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Minireview

Discovery and characterization of electron transfer proteins in the photosynthetic bacteria[1]

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Abstract

Research on photosynthetic electron transfer closely parallels that of other electron transfer pathways and in many cases they overlap. Thus, the first bacterial cytochrome to be characterized, called cytochrome c_2, is commonly found in non-sulfur purple photosynthetic bacteria and is a close homolog of mitochondrial cytochrome *c*. The cytochrome bc_1 complex is an integral part of photosynthetic electron transfer yet, like cytochrome c_2, was first recognized as a respiratory component. Cytochromes c_2 mediate electron transfer between the cytochrome bc_1 complex and photosynthetic reaction centers and cytochrome *a*-type oxidases. Not all photosynthetic bacteria contain cytochrome c_2; instead it is thought that HiPIP, auracyanin, *Halorhodospira* cytochrome *c*551, *Chlorobium* cytochrome *c*555, and cytochrome c_8 may function in a similar manner as photosynthetic electron carriers between the cytochrome bc_1 complex and reaction centers. More often than not, the soluble or periplasmic mediators do not interact directly with the reaction center bacteriochlorophyll, but require the presence of membrane-bound intermediates: a tetraheme cytochrome *c* in purple bacteria and a monoheme cytochrome *c* in green bacteria. Cyclic electron transfer in photosynthesis requires that the redox potential of the system be delicately poised for optimum efficiency. In fact, lack of redox poise may be one of the defects in the aerobic phototrophic bacteria. Thus, large concentrations of cytochromes c_2 and c' may additionally poise the redox potential of the cyclic photosystem of purple bacteria. Other cytochromes, such as flavocytochrome *c* (FCSD or SoxEF) and cytochrome *c*551 (SoxA), may feed electrons from sulfide, sulfur, and thiosulfate into the photosynthetic pathways via the same soluble carriers as are part of the cyclic system.

Abbreviations: FCSD – flavocytochrome *c* sulfide dehydrogenase; FNR – ferredoxin NADP reductase; HiPIP – high potential iron–sulfur protein; RBC – Rieske iron–sulfur protein/cytochrome *b*/cytochrome *c* complex; RC – reaction center; SQR – sulfide quinone reductase

Introduction

Space does not permit complete coverage, but the focus of this review will be on the discovery and characterization of redox proteins that are involved in photosynthetic electron transfer. We first give a broad overview, followed by detailed sections which contain citations in support of this introductory summary.

The history of the redox proteins involved in photosynthetic electron transfer began with observations using the hand spectroscope (1880s to the 1930s), progressed to the scanning spectrophotometer (in the 1940s), to isolation and purification of proteins (begin-

[1] This article is dedicated to the memory of our mentor and friend, Martin Kamen, who passed away 31 August 2002.

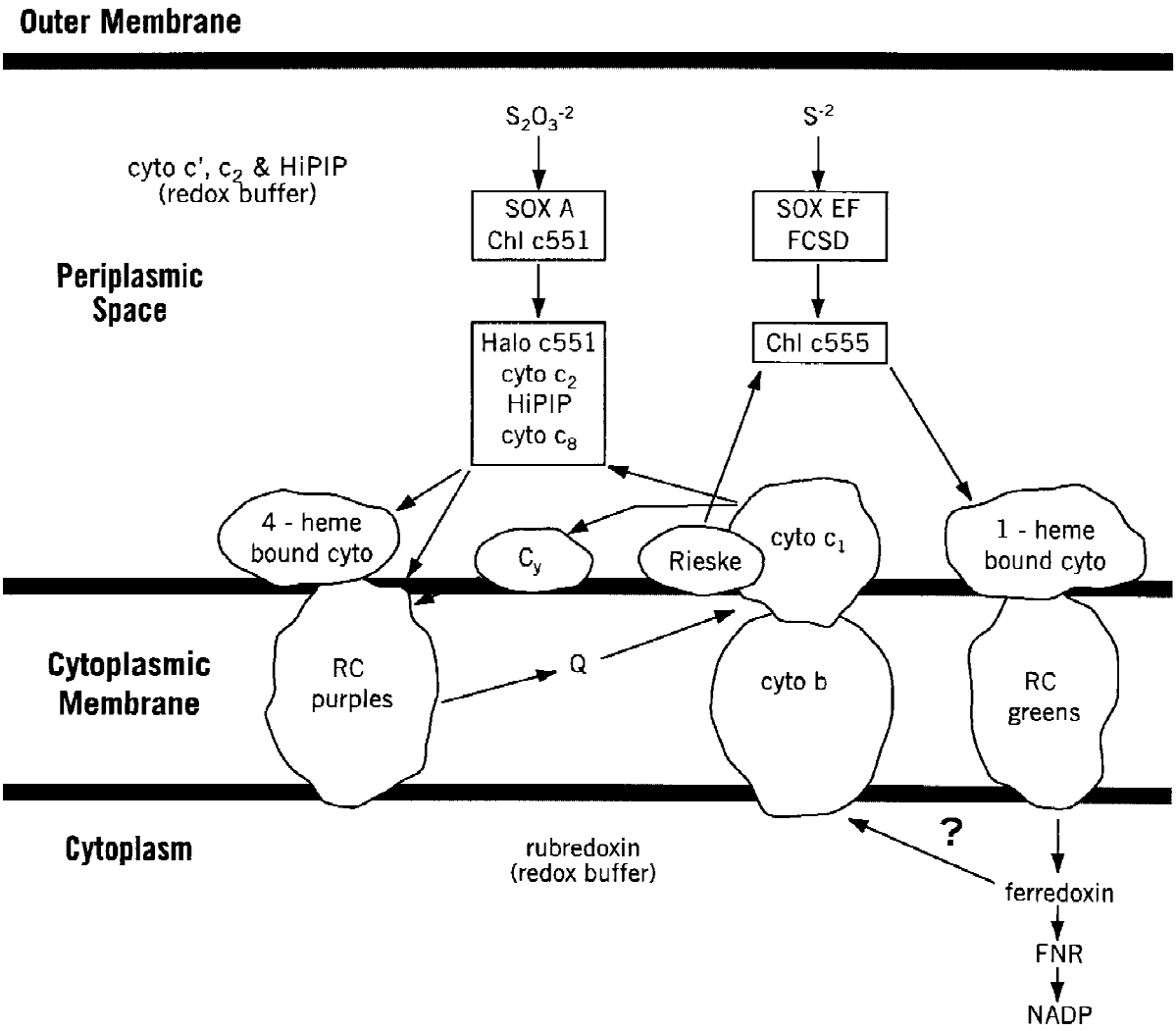

Figure 1. Composite photosynthetic bacterial electron transfer pathway indicating the variety of possible interactions in purple and green bacterial species. Arrows indicate direction of electron transfer. *Chloroflexus, Heliobacterium*, and cyanobacteria were omitted. For abbreviations see p. 111.

ning in the 1950s), which led to sequence determination (in the 1960s), to three-dimensional structural determination (in the 1970s), to genetics (in the 1980s), and to whole genome analysis (in the 1990s). Each of these developments improved our understanding and represent stages in characterization of the structure and function of redox proteins which continues to this day. It is thus only recently that we have developed an overall view of photosynthetic electron transfer pathways as shown in Figure 1. This is a greatly simplified view which combines aspects of photosynthesis from the purple and green bacterial families. Whereas plant photosynthesis and animal respiration are fairly uniform in their redox protein composition, bacterial metabolic processes exhibit great diversity which is not yet fully defined. There are certain aspects of photosynthetic electron transfer that are the same in virtually all species but there are others that can best be described as mix-and-match. The most variable element appears to be in the soluble mediator connecting the membrane-bound carriers. These can be any of several cytochromes *c*, iron–sulfur proteins, or copper proteins. The Rieske iron–sulfur protein/cytochrome *b*/cytochrome c_1 (RBC) complex is also variable with respect to the presence or absence of a *c*-type cytochrome and in the family to which that cytochrome

Table 1. Distribution of redox proteins in the seven families of photosynthetic bacteria

Protein[a]	Rhodo.	Chlflx	Chrom.	Ecto.	Chloro.	Helio.	Cyano.
Photosystem	II	II	II	II	I	I	I and II
cyt. c_2	Most	No			No		No
cyt. c_4	Some	No	Some	Some	No		No
cyt. c_5/cyt c_6		No		All	All		Most
cyt. c_8	Some	No	Some		No		No
RC cyt.	4 heme[b]	4 heme	4 heme	4 heme	1 heme	1 heme	No
RBC complex	All		All	All	No cyt *c*	Diheme c	Cyt *f*
FCSD	Some	No	Most	Some	Some		No
Chl. *c*551/SoxA	Some	No			Some		No
cyt. c'	Most	No	All	Most	No		No
HIPIP	Some	No	All	Most	No		No
8Fe–8S ferredoxin	All		All		All		
2Fe–2S ferredoxin							All
Rubredoxin	All?	No			All		No
Blue copper	Some	yes			No		All

[a]A blank cell is unknown. A 'yes' or a 'no' is certain only where there are genome sequences. Rhodo. – Rhodospirillaceae, Chlflx. – Chloroflexus, Chrom. – Chromatiaceae, Ecto. – Ectothiorhodospiraceae, Chloro. – Chlorobiaceae, Helio. – Heliobacteriaceae, Cyano. – cyanobacteria.
[b]Most but not all purple bacteria have the tetraheme RC cyt.

belongs. The utilization of either or both of the two types of reaction center (RC, Photosystems I or II) is also an important difference among species.

The greatest variability among species is from one photosynthetic family to another as shown in Table 1, but there is greater photosynthetic diversity in the non-sulfur purple bacteria than in the other families. The non-sulfur purple bacteria are represented by more species and are better characterized because they are easy to grow, they are metabolically more versatile, they can tolerate oxygen, they are simple to plate, and they are genetically amenable. The electron carriers are either imbedded in the cytoplasmic membrane or located primarily on the outside of the membrane. In Gram-positive species, those components on the outside must be tethered because there is no outer membrane or periplasmic compartment to prevent diffusion into the medium. In Gram-negative species, they can also be tethered but are more often soluble in the periplasmic space.

