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Breakthroughs and Views

Protein prenylation: a pivotal posttranslational process☆

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The joining of the 15-carbon farnesyl group ($C_{15}H_{25}$) and the 20-carbon geranylgeranyl group ($C_{20}H_{33}$) to protein–cysteines at or near their carboxy-termini is catalyzed by protein farnesyltransferase (FTase) and protein geranylgeranyltransferase-I and II (GGTase-I and GGTase-II) [1]. The prenyltransferases are heterodimers consisting of α- and β-subunits with combined molecular masses ranging from 91 to 98 kDa. The α-subunits of FTase and GGTase-I are the same, and the β-subunits differ. The β-subunits of the three enzymes are homologous to the α-subunits and to each other. The overall reactions are shown by the following chemical equations:

$$\text{FPP} + \text{HS-peptide} \rightarrow \text{farnesyl-}S\text{-peptide} + \text{PP}$$

$$\text{GGPP} + \text{HS-peptide} \rightarrow \text{geranylgeranyl-}S\text{-acceptor} + \text{PP}$$

The isoprenoid groups become linked to polypeptidic cysteines through thioether (C–S–C) bonds.

The importance of the prenyltransferases is underscored by the nature of their substrates, many of which participate in signal transduction pathways related to cell growth, differentiation, cytoskeletal function, and vesicle trafficking (Table 1) [2–4]. Of added significance, Ras is a substrate for FTase and mutants of *ras* occur in 10–15% of all human cancers [5]. Conversion of the protein–cysteine acceptor site to protein-serine in oncogenic H-Ras prevents prenylation and abolishes its malignant transforming ability [6]. FTase inhibitors are effective in the treatment of animal models of cancer and inhibition of Ras farnesylation represents a possible strategy for the treatment of human tumors [7]. FTase inhibitors may be effective in tumor cells containing non-mutant Ras proteins that are activated by upstream oncoproteins. Moreover, it is likely that inhibition of the farnesylation of a combination of substrates constitutes the mechanism of anti-neoplastic action of FTase inhibitors [3,7]. FTase inhibitors are currently used in clinical trials for the treatment of human cancer [7–12].

FTase and GGTase-I catalyze the prenylation of substrates with a carboxy-terminal tetrapeptide sequence called a CaaX box, where C refers to cysteine, a refers to an aliphatic residue, and X typically refers to methionine, serine, alanine, or glutamine for FTase or to leucine for GGTase-I (Table 1). Following prenylation of physiological substrates, the terminal three residues (aaX) are subsequently removed by a CaaX endoprotease and the carboxyl group of the terminal cysteine is methyl esterified [1]. Protein geranylgeranyltransferase-II (GGTase-II), or Rab geranylgeranyltransferase, catalyzes the geranylgeranylation of Rab proteins that terminate in CC or CXC sequences. Rab proteins ending with CXC residues are methyl esterified; those ending with CC are not. FTase and GGTase-I can catalyze the prenylation of tetrapeptides, polypeptides, and proteins containing appropriate CaaX box sequences. GGTase-II, in contrast, cannot catalyze the prenylation of peptides; it uses a Rab–Rab escort protein heterodimer as substrate [1,13].

There are a few exceptions to the substrate specificity rules for FTase and GGTase-I noted above. K-RasB, which has a classical FTase CaaX box (CVIM), is a substrate for FTase. Following inhibition of cellular FTase, K-RasB becomes a substrate for geranylgeranylation by GGTase-I [7,14]. The latter reaction is made possible by an upstream polybasic sequence that alters GGTase-I substrate specificity. Furthermore, RhoB, which contains a GGTase-I CaaX box (CKVL),

☆ *Abbreviations:* BZA-2B, Cys-(*N*-methyl)-3-amino-1-carboxymethyl-2,3-dihydro-5-phenyl-1H-1,4-benzodiazepin-2-one-Met; F-peptide, farnesylpeptide; FPP, farnesyl diphosphate; FTase, protein farnesyltransferase; GGPP, geranylgeranyl diphosphate; GGTase, protein geranylgeranyltransferase; PP, inorganic diphosphate.

