# Biosynthesis of Natural Products

## *Biosynthesis of Fatty Acids & Polyketides*

Alan C. Spivey
a.c.spivey@imperial.ac.uk

Imperial College London

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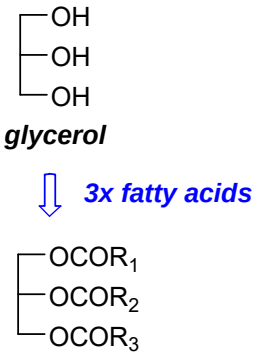
# Format & Scope of Lecture

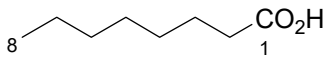
- ***What are fatty acids?***
  - 1° metabolites: fatty acids; 2° metabolites: their derivatives
  - biosynthesis of the building blocks: acetyl CoA & malonyl CoA
- ***Fatty acid synthesis by Fatty Acid Synthases (FASs)***
  - the chemistry involved
  - the FAS protein complex & the dynamics of the iterative synthesis process
- ***Fatty acid secondary metabolites***
  - eiconasiods: prostaglandins, thromboxanes & leukotrienes
- ***What are polyketides?***
  - definitions & variety
- ***Polyketide synthesis by PolyKetide Synthases (PKSs)***
  - the chemistry involved
  - the PKS protein complexes & the dynamics of the iterative synthesis process
- ***Polyketide secondary metabolites***
  - Type I modular metabolites: macrolides – *e.g.* erythromycin
  - Type I iterative metabolites: *e.g.* mevinolin (=lovastatin®)
  - Type II iterative metabolites: aromatic compounds and polyphenols: *e.g.* actinorhodin

# Fatty Acid Primary Metabolites

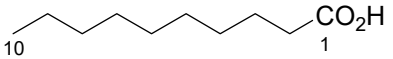
- ***Primary metabolites:***
  - ***fully saturated, linear carboxylic acids*** & derived ***(poly)unsaturated derivatives:***
    - constituents of essential natural waxes, seed oils, ***glycerides*** (fats) & phospholipids
    - ***structural role*** – ***glycerides*** & phospholipids are essential constituents of cell membranes
    - ***energy storage*** – ***glycerides*** (fats) can also be catabolised into acetate → citric acid cycle
    - ***biosynthetic precursors*** – for elaboration to secondary metabolites



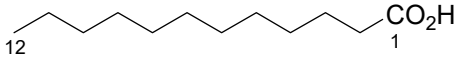

***SATURATED ACIDS [$MeCH_2(CH_2CH_2)_nCH_2CO_2H$ (n = 2-8)]***
***e.g.***



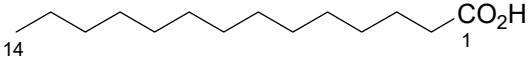

***caprylic acid (C8, n = 2)***



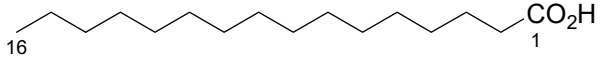

***capric acid (C8, n = 3)***



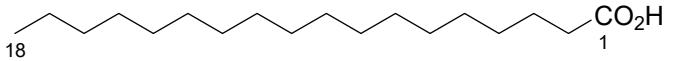

***lauric acid (C12, n = 4)***




***myristic acid (C14, n = 5)***

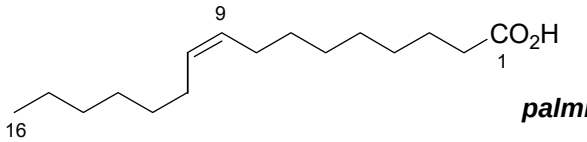



***palmitic acid (C16, n = 6)***

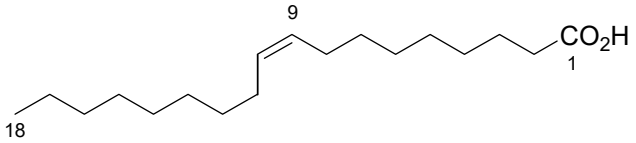



***stearic acid (C18, n = 7)***

***MONO-UNSATURATED ACID DERIVATIVES (MUFAs) e.g.***

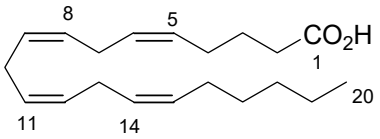



***palmitoleic acid (C16, Z -$\Delta^9$)***

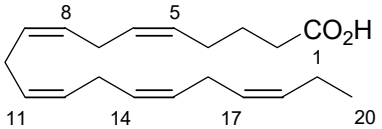



***oleic acid (C18, Z-$\Delta^9$)***
***(>80% of fat in olive oil )***

***POLY-UNSATURATED ACID DERIVATIVES (PUFAs)***
***e.g.***




***arachidonic acid (AA)***
***(C20, Z -$\Delta^5$, Z -$\Delta^8$, Z -$\Delta^{11}$, Z -$\Delta^{14}$)***

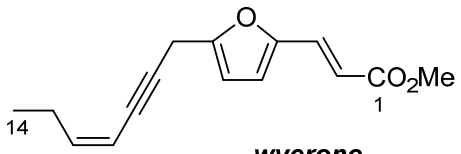



***eicosapentaenoic acid (EPA)***
***(C20, Z-$\Delta^5$, Z -$\Delta^8$, Z-$\Delta^{11}$, Z -$\Delta^{14}$, Z -$\Delta^{17}$)***
***(in cod liver oil)***

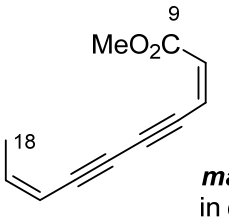
# Fatty Acids Derivatives – *Secondary Metabolites*

- ***Secondary metabolites***
  - further ***elaborated*** derivatives of ***polyunsaturated fatty acids (PUFAs)***
    - *e.g.* polyacetylenes & 'eicosanoids' (prostaglandins, thromboxanes & leukotrienes)

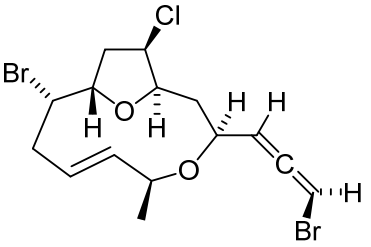
***POLYACETYLENES***
***e.g.***




***wyerone***
anti-fungal

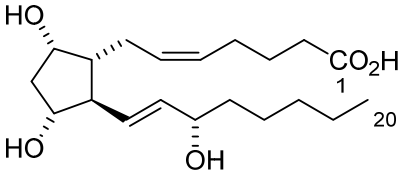



***matricaria ester***
in chamomile tea



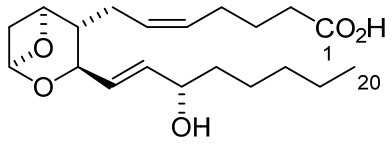

***obtusallene II***
marine natural product

***PROSTAGLANDINS***
***e.g.***



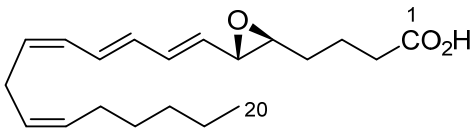

***prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$)***

***THROMBOXANES***
***e.g.***




***thromboxane $A_2$ ($TXA_2$)***

***LEUKOTRIENES***
***e.g.***

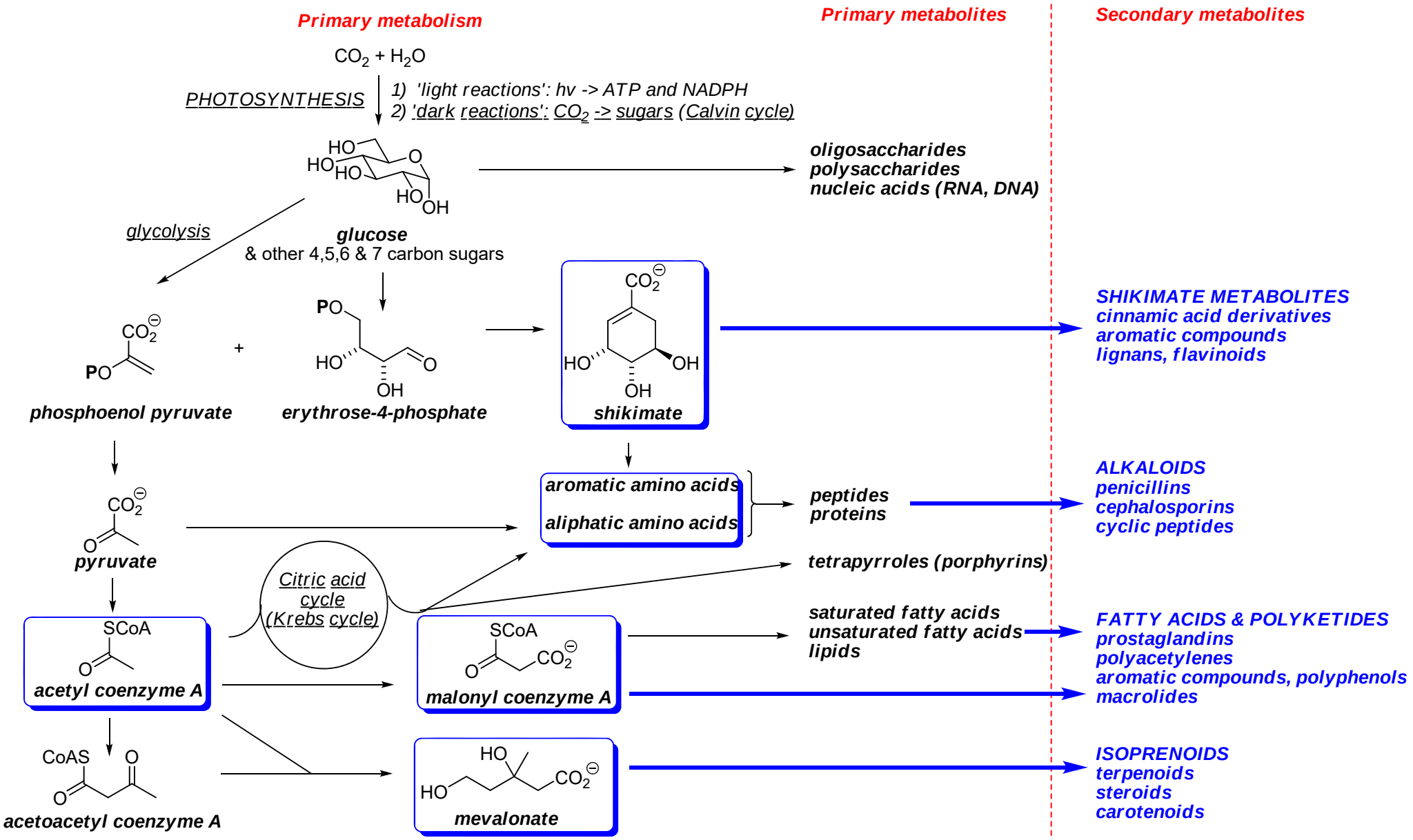



***leukotriene $A_4$ ($LTA_4$)***

***leukotriene $A_4$($LTA_4$)***

***EICOSANOIDS***

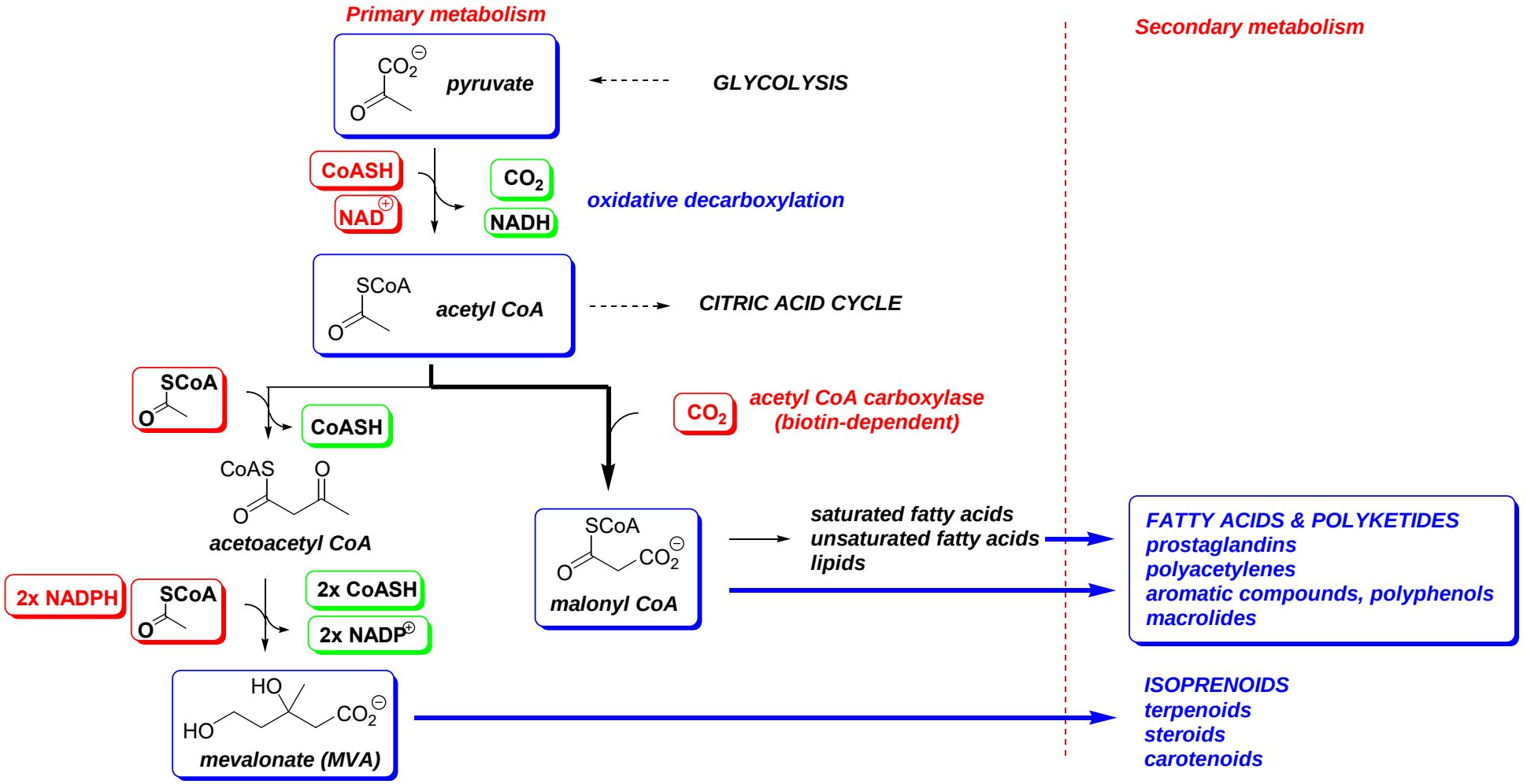
# Primary Metabolism - *Overview*




For interesting animations' of *e.g.* photosynthesis see: **http://www.johnkyrk.com/index.html**

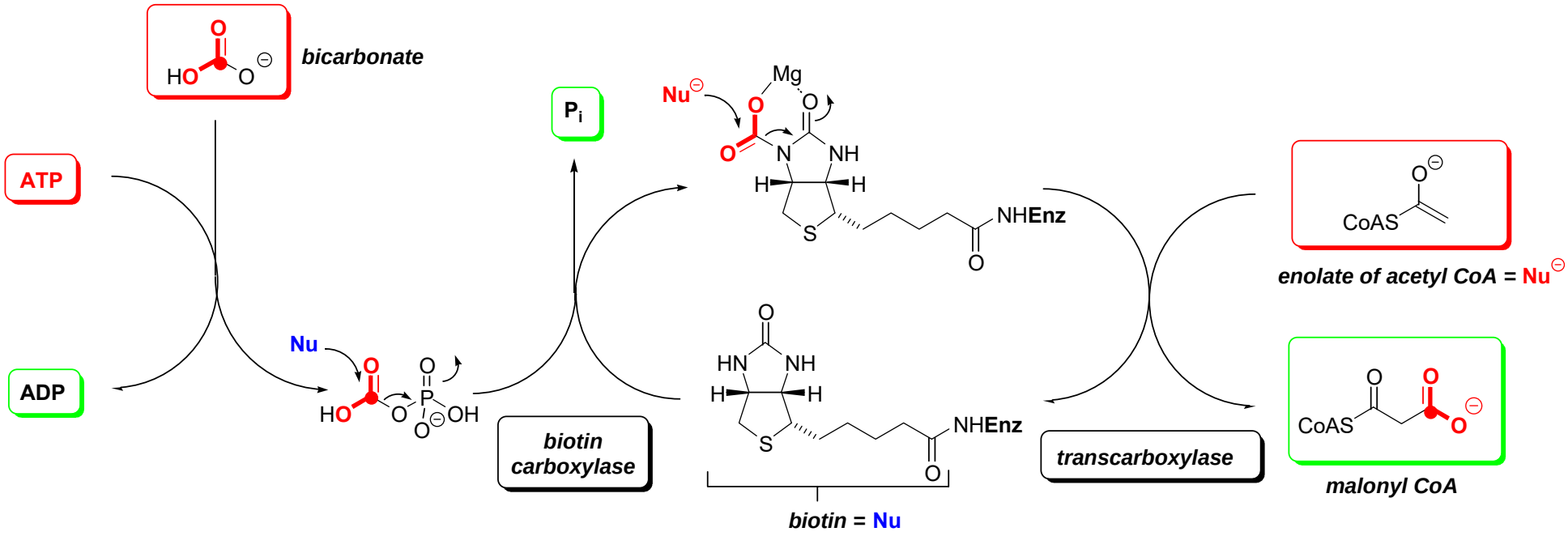
# Biosynthesis of Malonyl Coenzyme A

- ***Malonyl coenzyme A*** is the key ***'extender unit'*** for the biosynthesis of ***fatty acids (& polyketides):***
  - is formed by the ***carboxylation*** of ***acetyl coenzyme A*** mediated by a ***biotin-dependent enzyme***
  - this is the ***first committed step of fatty acid/polyketide biosynthesis*** (& is a rate controlling step)