Class I cytochromes *c* are defined as those which are structurally related to the familiar mitochondrial cytochrome *c* and are the most commonly occurring of the soluble electron carriers. The various subfamilies of the class I *c*-type cytochromes are defined based upon a combination of sequence identity, specific insertions and deletions, and presence or absence of rare amino acid residues which are thought to have functional significance such as cysteine and tryptophan (Meyer 1996). Cytochrome c_2 is the nearest bacterial homolog of mitochondrial cytochrome *c*. The other class I cytochromes *c* are generally smaller and are more divergent in that the sequences cannot be uniquely aligned with mitochrondrial cytochrome *c* although they are obviously related in three-dimensional structure. Suffice it to say, the class I *c*-type cytochromes radiated from a common precursor, they were duplicated and transferred to various species where they were modified for new roles, and eventually made their way back to the source where they occasionally replaced the original cytochrome *c*. Thus, we now have a variety of class I cytochromes that function in photosynthesis.

The role of redox proteins in the photosynthetic bacteria has only been addressed in the latter half of the 20th century with the bulk of the progress in the last 20 years. The photosynthetic bacteria currently include seven families (Table 1), including: non-sulfur purple bacteria (Rhodospirillaceae), purple sulfur bacteria (Chromatiaceae and Ectothiorhodospiraceae), green sulfur bacteria (Chlorobiaceae), heliobacteria (Heliobacteriaceae), *Chloroflexus* (Chloroflexaceae), and cyanobacteria. The subfamilies of class I *c*-type cytochromes in photosynthetic bacteria include: cytochromes c_1, c_2, c_4, c_5, c_6, c_8, *Chlorobium* cytochrome *c*555, *Halorhodospira* cytochrome *c*551,

and *Chlorobium* cytochrome *c*551 (SoxA). In addition, there are examples of divergent *c*-type cytochromes such as cytochrome c' and cytochrome f, which are not structurally related to the class I cytochromes. Finally, there are several, more complicated, proteins including: the RBC complex, flavocytochrome *c*, and RC cytochrome. In addition to *c*-type cytochromes, the photosynthetic bacteria contain a variety of iron–sulfur proteins (e.g., HIPIP, rubredoxin, and low potential ferredoxins), copper proteins, and *b*-type cytochromes. Electron transfer in the photosynthetic bacteria involves: the RC complex, the RBC complex, quinone, and what is usually a soluble protein mediator at a minimum. There are two basic types of RCs: 1) those related to Photosystem II (in purple bacteria and *Chloroflexus*) where the reaction center directly reduces quinone and 2) those related to Photosystem I (in green sulfur bacteria and Heliobacteria) where the reaction center directly reduces ferredoxins. Cyanobacteria contain both types of RCs. Table 1 summarizes the distribution of redox proteins in the seven families of photosynthetic bacteria. As more functions are determined and new cytochromes identified, a more complete picture of electron transfer in the photosynthetic bacteria will evolve in the 21st century.

Figure 2. Professor Martin Kamen at the Fogarty Center of the National Institutes of Health on the occasion of the International Workshop on Non-Mitochondrial Cytochromes *c* organized by him in August 1990 during his tenure as Scholar in Residence. The proceedings of the conference were published in a special issue of *Biochimica Biophysica Acta*, Volume 1058, 1991. Photo by T.E. Meyer.

Cytochrome bc_1 and b_6f Complexes

One of the more interesting aspects of the history of photosynthesis is that the RBC complex turned out to be an integral part of photosythesis as well as respiration (Wood 1980; Gabellini et al. 1982; Gabellini and Sebald 1986). In fact, it is at least as important as the RC complex itself. Cytochrome c_1 was spectroscopically identified by Yakushiji and Okunuki (1940) in respiratory tissues although it was much later that this membrane-bound protein was isolated and characterized as a single component (Robinson and Talbert 1980; Konig et al. 1980).

One of the landmark discoveries in biochemistry was the mechanism of proton pumping inferred from the three-dimensional structure of the RBC complex (Zhang et al. 1998). Although the cytochrome *b* and Rieske iron–sulfur proteins of the cytochrome bc_1 complex are homologous to those of the cytochrome b_6f complex, cytochrome c_1 is unrelated to the cytochrome f of plants and cyanobacteria (Hill and Scarisbrick 1951; Martinez et al. 1994).

Heliobacteria contain an RBC complex that includes a divergent diheme cytochrome *c* component instead of c_1 or f (Xiong et al. 1998) and a membrane-bound monoheme cytochrome *c*553 is part of the RC complex (Albert et al. 1998; Xiong et al. 1998). Because the heliobacteria are Gram-positive and lack an outer membrane or periplasmic compartment, it is unlikely that there is a soluble component that mediates electron transfer between the two complexes. Thus, the diheme cytochrome *c* transfers electrons directly to the cytochrome *c*553 or there is another as yet undescribed membrane-tethered component that serves as a mediator.

Green sulfur bacteria, such as *Chlorobium limicola*, have cytochrome *b* and Rieske proteins related to those of purple bacteria, but it appears that there is no cytochrome *c* of any kind in the RB complex (Schütz et al. 1994). This has been recently confirmed by the whole genome sequence of *Chlorobium tepidum* (Eisen et al. 2002). A truncated complex III also is characteristic of *Halobacterium* and other aerobic archaebacteria (which are all Gram-positive); there are no *c*-type cytochromes of any kind in these species which appear to utilize membrane-bound blue cop-

per proteins as mediators to the oxidase complex in respiration (Ng et al. 2000).

Mediators between the reaction center and bc_1 complexes

Cytochrome c_2

The first bacterial cytochrome *c* to be discovered was isolated from *Rhodospirillum rubrum* and was named cytochrome c_2 by Vernon (1953); it was completely purified and characterized by Vernon and Kamen (1954). Martin Kamen (Figure 2) began his career with the discovery of carbon 14 and the use of radioisotopes in biochemistry and particularly in photosynthesis. Kamen's proof that cytochromes *c* occur in anaerobic cells overturned the prevailing opinion of the time that cytochromes only functioned in aerobic respiration. Kamen never looked back and devoted the remainder of his career to the characterization of cytochromes, especially those of the photosynthetic bacteria. No less than 10 distinct kinds of *c*-type cytochromes were discovered in numerous species surveyed in the Kamen lab. Initially, cytochrome c_2 was thought to be structurally and functionally distinct from mitochondrial cytochrome *c* because it had an acidic isoelectric point (less than 7 vs 10 for mitochondrial cytochrome *c*), it had a much higher redox potential (330 vs 260 mV), it was abundant and readily isolated from anaerobically grown cells, and it was photoxidized by the bacteriochlorophyll in whole cells and membranes. However, following completion of the amino acid sequence (Dus et al. 1968), it was apparent that *Rsp. rubrum* cytochrome c_2 was closely related to the familiar mitochondrial cytochrome *c* discovered by Keilin (1925) who was the key cytochrome researcher of his day. Keilin built upon the extensive spectroscopic observations of MacMunn (1886) which suggested that tissues contain heme pigments other than hemoglobin, a view that was very unpopular at the time. Theorell and Åkesson (1939) were the first to purify and characterize cytochrome *c*. The general occurrence of cytochrome c_2 in non-sulfur purple bacteria was confirmed by comparative sequence analysis of 12 species of this cytochrome by Ambler et al. (1979a). The three-dimensional structures of mitochondrial cytochrome *c* (Dickerson et al. 1971) and of cytochrome c_2 (Salemme et al. 1973) showed that they folded similarly and that both had a concentration of positive charge near the edge of the heme exposed to solvent, independent of the net protein charge. It was thus realized that positive charge localized at the site of binding and electron transfer was more important than the isoelectric point in determining reactivity of cytochromes *c* and c_2. Functionally, cytochrome c_2 mediates electron transfer between the RBC and RC complexes (during anaerobic growth) and between the RBC and cytochrome oxidase complexes (during aerobic growth) in many non-sulfur purple bacteria as established genetically (Daldal et al. 1986; Donohue et al. 1988; Jenney et al. 1994). There are often isozymes of cytochrome c_2 in purple bacteria, both soluble and membrane-bound, that might have a role in photosynthesis under some conditions or may be specialized for reaction with cytochrome oxidase during aerobic growth depending in part on their redox potentials. The regulation of cytochrome c_2 and its isozymes remains an active area of research. A membrane-bound tetraheme cytochrome *c* mediates electron transfer between the cytochrome c_2 and RC (see below, Kennel and Kamen 1971; Deisenhofer et al. 1985) in a number of purple photosynthetic bacteria, but there are several species, such as *Rhodobacter capsulatus, Rhodobacter sphaeroides*, and *Rsp. rubrum*, where cytochrome c_2 reacts directly with the RC. Presumably, the high redox potentials of most photosynthetic cytochromes c_2 (Pettigrew et al. 1975, 1978) are a specific adaptation to photosynthetic electron transfer since it has been shown that the potentials of RCs (Wachtveitl et al. 1993) are in the region of 450–500 mV, which are considerably higher than those of the cytochrome oxidase complexes. The photosynthetic cytochromes c_2 are probably not very reactive with cytochrome oxidase specifically because of their high potentials (Yamanaka and Okunuki 1968; Errede and Kamen 1978). Exploiting the three-dimensional structures of both the photosynthetic RC (Deisenhofer et al. 1985; Allen et al. 1987) and cytochrome c_2, their interaction has been extensively studied beginning with the work of Prince et al. (1974), Overfield et al. (1979) and Rickle and Cusanovich (1979) and most recently in site-directed mutagenesis exemplified by the work of Tetreault et al. (2001, 2002). The three-dimensional structure of the RC–c_2 complex (Axelrod et al. 2002) is in good agreement with the conclusions of the mutatagenesis work. Although much remains to be learned about cytochrome c_2, it has become a prototype for investigating interactions mediating physiologically relevant electron transfer. Having said that, it is clear that not all species of purple bacteria utilize cytochrome c_2 and that there are other proteins

that mediate electron transfer between the RBC and RC complexes although their interactions are not as well characterized.

