of metallo-porphinoids and -corrinoids, and their functions as coenzymes.

Corrins and the Acetyl-CoA Path of Autotrophic Fixation of Carbon Dioxide in Bacteria**

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Autotrophic fixation of carbon dioxide in bacteria proceedes not only via the well-known «Calvin cycle», but also via the recently established «acetyl-coenzyme A pathway», meanwhile found to be widely distributed in bacterial anaerobes. In this linear path of carbon dioxide assimilation, a series of transition metal enzymes are engaged in the build-up of (the acetyl group of) acetyl-coenzyme A from carbon dioxide and molecular hydrogen. Corrins are given a central, if chemically still incompletely defined role in the crucial carbon-carbon bond forming step and correspondingly are abundant cofactors in the bacteria that follow this pathway. Detailed knowledge on the chemistry of this remarkable «organometallic» way for the fixation of carbon dioxide in nature not only is of interest in its own right, but also in view of the search for economic syntheses of basic organic chemicals and «fuels» from carbon dioxide.

1. A New Path for Carbon Dioxide Fixation

For the autotrophic fixation of carbon dioxide, the ultimate source of all cell carbon, only the rather intricate mechanism of the «Calvin cycle»[1] was established until recently (e.g. in plants[2] and bacteria[3]). However, with the discoveries in the microbiological laboratories of *Thauer, Wolfe, Gottschalk* and others, that methanogens (such as *Methanobacterium thermoautotrophicum*[4])[5] and acetogens (such as *Acetobacterium woodii*[4b] and *Clostridium thermoautotrophicum*[6]) can grow autotrophically from carbon dioxide and molecular hydrogen, the earlier picture eventually changed dramatically. Indeed in the last years three mechanisms for autotrophic fixation of carbon dioxide other than the «Calvin cycle» have been recognized:

(a) the «acetyl-coenzyme A pathway»[7];
(b) the «reductive tricarboxylic acid cycle»[8];
(c) the «glycine pathway»[9].

Fixation of carbon dioxide via the «acetyl-coenzyme A pathway» (see Fig. 1)[7,10,11] is now known to be widely distributed and to operate not only in acetogens, but also in autotrophic methanogens and in most autotrophic sulfate reducing bacteria[7b]. In its course, the acetyl group of acetyl-coenzyme A is first built up from carbon dioxide. Acetyl-coenzyme A then is transformed via reductive carboxylation and phosphoenol-pyruvate into glyceraldehyde-3-phosphate[7], a triosephosphate also of the «Calvin cycle»[1].

Central to this path of autotrophic fixation of carbon dioxide, is the assembly of the acetyl group of acetyl-coenzyme A, which involves a series of unusual (transition)metallo-enzymes (see Fig. 2)[7,10]: As concerns acetogenic bacteria, such as *Clostridium thermoaceticum* in particular, pioneering work on the assembly of acetyl-coenzyme A was done by *Wood* et al.[7a,11]. As is known now, in acetogens carbon dioxide is reduced to formate by the oxygen sensitive enzyme «formate dehydrogenase», containing in its active site a tungsten complex, that appears to be structurally related to the «molybdopterins»[10,12]. With the formate (produced this way or exogeneously supplied) tetrahydrofolate is formylated to give, after dehydration, N^5,N^{10}-methenyl-tetrahydrofolate. The latter is reduced to N^5-methyl-tetrahydrofolate[10,11]. With the methyl group of N^5-methyl-tetrahydrofolate, the cobalt center of an enzyme-bound corrin (5'-methoxybenzimidazolyl-cobamide) is methylated[11]. In a second branch of the pathway, carbon dioxide is reduced to carbon monoxide by the oxygen sensitive enzyme «carbon monoxide dehydrogenase», whose nickel-containing active site still is little characterized[13].