# Biosynthesis of Malonyl Coenzyme A

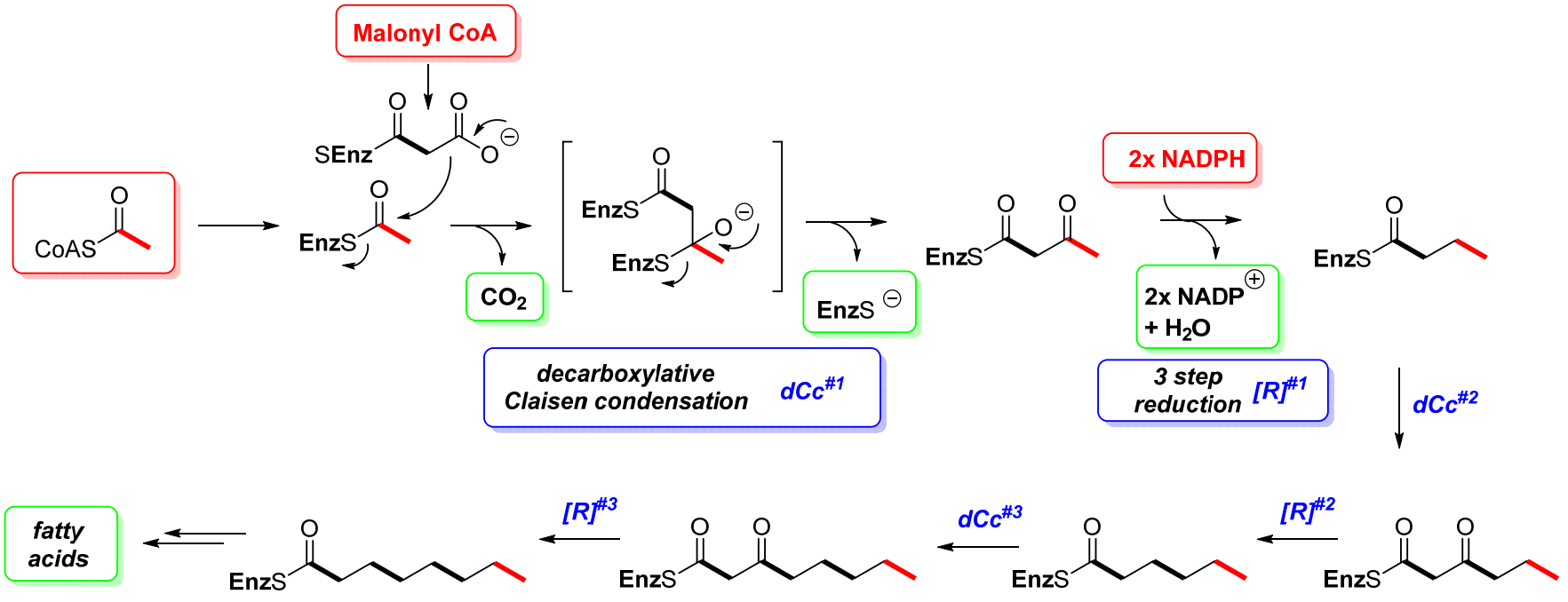
- ***Bicarbonate*** is the source of the ***$CO_2$***:
  - the bicarbonate is first ***activated*** via ***phosphorylation*** by ***ATP***
  - then the ***phosphorylated bicarbonate*** carboxylates ***biotin*** to give ***carboxybiotin***
  - then the ***carboxybiotin*** carboxylates the enolate of ***acetyl CoA*** to give ***malonyl CoA:***




  - the carboxylation of biotin & acetyl CoA are mediated by a ***single biotin-dependent enzyme (complex)*** having both ***biotin carboxylase*** and ***transcarboxylase active sites***
  - *NB.* coupling to ATP 'hydrolysis' provides ***energy*** to drive carboxylation processes

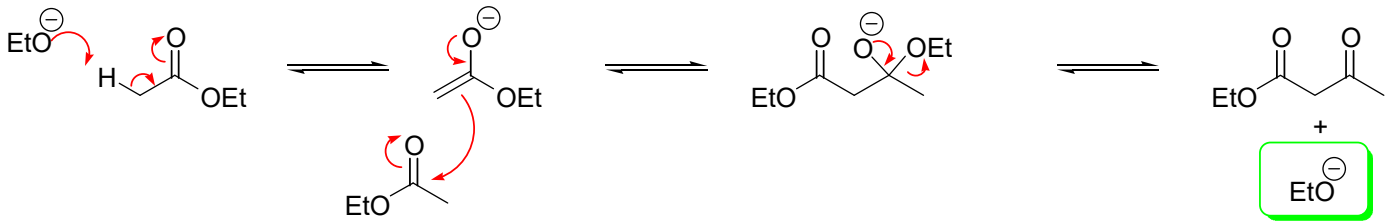
# Biosynthesis of Fatty Acids – *Iterative Oligomerisation*

- ***fatty acids*** are biosynthesised from ***acetyl CoA*** as a ***starter unit*** by ***iterative*** 'head-to-tail' ***oligomerisation*** involving:
  - condensation with ***malonyl CoA*** as an ***extender unit*** (with loss of $CO_2$) – a ***decarboxylative Claisen condensation***
  - 3-step ***reduction*** of the resulting ***ketone → methylene***
- after **n = 2-8 iterations** the ***C8-20 saturated fatty acid*** is released from the enzyme(s):

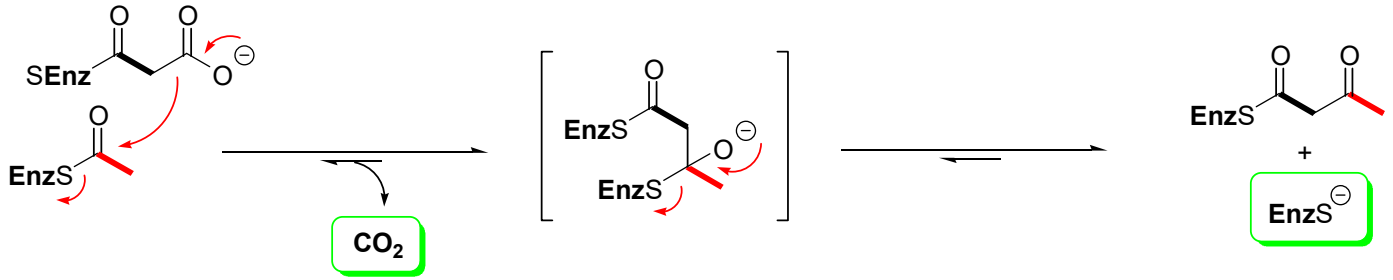


# The Decarboxylative Claisen Condensation (dCc)

- *in vitro* – the classical ***Claisen condensation***:



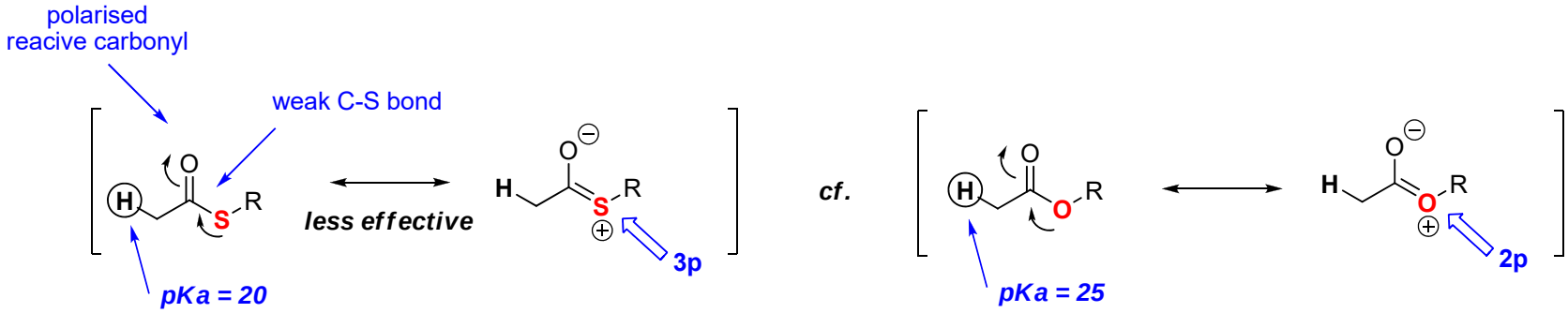

- *in vivo* - the ***decarboxylative Claisen condensation*** catalysed by a ***ketosynthase (KS)***




  - the energy released upon loss of $CO_2$ provides a driving force for the condensation
  - thioesters are also particularly reactive partners in this type of condensation...

# The Claisen Condensation - *Why Thioesters*?

- recall the chemistry of ***coenzyme A*** (1st lecture) – properties of ***alkyl thioesters*** (*cf.* alkyl esters)
  - ***highly electrophilic carbonyl*** (~ ketone)
  - ***high acidity of protons $\alpha$ to the carbonyl of thioesters*** (*cf.* ester)
  - ***weak C-S bond*** (*cf.* C-O bond):
    - due to poor orbital overlap between the p-orbital lone pair on sulfur ($n_S$) [*cf.* $n_O$] and the carbonyl anti bonding orbital $\pi^*_{C=O}$; (*i.e.* minimal 'resonance' $n_S \rightarrow \pi^*_{C=O}$)

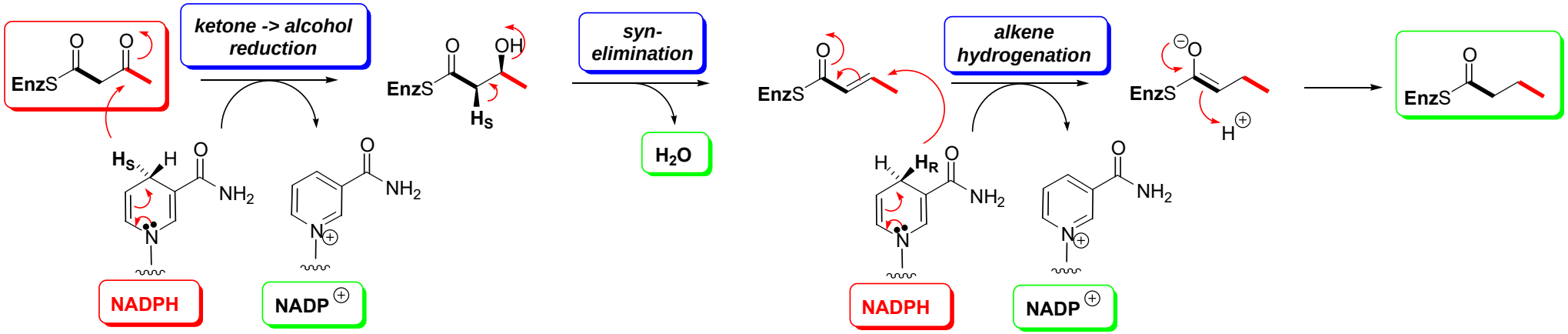



  - ***good leaving group ability of $RS^-$*** (*cf.* $RO^-$)
    - due to $pK_a$ (RSH) ~10 *cf.* $pK_a$ (ROH) ~16

# Ketone → Methylene - *Reduction*

- ***ketone*** → ***methylene*** reduction is achieved *via* a ***3-step process:***

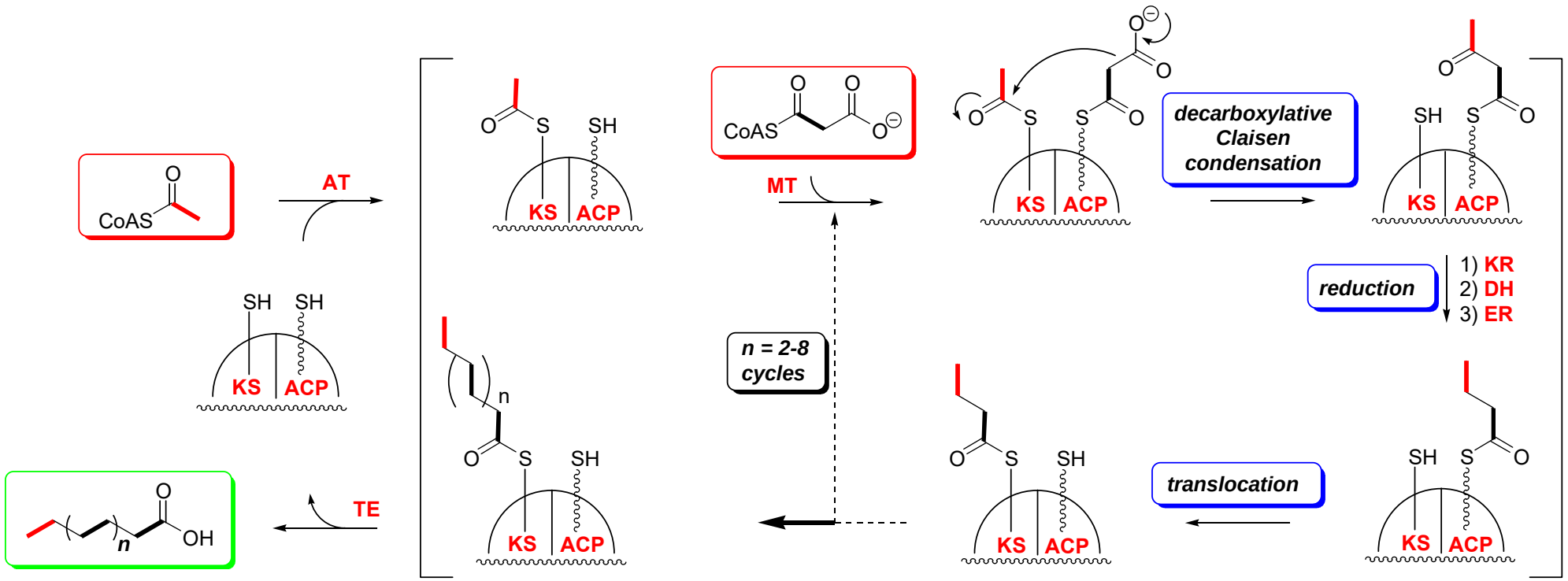
  1. ***NADPH***-mediated ***ketone*** → ***alcohol reduction*** catalysed by a ***keto reductase (KR)***
  2. ***syn-eliminataion*** of water catalysed by a ***dehydratase (DH)***
  3. ***NADPH***-mediated ***hydrogenation*** of the double bond catalysed by an ***enoyl reductase (ER)***




- all steps are generally stereospecific but stereospecificity varies from organism to organism
  - indicated specificities are for ***human FAS***