High potential iron–sulfur protein (HiPIP)

Another electron transfer protein discovered early in the history of photosynthesis research in the Kamen lab is HiPIP (Bartsch 1963), which, although it has the same 4Fe–4S cluster as bacterial ferredoxin, it has a redox potential of +330 mV that permits it to function in many of the same reactions as cytochrome c_2. It has been found in nearly all of the purple sulfur bacteria (90% of the species examined), in a number of non-sulfur purple bacteria (about 40% of the species), and even in a few non-photosynthetic species (Meyer 1994). In purple sulfur bacteria such as *Chromatium*, it is an abundant soluble protein and its concentration is more than an order of magnitude greater than that of the high redox potential cytochromes c_4 and c_8 (see below). Little attention was paid to the possible functional roles of HiPIP until relatively recently. By virtue of its abundance and the apparent lack of cytochrome c_2, it was felt that it serves the same functional role as cytochrome c_2 in *Chromatium* and other purple sulfur bacteria. It was shown by Kennel et al. (1972) that HiPIP could serve as an electron donor to the RC. This functional role was confirmed when spectroscopic and kinetic evidence showed that HiPIP could mediate electron transfer to the photosynthetic RC in at least seven purple bacteria in the families *Chromatiaceae* and *Rhodospirillaceae*, much the same as cytochrome c_2 in other species of the *Rhodospirillaceae* (Schoepp et al. 1995; Hochkoeppler et al. 1995, 1996; Menin et al. 1997, 1998). There is as yet no direct genetic evidence for the involvement of HiPIP in cyclic electron transfer and there are some purple sulfur bacteria that apparently do not have HiPIP, for example *Halorhodospira halochloris* and *Halorhodospira abdelmalekii*. Thus, it is still necessary to solidify the role of HiPIP as photosynthetic electron donor to RC. In addition to a direct role in cyclic electron transfer, HiPIP could also serve to buffer the redox potential of the cyclic photosystem (see below). Finally, in *Chromatium*, it was found that HiPIP could serve as electron acceptor for a thiosulfate: tetrathionate oxidoreductase (Fukumori and Yamanaka 1979b), suggesting that, in at least some cases, HiPIP can provide electrons to the photosystem from metabolites.

Cytochromes c_4, c_5, c_6, *and* c_8

Cytochromes c_4, c_5, and c_8 were first characterized in non-photosynthetic *Pseudomonas* and *Azotobacter* species (Kamen and Takeda 1956; Tissieres 1956; Horio et al. 1960; Swank and Burris 1969). In photosynthetic bacteria, cytochrome c_4 was initially isolated from *Chromatium vinosum* and is apparently a membrane protein, solubilized by treatment with organic solvents as in the pseudomonads (Cusanovich and Bartsch 1969) but which is also present at very low concentrations in the soluble state. Cytochrome c_4 is particularly interesting in that it contains two hemes, it has a cleavable signal sequence, and has no membrane anchor (Ambler et al. 1984; Ng et al. 1995). Thus, it is likely to be tightly bound to another protein in a membrane complex. In this regard, we now know from the amino acid sequence (Van Beeumen 1991) that *Chromatium vinosum* cytochrome c_4 is related to the diheme cytochrome subunit of flavocytochrome *c* (Van Beeumen et al. 1991; Chen et al. 1994; Kadziola and Larsen 1997). One of the most commonly occurring cytochrome genes encountered in whole genome sequence analyses of bacteria encodes cytochrome c_4 (represented as both diheme and monoheme variants in about twice as many species as is cytochrome c_2), yet it is usually not associated with any other genes that might give a clue as to its functional interactions. Although a function for cytochrome c_4 has not been directly demonstrated in any species, the protein could be more widespread in the purple bacteria than is currently apparent because of its predominantly membrane location. Cytochrome c_4 (also called *c*554(549)) appears to be a soluble protein in the *Ectothiorhodospira* species such as *E. vacuolata, E. mobilis*, and *E. shaposhnikovii* (Küsche and Trüper 1984) and it is possible that it might have a role as a photosynthetic electron transfer mediator in these species under some conditions.

Cytochrome c_8 is best known as an electron donor to the *Pseudomonas* cytochrome cd_1 type of nitrite reductase (Horio 1958ab). Ambler (1963) determined the first amino acid sequence and Almassy and Dickerson (1978) solved the three-dimensional structure. There are two interesting aspects to cytochrome c_8, it is much smaller than cytochrome c_2 and there is no concentration of positive charge at the edge of the heme where electron transfer is thought to take place. The first photosynthetically relevant cytochrome c_8 isolated was from *Rubrivivax gelatinosus* (Tedro et al. 1976) and is normally a soluble protein that is pro-

duced at a low level. There are no other soluble high potential electron carriers in *Rc. purpureus*, including HiPIP and c_2, therefore c_8 may be the sole electron donor to RC in this species. Cytochrome c_8 has good activity as electron donor to *Chromatium* RCs *in vitro* (van Grondelle et al. 1977) and forms a complex with flavocytochrome *c*–sulfide dehydrogenase (Davidson et al. 1985). The *Chromatium* cytochrome was identified as a c_8 because of its distinctive sequence (Samyn et al. 1996). Cytochrome c_8 appears to be present only in a small number of non-sulfur purple bacteria as shown by its amino acid sequence (Ambler et al. 1979b) but is abundant in *Rhodocyclus tenuis* and *Rhodocyclus purpureus*. It is therefore not surprising that *Rc. tenuis* cytochrome c_8 may effectively compete with HiPIP as electron donor to RC (Menin et al. 1997). However, there is no evidence that any other known cytochrome c_8 might function directly in photosynthesis. There are isozymes of c_8 in *Rc. gelatinosus* that are separately regulated, they have distinctive redox potentials, and may have other roles such as in aerobic electron transfer (Menin et al. 1999).

Pseudomonas and *Azotobacter* cytochromes c_5 are characterized by a primarily membrane location (due to an uncleaved signal peptide) and by the presence of a rare disulfide bridge (Carter et al. 1985; Ambler 1991; Rey and Maier 1997). A cytochrome c_5, as such, has not yet been encountered in photosynthetic bacteria, but the soluble *Chlorobium* cytochrome *c*555 is a close homolog based on amino acid sequence (Van Beeumen et al. 1976) as is the algal cytochrome c_6, which functions interchangeably with the copper protein, plastocyanin, as electron donor to the RC in cyanobacteria (Wood 1978). Cytochrome c_6 was first purified and characterized by Katoh (1959). Laycock (1972) showed that cytochrome c_6 had a distinct amino acid sequence related to that of cytochrome c_5 and was not a soluble form of cytochrome f, which was the prevailing opinion at the time. We note that Yakushiji (1935) had spectroscopic evidence for the presence of cytochrome c_6 in algae rather early in the history of photosynthesis research, yet it never excited the interest that cytochromes c_1 and f did. *Chlorobium* cytochrome *c*555 was spectroscopically identified by Kamen and Vernon (1954); it was purified and characterized by Gibson (1961). Although there is no evidence to link *Chlorobium* cytochrome *c*555 to photosynthetic electron transfer at this time, it is possible that it could function in this role (Menin et al. 1998). Cytochrome *c*555 is the only cytochrome that has been found in every species of green sulfur bacteria studied to date (Fischer 1988) which suggests that it has an essential role.

A somewhat more obscure soluble cytochrome *c*551 was discovered in *H. abdelmalekii, H. halochloris* (Then and Trüper 1983, 1984) and *H. halophila* (Meyer 1985). It was found to be yet another distinct member of the greater cytochrome c_5 family of proteins through sequence and structural analysis (Ambler et al. 1993; Bersch et al. 1996). There are apparently no other soluble high potential cytochromes or HiPIP in *H. halochloris* or *H. abdelmalekii*, thus it is possible that the *Halorhodospira* cytochrome *c*551 donates electrons to the RC in these species.

An evolutionary progression from *Pseudomonas* cytochrome c_5 to *Chlorobium* cytochrome *c*555 to *Halorhodospira* cytochrome *c*551 to cyanobacterial cytochrome c_6 has been proposed (Meyer et al. 1997), which suggests that pseudomonads predate the evolution of green and purple sulfur bacteria, and that cyanobacteria evolved more recently.

Copper proteins

In green plants, it is well known that the blue copper protein, plastocyanin, discovered by Katoh (1960) is the sole mediator between the b_6f complex (which is analogous to bc_1) and RC. It is less well known that cytochrome c_6 can function interchangeably with plastocyanin in algae and cyanobacteria (Wood and Bendall 1975; Wood 1978; Merchant and Bogorad 1986) and may be the sole electron donor in some species. In those species which have both proteins, they are regulated by copper and plastocyanin is the preferred electron donor. Plastocyanin is homologous to the blue copper protein azurin, which was first discovered in *Pseudomonas* by Horio (1958a) and found to function interchangeably with cytochrome c_8 as the electron donor to cytochrome cd_1–nitrite reductase (Horio 1958b).

Chloroflexus is generally known as a non-sulfur green bacterium because of the presence of light-harvesting bacteriochlorophyll complexes (chlorosomes) that resemble those of the green sulfur bacteria (Feick et al. 1982), but it has a reaction center resembling those of the purple bacteria (Pierson and Thornber 1983; Shiozawa et al. 1989). Thus, it should be recognized as a chimera. *Chloroflexus* appears not to have any truly soluble electron donors (such as cytochromes), but there are two normally membrane-bound blue copper proteins, both called auracyanin,

which have been isolated from soluble extracts that are likely to donate electrons to the RC (Trost et al. 1988; McManus et al. 1992; Van Driessche et al. 1999). The auracyanins are more closely related to the azurins of pseudomonads than they are to the plastocyanins of plants and cyanobacteria (Bond et al. 2001). The genome sequence of *Chloroflexus* indicates that there are actually three related auracyanin type copper proteins and several cytochromes *c*, any of which could function in photosynthesis (DOE Joint Genome Institute: http://spider.jgi-psf.org). An evolutionary scheme for the copper proteins shows an interesting parallel to that of the cytochrome c_5 family described above.

Photosynthetic reaction center cytochrome

Up to this point, we have discussed generally soluble proteins that interact with photosynthetic RCs. However, frequently, the immediate electron donor to the RC is a membrane-bound cytochrome. The first such protein to be discovered was in *Chromatium vinosum* based upon spectroscopic observations (Olson and Chance 1960a, b). Following the initial report, there were many others on light-induced absorbance measurements in *Chromatium*, but it was generally assumed that they were due to a membrane-bound form of the familiar soluble flavocytochrome *c*. There were two observations that strongly indicated that this membrane-bound cytochrome was unique: (1) Parson and Case (1970) found that this cytochrome could be photoxidized at liquid nitrogen temperatures, indicating an intimate association with the RC, (2) Kennel and Kamen (1971) purified this membrane cytochrome using detergent and characterized it as a tetraheme protein of 40 kDa which had two widely spaced redox potentials. Subsequent to these reports, it was found that the tetraheme RC cytochrome was present in the majority of purple sulfur and non-sulfur purple species and also in *Chloroflexus* (Freeman and Blankenship 1990; Dracheva et al. 1991). The three-dimensional structure of the *Rps. viridis* RC established that the tetraheme cytochrome was an integral subunit of the RC (Deisenhofer et al. 1985), and that a high potential heme is virtually in contact with the special pair bacteriochlorophyll as expected from the spectroscopic observations. Purple species which lack the tetraheme cytochrome, such as, *Rsp. rubrum, Rb. capsulatus, Rps. palustris*, and *Rb. sphaeroides* appear to be in the minority. *Chloroflexus* is an anomaly because it has a purple bacterial type of RC but green bacterial light-harvesting bacteriochlorophyll. Perhaps it is the tetraheme cytochrome that allows a diverse group of proteins to mediate electron transfer to the RC, including cytochromes c_2, c_8, *Halorhodospira* *c*551, HiPIP, and auracyanin.