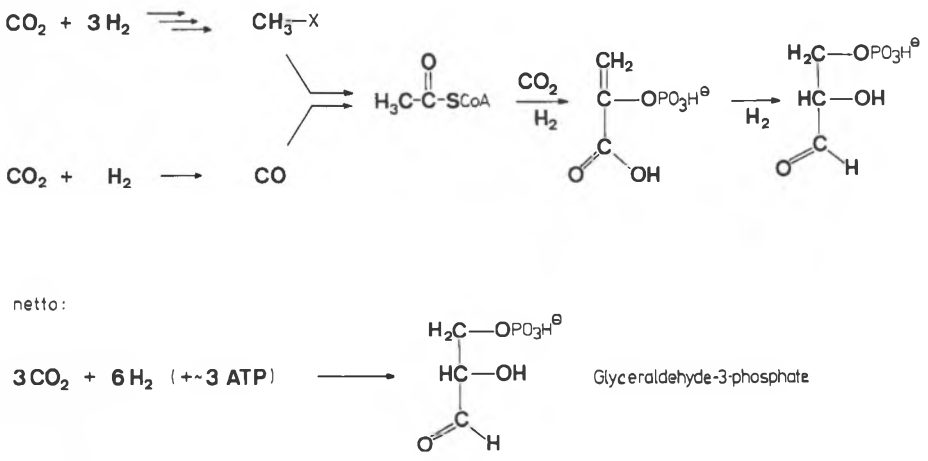

Fig. 1. Schematic representation of the «acetyl-coenzyme A pathway» for autotrophic fixation of carbon dioxide in anaerobic bacteria[7].

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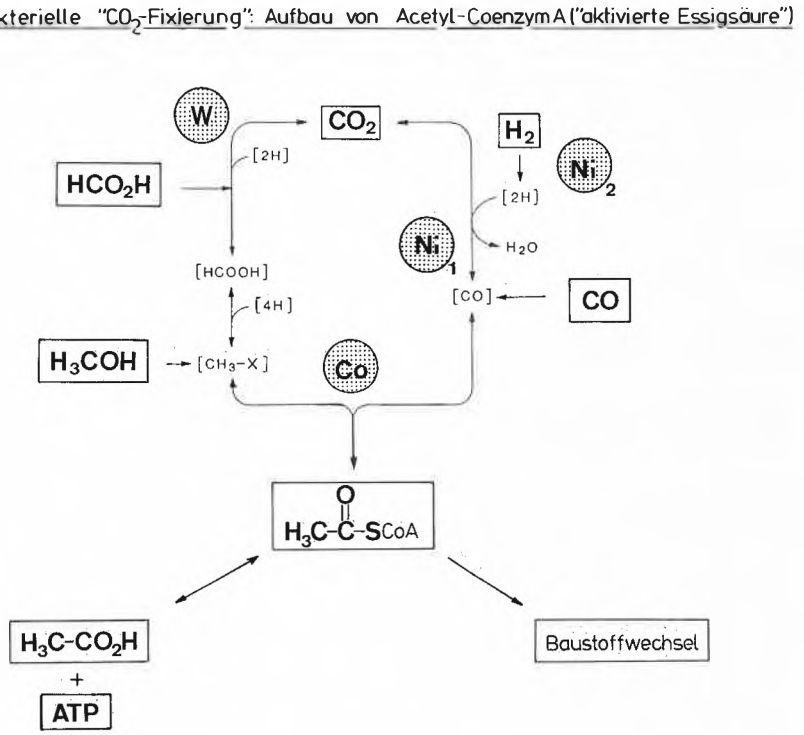

Fig. 2. Flow diagramm illustrating the assembly of acetyl-coenzyme A («activated acetic acid») from carbon dioxide and molecular hydrogen in anaerobic acetogens (symbols for the metallo-enzymes: **W** *= formate dehydrogenase;* **Ni_1** *= carbon monoxide dehydrogenase;* **Ni_2** *= hydrogenase:* **Co** *= corrinoid enzyme)* [7,10].