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Table 1
Selected prenyltransferase substrates[a]

Protein	GTPase	Protein function	Sequence[b]	Prenyl group[c]	Transferase[c]
H-ras	+	Growth, differentiation	CVLS	15C	FTase
K-rasA	+	Growth, differentiation	CIIM	15C	FTase
K-rasB	+	Growth, differentiation	CVIM	15C	FTase
N-ras	+	Growth, differentiation	CVVM	15C	FTase
2′, 5′ Oligoadenylate synthetase 1	0	Growth, differentiation, and apoptosis	CTIL	20C	GGTase-I
RaplA	+	Regulation of cell adhesion	CLLL	20C	GGTase-I
RaplB	+	Activation of the MEK-ERK cascade	CQLL	20C	GGTase-I
Rac1	+	Secretion at plasma membrane	CLLL	20C	GGTase-I
RalA	+	Regulation of actin cytoskeleton	CCIL	20C	GGTase-I
Cdc42/G25K	+	Filopodia formation	CCIF	20C	GGTase-I
RhoA	+	Assembly of actin stress fibers and focal adhesion sites	CLVL	20C	GGTase-I
RhoB	+	Assembly of actin stress fibers and focal adhesion sites; gene transcription via SRF	CKVL	20C and 15C	GGTase-I
Rab2	+	Vesicular trafficking	GGCC	20C	GGTase-II
Rab3a	+	Vesicular trafficking	DCAC	20C	GGTase-II
HDJ2	0	Protein import into mitochondria, co-chaperone of Hsc70.	CQTS	15C	FTase
Inositol-1,4,5-trisphosphate-5-phosphatase II	0	Inactivates inositol trisphosphate	CPNL	20C	GGTase-I
Ptp4a1	0	Protein–tyrosine phosphatase	CCIQ	15C	FTase
S. cerevisiae RAS2	+	Adenylyl cyclase activation	CIIS	15 C	FTase
Heterotrimeric G-protein (γ-subunit)	0	Serpentine receptor linked	CAIL	20C	GGTase-I
S. cerevisiae a-factor	0	Mating pheromone	CVIA	15C	FTase
R. toruloides Rhodotorucine A	0	Mating pheromone	CTVA	15C	FTase
Lamin A	0	Nuclear membrane component	CSIM	15 C	FTase
Lamin B	0	Nuclear membrane component	CYVM	15C	FTase
Cenp-F	0	Centromere (kinetochore) protein for G_2/M transition	CKVQ	15C	FTase
Phosphorylase kinase, α-subunit	0	Muscle glycogen metabolism	CAMQ	15C	FTase
Phosphorylase kinase, α-subunit	0	Liver glycogen metabolism	CQMQ	15C	FTase
Phosphorylase kinase, β-subunit	0	Muscle glycogen metabolism	CLIS	15C	FTase
Transducin (γ-subunit)	0	Vision	CVIS	15C	FTase
Retinal cGMP phosphodiesterase α-subunit	0	Vision	CCIQ	15C	FTase
Retinal cGMP phosphodiesterase β-subunit	0	Vision	CCIL	20C	GGTase-I
Rhodopsin kinase	0	Vision	CVLS	15C	FTase
RhoE	0	Regulation of the actin cytoskeleton	CTVM	15C	FTase
Rap2a	+	Function unknown	CNIQ	15C	FTase
Rap2b	+	Function unknown	CVIL	20C	GGTase-I
Rheb	+	Function unknown	CSVM	15C	FTase
PxF	0	Peroxisome assembly	CLIM	15C	FTase
Interferon induced guanylate binding protein-1	?	Binds GMP, GDP, and GTP in macrophages	CTIS	15C	FTase
Interferon induced guanylate binding protein-2	?	Binds GMP, GDP, and GTP in macrophages	CNIL	20C	GGTase-I

[a] Sources of the information in Table 1 are from [2–4].
[b] Human sequences are given unless otherwise noted.
[c] The nature of the prenyl group and prenyltransferase used is often inferred.

is found in both farnesylated and geranylgeranylated forms in cells. This is due to the ability of GGTase-I to both geranylgeranylate and farnesylate this substrate [15]. It appears that upstream sequences (as yet uncharacterized) are responsible for this altered substrate specificity. Moreover, Cdc42, which contains a carboxy-terminal CCIF sequence, undergoes geranylgeranylation. Ordinarily GGTase-I substrates contain leucine in the X position of the CaaX box, but Cdc42 represents an exception to the leucine rule.