The green sulfur bacteria also have a membrane cytochrome associated with the RCs which was discovered relatively early by spectroscopic means (Fowler et al. 1971; Prince and Olson 1976; Bruce et al. 1982; Hurt and Hauska 1984) and which was genetically characterized more recently (Okkels et al. 1992). The *Chlorobium* RC-associated cytochrome *c*551 has only a single heme in a 20 kDa peptide chain. Heliobacteria appear to have a related monoheme reaction center cytochrome *c*553 (Xiong et al. 1998; Albert et al. 1998) although it remains to be seen if it is structurally homologous to that of the *Chlorobiaceae*.

A need to poise the redox potential in cyclic photosynthesis

Cytochrome c′

Cytochrome c' was discovered and characterized at the same time as cytochrome c_2 (Vernon and Kamen 1954). It is an unusual high-spin protein spectroscopically similar to hemoglobin, but which has covalently bound heme that does not bind oxygen. The amino acid sequences of 12 species of cytochrome c' (Ambler et al. 1981) and the three-dimensional structure (Weber et al. 1980) demonstrate that there is no relationship to cytochrome c_2 or to hemoglobin, but that the heme is covalently bound to the C-terminus of the approximately 130-residue peptide chain. Amino acid sequence determination also showed that there are naturally occurring variants of cytochrome c', such as the cytochromes *c*554 and *c*556 that are low-spin due to the presence of a methionine sixth ligand. The genetic context of cytochrome c' is interesting in that it may provide some indication of possible functional roles. In *Chromatium*, it was found that a gene for a membrane-spanning cytochrome *b* is adjacent to that for cytochrome c', but is in the opposite orientation (Even et al. 1995). In *Rb. sphaeroides*, a gene for the low spin isozyme of c', called cytochrome *c*554, is adjacent to a gene for a diheme cytochrome *c* (Flory and Donohue 1995). Similar genetic associations have been observed in other species, suggesting that they are not chance occurrences, but that closely linked genes may encode proteins that are interact-

ing redox partners. However, the significance of these interactions is not yet clear.

We proposed some time ago that the redox potential of the cyclic photosystem should be poised between that of the photosynthetic RC and the quinone electron acceptor for optimal electron transfer, i.e., between +450 and −50 mV (Meyer and Kamen 1982). If the cells are too highly reduced, the concentration of catalytically active electron acceptor will be effectively nil and the same for the donor if the potential is too high. Both cytochromes c_2 and c' are present at very high cellular levels and are capable of performing this function at either end of this range. The aerobic phototrophic bacteria, such as *Roseobacter*, are an interesting case in point since no defect in the cyclic photosystem has been found, yet the cells will not grow photosynthetically under anaerobic conditions. It has recently been proposed that they lack redox buffering capacity (Candela et al. 2001). If in fact cytochrome c' does function to poise the redox potential near 0 mV in most purple bacteria, what of the species that lack cytochrome c'? Purple bacteria such as *Rps. viridis, Rm. vannielii, Rps. acidophila*, and *Rp. globiformis* appear to lack a soluble cytochrome c' and produce no other soluble components that could fill the proposed role of cytochrome c' as a redox buffer. It will be interesting to determine whether such a cytochrome is in fact completely absent or is membrane-bound in these species.

Rubredoxin and ferredoxin

None of the green sulfur bacteria produce cytochrome c', but they have large amounts of cytochrome c555 (Em = 145 mV) that might play a similar role as cytochrome c_2 in buffering the photosystem at the high potential end. At the low redox potential end, the green bacteria produce large amounts of rubredoxin (Meyer et al. 1971) which has a redox potential similar to that of cytochrome c' (near 0 mV) and could fulfill the role of a redox buffer, although it is located in the cytoplasm rather than in the periplasm. Rubredoxin was first discovered in *Clostridium pasteurianum* by Lovenberg and Sobel (1965) and is also found in *Heliobacillus mobilis* (Lee et al. 1995). Rubredoxin has a specific role as electron acceptor for pyruvate ferredoxin oxidoreductase in *Chlorobium* (Yoon et al. 1999). Purple bacteria are capable of producing rubredoxin, but it is part of the hydrogenase gene cluster and presumably regulated differently than in green bacteria (Colbeau et al. 1994).

The green sulfur bacteria have a different photosystem than the purple bacteria, one that is related to plant Photosystem I, which has a tightly bound 8Fe–8S ferredoxin subunit that is the electron acceptor for the RC (Büttner et al. 1992). In plants and cyanobacteria, the bound 8Fe–8S ferredoxin (called PsaC) passes electrons on to an abundant soluble 2Fe–2S ferredoxin which ultimately reduces NADP via the flavoprotein, ferredoxin–NADP reductase (FNR). The bound ferredoxin is homologous to the bacterial 8Fe–8S ferredoxin discovered by Mortenson et al. (1962) in *Clostridium pasteurianum*. The 2Fe–2S ferredoxin was discovered by Tagawa and Arnon (1962) and FNR by Shin et al. (1963). Multiple forms of bacterial ferredoxin have been found in most of the purple and green bacteria examined, where it is associated with nitrogen fixation (Schatt et al. 1989). *Chlorobium* has the bound 8Fe–8S ferredoxin but does not have a 2Fe–2S ferredoxin like that of cyanobacteria. Instead, the small soluble 8Fe–8S ferredoxin may accept electrons from the bound protein. Kusai and Yamanaka (1973c) purified FNR enzyme activity in *Chlorobium*, but there is no gene homologous to plant FNR in the *Chlorobium tepidum* genome sequence (Eisen et al. 2002). *Chloroflexus* has a homolog of FNR, but apparently has no small soluble ferredoxins that might interact with it. Neither bound nor soluble ferredoxins corresponding to the above have been found in *Chloroflexus* or heliobacteria. It is likely that there could be more than one coupling site for proton translocation, utilizing complex I-NADH dehydrogenase as well as complex III or the RBC. Nevertheless, the redox potential of the system should still be poised higher than that of the quinone, but lower than that of the special pair bacteriochlorophyll, which has been reported to be about +240 mV in green bacteria (Fowler et al. 1971).

Oxidation of reduced sulfur compounds

Flavocytochrome c*–sulfide dehydrogenase (FCSD) and sulfide quinone reductase (SQR)*

Although apparently not directly involved in photosynthesis as part of the cyclic electron transfer pathway, there are several well-known electron transfer proteins in photosynthetic bacteria that deserve attention. This is because of their involvement in oxidation of reduced sulfur compounds such as sulfide, sulfur, thiosulfate, and sulfite. In the autotrophic species, such

as some of the purple sulfur bacteria and all green sulfur bacteria, electrons for carbon dioxide fixation come from oxidation of reduced sulfur compounds. The enzymes that oxidize reduced sulfur compounds pass electrons on to higher potential mediators, and the electrons eventually enter the photosystem where they are used to reduce NAD. Flavocytochrome *c* from *Chromatium vinosum* was characterized by Bartsch and Kamen (1960) as a relatively abundant soluble protein. A similarly abundant flavocytochrome *c* was found in *Chlorobium* by Meyer et al. (1968). Both proteins were found to have sulfide dehydrogenase activity *in vitro* (Kusai and Yamanaka 1973a; Yamanaka and Kusai 1976; Fukumori and Yamanaka 1979a). This was not thought to be remarkable at the time because sulfide is a strong reducing agent that is capable of reducing a variety of cytochromes, although at a rate that is an order of magnitude less than the enzymatic reaction. However, the balance of opinion now favors enzymatic oxidation of sulfide. Hence the name flavocytochrome *c*–sulfide dehydrogenase (FCSD). The flavin of the FCSDs has a relatively high redox potential which makes it reactive with a variety of ligands including sulfite, cyanide, thiosulfate, and mercaptans. But, these compounds generally do not reduce the protein (Meyer and Bartsch 1976) and are not substrates although they do act as inhibitors of sulfide oxidation. The amino acid sequence of FCSD and three-dimensional structure show that the flavoprotein subunit is related to glutathione reductase, but has a different active site disulfide on the opposite side of the flavin (Dolata et al. 1993; Chen et al. 1994; Van Driessche et al. 1996). *Chlorobium* cytochrome *c*555 forms a complex with FCSD and serves as electron acceptor (Davidson et al. 1986), and a similar relationship was observed between cytochrome c_8 and FCSD in *Chromatium* (Davidson et al. 1985). HiPIP might also serve as mediator between FCSD and the photosystem. There is conflicting evidence as to whether FCSD is actually a sulfide dehydrogenase *in vivo*, since knockout mutants of FCSD in *Chromatium* do not impair the ability of cells to grow on or to utilize sulfide (Reinartz et al. 1998) and FCSD has not been found in all species of purple and green sulfur bacteria.

Another sulfide-oxidizing enzyme, initially isolated from the cyanobacterium *Oscillatoria limnetica* (Arieli et al. 1994), called sulfide quinone reductase (SQR), has since been found in several other species, including *Rb. capsulatus, Chlorobium*, and *Aquifex* (Shahak et al. 1999). However, the sequences of SQRs (Schütz et al. 1999; Bronstein et al. 2000) indicate that they are homologous to the flavoprotein subunit of FCSD and they have the same unusual disulfide. SQR is a membrane-bound periplasmic protein but lacks a signal peptide for translocation (Schütz et al. 1999) suggesting that it is transported across the membrane by association with a reaction partner protein which is as yet unidentified. A membrane-bound form of FCSD has been found in *E. vacuolata* (Kostanjevecki et al. 2000) and a second gene for FCSD has been found in the *Chlorobium* genome as part of the thiosulfate utilization gene cluster (Verté et al. 2002; Eisen et al. 2002). Purification of sulfide oxidizing activity in a *Thiobacillus* resulted in the isolation of a flavocytochrome *c* (Visser et al. 1997). Thus, our understanding of the role of the sulfide dehydrogenase *in vivo* is uncertain. It is likely that FCSD, its isozymes, and SQR may all be involved depending on growth conditions.