2. On the Role of Corrins

In a complex process, the intact methyl group from the corrin, carbon monoxide, and coenzyme A finally combine to acetyl-coenzyme A. Current interest centers on this latter set of reactions and on the question, in particular, on which metal site the hypothetical and crucial carbon-carbon bond forming step takes place[11,13] (see Fig. 3): On one hand, (e.g.) cyanide inhibition studies have suggested, that the nickel enzyme need not be directly involved in the carbon-carbon bond forming reaction, a carbonylation of the methyl-corrinoid[13] (which would produce an acetyl-corrinoid of suitable reactivity for the thiolytic formation of acetyl-coenzyme A[14]). The alternative scheme, where the cobalt-bound methyl group is first transferred to the nickel center of the «carbon monoxide dehydrogenase», likewise has some experimental support from a carbonyl exchange reaction of acetyl-coenzyme A, that is catalyzed (albeit with low activity) by the nickel enzyme (alone)[11,15].

Chemical precedent for the assembly of an acetyl group by a carbon-carbon bond forming reaction with the methyl group from a methyl-corrinoid so far has only been provided in the photoinduced carbonylation of methyl-cobalamin (**1**) to acetyl-cobalamin (**2**) (see Fig. 3 and 4)[14,16]. However, this chemical carbonyl-insertion reaction at the corrin-bound cobalt center presumably follows a free radical mechanism, whose operation meanwhile has been made unlikely in the assembly of acetyl-coenzyme A in *Clostridium thermoaceticum*[17]: intact incorporation with (predominant) overall retention of configuration of the chiral methyl group (chiral by H,D,T-isotope label) from methyl-tetrahydrofolate into acetate was obtained by cell free extracts of this bacterium, which rather indicated the carbonylation to occur stereocontrolled and with an even number of inversions at the methyl-carbon. Such a stereochemical outcome would be plausible for the second mechanism, where a methyl group transfer from the corrin to the nickel enzyme would occur[11]. Further experimental work is required to this point.

Interestingly, the enzymes that are characteristic of the «acetyl-coenzyme A pathway» for autotrophic fixation of carbon dioxide have been shown by *Fuchs, Stupperich* and others to operate correspondingly not only in acetogens, but also in a series of autotrophic methanogenic and sulfate reducing bacteria[7b,8]. In addition, in the acetate degradation to methane and carbon dioxide in methanogens and to carbon dioxide in sulfate reducing heterotrophs the same set of reactions appears to be involved, but in reverse sense[7,18].

In a wide range of anaerobic bacteria, a central role is given to the corrins in the autotrophic fixation of carbon dioxide and in the acetate degradation via the organometallic «acetyl-coenzyme A pathway»[7]. Indeed, in the acetogens, the organisms that foremost use this mechanism, corrinoids characteristically are abundant and, in some cases are found to make up for > 1‰ of the bacterial dry weight ([10,19], see Table 1). The corrinoids from acetogens, when isolated in their Co_β-cyano form, were predominantly found not to be vitamin B_{12} (**3**), but rather the related 5'-methoxybenzimidazolyl-cobamides (such as **4**)[7c], the newly found 5'-methoxy-6'-methylbenzimidazolyl-cobamides (such as **5**), or apparently even *p*-cresolyl-cobamides (such as **6**), all differing from vitamin B_{12} by their nucleotide base[19]. In several methanogens likewise large amounts of corrinoids were found, again not vitamin B_{12}, but rather the related 5'-hydroxybenzimidazolyl-cobamides (such as **7**) or 7'α-adeninyl-cobamides (such as **8**)[20]. These corrinoids were analyzed as membrane-bound in part, where they appear to function in methanogenesis, rather than in acetyl-coenzyme A synthesis[20c]. Also in some (but not all) of the investigated sulfur-metabolizing bacteria, significant amounts of corrinoids were found (mostly 5'-methylbenzimidazolyl-cobamides, such as **9**[21]). In several of these