The role of metals in prenyltransferase reactions

All three prenyltransferases require Zn^{2+}, and FTase and GGTase-II require Mg^{2+} for activity [13,14,16–20]. Zn^{2+} greatly enhances the productive binding of protein or peptide substrates to FTase and GGTase-I, but it is not required for the binding of prenyl diphosphate. Furthermore, interaction of the CaaX sulfur with the FTase Zn^{2+} lowers the pK_a of the thiol by approximately two pH units, suggesting that the Zn^{2+}-coordinated

thiolate ($RS:^-$) is present at physiological pH. Activity of FTase or GGTase-I can be restored by reconstitution with Co^{2+} or Cd^{2+}. Initial evidence for a direct interaction of a CaaX cysteine sulfur with metal came from optical absorbance spectra of Co^{2+} substituted FTase. An increase in absorbance at 340 nm occurs upon addition of a CaaX peptide substrate to an FTase · isoprenoid complex showing that the sulfur from the peptide is coordinated to the metal as indicated by this Co^{2+}-sulfur charge transfer band [19]. X-ray crystallographic studies amply confirm the catalytic role of Zn^{2+} for FTase [17].

The role of Mg^{2+} in the reaction catalyzed by FTase is less well understood than the role of Zn^{2+}. Mg^{2+} is not strictly required for catalysis, but millimolar concentrations are required for maximal rates of product formation. Mg^{2+} increases the rate of peptide farnesylation by 700-fold. By measuring the pH dependence of the chemical step of product formation under single-turnover conditions, Saderholm et al. [21] demonstrated that the farnesylation rate constant is enhanced by two deprotonations. The first ionization reflects deprotonation of the metal-coordinated thiol of the peptide CaaX motif with a pK_{a1} of 6.0. The second ionization was assigned to a hydroxyl on the diphosphate of FPP with a pK_{a2} of 7.4. Deprotonation of this group is important for binding of Mg^{2+}. This second ionization effect is not observed for catalysis in the absence of Mg^{2+} or when the substrate is farnesyl monophosphate. These data indicate that the maximal rate of farnesylation involves a Zn^{2+}-coordinated thiolate nucleophile and a Mg^{2+}-coordinated diphosphate leaving group. Mn^{2+} will substitute for Mg^{2+}, and X-ray studies show that the Mn^{2+} coordinates the diphosphate of the isoprenoid [17]. In contrast to FTase and GGTase-II, Zhang and Casey [22] showed that GGTase-I does not require Mg^{2+} for activity.

Both electrophilic and nucleophilic mechanisms have been proposed for FTase [23,24]. Huang et al. [24] studied the detailed catalytic mechanism of mammalian protein farnesyltransferase by measuring the effect of metal substitution and substrate modifications on the rate constant of the chemical step. Substitution of Cd^{2+} for the active site Zn^{2+} enhances peptide affinity approximately 5-fold and decreases the rate constant for the formation of the thioether product approximately 6-fold, indicating changes in the metal–thiolate coordination in the catalytic transition state. In addition, the observed rate constant for product formation decreases for C-3 fluoromethyl farnesyl diphosphate substrates, paralleling the number of fluorine atoms at the C-3 methyl position and indicating that a rate-contributing transition state has carbocation character. These data suggest that FTase catalyzes protein farnesylation by an associative mechanism with a transition state where the metal-bound peptide/protein sulfur has a partial negative charge, the C-1 of farnesyl diphosphate has a partial positive charge, and the bridge oxygen between C-1 and the α-phosphate of farnesyl diphosphate has a partial negative charge. This proposed transition state suggests that stabilization of the developing charge on the carbocation and diphosphate oxygens is an important catalytic feature.

The kinetic mechanism

Prenyltransferases are slow enzymes with turnover numbers of the order of $3\,min^{-1}$, depending upon the substrate. Steady-state kinetic data suggested that the reaction catalyzed by FTase is sequential with both substrates binding prior to the release of product. Furfine et al. [25] used pre-steady-state protein fluorescence spectroscopy to show that farnesyl diphosphate binds to the free enzyme in a two-step process with an overall K_d of about 5 nM. Further studies showed that the reaction of FTase · FPP with peptide to form products is functionally irreversible. Using rapid quench methodology, these workers found that the rate of formation of the enzyme-bound product (k_2) was much larger than k_{cat} (17 versus $0.05\,s^{-1}$) as indicated in Fig. 1A. They conclude that product release is the rate-limiting step in the reaction mechanism.