# Biosynthesis of Fatty Acids – *Overview of FAS*

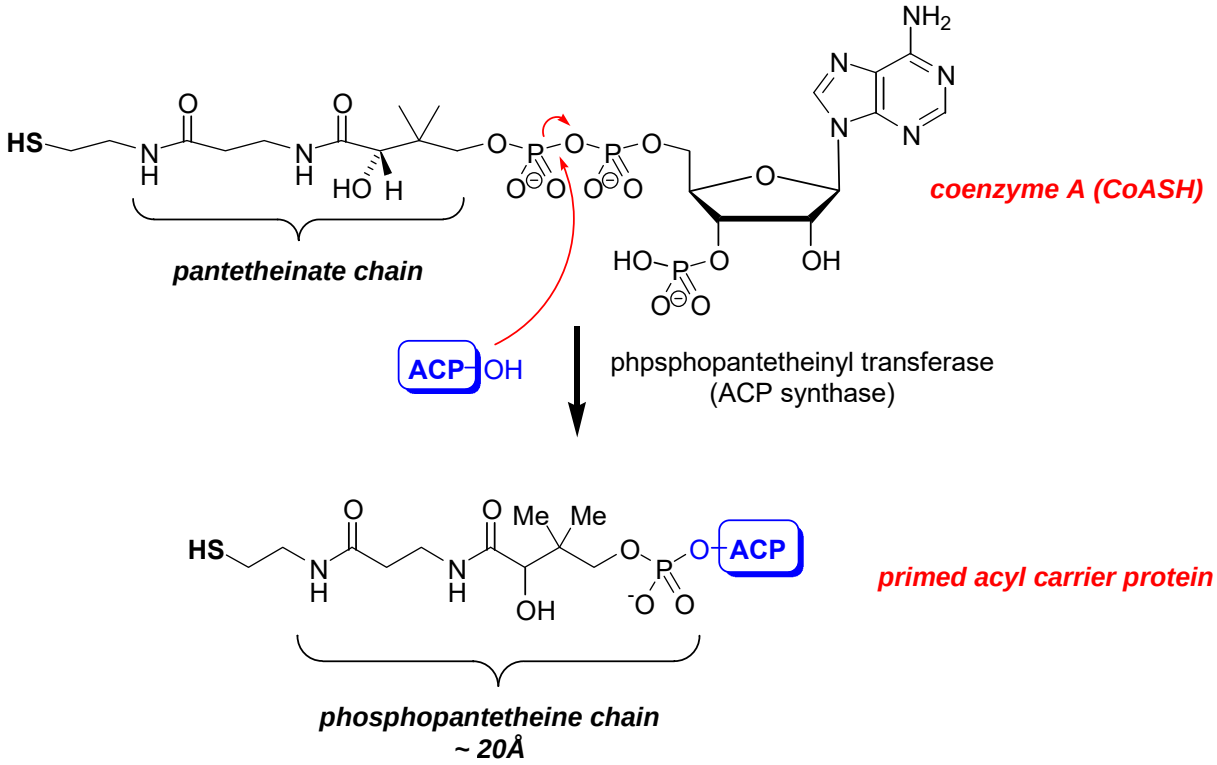
- The *in vivo* process by which all this takes place involves a 'molecular machine' - ***Fatty Acid Synthase (FAS)***
  - ***Type I FAS: single multifunctional protein complex*** (*e.g.* in mammals incl. humans)
  - ***Type II FAS: set of discrete, dissociable single-function proteins*** (*e.g.* in bacteria)
  - ***All FASs*** comprise ***8 components*** (ACP & 7× catalytic activities): ***ACP***, ***KS***, ***AT***, ***MT***, ***KR***, ***DH***, ***ER*** & [***TE***] :




**KS** = keto synthase (also known as **CE** = condensing enzyme); **AT** = acetyl transferase; **MT** = malonyl transferase; **KR** = keto reductase; **DH** = dehydratase; **ER** = enoyl reductase; **TE** = thioesterase; **ACP** = acyl carrier protein

# The Acyl Carrier Protein (ACP)

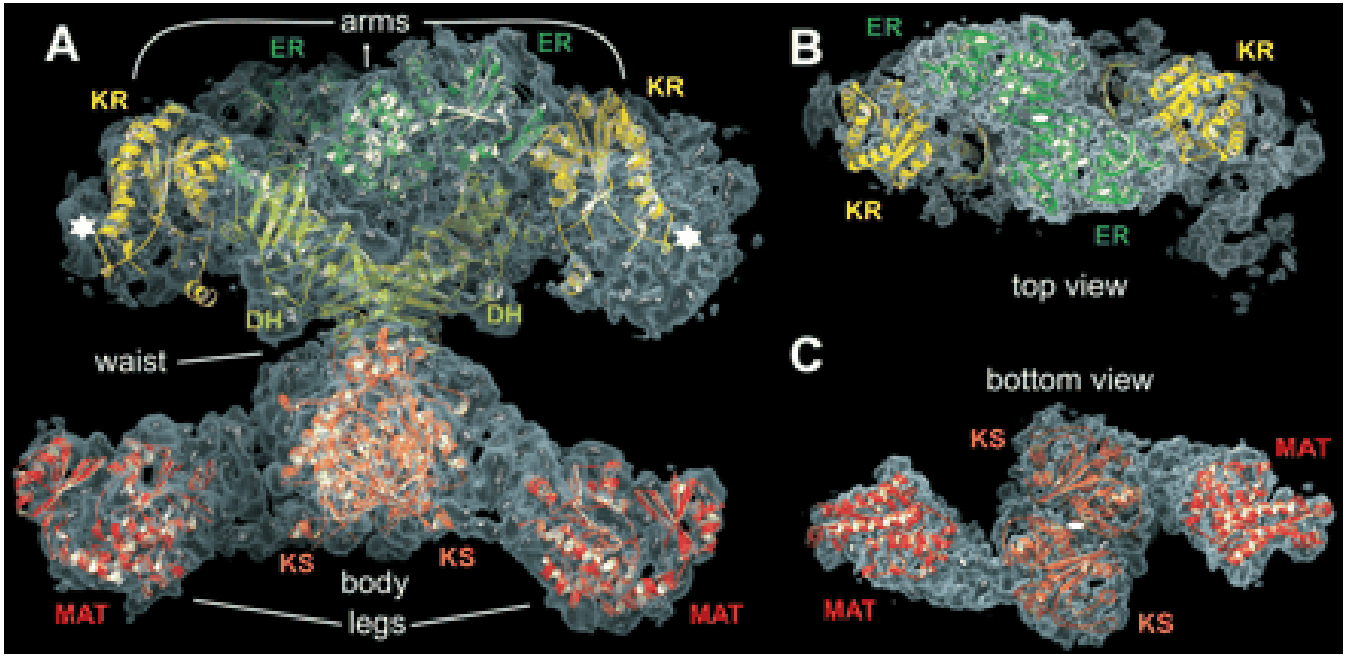
- the ***Acyl Carrier Protein (ACP)*** is the key protein that allows the growing oligomer to access the appropriate active sites
- The ACP is first ***primed*** by the post-translational modification of one of its serine hydroxyl groups:
  - the introduction of a ***phosphopantetheine 'swinging-arm'*** by reaction with ***acetyl coenzyme A:***




  - this swinging-arm provides ***flexibility*** for module-module acyl transfer & provides ***binding energy*** for catalysis
  - the ACP is inactive prior to priming

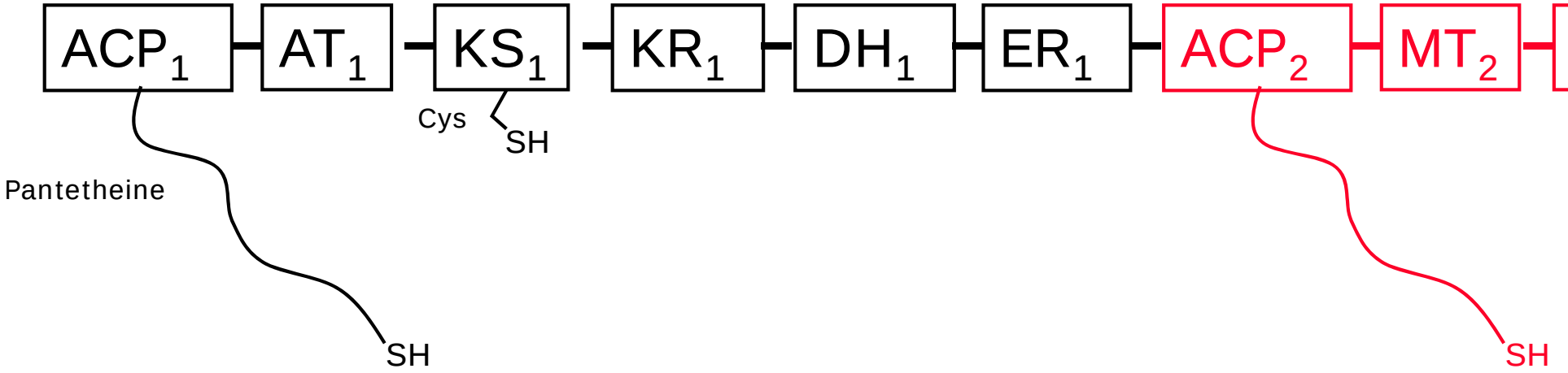
# Human Fatty Acid Synthase (FAS)

- the first three-dimensional structure of human fatty acid synthase (272 kDa) at 4.5 Å resolution by X-ray crystallography:
  - Maier, Jenni & Ban *Science* **2006**, *311*, 1258 (DOI) ; also Fungal FAS @ 3.1 Å resolution see: Jenni *et al. Science* **2007**, *316*, 254 & 288



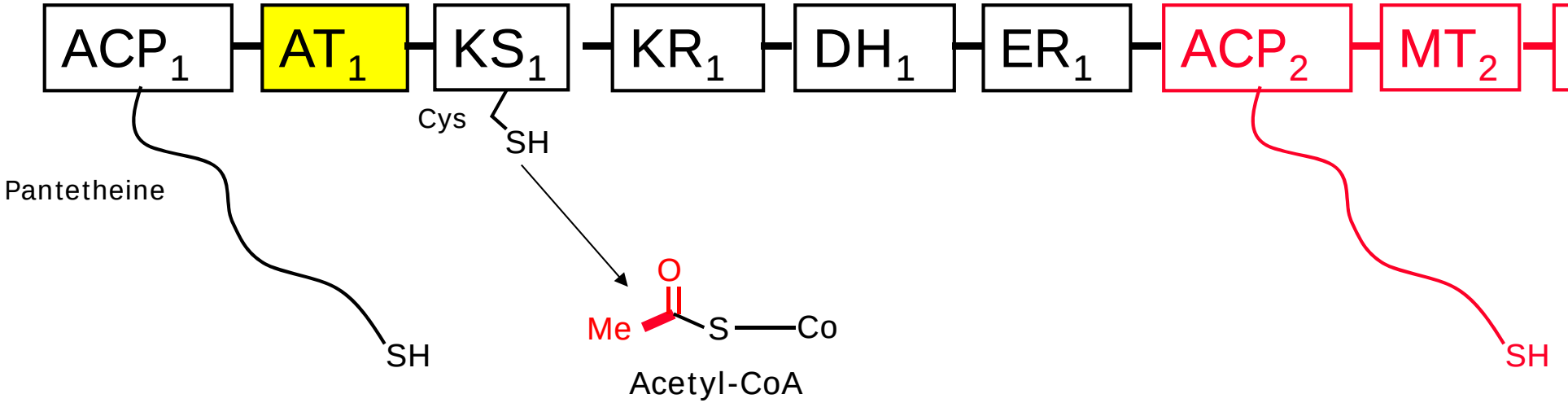

Structural overview. (**A**) Front view: FAS consists of a lower part comprising the KS (lower body) and MAT domains (legs) connected at the waist with an upper part formed by the DH, ER (upper body), and KR domains (arms). (**B**) Top view of FAS with the ER and KR domains resting on the DH domains. (**C**) Bottom view showing the arrangement of the KS and MAT domains and the continuous electron density between the KS and MAT domains

# FATTY ACID BIOSYNTHESIS (type II FAS)



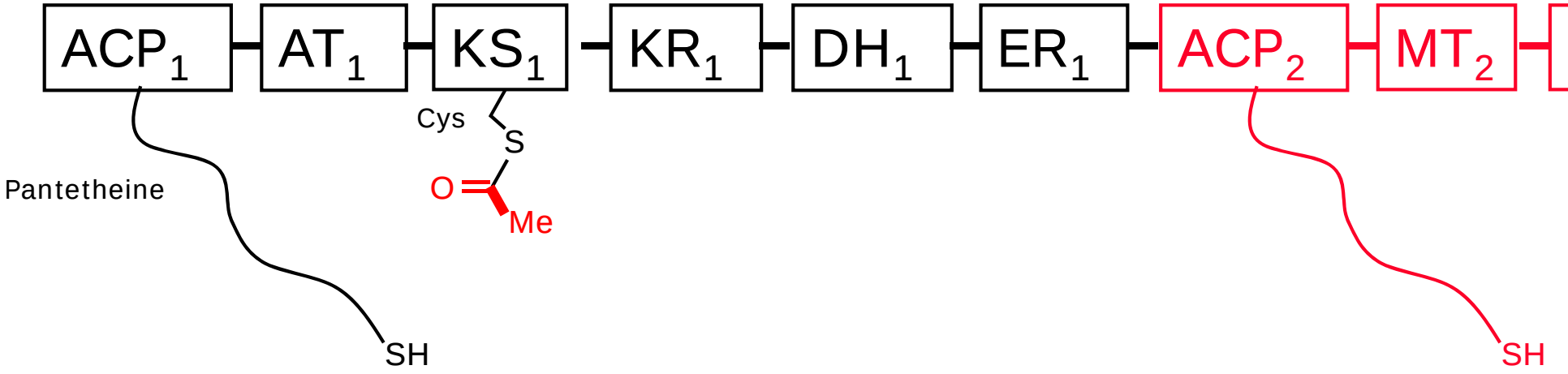

*NB*. the following sequence of slides have been adapted from: http://www.courses.fas.harvard.edu/%7echem27/