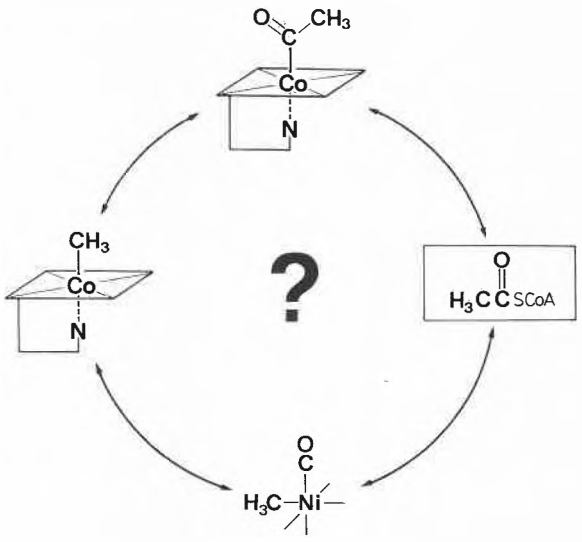

Fig. 3. Schematic illustration to the question concerning the carbon-carbon bond forming step of the «acetyl-coenzyme A pathway» [13,16].

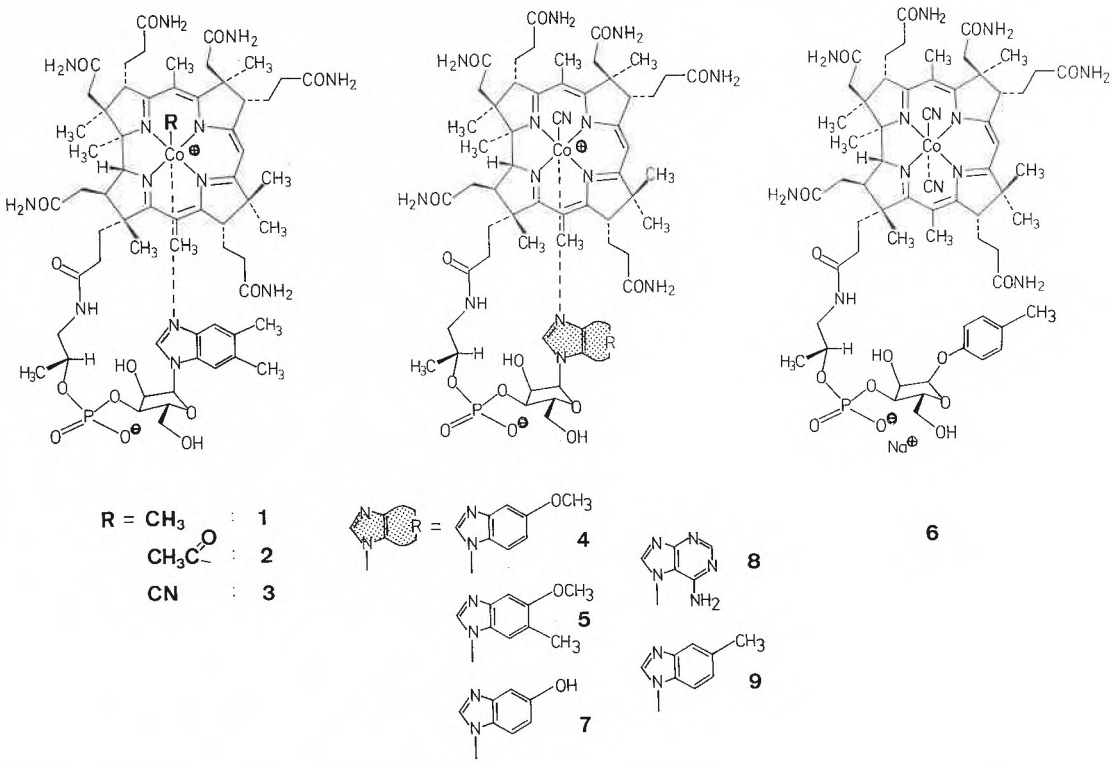

Fig. 4. Structural formulae of Co_β-cyano forms of the corrins from methanogenic, acetogenic, and sulfur-metabolizing bacteria [19–21].

bacteria, the quantity of the corrins found is large, but below detection in others (see Table 1 [20c, 21]), apparently related to the operation (such as in *Desulfobacterium autotrophicum* [20c]) or absence (such as in *Desulfobacter hydrogenophilus* [20c] or *Thermoproteus neutrophilus* [22]) of the «acetyl-coenzyme A pathway».