The following reaction scheme is supported by pre-steady-state, steady-state and X-ray crystallographic data [16,17,20,25–27] where the bold numbers in parentheses refer to structures solved by X-ray crystallography, the dash designates a covalent bond, and a dot designates a non-covalent bond.

$$\text{FTase} \cdot \text{FPP}\ (\mathbf{1}) + \text{peptide} \rightarrow \text{FTase} \cdot \text{FPP} \cdot \text{peptide}\ (\mathbf{2}) \quad (1)$$

$$\text{FTase} \cdot \text{FPP} \cdot \text{peptide}\ (\mathbf{2}) \rightarrow \text{FTase} \cdot \text{F-peptide} \cdot \text{PP} \quad (2)$$

$$\text{FTase} \cdot \text{F-peptide} \cdot \text{PP} \rightarrow \text{FTase} \cdot \text{F-peptide}\ (\mathbf{3}) + \text{PP} \quad (3)$$

$$\text{FTase} \cdot \text{F-peptide}\ (\mathbf{3}) + \text{FPP} \rightarrow \text{FTase} \cdot \text{F-peptide} \cdot \text{FPP}\ (\mathbf{4}) \quad (4)$$

$$\text{FTase} \cdot \text{F-peptide} \cdot \text{FPP}\ (\mathbf{4}) \rightarrow \text{FTase} \cdot \text{FPP}\ (\mathbf{1}) + \text{F-peptide} \quad (5)$$

The first step of the pathway involves the formation of a ternary complex from FTase · FPP and the peptide. This is followed by the farnesylation of the peptide, the actual chemical transformation (corresponding to k_2). Next, diphosphate is released, yielding the FTase · farnesylpeptide complex.

The nature of product release from the enzyme is unclear. Under single turnover conditions, Tschantz et al. [26] found that the farnesylpeptide product does