# FATTY ACID BIOSYNTHESIS

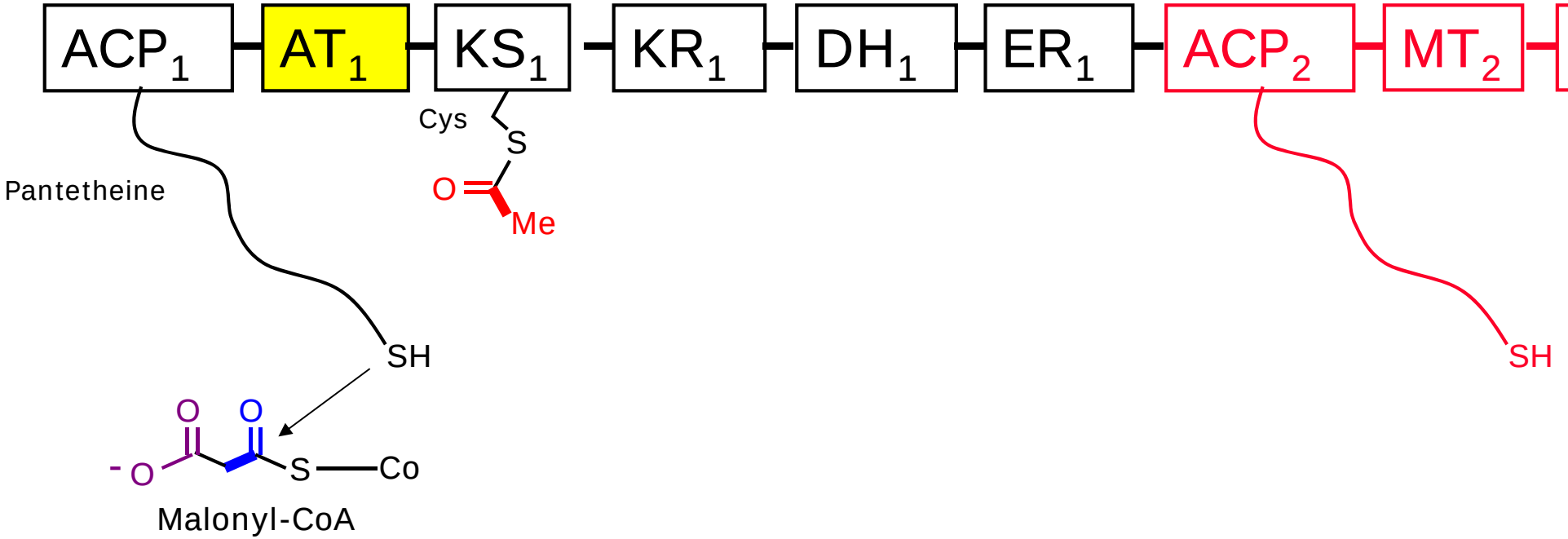



- $AT_1$ loads acetyl group onto $KS_1$

# FATTY ACID BIOSYNTHESIS

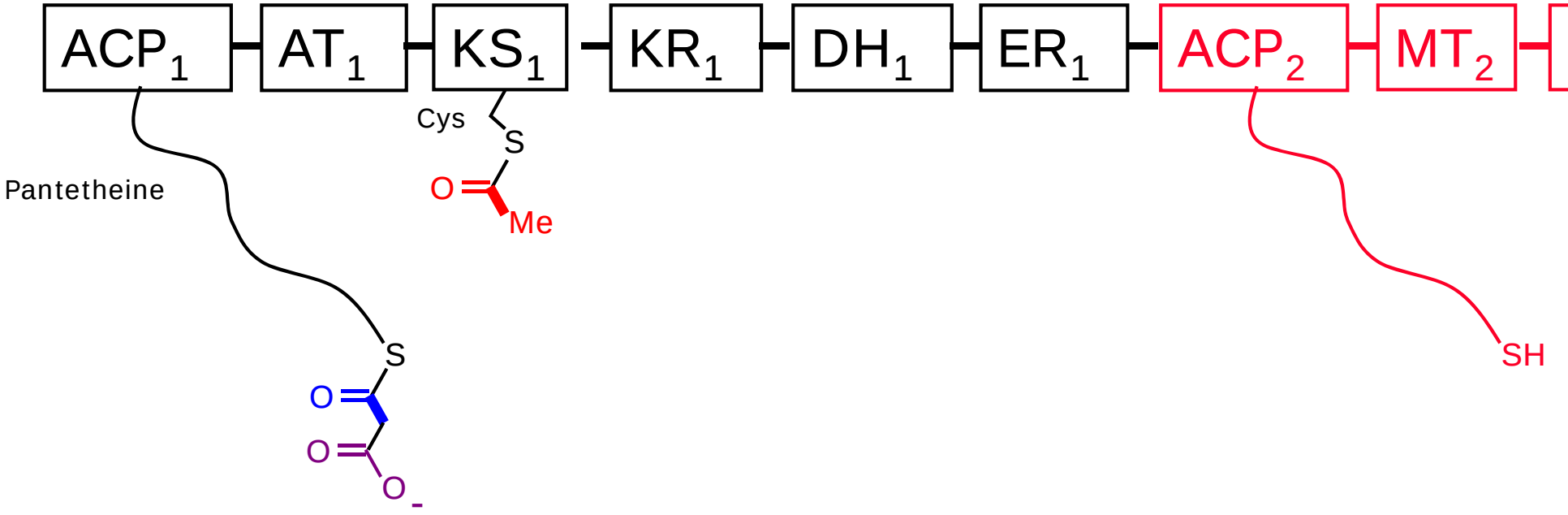


# FATTY ACID BIOSYNTHESIS

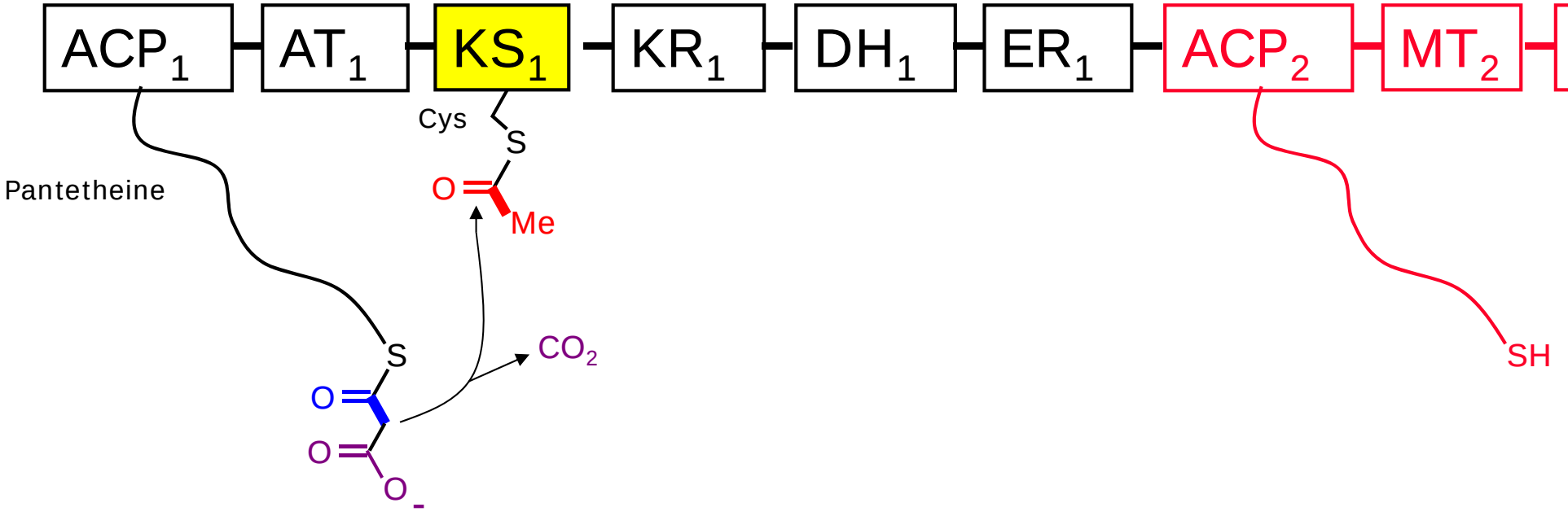



- $AT_1$ loads malonyl group onto $ACP_1$

# FATTY ACID BIOSYNTHESIS

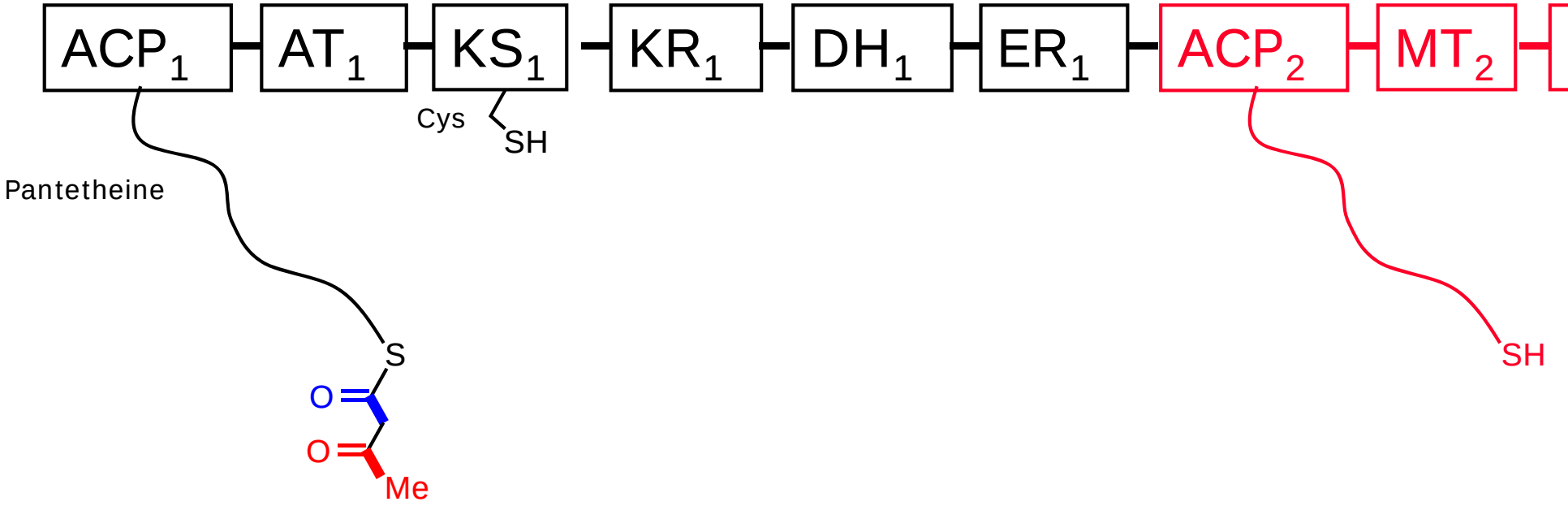


# FATTY ACID BIOSYNTHESIS

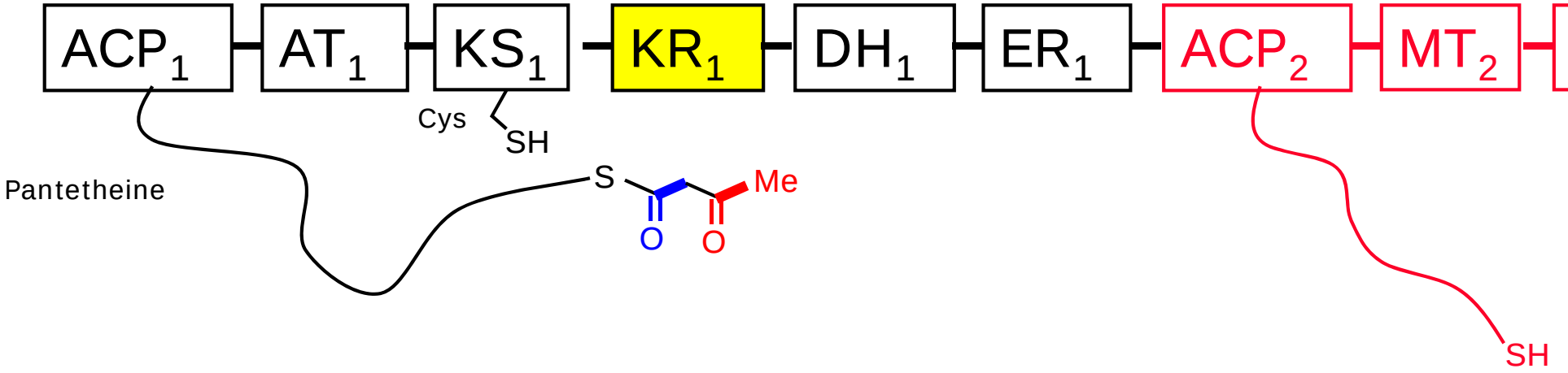



- $KS_1$ catalyzes Claisen condensation

# FATTY ACID BIOSYNTHESIS

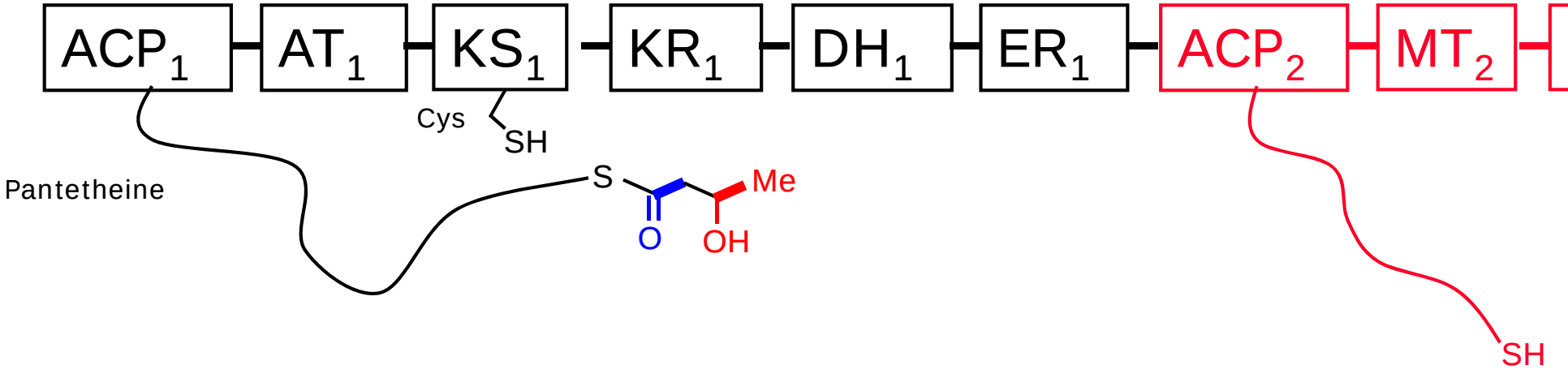


# FATTY ACID BIOSYNTHESIS




- $KR_1$ catalyzes reduction of ketone

# FATTY ACID BIOSYNTHESIS



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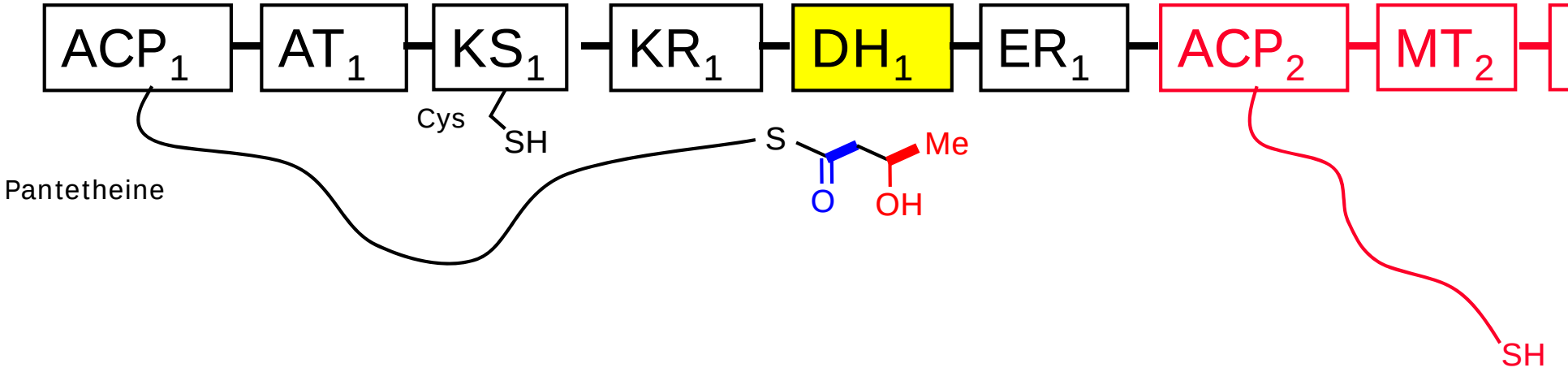
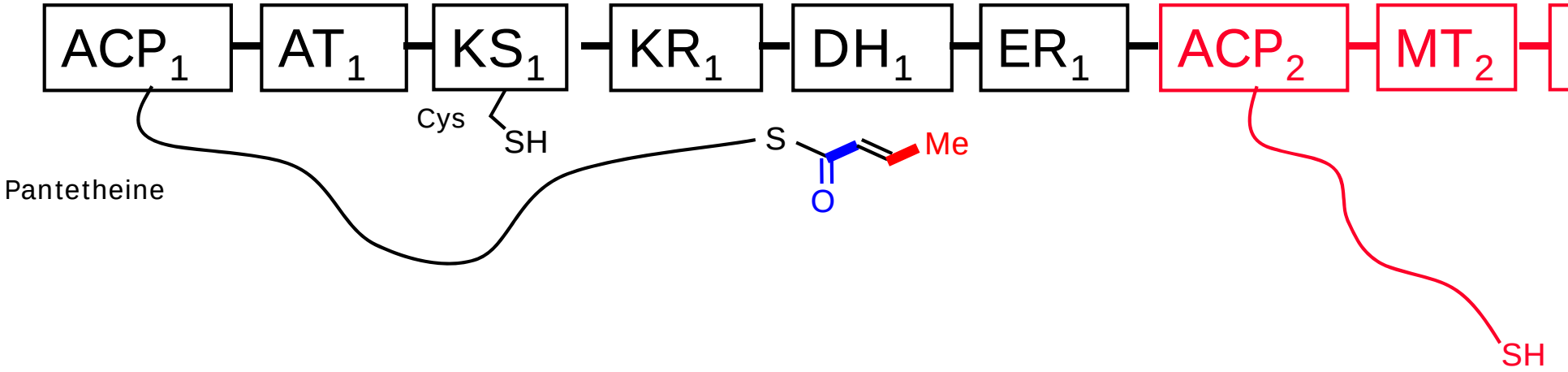
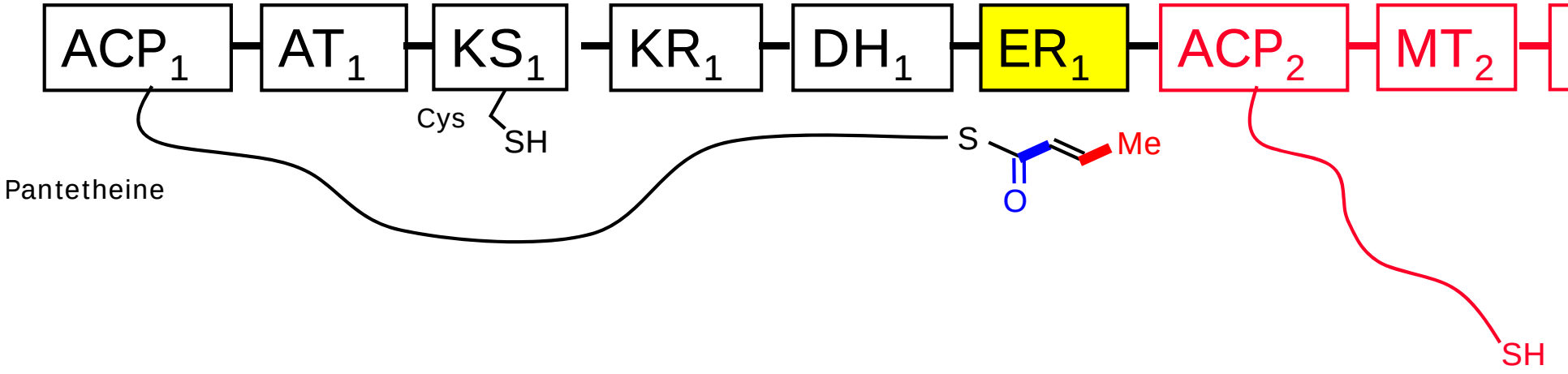