Chlorobium soluble cytochrome c*551 (SoxA)*

An unusual cytochrome *c*551 was isolated from *Chlorobium* by Meyer et al. (1968), which was found to be involved in oxidation of thiosulfate although it was not believed to be the thiosulfate oxidase enzyme (Kusai and Yamanaka 1973b). The amino acid sequence shows that it is a monoheme protein of about 30 kDa that also contains a disulfide (Klarskov et al. 1998). Homologs have been found in *Paracoccus pantotrophus, Aquifex aeolicus, Rhodopseudomonas palustris, Rhodobacter sulfidophilus*, and *Alcaligenes eutrophus* (Appia-Ayme et al. 2001 and references therein) where they are known as SoxA. Thus, SoxA is not unique to the photosynthetic bacteria or even to the green bacteria. Most of the SoxA proteins have two hemes. The three-dimensional structure of the *Rb. sulfidophilum* SoxAX complex indicates that SoxA is a class I cytochrome *c* with histidine/cysteine heme ligation (Bamford et al. 2002). The gene for *Chlorobium* cytochrome *c*551 was cloned and found to be regulated by thiosulfate as part of a cluster of eight genes including an isozyme of flavocytochrome *c*–sulfide dehydrogenase (FCSD) (Verté et al. 2002). The homolog of cytochrome *c*551 from Paracoccus is also part of a cluster of genes (Friedrich 1998; Friedrich et al. 2000, 2001), most of which are related to those of *Chlorobium*, including FCSD (called SoxEF). Similar gene clusters have been found in *Rhodobacter sulfidophilus* and the other species that have a cytochrome *c*551 (Appia-Ayme et al. 2001). Although the enzymolgy of thiosufate utilization has been studied more in *Paracoccus* than in *Chlorobium*,

Figure 3. The authors: Terry Meyer (left) and Mike Cusanovich (right).

the SoxYZ protein is present in both species where it appears to be a sulfur-binding protein, SoxB is a nucleotidase homolog, and SoxCD is a sulfite dehydrogenase (Friedrich 1998). Thiosulfate and sulfur oxidation are active areas of research and it is not yet certain exactly how the proteins produced by the thiosulfate gene cluster (including SoxA) interact to oxidize reduced sulfur compounds. It was shown that *Chlorobium* cytochrome *c*555 stimulates the oxidation of thiosulfate (Kusai and Yamanaka 1973b) and is likely to transfer electrons between SoxA and the photosystem.

Conclusions

The 20th century has seen the discovery of an incredibly large and diverse number of redox proteins in the photosynthetic bacteria, only a few of which have been covered by this review. Although in some cases the redox proteins are well characterized and their function clearly understood, in many cases the functional importance has eluded us to date. Looking to the future, it is clear that genome sequences are now revolutionizing the study of electron transfer including that of photosynthesis. The genomes of *Chlorobium tepidum* (Eisen et al. 2002), *Chloroflexus aurantiacus* (Joint Genome Institute), *Rhodobacter capsulatus* (Integrated Genomics; Haselkorn et al. 2001), *Rhodobacter sphaeroides* (Joint Genome Institute; Mackenzie et al. 2001), *Rhodopseudomonas palustris* (Joint Genome Institute), *Heliobacillus mobilis* (Integrated Genomics), and *Chromatium tepidum* (Integrated Genomics) as well as of a half-dozen cyanobacteria (Joint Genome Institute; Kasuza DNA Research Institute; Kaneko et al. 1996, 2001; Nakamura et al. 2002) have been or are now being completed. These sequences have already indicated that there are many more potentially photosynthetically relevant cytochromes than previously imagined. There are at least 12 *c*-type cytochromes in *Rb. capsulatus*, 21 each in *Rps. palustris* and *Rb. sphaeroides*, 8 in *Ch. tepidum*, 7 in *Cfx. aurantiacus*, and 3–7 in the cyanobacteria. This may be compared and contrasted with the 7 in *E. coli*, approximately 32 in *Pseudomonas aeruginosa*, and 42 in *Shewanella oneidensis*. Some of these are genuine cytochrome genes that have functions well-characterized in other species, such as bacterial cytochrome *c* peroxidase (BCCP), soluble nitrate reductase (Nap), nitrite reductase (Nrf), nitric oxide reductase (Nor), trimethylamine N-oxide reductase (Tor), specialized cytochrome oxidase (Fix), and the more usual cytochrome oxidase (Cox). However, the remainder of these genes provide a challenge to future biochemists to determine if and how they fit into photosynthetic electron transfer and the metabolism of the photosynthetic bacteria.

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References

Albert I, Rutherford AW, Grav H, Kellermann J and Michel H (1998) The 18 kDa cytochrome *c*553 from *Heliobacterium gestii*: gene sequence and characterization of the mature protein. Biochemistry 37: 9001–9008

Allen JP, Feher G, Yeates TO, Komiya H and Rees DC (1987) Structure of the reaction center from *Rhodobacter sphaeroides* R-26: the protein subunits. Proc Natl Acad Sci USA 84: 6162–6166

Almassy RJ and Dickerson RE (1978) *Pseudomonas* cytochrome *c*551 at 2.0 Å resolution: enlargement of the cytochrome *c* family. Proc Natl Acad Sci USA 75: 2674–2678

Ambler RP (1963) The amino acid sequence of *Pseudomonas* cytochrome *c*551. Biochem J 89: 349–378

Ambler RP (1991) Sequence variability in bacterial cytochromes *c*. Biochim Biophys Acta 1058: 42–47

Ambler RP, Daniel M, Hermoso J, Meyer TE, Bartsch RG and Kamen MD (1979a) Cytochrome c_2 sequence variation among the recognized species of purple non-sulfur photosynthetic bacteria. Nature (London) 278: 659–660

Ambler RP, Meyer TE and Kamen MD (1979b) Anomalies in amino acid sequences of small cytochromes *c* and cytochromes c' from two species of purple photosynthetic bacteria. Nature (London) 278: 661–662

Ambler RP, Bartsch RG, Daniel M, Kamen MD, McLellan L, Meyer TE and Van Beeumen JJ (1981) Amino acid sequences of bacterial cytochromes c' and *c*-556. Proc Natl Acad Sci USA 78: 6854–6857

Ambler RP, Daniel M, Melis K and Stout CD (1984) The amino acid sequence of the dihaem cytrochrome c_4 from the bacterium *Azotobacter vinelandii*. Biochem J 222: 217–227

Ambler RP, Meyer TE and Kamen MD (1993) Amino acid sequences of cytochromes *c*-551 from the halophilic purple phototrophic bacteria *Ectothiorhodospira halophila* and *E. halochloris*. Arch Biochem Biophys 306: 83–93

Appia-Ayme C, Little PJ, Matsumoto Y, Leech AP and Berks BC (2001) Cytochrome complex essential for photosynthetic oxidation of both thiosulfate and sulfide in *Rhodovulum sulfidophilum*. J Bacteriol 183: 6107–6118

Arieli B, Shahak Y, Taglicht D, Hauska G and Padan E (1994) Purification and characterization of sulfide-quinone reductase, a novel enzyme driving anoxygenic photosynthesis in *Oscillatoria limnetica*. J Biol Chem 269: 5705–5711

Axelrod HL, Abresch EC, Okamura MY, Yeh AP, Rees DC and Feher G (2002) X-Ray structural determination of the cytochrome c_2: reaction center electron transfer complex from *Rhodobacter sphaeroides*. J Mol Biol 319: 501–515

Bamford BA, Bruno S, Rasmussen T, Appia-Ayme C, Cheesman MR, Berks BC and Hemmings AM (2002) Structural basis for the oxidation of thiosulfate by a sulfur cycle enzyme. EMBO J 21: 5599–5610

Bartsch RG (1963) Nonheme iron proteins and *Chromatium* iron protein. In: Gest H, San Pietro A and Vernon LP (eds) Bacterial Photosynthesis, pp 315–326. Antioch Press, Yellow Springs, Ohio

Bartsch RG and Kamen MD (1960) Isolation and properties of two soluble heme proteins in extracts of the photoanaerobe *Chromatium*. J Biol Chem 235: 825–831

Bersch B, Blackledge MJ, Meyer TE and Marion D (1996) *Ectothiorhodospira halophila* ferrocytochrome *c*551: NMR solution structure and comparison with bacterial cytochromes *c*. J Mol Biol 264: 567–584

Bond CS, Blankenship RE, Freeman HC, Guss JM, Maher MJ, Selvaraj FM, Wilce MCJ and Willingham KM (2001) Crystal structure of auracyanin, a 'blue' copper protein from the green thermophilic photosynthetic bacterium *Chloroflexus aurantiacus*. J Mol Biol 306: 47–67

Bronstein M, Schütz M, Hauska G, Padan E and Shahak Y (2000) Cyanobacterial sulfide-quinone reductase: cloning and heterologous expression. J Bacteriol 182: 3336–3344

Bruce BD, Fuller RC and Blankenship RE (1982) Primary photochemistry in the facultatively aerobic green photosynthetic bacterium *Chloroflexus aurantiacus*. Proc Natl Acad Sci USA 79: 6532–6536

Büttner B, Xie D-L, Nelson H, Pinther W, Hauska G and Nelson N (1992) Photosynthetic reaction center genes in green sulfur bacteria and in Photosystem 1 are related. Proc Natl Acad Sci USA 89: 8135–8139

Candela M, Zaccherini E and Zannoni D (2001) Respiratory electron transport and light-induced energy transduction in membranes from the aerobic photosynthetic bacterium *Roseobacter denitrificans*. Arch Microbiol 175: 168–177

Carter DC, Melis KA, O'Donnell SE, Burgess BK, Furey WF Jr, Wang B-C and Stout CD (1985) Crystal structure of *Azotobacter* cytochrome c_5 at 2.5 Å resolution. J Mol Biol 184: 279–295

Chen Z, Koh M, Van Driessche G, Van Beeumen JJ, Bartsch RG, Meyer TE, Cusanovich MA and Mathews FS (1994) The structure of flavocytochrome *c*–sulfide dehydrogenase from the purple phototrophic bacterium, *Chromatium vinosum*. Science 266: 430–432

Colbeau A, Kovacs KL, Chabert J and Vignais PM (1994) Cloning and sequencing of the structural genes (hupSLC) and accessory genes (hupDHI) for hydrogenase biosynthesis in *Thiocapsa roseopersicina*. Gene 140: 25–31