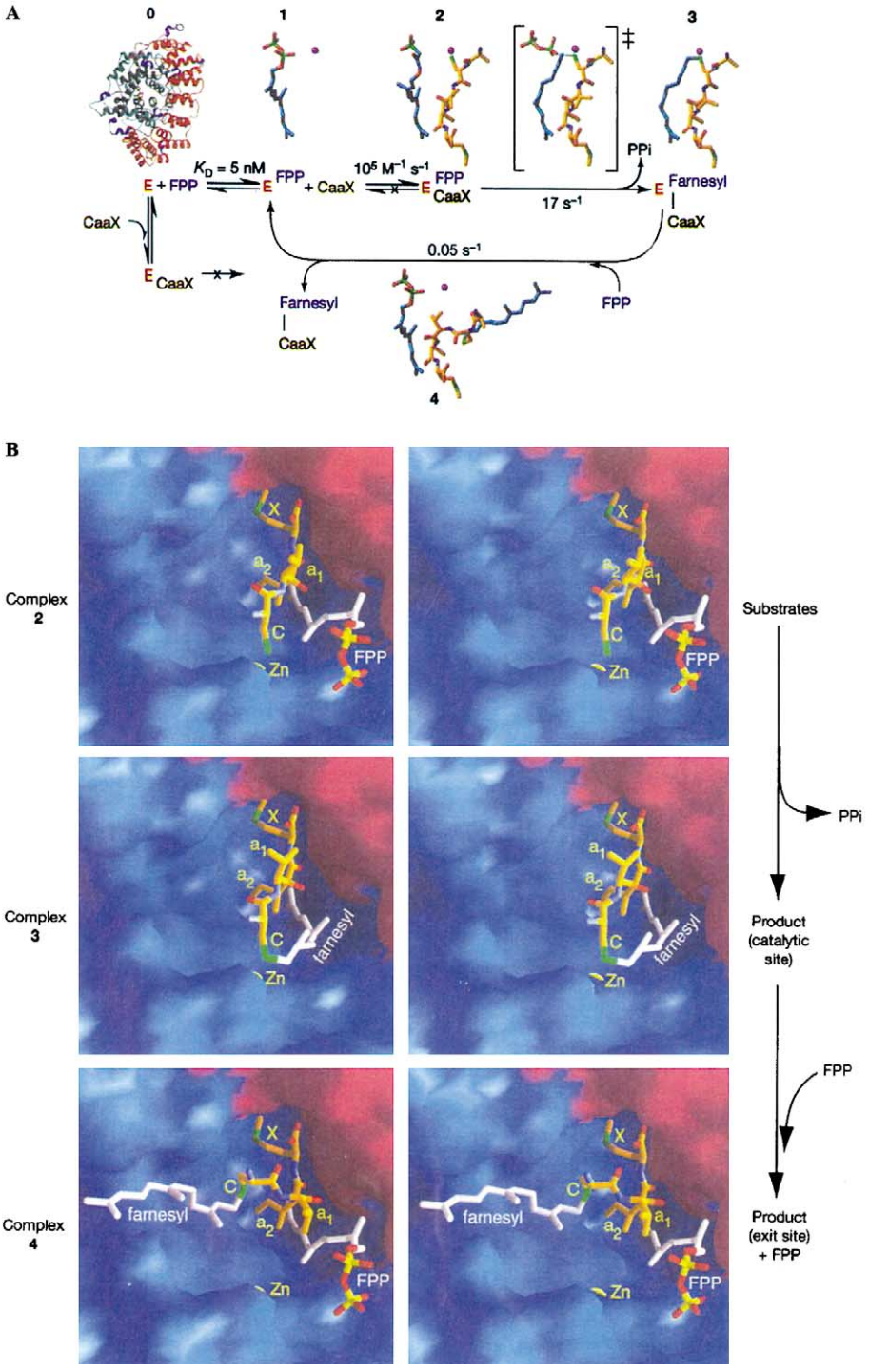

Fig. 1. The FTase reaction path structures. (A) Kinetic scheme [25]. **0**, Structure of apo-FTase (PDB number 1FT1); red, α-subunit, blue, β-subunit. **1**–**4**, Observed substrate conformations in FTase crystal structures. **1**, FPP (blue) bound to FTase (PDB number 1FT2). **2**, CaaX peptide substrate (yellow) and an FPP analogue (blue) representing the ternary complex (PDB number 1D8D). **3**, Farnesylpeptide product following catalysis. **4**, Both the farnesylpeptide product and FPP substrate bound following the addition of fresh FPP. Dissociation of the farnesylpeptide regenerates complex **1**. Double dagger symbol. A modeled transition state along the reaction coordinate between **2** and **3**. The Zn^{2+} is shown as a purple sphere. (B) Complexes **2**–**4** shown (stereo) in the context of the molecular surface of the active site (red, α-subunit; blue, β-subunit; yellow, peptide; and white, isoprenoid). In complex **4**, the isoprenoid of the product now occupies a shallow solvent-accessible groove. Only the CaaX motifs of the peptides are drawn. This figure is reproduced from [27] by copyright permission of the Nature Publishing Group.

not dissociate from the enzyme unless an additional substrate is provided. Farnesyl diphosphate is more effective than the acceptor peptide (CVIM) in displacing farnesylpeptide. A consequence of the addition of a fresh farnesyl diphosphate is that the free enzyme does not take part in the reaction. This reaction pathway is similar

to that of glyceraldehyde 3-phosphate dehydrogenase: $NAD^+ + \text{glyceraldehyde 3-phosphate} + P_i \rightarrow NADH + 1,3\text{-bisphosphoglycerate}$. Addition of fresh NAD^+ is required prior to the release of 1,3-bisphosphoglycerate and free enzyme is not part of the main reaction sequence [28].

Reaction pathway for FTase

X-ray crystallographic studies have been performed with unliganded or apo-FTase (**0**), FTase · FPP (**1**), a non-reactive isoprenoid bound to FTase · peptide (**2**), FTase · farnesylpeptide (**3**), and FTase · farnesylpep tide · FPP (**4**), all shown in Fig. 1. The α- and β-subunits are mainly α-helical (15 in the α-subunit and 14 in the β-subunit) [17]. The crystal structures show a single Zn^{2+} bound to the β-subunit near the subunit interface marking the location of the active site. In the apoenzyme (structure **0**), the Zn^{2+} is coordinated to D297β (a bidentate ligand), C299β, H362β, and a water molecule. Mutagenesis studies confirm a Zn^{2+}-binding role for these residues [17,18]. In structure **2**, the cysteine thiol of the CaaX substrate coordinates with Zn^{2+}, displacing the water molecule. Structure **1** shows a farnesyl diphosphate bound to the enzyme by several conserved tyrosine and tryptophan residues, and the aromatic nature of these residues accounts for the increase in intrinsic enzyme fluorescence following farnesyl diphosphate binding observed by Furfine et al. [25]. The diphosphate of FPP is adjacent to the essential Zn^{2+} ion. The length of the hydrophobic cavity that binds farnesyl diphosphate corresponds to the length of FPP, and a molecular ruler hypothesis accounts for the exclusion of the longer GGPP as a substrate [17]. Note that the apoenzyme (structure **0**) is not thought to be on the main reaction pathway in vivo.

An important and unusual property of FTase is that the first substrate, farnesyl diphosphate, contributes significantly to the binding site for the second substrate, the peptide, as the second and third isoprene units make van der Waals contact with the CaaX sequence. Although the notion that the binding of one substrate can influence the binding of a second substrate is common, it is unusual for one substrate to actually form part of the binding site for another substrate. The peptide binds to the isoprenoid in a hydrophobic cavity of the enzyme, and the peptide sequesters much of the second and all of the third isoprene units from the solvent. That farnesyl diphosphate forms a portion of the peptide binding site accounts for the ordered reaction with farnesyl diphosphate functioning as the leading substrate. The R-group of the a_1 residue (of Ca_1a_2X) faces the solvent and accounts for the ability of FTase to mediate the farnesylation of the centromere binding protein (Cenp-F) with a lysine (and not an aliphatic residue) in this position.