- $DH_1$ catalyzes dehydration of alcohol

# FATTY ACID BIOSYNTHESIS

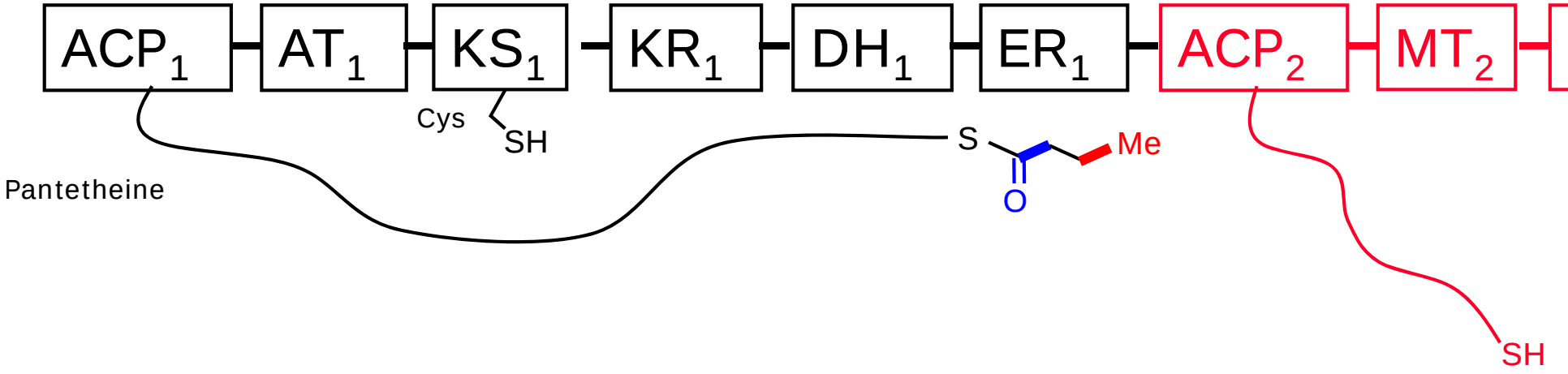


# FATTY ACID BIOSYNTHESIS

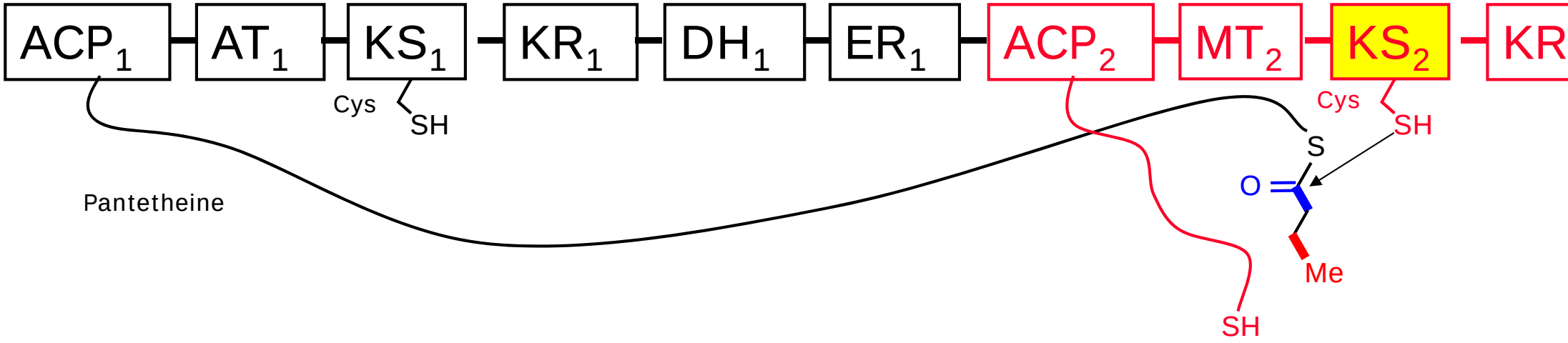



- $ER_1$ catalyzes reduction of alkene

# FATTY ACID BIOSYNTHESIS

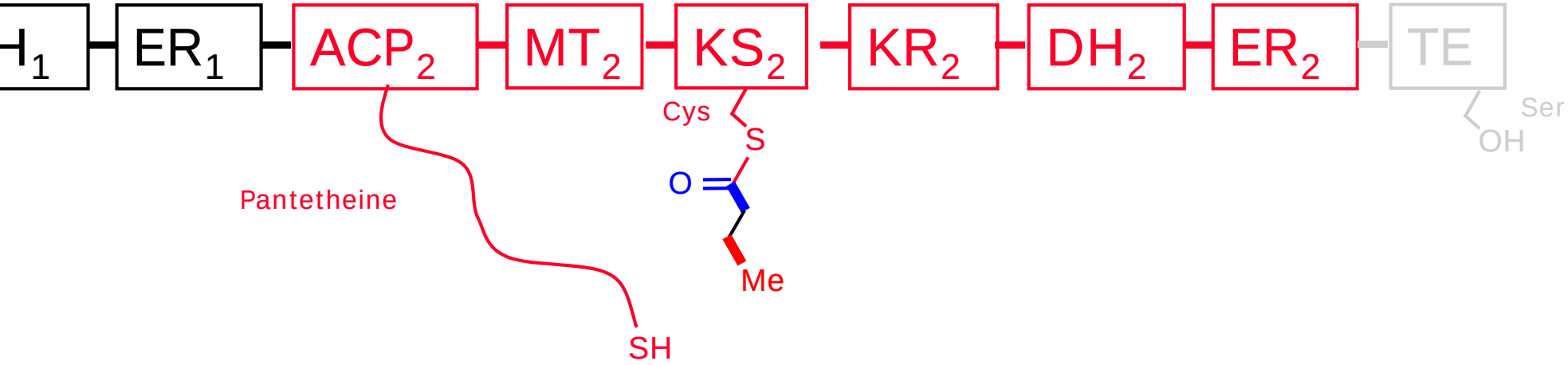


# FATTY ACID BIOSYNTHESIS

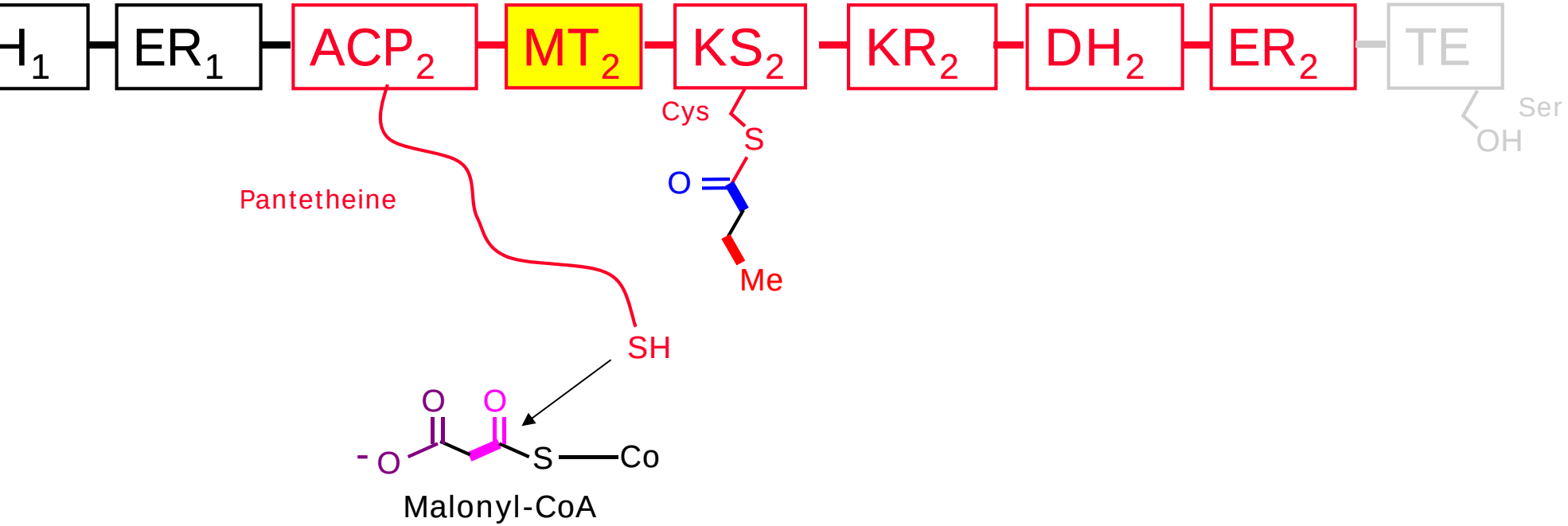



- $KS_2$ catalyzes translocation to module 2

# FATTY ACID BIOSYNTHESIS

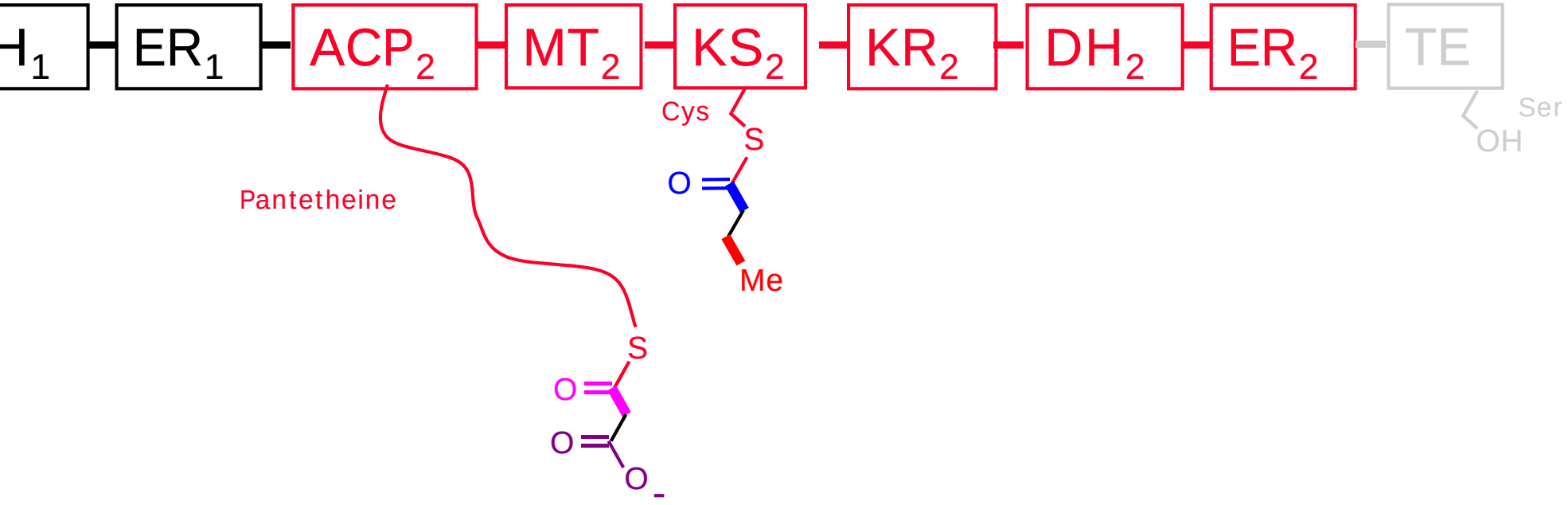


# FATTY ACID BIOSYNTHESIS




- $MT_2$ loads malonyl group onto $ACP_2$

# FATTY ACID BIOSYNTHESIS



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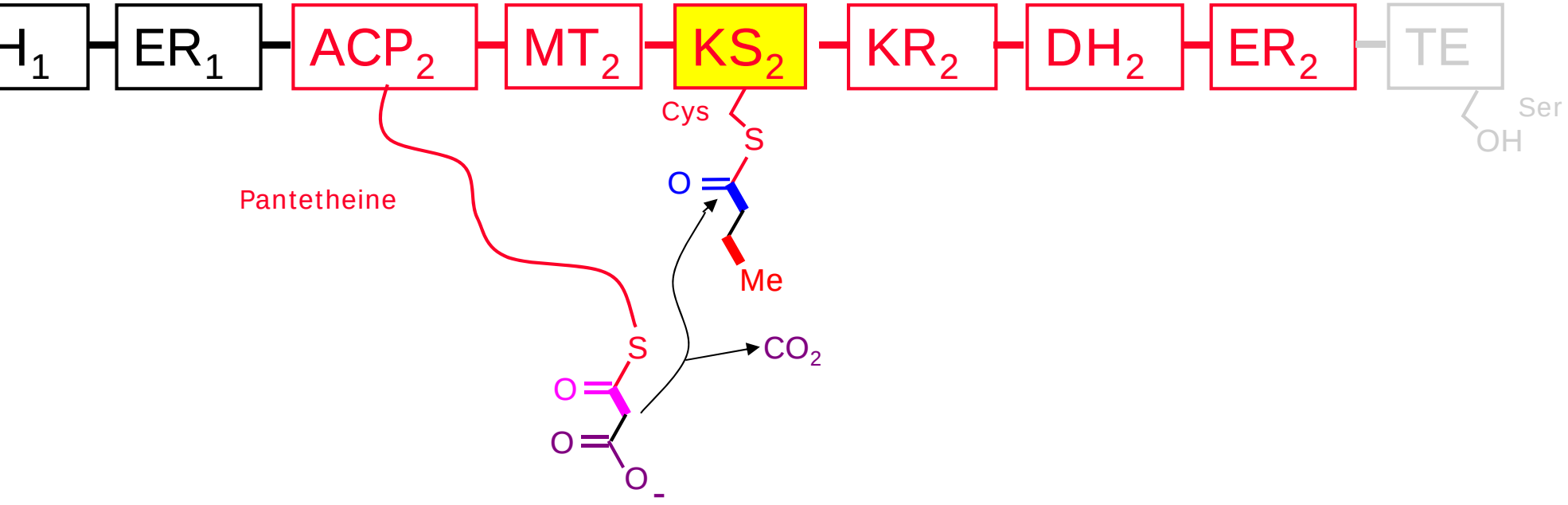
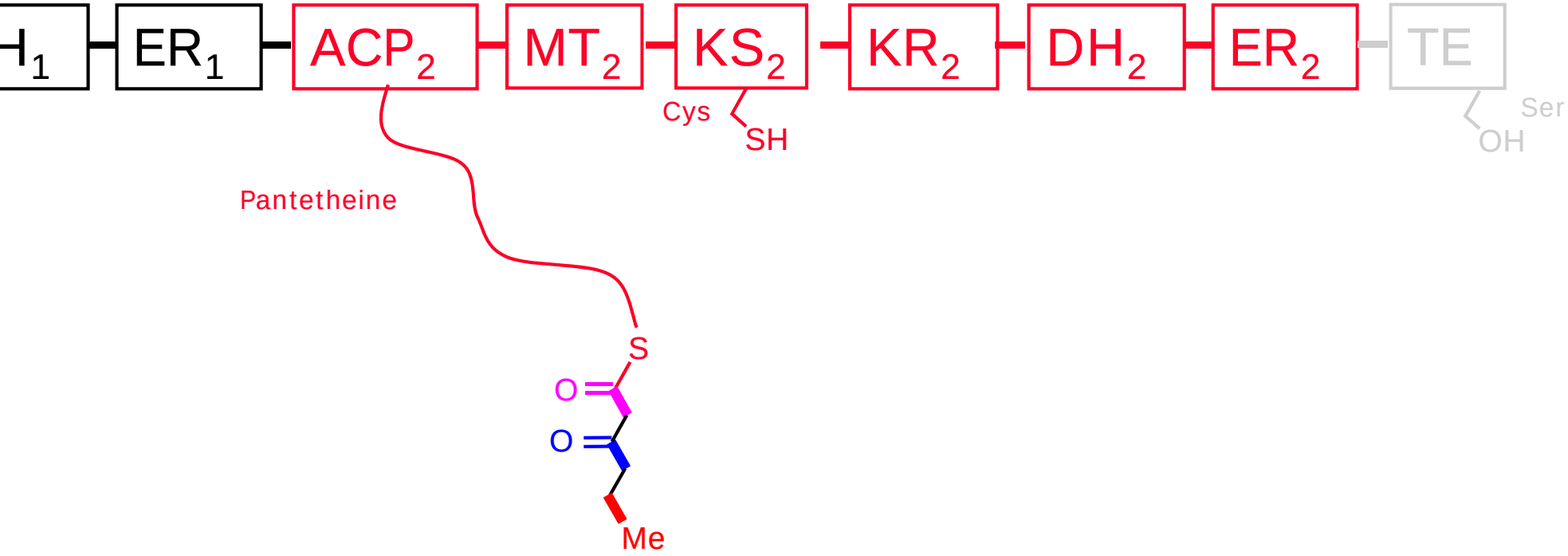
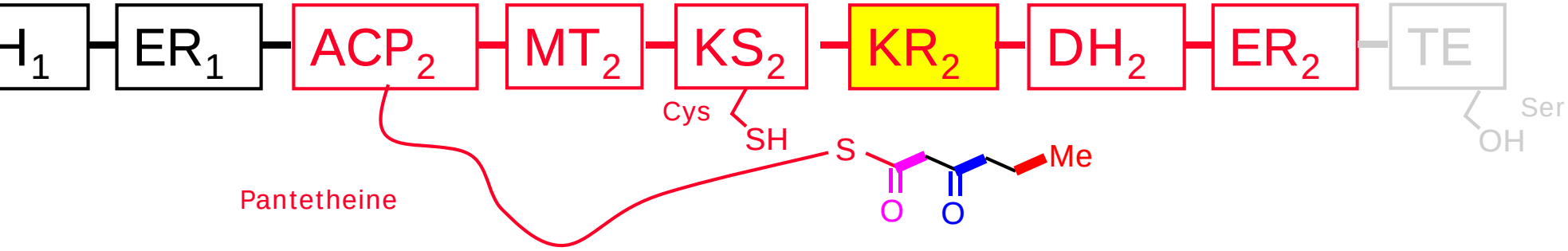