Cusanovich MA and Bartsch RG (1969) A high potential cytochrome *c* from *Chromatium* chromatophores. Biochim Biophys Acta 189: 245–255

Daldal F, Cheng S, Applebaum J, Davidson E and Prince RC (1986) Cytochrome c_2 is not essential for photosynthetic growth of *Rhodopseudomonas capsulata*. Proc Natl Acad Sci USA 83: 2012–2016

Davidson MW, Gray GO and Knaff DB (1985) Interaction of *Chromatium vinosum* flavocytochrome *c*-552 with cytochromes *c* studied by affinity chromatography. FEBS Lett 187: 155–159

Davidson MW, Meyer TE, Cusanovich MA and Knaff DB (1986) Complex formation between *Chlorobium limicola* f. *thiosulfatophilum c*-type cytochromes. Biochim Biophys Acta 850: 396–401

Deisenhofer J, Epp O, Miki K, Huber R and Michel H (1985) Structure of the protein subunits in the photosynthetic reaction centre of *Rhodopseudomonas viridis* at 3 Å resolution. Nature (London) 318: 618–624

Dickerson RE, Takano T, Eisenberg D, Kallai OB, Samson L, Cooper A and Margoliash E (1971) Ferricytochrome *c*. 1. General features of the horse and bonito proteins at 2.8 Å resolution. J Biol Chem 246: 1511–1535

Dolata MM, Van Beeumen JJ, Ambler RP, Meyer TE and Cusanovich MA (1993) Nucleotide sequence of the flavocytochrome *c* heme subunit from the purple phototrophic bacterium, *Chromatium vinosum*. A 2.6 kilobase pair DNA fragment contains two multiheme cytochromes, a flavoprotein and a homolog of human ankyrin. J Biol Chem 268: 14426–14431

Donohue TJ, McEwan AG, Van Doren S, Crofts AR and Kaplan S (1988) Phenotypic and genetic characterization of cytochrome c_2 deficient mutants of *Rhodobacter sphaeroides*. Biochemistry 27: 1918–1925

Dracheva S, Williams JC, Van Driessche G, Van Beeumen JJ and Blankenship RE (1991) The primary structure of cytochrome *c*-554 from the green photosynthetic bacterium *Chloroflexus aurantiacus*. Biochemistry 30: 11451–11458

Dus K, Sletten K and Kamen MD (1968) Cytochrome c_2 of *Rhodospirillum rubrum* II. Complete amino acid sequence and phylogenetic relationships. J Biol Chem 243: 5507–5518

Eisen JA, Nelson KE, Paulsen IT, Heidelberg JF, Wu M, Dodson RJ, Deboy R, Gwinn ML, Nelson WC, Haft DH, Hickey EK, Peterson JD, Durkin AS, Kolonay JL, Yang F, Holt I, Umayam LA, Mason T, Brenner M, Shea TP, Parksey D, Nierman WC, Feldblyum TV, Hansen CL, Craven MB, Radune D, Vamathevan J, Khouri H, White O, Venter JC, Gruber TM, Ketchum KA, Tettelin H, Bryant DA and Fraser CM (2002) The complete genome sequence of *Chlorobium tepidum* TLS, a photosynthetic, anaerobic, green-sulfur bacterium. Proc Natl Acad Sci USA 99: 9509–14.

Errede B and Kamen MD (1978) Comparative kinetic studies of cytochromes *c* in reactions with mitochondrial cytochrome *c* oxidase and reductase. Biochemistry 17: 1015–1027

Even MT, Kassner RJ, Dolata M, Meyer TE and Cusanovich MA (1995) Molecular cloning and sequencing of cytochrome c' from the photosynthetic purple sulfur bacterium, *Chromatium vinosum*. Biochim Biophys Acta 1231: 220–222

Feick RG, Fitzpatrick M and Fuller RC (1982) Isolation and characterization of cytoplasmic membranes and chlorosomes from the green bacterium *Chloroflexus aurantiacus*. J Bacteriol 150: 905–915

Fischer U (1988) Soluble electron-transfer proteins of *Chlorobiaceae*. In: Olson JM, Ormerod JG, Amesz J, Stackebrandt E and Trüper HG (eds) Green Photosynthetic Bacteria, pp 127–131. Plenum Press, New York

Flory JE and Donohue TJ (1995) Organization and expression of the *Rhodobacter sphaeroides* cycFG operon. J Bacteriol 177: 4311–4320

Fowler CF, Nugent NA and Fuller RC (1971) The isolation and characterization of a photochemically active complex from *Chloropseudomonas ethylica*. Proc Natl Acad Sci USA 68: 2278–2282

Freeman JC and Blankenship RE (1990) Isolation and characterization of the membrane-bound cytochrome *c*-554 from the thermophilic green photosynthetic bacterium *Chloroflexus aurantiacus*. Photosynth Res 23: 29–38

Friedrich CG (1998) Physiology and genetics of sulfur-oxidizing bacteria. Adv Microbial Physiol 39: 235–289.

Friedrich CG, Quentmeier A, Bardischewsky F, Rother D, Kraft R, Kostka S and Prinz H (2000) Novel genes coding for lithotrophic sulfur oxidation of *Paracoccus pantotrophus*. J Bacteriol 182: 4677–4687

Friedrich CG, Rother D, Bardischewsky F, Quentmeier A and Fischer J (2001) Oxidation of reduced inorganic sulfur compounds by bacteria: emergence of a common mechanism. Appl Environ Microbiol 67: 2873–2882

Fukumori Y and Yamanaka T (1979a) Flavocytochrome *c* of *Chromatium vinosum*. Some enzymatic properties and subunit structure. J Biochem 85: 1405–1414

Fukumori Y and Yamanaka T (1979b) A high-potential nonheme iron protein (HiPIP)-linked, thiosulfate-oxidizing enzyme derived from *Chromatium vinosum*. Curr Microbiol 3: 117–120

Gabellini N and Sebald W (1986) Nucleotide sequence and transcription of the *fbc* operon from *Rhodopseudomonas sphaeroides*. Eur J Biochem 154: 569–579

Gabellini N, Bowyer JR, Hurt E, Melandri BA and Hauska G (1982) A cytochrome b/c_1 complex with ubiquinol-cytochrome c_2 oxidoreductase activity from *Rhodopseudomonas sphaeroides* GA. Eur J Biochem 126: 105–111

Gibson J (1961) Cytochrome pigments from the green photosynthetic bacterium *Chlorobium thiosulphatophilum*. Biochem J 79: 151–158

Haselkorn R, Lapidus A, Kogan Y, Vlcek C, Paces J, Paces V, Ulbrich P, Pecenkova T, Rebrekov D, Milgram A, Mazur M, Cox R, Kyrpides N, Ivanova N, Kapatral V, Los T, Lykidis A, Mikhailova N, Reznik G, Vasieva O, Fonstein M (2001) The *Rhodobacter capsulatus* genome. Photosynth Res 70: 43–52.

Hill R and Scarisbrick R (1951) The haematin compounds of leaves. New Phytol 50: 98–111

Hochkoeppler A, Ciurli S, Venturoli G and Zannoni D (1995) The high potential iron–sulfur protein (HIPIP) from *Rhodoferax fermentans* is competent in photosynthetic electron transfer. FEBS Lett 357: 70–74

Hochkoeppler A, Zannoni D, Ciurli S, Meyer TE, Cusanovich MA and Tollin G (1996) Kinetics of photo-induced electron transfer from high-potential iron–sulfur protein to the photosynthetic reaction center of the purple phototroph *Rhodoferax fermentans*. Proc Natl Acad Sci USA 93: 6998–7002

Horio T (1958a) Terminal oxidation system in bacteria. I. Purification of cytochromes from *Pseudomonas aeruginosa*. J Biochem 45: 195–205

Horio T (1958b) Terminal oxidation system in bacteria II. Some physical and physiological properties of purified cytochromes of *Pseudomonas aeruginosa*. J Biochem 45: 267–279

Horio T, Higashi T, Sasagawa M, Kusai K, Nakai M and Okunuki K (1960) Preparation of crystalline *Pseudomonas* cytochrome *c*-551 and its general properties. Biochem J 77: 194–201

Hurt EC and Hauska G (1984) Purification of membrane-bound cytochromes and a photoactive P840 protein complex of the green sulfur bacterium *Chlorobium limicola* f. *thiosulfatophilum*. FEBS Lett 168: 149–154

Jenney FE Jr, Prince RC and Daldal F (1994) Roles of the soluble cytochrome c_2 and membrane-associated cytochrome cy of *Rhodobacter capsulatus* in photosynthetic electron transfer. Biochemistry 33: 2496–2502

Kadziola A and Larsen S (1997) Crystal structure of the dihaem cytochrome c_4 from *Pseudomonas stutzeri* determined at 2.2 Å resolution. Structure 5: 203–216

Kamen MD and Takeda Y (1956) A comparative study of bacterial and mammalian cytochrome *c*. Biochim Biophys Acta 21: 518–523

Kamen MD and Vernon LP (1954) Existence of haem compounds in a photosynthetic obligate anaerobe. J Bacteriol 67: 617–618

Kaneko T, Sato S, Kotani H,Tanaka A, Azamizu E, Nakamura Y, Miyajima N, Hirosawa M, Sugiura M, Sasamoto S, Kimura T, Hosouchi T, Matsuno A, Muraki A, Nakazaki N, Naruo K, Okumura S, Shimpo S, Takeuchi C, Wada T, Watanabe A, Yamada M, Yasuda M and Tabata S (1996) Sequence analysis of the genome of the unicellular cyanobacterium *Synechocystis* sp. strain PCC 6803. II. Sequence determination of the entire genome and assignment of potential protein-coding regions. DNA Res 3: 109–136

Kaneko T, Nakamura Y, Wolk CP Kuritz T, Sasamoto S, Watanabe A, Iriguchi M, Ishikawa A, Kawashima K, Kimura T, Kishida Y, Kohara M, Marsumoto M, Matsono A, Muraki A, Nakazaki N, Shimpo S, Sugimoto M, Takazawa M, Yamada M, Yasuda M and Tabata S (2001) Complete genomic sequence of the filamentous nitrogen-fixing cyanobacterium *Anabaena* sp. strain PCC 7120. DNA Res 8: 205–213

Katoh S (1959) Studies on the algal cytochrome of *c*-type. J Biochem 46: 629–632

Katoh S (1960) A new copper protein from *Chlorella ellipsoidea*. Nature (London) 186: 533–534