Structure **2** shows that the peptide is bound in an extended conformation with the X residue (methionine) near the bottom of a cleft and the a_2 residue is closely positioned near the isoprenoid. Modeling studies of various CaaX peptides and FTase allow methionine, serine, glutamine, and alanine to bind to the X subsite, but there is steric restriction when leucine is the X residue [17]. Using peptides with the sequence KKSSCVLX, the best substrates for FTase are those where X is methionine, serine, glutamine, alanine, and cysteine [29]. The specificity constant (k_{cat}/K_m) of the peptide with leucine in the X position is 1/30th to 1/70th that of the optimal substrates.

In structure **2**, the C-1 of farnesyl diphosphate is 7 Å from the cysteine sulfur atom, much too far for a chemical reaction to occur. Movement of the first two isoprene units of farnesyl diphosphate places C-1 adjacent to the reactive thiol and allows the reaction to proceed, as indicated in Fig. 1. In structure **3**, the FTase · F-peptide complex, the interaction of the hydrophobic farnesyl group and the peptide is more pronounced than that before the reaction.

Kinetic studies indicate that product release for FTase and GGTase-I is rate-limiting. For FTase, release of a product requires the addition of fresh substrate(s). Long et al. [27] determined the structure of FTase · F-peptide · FPP (Structure **4**). In this complex, the fresh farnesyl diphosphate displaces the farnesyl group of the farnesylpeptide to a hydrophobic site labeled the exit groove. This is accompanied by a conformational change in the CaaX sequence. There are thus two CaaX peptide conformations in the product: an initial extended conformation and a β-turn. The authors point out that this mechanism may explain the dual, or processive, situation in GGTase-II where the enzyme adds two geranylgeranyl groups to the acceptor protein prior to the release of a product. The X-ray structure of GGTase-II has been determined [13] and it contains an analogous exit groove [27].

The reaction pathway and kinetic scheme Eqs. (1)–(5) may be even more intricate than that those described thus far. Long et al. [27] report that the crystalline FTase · F-peptide · FPP complex is stable and requires the addition of fresh peptide to release the farnesylpeptide. This might occur as one person enters the front of a revolving door and displaces another person through the rear. As an alternative for the requirement of both fresh farnesyl diphosphate and peptide for farnesylpeptide release, these workers hypothesize that the enzyme-bound farnesylpeptide may interact with the CaaX endoprotease, and this interaction triggers product release physiologically. Additional experiments are required to sort out the steps involved in farnesylpeptide release.

Long et al. [27] suggest that FTase inhibitors may bind in an extended conformation or in a turn confor-

mation. CVFM is flexible and lacks a rigid turn conformation, and Cys-4-aminobenzoate-Met is rigid, but it assumes an extended and not turn conformation. These compounds are classical and instantaneous inhibitors of FTase. BZA-2B, a benzodiazepine peptidomimetic, has a hydrophobic benzodiazepine scaffold from which cysteine and methionine residues emanate to produce an overall configuration that resembles a β-turn. BZA-2B is a time-dependent inhibitor of FTase [30]. The ability of BZA-2B to inhibit FTase in a time-dependent fashion may be related in part to its β-turn conformation, and it may bind preferentially in the exit groove.

The three C-terminal amino acids of the farnesylpeptide product make extensive van der Waals contacts with a new farnesyl diphosphate substrate, and Long et al. [27] suggest that the amino acid sequence of the CaaX motif may modulate product release. Furfine et al. [25], used a biotinylated peptide with a CVVM CaaX box, and this is the source of data corresponding to a k_{cat} of $0.05\,s^{-1}$ at 25 °C. Steady-state kinetic studies indicate that a peptide ending with CVVM exhibits k_{cat} values of $0.31\,s^{-1}$ at 37 °C [29]. However, a peptide ending in CVVS exhibits a k_{cat} of $1.1\,s^{-1}$. These data are consistent with the notion that the amino acid sequence of the CaaX motif can alter the rate of product release, the rate-limiting step in the FTase catalyzed reaction at saturating substrate concentrations. Peptides containing serine in the X position generally have larger k_{cat} values than the corresponding methionine derivatives [29].