- $KS_2$ catalyzes Claisen condensation

# FATTY ACID BIOSYNTHESIS

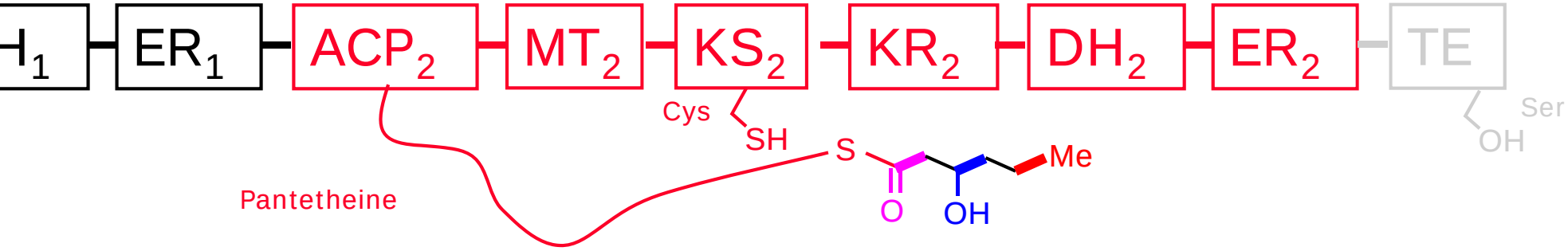


# FATTY ACID BIOSYNTHESIS




- $KR_2$ catalyzes reduction of ketone

# FATTY ACID BIOSYNTHESIS



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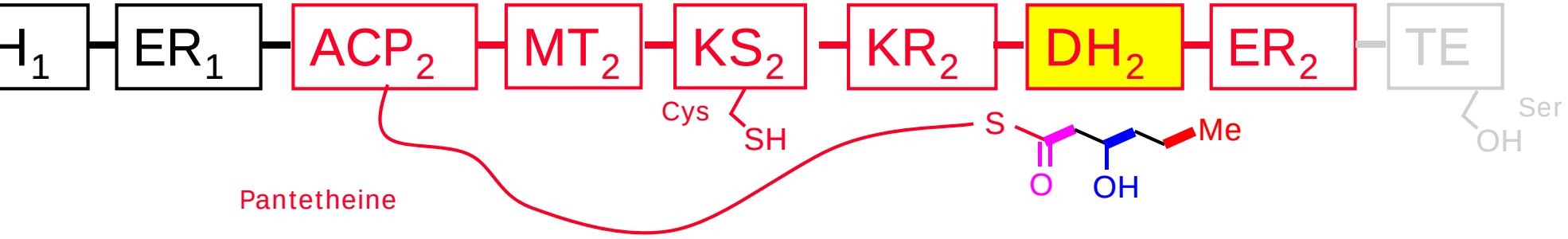
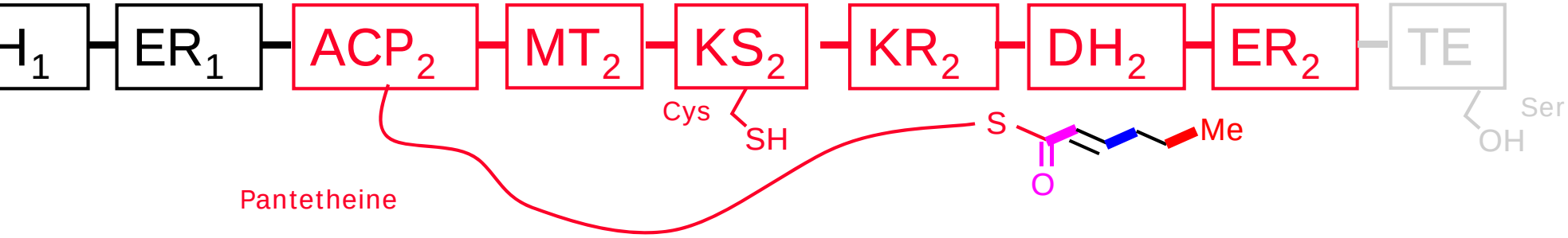



- $DH_2$ catalyzes dehydration of alcohol

# FATTY ACID BIOSYNTHESIS



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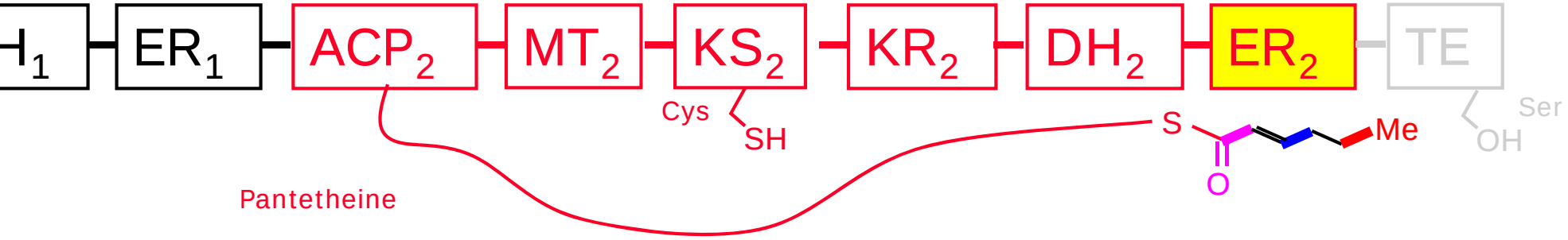
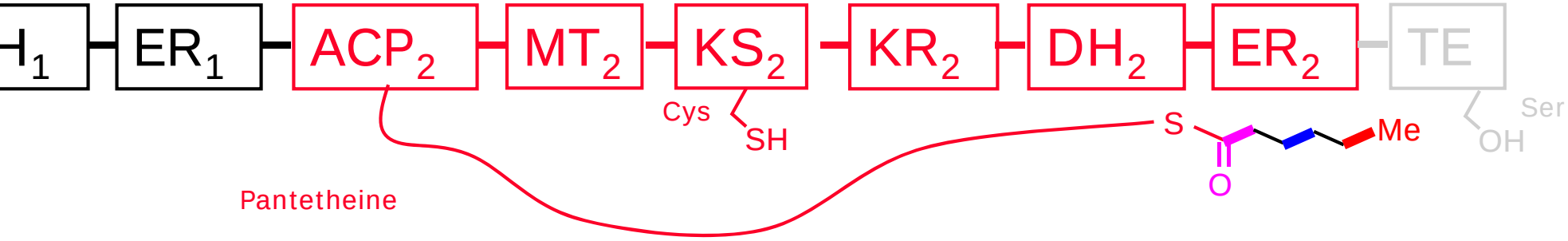



- $ER_2$ catalyzes reduction of alkene

# FATTY ACID BIOSYNTHESIS



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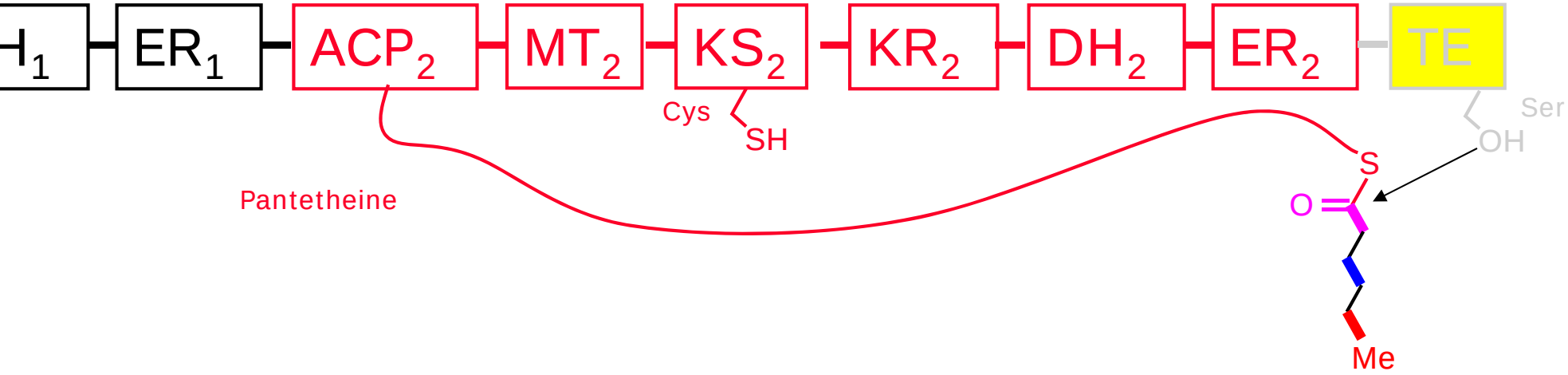
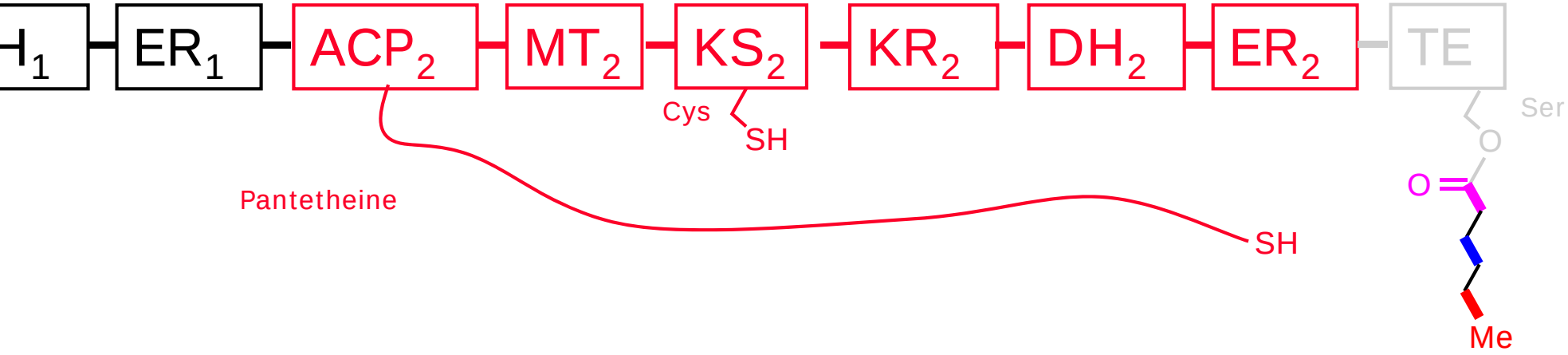
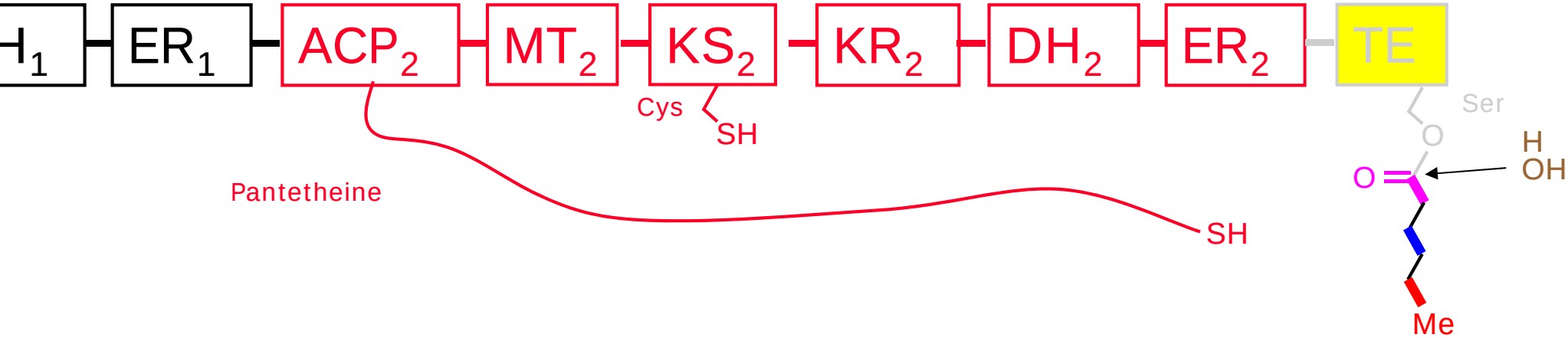



- TE catalyzes transesterification

# FATTY ACID BIOSYNTHESIS

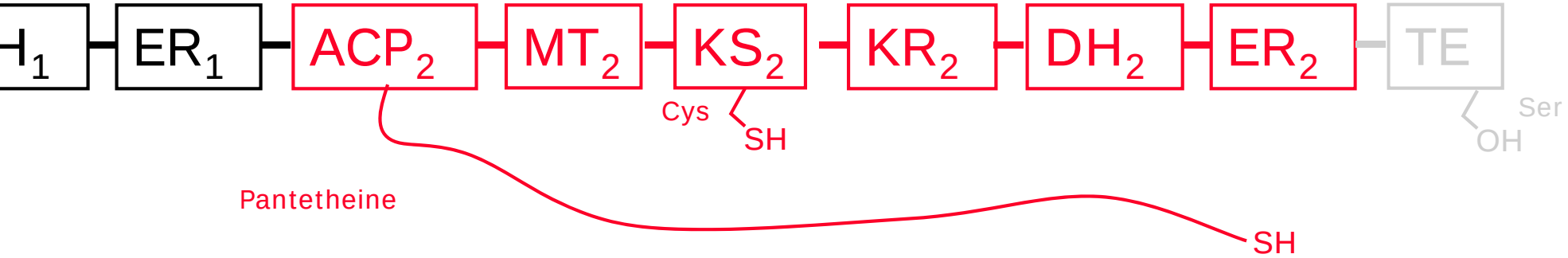


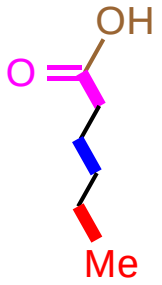
# FATTY ACID BIOSYNTHESIS




- TE catalyzes hydrolysis

# FATTY ACID BIOSYNTHESIS

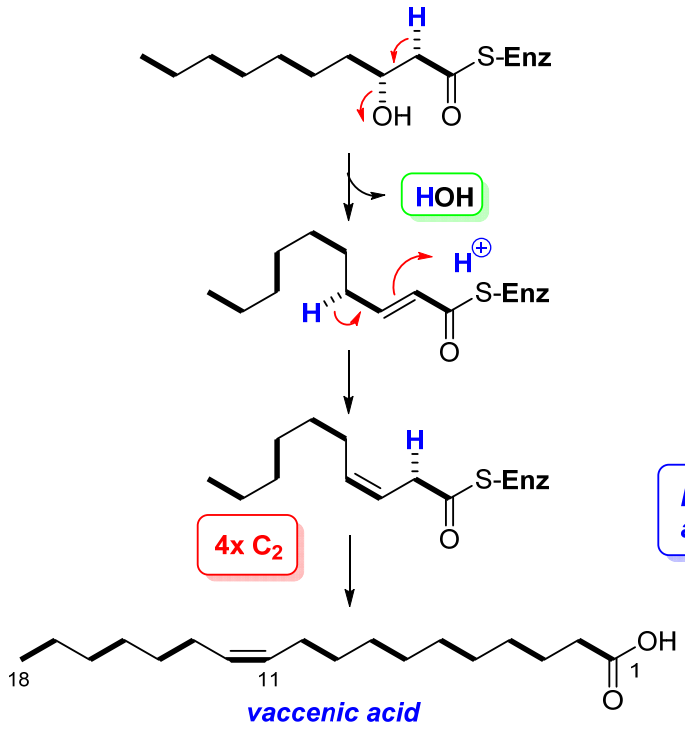


# Biosynthesis of Unsaturated Fatty Acids

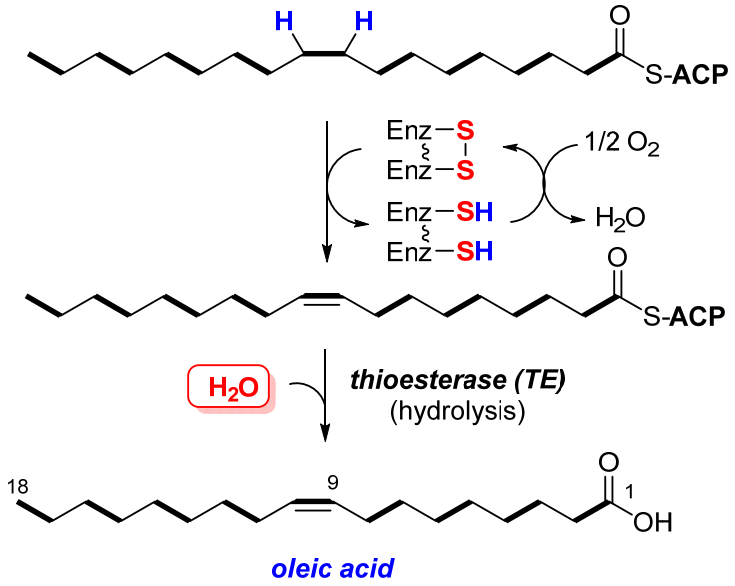
- ***two mechanisms*** are known for the introduction of double bonds into fatty acids:
  - in ***BACTERIA: anaerobic [O]*** → monounsaturated FAs (***MUFAs***)
  - in ***MAMMALS***, ***INSECTS*** & ***PLANTS: aerobic [O]*** → ***MUFAs*** & polyunsaturated FAs (***PUFAs***)