Keilin D (1925) On cytochrome, a respiratory pigment, common to animals, yeast and higher plants. Proc R Acad Sci London Ser B 98: 312–339

Kennel SJ and Kamen MD (1971) Iron-containing proteins in *Chromatium* II. Purification and properties of cholate-solubilized cytochrome complex. Biochim Biophys Acta 234: 458–467

Kennel SJ, Bartsch RG and Kamen MD (1972) Observations on light-induced oxidation reactions in the electron transfer system of *Chromatium*. Biophys J 12: 882–896

Klarskov K, Verté F, Van Driessche G, Meyer TE, Cusanovich MA and Van Beeumen JJ (1998) The primary structure of soluble cytochrome *c*-551 from the phototrophic green sulfur bacterium, *Chlorobium limicola*. Biochemistry 37: 10555–10568

Konig BW, Schilder LTM, Tervoort MJ and Van Gelder BF (1980) The isolation and purification of cytochrome c_1 from bovine heart. Biochim Biophys Acta 621: 283–295

Kostanjevecki V, Brigé A, Meyer TE, Cusanovich MA and Van Beeumen JJ (2000) Sequence of the flavocytochrome *c*–sulfide dehydrogenase operon from the purple phototrophic bacterium, *Ectothiorhodospira vacuolata*. J Bacteriol 182: 3097–3103

Kusai A and Yamanaka T (1973a) Cytochrome *c*553, *Chlorobium thiosulfatophilum*, is a sulfide cytochrome *c* reductase. FEBS Lett 34: 235–237

Kusai A and Yamanaka T (1973b) The oxidation mechanisms of thiosulfate and sulfide in *Chlorobium thiosulphatophilum*: roles of cytochrome *c*-551 and cytochrome *c*-553. Biochim Biophys Acta 325: 304–314

Kusai A and Yamanaka T (1973c) An NADP reductase derived from *Chlorobium thiosulfatophilum*: purification and some properties. Biochim Biophys Acta 292: 621–633

Küsche WH and Trüper HG (1984) Cytochromes of the photosynthetic sulfur bacterium *Ectothiorhodospira shaposhnikovii*. Z Naturforsch 39c: 894–901

Laycock MV (1972) The amino acid sequence of cytochrome *c*-553 from the chrysophycean alga *Monochrysis lutheri*. Can J Biochem 50: 1311–1325

Lee WY, Brune DC, LoBrutto R and Blankenship RE (1995) Isolation, characterization, and primary structure of rubredoxin from the photosynthetic bacterium, *Heliobacillus mobilis*. Arch Biochem Biophys 318: 80–88

Lovenberg W and Sobel BE (1965) Rubredoxin: a new electron transfer protein from *Clostridium pasteurianum*. Proc Natl Acad Sci USA 54: 193–199

Mackenzie C, Choudhary M, Larimer FW, Predki PF, Stilwagen S, Armitage JP, Barber RD, Donohue TJ, Hosler JP, Newman JE, Shapleigh JP, Sockett RE, Zeilstra-Ryalls J and Kaplan S (2001) The home stretch, a first analysis of the nearly completed genome of *Rhodobacter sphaeroides* 2.4.1. Photosynth Res 70: 19–41.

MacMunn CA (1886) VI. Researches on myohaematin and the histohaematins. Phil Trans R Soc London 177: 267–298

Martinez SE, Huang D, Szczepaniak A, Cramer WA and Smith JL (1994) Crystal structure of chloroplast cytochrome *f* reveals a novel cytochrome fold and unexpected heme ligation. Structure 2: 95–105

McManus JD, Brune DC, Han J, Sanders-Loehr J, Meyer TE, Cusanovich MA, Tollin G and Blankenship RE (1992) Isolation, characterization, and amino acid sequences of auracyanins, blue copper proteins from the green photosynthetic bacterium *Chloroflexus aurantiacus*. J Biol Chem 267: 6531–6540

Menin L, Schoepp B, Parot P and Verméglio A (1997) Photoinduced cyclic electron transfer in *Rhodocyclus tenuis* cells: participation of HiPIP or cyt c_8 depending on the ambient redox potential. Biochemistry 36: 12183–12188

Menin L, Gaillard J, Parot P, Schoepp B, Nitschke W and Verméglio A (1998) Role of HiPIP as electron donor to the RC-bound cytochrome in photosynthetic purple bacteria. Photosynth Res 55: 343–348

Menin L, Yoshida M, Jaquinod M, Nagashima KVP, Matsuura K, Parot P and Verméglio A (1999) Dark aerobic growth conditions induce synthesis of a high midpoint potential cytochrome c_8 in the photosynthetic bacterium *Rubrivivax gelatinosus*. Biochemistry 38: 15238–15244

Merchant S and Bogorad L (1986) Regulation by copper of the expression of plastocyanin and cytochrome *c*552 in *Chlamydomonas reinhardi*. Mol Cell Biol 6: 462–469

Meyer TE (1985) Isolation and characterization of soluble cytochromes, ferredoxins, and other chromophoric proteins from the halophilic phototrophic bacterium, *Ectothiorhodospira halophila*. Biochim Biophys Acta 806: 175–183

Meyer TE (1994) Purification and properties of high-potential iron–sulfur proteins. Method Enzymol 243: 435–447

Meyer TE (1996) Evolution and classification of *c*-type cytochromes. In: Scott RA and Mauk AG (eds) Cytochrome *c*. A Multidisciplinary Approach, pp 33–99. University Science Books, Sausalito, California

Meyer TE and Bartsch RG (1976) The reaction of flavocytochrome *c* of the phototrophic sulfur bacteria with thiosulfate, sulfite, cyanide, and mercaptans. In: Singer TP (ed) Flavins and Flavoproteins, pp 312–317. Elsevier, Amsterdam

Meyer TE and Kamen MD (1982) New perspectives on *c*-type cytochromes. Adv Prot Chem 35: 105-212

Meyer TE, Bartsch RG, Cusanovich MA and Mathewson JH (1968) The cytochromes of *Chlorobium thiosulfatophilum*. Biochim Biophys Acta 153: 854–861

Meyer TE, Sharp JJ and Bartsch RG (1971) Isolation and properties of rubredoxin from the photosynthetic green sulfur bacteria. Biochim Biophys Acta 234: 266–269

Meyer TE, Van Beeumen JJ, Ambler RP and Cusanovich MA (1997) The evolution of electron transfer proteins in phototrophic bacteria and denitrifying pseudomonads. In: Baltscheffsky H (ed.) Origin and Evolution of Biological Energy Conversion, pp 71–108. VCH Publishers, New York

Mortenson LE, Valentine RC and Carnahan JE (1962) An electron transport factor from *Clostridium pasteurianum*. Biochem Biophys Res Comm 7: 448–452

Nakamura Y, Kaneko T, Sato S, Ikeuchi M, Katoh H, Sasamoto S, Watanabe A, Iriguchi M, Kawashima K, Kimura T, Kishida Y, Kiyokawa C, Kohara M, Matsumoto M, Matsuno A, Nakazaki N, Shimpo S, Sugimoto M, Takeuchi C, Yamada M and Tabata S (2002) Complete Genome Structure of the Thermophilic Cyanobacterium *Thermosynechococcus elongatus* BP-1. DNA Res. 9: 123–130.

Ng TCN, Laheri AN and Maier RJ (1995) Cloning, sequencing, and mutagenesis of the cytochrome c_4 gene from *Azotobacter vinelandii*: characterization of the mutant strain and a proposed new branch in the respiratory chain. Biochim Biophys Acta 1230: 119–129

Ng WV, Kennedy SP, Mahairas GG, Berquist B, Pan M, Shukla HD, Lasky SR, Baliga NS, Thorsson V, Sbrogna J, Swartzell S, Weir D, Hall J, Dahl TA, Welti R, Goo YA, Leithauser B, Keller K, Cruz R, Danson MJ, Hough DW, Maddocks DG, Jablonski PE, Krebs MP, Angevine CM, Dale H, Isenbarger TA, Peck RF, Pohlschroder M, Spudich JL, Jung KW, Alam M, Freitas T, Hou S, Daniels CJ, Dennis PP, Omer AD, Ebhardt H, Lowe TM, Liang P, Riley M, Hood L and DasSarma S (2000) Genome sequence of *Halobacterium* species NRC-1. Proc Natl Acad Sci U S A. 97: 12176–81.

Okkels JS, Kjær B, Hansson O, Svendsen I, Møller B and Scheller HV (1992) A membrane-bound monoheme cytochrome *c*551 of a novel type is the immediate electron donor to P840 of the

Chlorobium vibrioforme photosynthetic reaction center complex. J Biol Chem 267: 21139–21145

Olson JM and Chance B (1960a) Oxidation-reduction reactions in the photosynthetic bacterium *Chromatium*. 1. Absorption spectrum changes in whole cells. Arch Biochem Biophys 88: 26–39

Olson JM and Chance B (1960b) Oxidation-reduction reactions in the photosynthetic bacterium *Chromatium*. 2. Dependence of light reactions on intensity of irradiation and quantum efficiency of cytochrome oxidation. Arch Biochem Biophys 88: 40–53

Overfield RE, Wraight CA and Devault D (1979) Microsecond photooxidation kinetics of cytochrome c_2 from *Rhodopseudomonas sphaeroides*: *in vivo* and solution studies. FEBS Lett 105: 137–142

Parson WW and Case GD (1970) In *Chromatium*, a single photochemical reaction center oxidizes both cytochrome *c*552 and cytochrome *c*555. Biochim Biophys Acta 205: 232–245

Pettigrew GW, Meyer TE, Bartsch RG and Kamen MD (1975) pH dependence of the oxidation-reduction potential of cytochrome c_2. Biochim Biophys Acta 430: 197–208

Pettigrew GW, Meyer TE, Bartsch RG and Kamen MD (1978) Redox potentials of the photosynthetic bacterial cytochromes c_2 and the structural bases for variability. Biochim Biophys Acta 503: 509–523

Pierson BK and Thornber JP (1983) Isolation and spectral characterization of photochemical reaction centers from the thermophilic green bacterium *Chloroflexus aurantiacus* strain J-10-fl. Proc Natl Acad Sci USA 80: 80–84

Prince RC and Olson JM (1976) Some thermodynamic and kinetic properties of the primary photochemical reactants in a complex from a green photosynthetic bacterium. Biochim Biophys Acta 423: 357–362

Prince RC, Cogdell RJ and Crofts AR (1974) The photooxidation of horse heart cytochrome *c* and native cytochrome c_2 by reaction centres from *Rhodopseudomonas sphaeroides* R26. Biochim Biophys Acta 347: 1–13