Epilogue

Although the prenyltransferases were discovered only a decade ago, progress in characterizing them has been rapid. Early studies on the α- and β-subunit composition of the prenyltransferases and the identification of essential amino acid residues (prior to the elucidation of the enzyme structure using X-ray crystallography) were aided by yeast genetics [1,18]. The richest source of FTase and GGTase-I is brain (a nondividing cell population), and purification of 60,000-fold (FTase) and 50,000-fold (GGTase-I) is required to obtain homogeneous proteins. Because purification of stoichiometric amounts of enzyme from natural sources would have been prohibitive, gene cloning and protein expression systems provide the enzymes that make single turnover kinetics and protein crystallography possible.

The reaction pathway for FTase is more intricate than that originally proposed [25] with the requirement for binding fresh substrate(s) to displace farnesylpeptide Eqs. (1)–(5) [26,27]. FTase exhibits substrate inhibition by peptides and proteins [16], and the mechanism of this inhibition is unclear. Perhaps the peptide binds to the FTase · F-peptide complex to form an abortive FTase · F-peptide · peptide complex that then releases F-peptide. The FTase · peptide presumably dissociates to form free FTase, which then binds with farnesyl diphosphate.

The understanding of GGTase-I and II is not as complete as that for FTase. That investigators focus on FTase stems largely because of the potential cancer chemotherapeutic possibilities of FTase inhibitors. However, inhibition of GGTase-I may be required to develop full anti-K-Ras activity in vivo. K-ras mutations are the most common ras mutations in human tumors [5] and K-ras is geranylgeranylated when FTase is inhibited in cells [7,14].

The number of proteins that are prenylated is greater than that anticipated in the early 1990s. The total number of likely substrates for FTase is about 50 [3] and for GGTase-I is about a hundred. Many of the GGTase-I substrates participate in the pivotal signal transduction pathways and these pathways may be dysregulated in tumorigenic cells. The Rab family of G proteins contains more than seventy members. Although the function of many of the prenylated proteins is known in broad strokes, there is much to learn about the functions of many of these proteins including unimagined nuances of signal transduction.

Despite the great progress in the enzymology of the prenyltransferases, these catalysts still lack enzyme commission numbers.

References

[1] F.L. Zhang, P.J. Casey, Protein prenylation: molecular mechanisms and functional consequences, Annu. Rev. Biochem. 65 (1996) 241–269, Reviews the early work on the discovery and properties of protein prenyltransferases, CaaX endoproteolysis, and methyl esterification.

[2] S. Clarke, Protein isoprenylation and methylation at carboxyl-terminal cysteine residues, Annu. Rev. Biochem. 61 (1992) 355–386, Traces the early work on the discovery of peptide and protein prenylation.

[3] A.D. Cox, C.J. Der, Farnesyltransferase inhibitors and cancer treatment: targeting simply Ras? Biochim. Biophys. Acta 1333 (1997) F51–F71.

[4] C. O'Donovan, M.J. Martin, A. Gattiker, E. Gasteiger, A. Bairoch, R. Apweiler, High-quality protein knowledge resource: SWISS-PROT and TrEMBL brief, Bioinformatics 3 (2002) 275–284, or http://us.expasy.org/sprot/, this is the source for the sequence and postulated functions of the various proteins depicted in Table 1.

[5] M. Barbacid, ras Genes, Annu. Rev. Biochem. 56 (1987) 779–827.

[6] D.R. Lowy, B.M. Willumsen, Function and regulation of Ras, Annu. Rev. Biochem. 62 (1993) 851–891.

[7] J.B. Gibbs, Farnesyltransferase inhibitors, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, third ed., Academic Press, San Diego, 2001, pp. 81–103.

[8] S.M. Sebti, A.D. Hamilton, Farnesyltransferase and geranylgeranyltransferase I inhibitors and cancer therapy: lessons from mechanism and bench-to-bedside translational studies, Oncogene 19 (2000) 6584–6593.

[9] F. Caponigro, Farnesyl transferase inhibitors: a major breakthrough in anticancer therapy?, Anticancer Drugs 13 (2002) 891–897.