***ANAEROBIC ROUTE (bacteria)***
***(dehydrogenation occurs during chain elongation)***
***mainly MUFAs but some PUFAs***




***NB. in both cases cis - alkenes are produced***

***AEROBIC ROUTE (mammals, insects & plants)***
***(dehydrogenation occurs after chain elongation)***
***MUFAs & PUFAs***



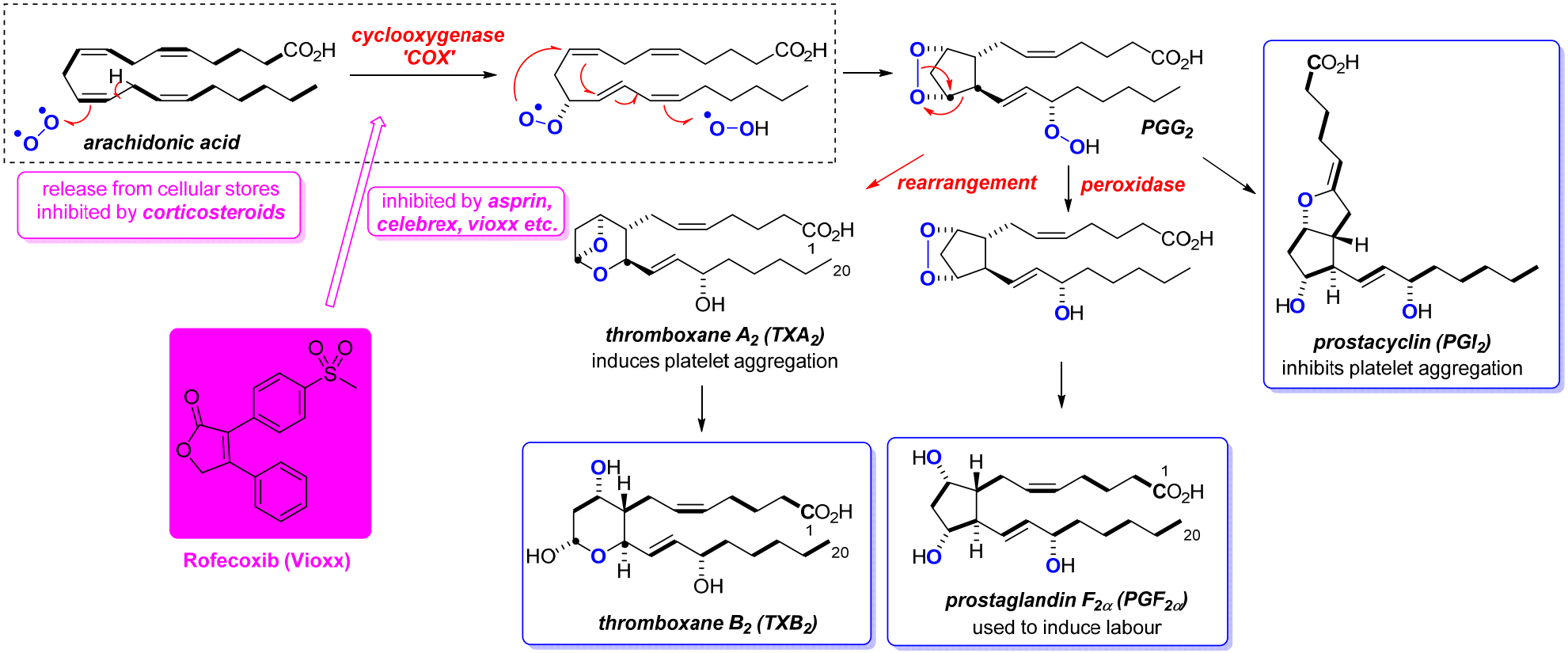

***Position of alkenes in PUFAs***

***animals*** ***1st alkene $C_{11}$-$C_{12}$***

***plants*** ***1st alkene $C_5$-$C_6$***

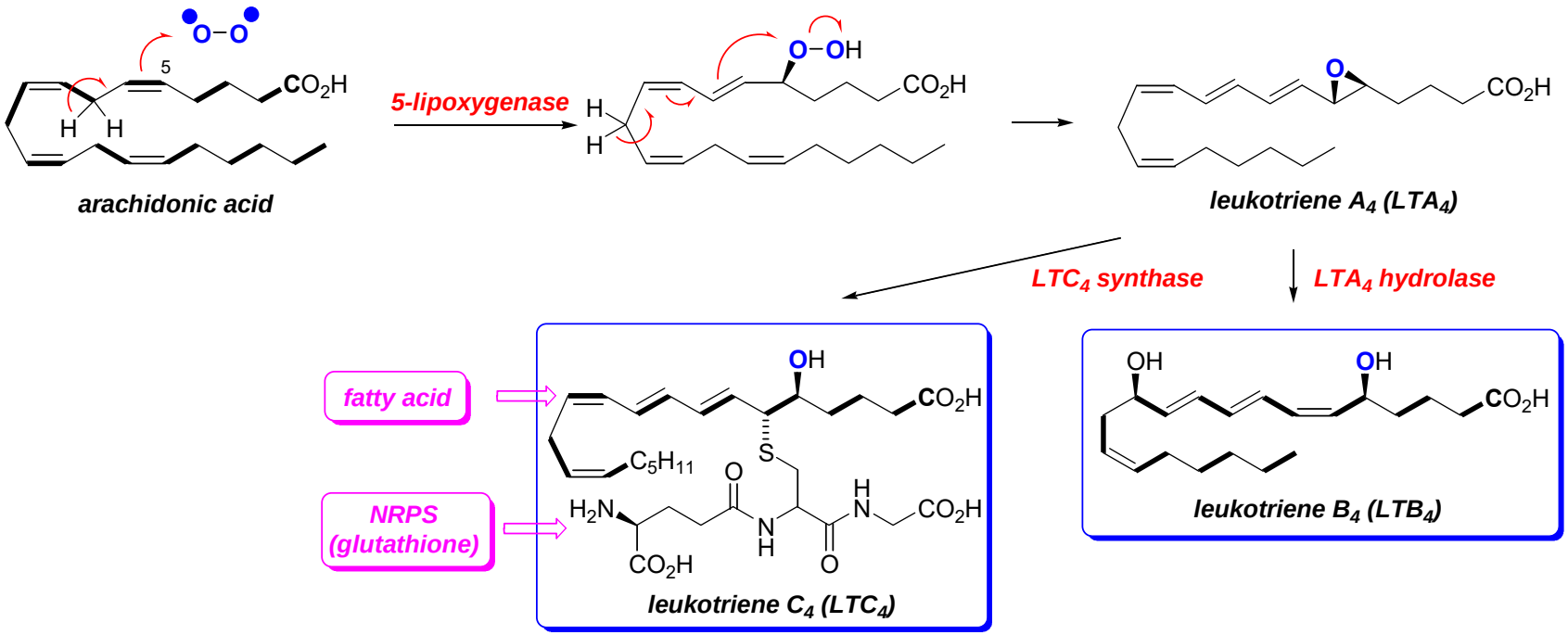
# Biosynthesis of Prostaglandins & Thromboxanes

- ***prostaglandins*** & ***thromboxanes*** are derived from further oxidative processing of arachiodonic acid
- both are important ***hormones*** which control *e.g.* smooth ***muscle contractility*** (blood pressure), ***gastric secretion***, ***platelet aggregation*** & ***inflammation*** (<nM activity)
  - various pharmaceuticals including ***corticosteroids*** & ***asprin*** inhibit biosynthethetic steps in these pathways



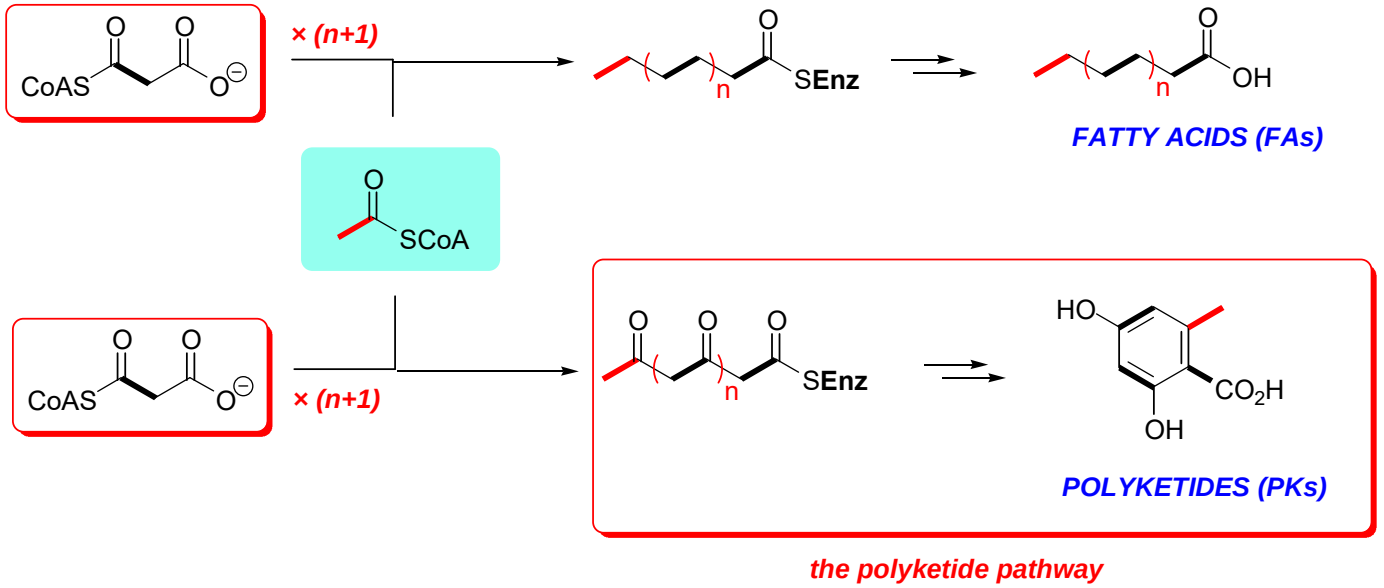
# Biosynthesis of Leukotrienes

- ***leukotrienes*** are the other main class of 2° metabolites derived from ***arachiodonic acid***
  - they are potent (<nM) ***inflammatory substances*** released during allergic reactions



# The Polyketide Pathway

- ***Polyketides*** are also sometimes known as ***acetogenins***
- ***acetyl CoA*** is also the starting point for the biosynthesis of ***polyketide*** secondary metabolites
- these metabolites are topologically very different to the fatty acid metabolites but are synthesised in a very similar fashion. The difference is that during the iterative cycle of chain extension ***the β-keto group is generally not completely reduced out***. This gives rise to huge structural diversity based around a 1,3-oxygenation pattern & cyclisation to give aromatic compounds

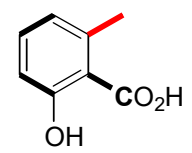



*the polyketide pathway*

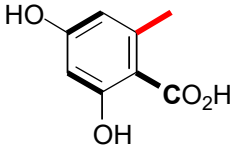
- *NB.* unlike fatty acids. polyketides are NOT biosynthesised by humans – only microorganisms (bacteria) & fungi

# Polyketides

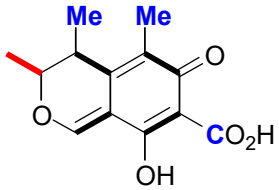
- the structural variety of ***polyketide secondary metabolites*** is very wide:
  - *NB.* starter units marked in red; extender units in bold black; post oligomerisation appended groups in blue



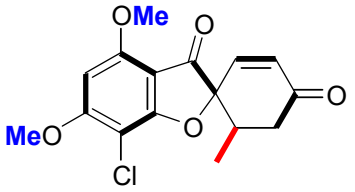
***6-methylsalicylic acid***
*(antibiotic)*



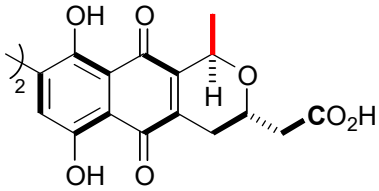
***orsellinic acid***



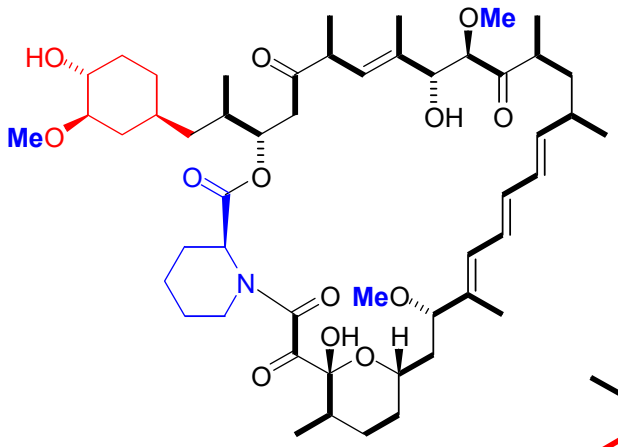
***citrinin***
*(kidney toxin*
*'yellow rice disease')*



***Griseofulvin***
*(treatment for ring*
*worm infections)*

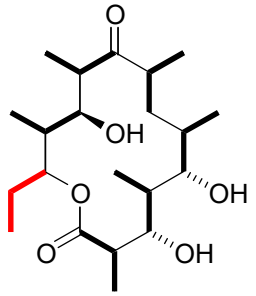


***actinorhodin***
*(antibiotic)*

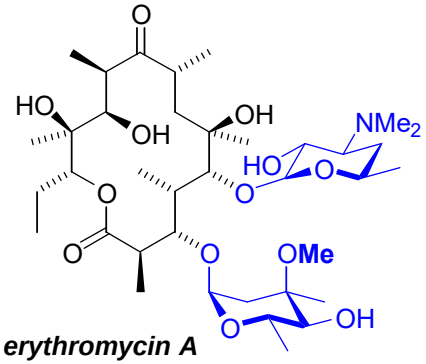


***rapamycin***
*(immunosuppressant)*
***NB. a mixed polypropionate/acetate***

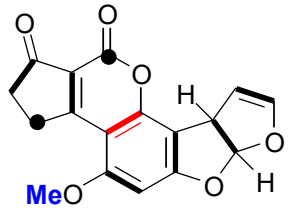
**POLYKETIDES**



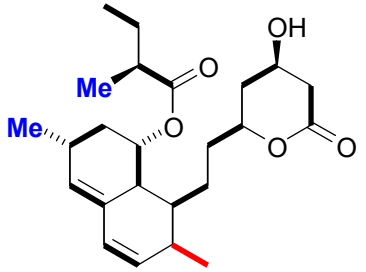
***6-deoxyerythronolide B***
***NB. a polypropionate***