Reinartz M, Tschäpe J, Brüser T, Trüper HG and Dahl C (1998) Sulfide oxidation in the phototrophic sulfur bacterium *Chromatium vinosum*. Arch Microbiol 170: 59–68

Rey L and Maier RJ (1997) Cytochrome *c* terminal oxidase pathways of *Azotobacter vinelandii*: analysis of cytochrome c_4 and c_5 mutants and up-regulation of cytochrome *c*-dependent pathways with N2 fixation. J Bacteriol 179: 7191–7196

Rickle GK and Cusanovich MA (1979) The kinetics of photooxidation of *c*-type cytochromes by *Rhodospirillum rubrum* reaction centers. Arch Biochem Biophys 197: 589–598

Robinson NC and Talbert L (1980) Isolation of bovine cytochrome c_1 as a single non-denatured subunit using gel filtration or high performance liquid chromatography in deoxycholate. Biochem Biophys Res Commun 95: 90–96

Salemme FR, Freer ST, Xuong NH, Alden RA and Kraut J (1973) The structure of oxidized cytochrome c_2 of *Rhodospirillum rubrum*. J Biol Chem 248: 3910–3921

Samyn B, De Smet L, Van Driessche G, Meyer TE, Bartsch RG, Cusanovich MA and Van Beeumen JJ (1996) A high-potential soluble cytochrome *c*-551 from the purple phototrophic bacterium *Chromatium vinosum* is homologous to cytochrome c_8 from denitrifying pseudomonads. Eur J Biochem 236: 689–696

Schatt E, Jouanneau Y and Vignais PM (1989) Molecular cloning and sequence analysis of the structural gene of ferredoxin I from the phototrophic bacterium, *Rhodobacter capsulatus*. J Bacteriol 171: 6218–6226

Schoepp B, Parot P, Menin L, Gaillard J, Richaud P and Verméglio A (1995) *In vivo* participation of a high potential iron–sulfur protein as electron donor to the photochemical reaction center of *Rubrivivax gelatinosus*. Biochemistry 34: 11736–11742

Schütz M, Zirngibl S, le Coutre J, Büttner M, Xie D-L, Nelson N, Deutzmann R and Hauska G (1994) A transcription unit for the Rieske FeS-protein and cytochrome *b* in *Chlorobium limicola*. Photosynth Res 39: 163–174

Schütz M, Maldener I, Griesbeck C and Hauska G (1999) Sulfide-quinone reductase from *Rhodobacter capsulatus*. Requirement for growth, periplasmic location, and extension of gene sequence analysis. J Bacteriol 181: 6516–6523

Shahak Y, Schütz M, Bronstein M, Griesbeck C, Hauska G and Padan E (1999) Sulfide-dependent anoxygenic photosynthesis in prokaryotes. In: Peschek GA, Loffelhardt W and Schmetterer (eds) The Phototrophic Prokaryotes, pp 217–228. Kluwer Academic Publishers/Plenum Press, New York

Shin M, Tagawa K and Arnon DI (1963) Crystallization of ferredoxin–TPN reductase and its role in the photosynthetic apparatus of chloroplasts. Biochem Z 338: 84–96

Shiozawa JA, Lottspeich F, Oesterhelt D and Feick R (1989) The primary structure of the *Chloroflexus aurantiacus* reaction-center polypeptides. Eur J Biochem 180: 75–84

Swank RT and Burris RH (1969) Purification and properties of cytochromes *c* of *Azotobacter vinelandii*. Biochim Biophys Acta 180: 473–489

Tagawa K and Arnon DI (1962) Ferredoxins as electron carriers in photosynthesis and in the biological production and consumption of hydrogen gas. Nature (London) 195: 537–543

Tedro SM, Meyer TE and Kamen MD (1976) Primary structure of a high potential iron–sulfur protein from the purple non-sulfur photosynthetic bacterium *Rhodopseudomonas gelatinosa*. J Biol Chem 251: 129–136

Tetreault M, Rongey SH, Feher G and Okamura MY (2001) Interaction between cytochrome c_2 and the photosynthetic reaction center from *Rhodobacter sphaeroides*: effects of charge modification mutants on binding and electron transfer. Biochemistry 40: 8452–8462

Tetreault M, Cusanovich M, Meyer T, Axelrod H and Okamura MY (2002) Double mutant studies identify electrostatic interactions that are important for docking cytochrome c_2 onto the bacterial reaction center. Biochemistry 41: 5807–5815

Then J and Trüper HG (1983) Sulfide oxidation in *Ectothiorhodospira abdelmalekii*. Evidence for the catalytic role of cytochrome *c*551. Arch Microbiol 135: 254–258

Then J and Trüper HG (1984) Utilization of sulfide and elemental sulfur by *Ectothiorhodospira halochloris*. Arch Microbiol 139: 295–298

Theorell H and Åkesson A (1939) Absorption spectrum of further purified cytochrome *c*. Science 90: 67

Tissières A (1956) Purification, some properties and the specific biological activity of cytochromes c_4 and c_5 from *Azotobacter vinelandii*. Biochem J 64: 582–589

Trost JT, McManus JD, Freeman JC, Ramakrishna BL and Blankenship RE (1988) Auracyanin, a blue copper protein from the green photosynthetic bacterium *Chloroflexus aurantiacus*. Biochemistry 27: 7858–7863

Van Beeumen JJ (1991) Primary structure diversity of prokaryotic diheme cytochromes *c*. Biochim Biophys Acta 1058: 56–60

Van Beeumen JJ, Ambler RP, Meyer TE, Kamen MD, Olson JM and Shaw EK (1976) The amino acid sequences of the cytochromes *c*-555 from two green sulfur bacteria of the genus *Chlorobium*. Biochem J 159: 757–774

Van Beeumen JJ, De Mol H, Samyn B, Bartsch RG, Meyer TE, Dolata MM and Cusanovich MA (1991) Covalent structure of the diheme cytochrome subunit and amino-terminal sequence of

the flavoprotein subunit of flavocytochrome *c* from *Chromatium vinosum*. J Biol Chem 266: 12921–12931

Van Driessche G, Koh M, Chen ZW, Mathews FS, Meyer TE, Bartsch RG, Cusanovich MA and Van Beeumen JJ (1996) Primary structure of the flavoprotein subunit of the flavocytochrome *c*–sulfide dehydrogenase from the purple phototrophic bacterium *Chromatium vinosum*. Prot Sci 5: 1753–1764

Van Driessche G, Hu W, Van de Werken G, Selvariaj F, McManus JD, Blankenship RE and Van Beeumen JJ (1999) Auracyanin A from the thermophilic green gliding photosynthetic bacterium *Chloroflexus aurantiacus* represents an unusual class of small blue copper proteins. Prot Sci 8: 947–957

van Grondelle R, Duysens LNM, van der Wel JA and van der Wal HN (1977) Function and properties of a soluble *c*-type cytochrome *c*-551 in secondary photosynthetic electron transport in whole cells of *Chromatium vinosum* as studied with flash spectroscopy. Biochim Biophys Acta 461: 188–201

Vernon LP (1953) Cytochrome *c* content of *Rhodospirillum rubrum*. Arch Biochem Biophys 43: 492–493

Vernon LP and Kamen MD (1954) Hematin compounds in photosynthetic bacteria. J Biol Chem 211: 643–663

Verté F, Kostanjevecki V, De Smet L, Meyer TE, Cusanovich MA and Van Beeumen JJ (2002) Identification of a thiosulfate utilization gene cluster from the green phototrophic bacterium *Chlorobium limicola*. Biochemistry 41: 2932–2945

Visser JM, de Jong GAH, Robertson LA and Kuenen JG (1997) A novel membrane-bound flavocytochrome *c* sulfide dehydrogenase from the colourless sulfur bacterium *Thiobacillus* sp. W5. Arch Microbiol 167: 295–301

Wachtveitl J, Farchaus JW, Das R, Lutz M, Robert B and Mattioli TA (1993) Structure, spectroscopic, and redox properties of *Rhodobacter sphaeroides* reaction centers bearing point mutations near the primary electron donor. Biochemistry 32: 12875–12886

Weber PC, Bartsch RG, Cusanovich MA, Hamlin RC, Howard A, Jordan SR, Kamen MD, Meyer TE, Weatherford DW, Xuong NH and Salemme FR (1980) Structure of cytochrome c': a dimeric, high-spin haem protein. Nature (London) 286: 302–304

Wood PM (1978) Interchangeable copper and iron proteins in algal photosynthesis, studies on plastocyanin and cytochrome *c*552 in *Chlamydomonas*. Eur J Biochem 87: 7–19

Wood PM (1980) Do photosynthetic bacteria contain cytochrome c_1? Biochem J 189: 385–391

Wood PM and Bendall DS (1975) The kinetics and spectroscopy of electron transfer from cytochromes and copper proteins to P700. Biochim Biophys Acta 387: 115–128

Xiong J, Inoue K and Bauer CE (1998) Tracking molecular evolution of photosynthesis by characterization of a major photosynthesis gene cluster from *Heliobacillus mobilis*. Proc Natl Acad Sci USA 95: 14851–14856

Yakushiji E (1935) Occurrence of cytochrome in higher plants and algae. Acta Phytochim (Japan) 8: 325–329

Yakushiji E and Okunuki K (1940) New cytochrome component and its function. Proc Imp Acad (Tokyo) 16: 299–302

Yamanaka T and Kusai A (1976) The function and some molecular features of cytochrome *c*-553 derived from *Chlorobium thiosulfatophilum*. In: Singer TP (ed) Flavins and Flavoproteins, pp 292–301. Elsevier, Amsterdam

Yamanaka T and Okunuki K (1968) Comparative studies on reactivities of cytochrome *c* with cytochrome oxidases. In: Okunuki K, Sekuzu I and Kamen MD (eds) Structure and Function of Cytochromes, pp 390–403. University Park Press, Baltimore, Maryland

Yoon KS, Hille R, Hemann C and Tabita FR (1999) Rubredoxin from the green suflur bacterium *Chlorobium tepidum* functions as an electron acceptor for pyruvate ferredoxin oxidoreductase. J Biol Chem 274: 29772–29778

Zhang Z, Huang L, Shulmeister VM, Chi YI, Kim KK, Hung LW, Crofts AR, Berry EA and Kim SH (1998) Electron transfer by domain movement in cytochrome bc_1. Nature (London) 392: 677–684