While acetogens produce a spectrum of complete corrins, so far the corrins from methanogens (**7** and **8**) and from sulfur-metabolizing bacteria (**9**, mostly) are less diverse. This structural variety is remarkable, since the complete corrins play such a central role in the bacterial fixation of carbon dioxide via the «acetyl-coenzyme A pathway». It appears to be a consequence first of all, of the biosynthetic availability of the corresponding nucleotide bases [23]. In addition, however, the nucleotide structure offers a twofold potential for adjusting the reactivity of the enzyme-bound corrin: firstly, the nucleotide coordination at the cobalt center directly affects the strength of the cobalt-carbon bond of corrins and their one-electron redox potentials [24]; secondly, the nucleotide can also contribute indirectly to the reactivity, by way of the structure-sensitive binding between the corrins and the apoenzyme (see e.g. [25]).

Table 1. Corrin-content of selected methanogenic, acetogenic, and sulfur-metabolizing bacteria.

Organism[a]	Corrin-content[b,c] [nmol/g]	Cobamide-form[d]	Ref.
Acetogens:			
C. thermoaceticum	440–1100	**4**	[7c, 10]
C. formicoaceticum	950	**3, 5**	[19]
A. woodii	460–870	**3**	[10, 19]
Sporomusa ovata	3100	**6**	[19]
Methanogens:			
M. thermoautotrophicum	75–120	**7**	[10, 20]
Mr. arboriphilus	270	**7**	[20b]
Ms. barkeri	310–560	**7**	[10, 20c]
Mc. aeolicus	600	**8**	[20b]
Mc. thermolithotrophicus	110	**8**	[20b]
Ml. tindarius	1400	**7**	[20b]
Sulfur-metabolizers:			
D. autotrophicum	40–200	**9**	[20c, 21]
Db. propionicus	150	**9**	[21]
Dr. hydrogenophilus	< 1	–	[20c]
Ar. fulgidus (VC-16)	100	**9**	[21]
Dl. ambivalens	15	**3***	[21]
Tl. acidophilum	8	**9***	[21]
Tr. neutrophilus	< 1	–	[21]

[a]) Abbreviations: *C.* = *Clostridium*, *A.* = *Acetobacterium*, *M.* = *Methanobacterium*, *Mr.* = *Methanobrevibacterium*, *Ms.* = *Methanosarcina*, *Mc.* = *Methanococcus*, *Ml.* = *Methanolobus*, *D.* = *Desulfobacterium*, *Db.* = *Desulfobulbus*, *Dr.* = *Desulfobacter*, *Ar.* = *Archeoglobus*, *Dl.* = *Desulfurolobus*, *Tl.* = *Thermoplasma*, *Tr.* = *Thermoproteus*. [b]) Amount (nmol/g dry bacterial weight) estimated UV/VIS-spectroscopically. [c]) Depends on nutritional conditions. [d]) Spectroanalytical analysis (*assignment tentative).

3. Unique Organometallic Chemistry in Nature

In several ways the «acetyl-coenzyme A pathway» is chemically unique among the known mechanisms for autotrophic fixation of carbon dioxide [1, 7–9]: It makes use of organometallic transformations, that are catalyzed by a series of oxygen-labile metallo-enzymes. Accordingly, it has been found only in anaerobic organisms, in anaerobic «archaebacteria» in particular [7, 24]. Secondly, it not only provides a means for autotrophic fixation of carbon dioxide, but, in acetogens, also for the production of adenosine-triphosphate (ATP) [7]. Thirdly, the synthesis of glyceraldehyde-3-phosphate from carbon dioxide and hydrogen via this pathway consumes only ca. 3 ATP, considerably less than that by the «Calvin cycle» (9 ATP) [8]. This is made possible in part, by organometallic reactions with transition metal ions: their metal-carbon bonds typically are weak [26] and correspondingly appear to have a potential to provide «energy-rich» organic (functional) groups in nature.

The course taken by the («ancient enzymatic machinery» [27] of the) «acetyl-coenzyme A pathway» thus also exemplifies a remarkable and efficient path for the reduction of carbon dioxide, of interest in view of the ongoing search for ways to synthesize basic organic chemicals and «fuels» from carbon dioxide in an economic way.

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