[10] S.M. Hahn, E.J. Bernhard, W. Regine, M. Mohiuddin, D.G. Haller, J.P. Stevenson, D. Smith, B. Pramanik, J. Tepper, T.F. DeLaney, K.E. Kiel, B. Morrison, P. Deutsch, R.J. Muschel, W.G. McKenna, A phase I trial of the farnesyltransferase inhibitor L-778,123 and radiotherapy for locally advanced lung and head and neck cancer, Clin. Cancer Res. 8 (2002) 1065–1072.
[11] R. Kurzrock, J. Cortes, H. Kantarjian, Clinical development of farnesyltransferase inhibitors in leukemias and myelodysplastic syndrome, Semin. Hematol. 39 (2002) 20–24.
[12] G.K. Dy, A.A. Adjei, Farnesyltransferase inhibitors in breast cancer therapy, Cancer Invest. 20 (Suppl. 2) (2002) 30–37.
[13] M.G. Seabra, Biochemistry of Rab geranylgeranyltransferase, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, 3rd ed., Academic Press, San Diego, 2001, pp. 131–154, Reviews the mechanistic and X-ray crystallographic studies of GGTase-II.
[14] K. Yokayama, M.H. Gelb, Protein geranylgeranyltransferase type I, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, 3rd ed., Academic Press, San Diego, 2001, pp. 105–130.
[15] S.A. Armstrong, V.C. Hannah, J.L. Goldstein, M.S. Brown, CAAX geranylgeranyltransferase transfers farnesyl as efficiently as geranylgeranyl to RhoB, J. Biol. Chem. 270 (1995) 7864–7868.
[16] R.A. Spence, P.J. Casey, Mechanism of catalysis by protein farnesyltransferase, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, 3rd ed., Academic Press, San Diego, 2001, pp. 1–18.
[17] K.L. Terry, S.B. Long, L.S. Beese, Structure of protein farnesyltransferase, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, 3rd ed., Academic Press, San Diego, 2001, pp. 19–46, reviews the X-ray crystallographic studies of FTase.
[18] J. Urano, W. Yang, F. Tamanoi, Mutational analysis of protein farnesyltransferase, in: F. Tamanoi, in: D.S. Sigman (Eds.), The Enzymes, vol. XXI, 3rd ed., Academic Press, San Diego, 2001, pp. 47–80.
[19] K.E. Hightower, C. Huang, P.J. Casey, C.A. Fierke, H-Ras peptide and protein substrates bind protein farnesyltransferase as an ionized thiolate, Biochemistry 37 (1998) 15555–15562.
[20] C.C. Huang, P.J. Casey, C.A. Fierke, Evidence for a catalytic role of zinc in protein farnesyltranferase, J. Biol. Chem. 272 (1997) 20–23.
[21] M.J. Saderholm, K.E. Hightower, C.A. Fierke, Role of metals in the reaction catalyzed by protein farnesyltransferase, Biochemistry 39 (2000) 12398–12405.
[22] F.L. Zhang, P.J. Casey, Influence of metal ions on substrate binding and catalytic activity of mammalian protein geranylgeranyltransferase type-I, Biochem. J. 320 (1996) 925–932.
[23] J.M. Dolence, C.D. Poulter, A mechanism for posttranslational modifications of proteins by yeast protein farnesyltransferase, Proc. Natl. Acad. Sci. USA 92 (1995) 5008–5011.
[24] C. Huang, K.E. Hightower, C.A. Fierke, Mechanistic studies of rat protein farnesyltransferase indicate an associative transition state, Biochemistry 39 (2000) 2593–2602.
[25] E.S. Furfine, J.J. Leban, A. Landavazo, J.F. Moomaw, P.J. Casey, Protein farnesyltransferases: kinetics of farnesyl pyrophosphate binding and product release, Biochemistry 34 (1995) 6857–6862.
[26] W.R. Tschantz, E.S. Furfine, P.J. Casey, Substrate binding is required for release of product from mammalian protein farnesyltransferase, J. Biol. Chem. 272 (1997) 9989–9993.
[27] S.B. Long, P.J. Casey, L.S. Beese, Reaction path of protein farnesyltransferase at atomic resolution, Nature 419 (2002) 645–650, Formulation of the reaction pathway for FTase and its relationship to the enzymology of GGTase-II.
[28] D.R. Trentham, Rate-determining processes and the number of simultaneously active sties of D-glyceraldehyde 3-phosphate dehydrogenase, Biochem. J. 122 (1971) 71–77.
[29] R. Roskoski Jr., P.A. Ritchie, Role of the carboxyterminal residue in peptide binding to protein farnesyltransferase and protein geranylgeranyltransferase, Arch. Biochem. Biophys. 356 (1998) 167–176.
[30] R. Roskoski Jr., P.A. Ritchie, Time-dependent inhibition of protein farnesyltransferase by a benzodiazepine peptide mimetic, Biochemistry 40 (2001) 9329–9335.