SAM systems speaks toward the evolutionary development of these enzymes and the requirement for a protein architecture that was inherently not complex and in regards to radical SAM proteins allowed for the diversification of chemical reactions through the acquisition of additional modular protein domains." Does this support the idea that radical SAM enzymes with their FeS clusters predate the B12 enzymes in protein-based catalysis?[19,20,228,229]

18.5.3 Types of SAM Radical Enzymes

At least a dozen types of radical SAM enzymes perform a wide variety of reactions.[20] These enzymes are involved in central metabolism, energy generation and transfer, in the synthesis of many essential cofactors and antibiotics, but also DNA repair, and RNA modifications that prevent inaccurate translation or generate antibiotic resistance[20,230] (see Table 18.1; data derived from ref. 20). Many enzymes synthesize antibiotics and toxins that may affect functioning of mitochondria and chloroplasts.[213,214,231–233] Well-known cofactors include modified tetrapyrroles, such as (bacterio)chlorophyll, heme, and cobalamins, to name just a few.[18] Anaerobic ribonucleotide reductase (aRNR) reduces CTP to dCTP, a rate-limiting step in DNA metabolism strictly dependent on SAM.[234,235] The sulfur-inserting enzyme biotin synthase (BS/BioB) uses two FeS clusters in a difficult final step in the synthesis of vitamin B1 (essential in the methionine cycle (Figure 18.2)). The reduced SAM-dependent $[4Fe–4S]^+$ cluster donates one electron to SAM producing a 5′-deoxyadenosine radical, which then requires a second half cluster, [2Fe–2S], to insert a sulfur atom into the biotin precursor.[20,236,237] PylB is involved in the synthesis of pyrrolysine, present in the in-frame UAG amber codon in SAM-dependent MTases in certain archaea that use these MTases to generate methane.[20] Complex changes involve formation of the pyrimidine ring and synthesis of vitamin B1. Nucleotide analogues act as antibiotics, neoplastic agents, enhance translational fidelity, and synthesize F420, the cofactor for hydride transfer in energy metabolism, antibiotic biosynthesis, and DNA repair. MqnC and MqnE are involved in the synthesis of vitamin K2, which serves as an electron shuttle between membrane-bound proteins in the respiratory chain; NosL/NocL are involved in antibiotics of clinical interest against drug-resistant bacterial pathogens (see ref. 20 for details).

Of particular interest are anaerobic sulfatase maturating enzymes (anSME) that can oxidize cysteine or serine residues in proteins to generate electrons, and play a key role in the microbiome (see Section 22.6.5). An SME is a group of at least 1400 enzymes with a SPASM domain and an amazing 7-cysteine motif ($CX_{9–15}GX_4C$-gap-$CX_2CX_5CX_3C$-gap-C), which coordinates additional FeS clusters in these enzymes.[20,238]

Synthesis of enzymes that form C–C, C–N, and C–S bonds includes enzymes that regulate (post)translational modifications and fidelity, and synthesis of antibiotics that target feared *Clostridium difficile* and other pathogens.[20,239] PqqE is involved in the synthesis of pyrroloquinoline quinone (PQQ), a cofactor found in alcohol dehydrogenases and other bacterial enzymes.[240–242] Not