***erythromycin A***
*(antibiotic)*



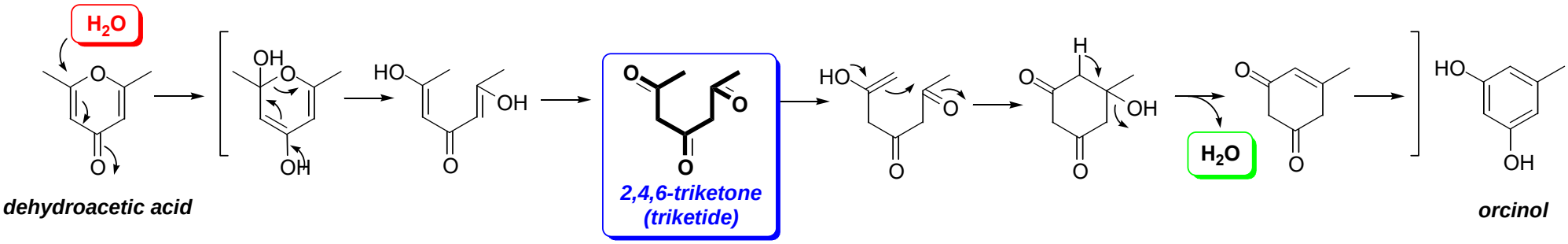
***aflatoxin B1***
*(mycotoxic carcingen)*



***mevinolin***
***(=lovastatin®)***
*(anti-cholesterol)*

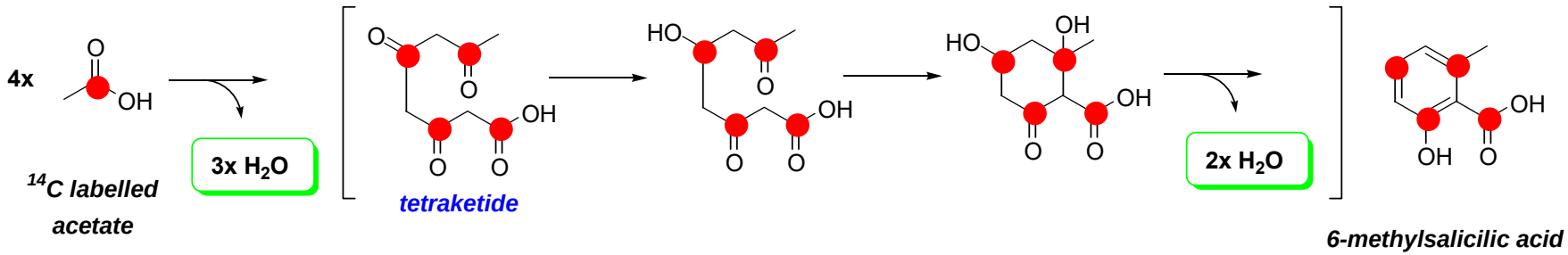
# Historical Perspective – *'The Acetate Hypothesis'*

- ***1907: James Collie*** (University of London) effects conversion of ***dehydroacetic acid*** to ***orcinol*** by boiling with $Ba(OH)_2$ (while trying to deduce the structure of the former):



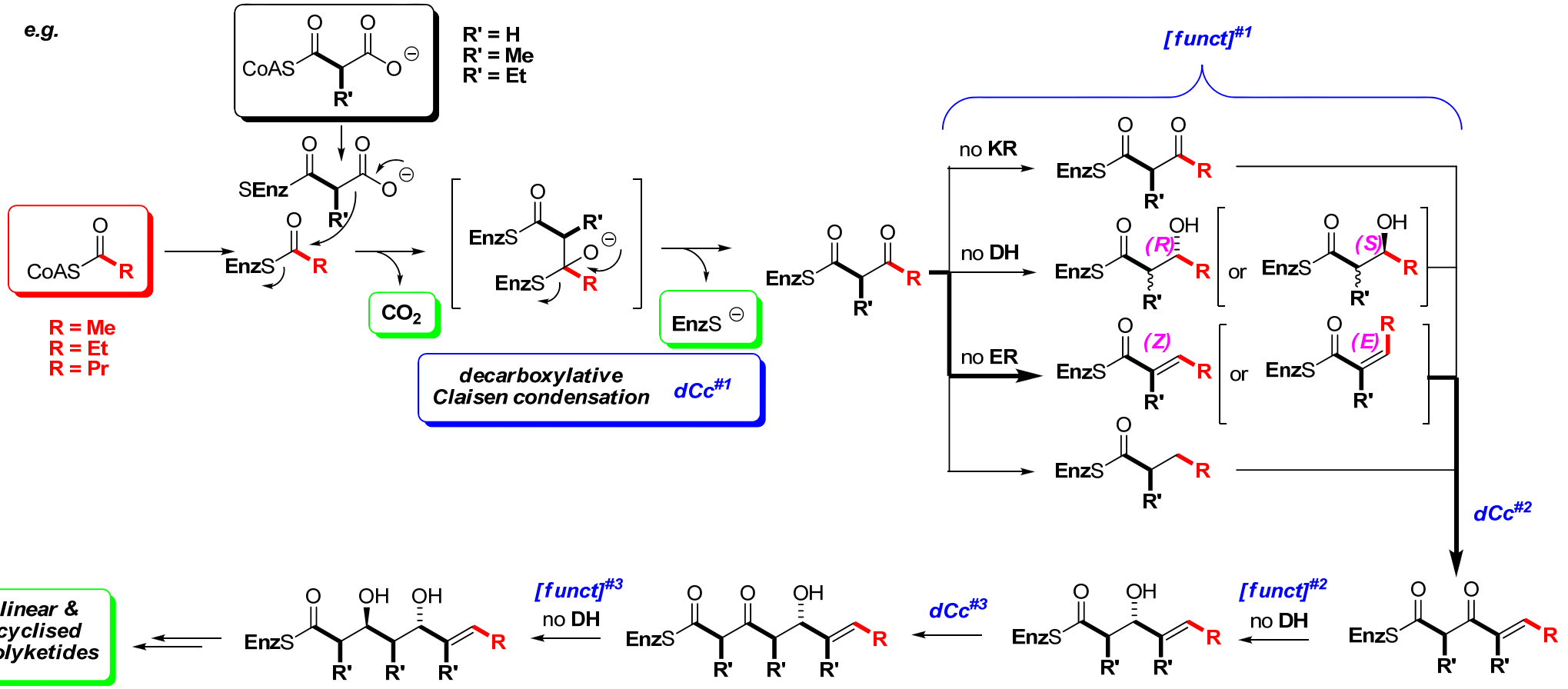

  - Collie perceptively postulated the ***triketone*** as an intermediate ***&*** suggetsed that this might also be an ***intermediate*** in the ***biosynthesis*** of ***orcinol*** (the 'polyketide hypothesis')

- ***1955: Arthur Birch*** used $^{14}C$ labelled acetate to show that 6-methylsalicylic acid (ex. *Penicillium patulum*) was biosynthesised by head-to-tail oligomerisation of ***4 × acetate units*** and proposed the following biogenesis – proceeding *via* a ***tetraketide intermediate*** (*cf.* Collie!):



# Biosynthesis of Polyketides – *Oligomerisation Steps*

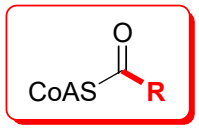
- ***polyketides*** are biosynthesised by a process very similar to that for ***fatty acids***
  - the key ***differences*** are:
    - ***greater variety*** of ***starter units, extender units*** & ***termination processes***
    - ***absent or incomplete reduction of the iteratively introduced β-carbonyl groups: ie. each cycle may differ in terms of KR, DH & ER modules & stereochemistry***



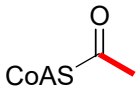

  - this leads to ***enormous diversity...***

# Polyketide Diversity

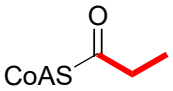
- ***starter units:***



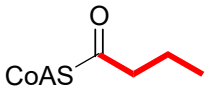

**R = Me**
**R = Et**
**R = $^{n}$Pr & $^{i}$Pr**



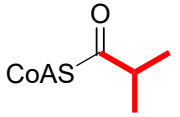

***acetyl CoA***




***propionyl CoA***

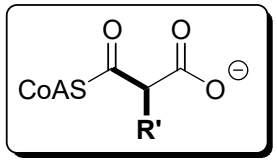



***butyryl CoA***

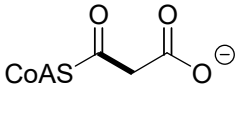



***isobutyryl CoA***

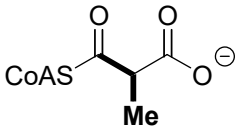
- ***extender units:***



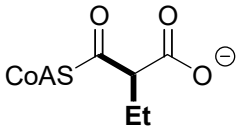

**R' = H**
**R' = Me**
**R' = Et**




**malonyl CoA ($C_2$)**

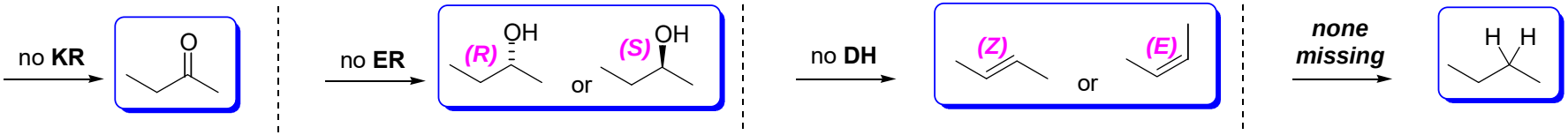



**Me-malonyl CoA ($C_3$)**




**Et-malonyl CoA ($C_4$)**

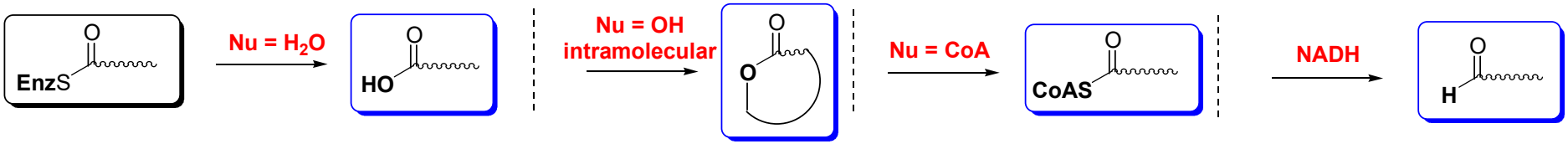
- ***non-functional or missing KR, DH, ER:***




- ***stereochemistry:***

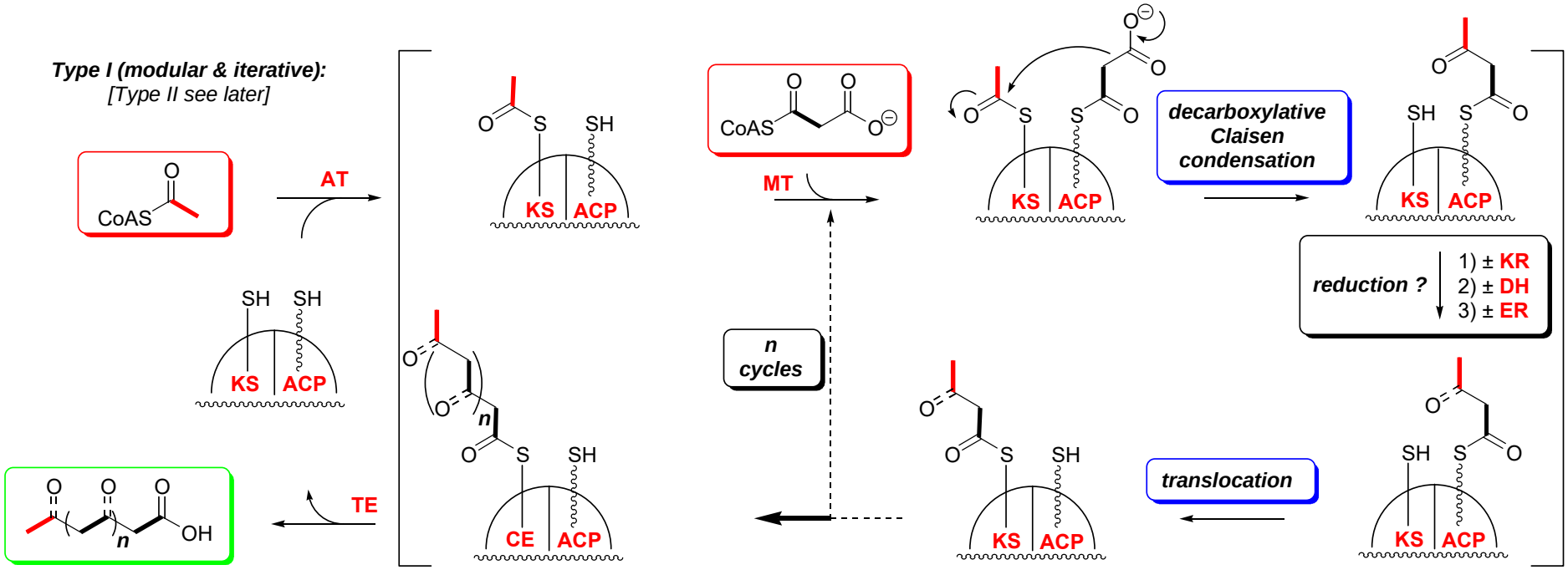
***1) side chain stereochemistry*** (determined by **$KS_n$**)
***2) OH stereochemistry*** (determined by **$KR_{n+1}$**)
***3) alkene stereochemistry*** (determined by **$DH_{n+1}$**)

- ***termination step:***
  - depends on nucleophile that releases product at ***TE*** stage:



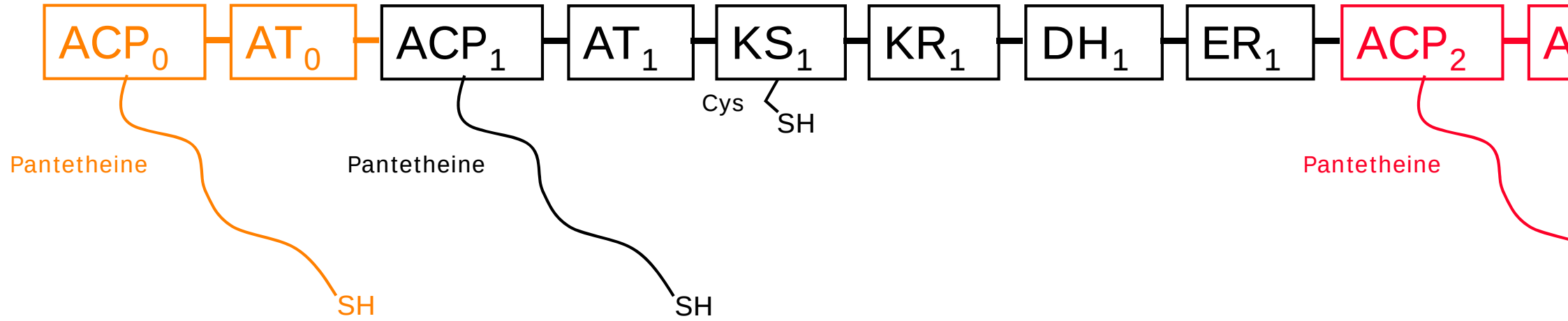
# Biosynthesis of Polyketides – *Overview of PKS*

- the *in vivo* process of polyketide synthesis involves ***PolyKetide Synthases (PKSs):***
  - ***PKSs*** (except Type II, see later) comprise the same ***8 components*** as ***FASs***. *i.e.* (ACP & 7× catalytic activities): ***ACP***, ***KS***, ***AT***, ***MT***, [***KR***, ***DH***, ***ER*** & ***TE]***
  - ***Type I PKSs: single (or small set of) multifunctional protein complex(es)***
    - ***modular (microbial)*** - each 'step' has a dedicated catalytic site (→ ***macrolides***)
    - ***iterative (fungal)*** – single set of catalytic sites, each of which *may* operate in each iteration (*cf.* FASs) (→ ***aromatics/polyphenols*** - generally)
  - ***Type II PKSs: single set of discrete, dissociable single-function proteins***
    - ***iterative (microbial***) - each catalytic module *may* operate in each iteration (*cf.* FASs) (→ ***aromatics/polyphenols***)



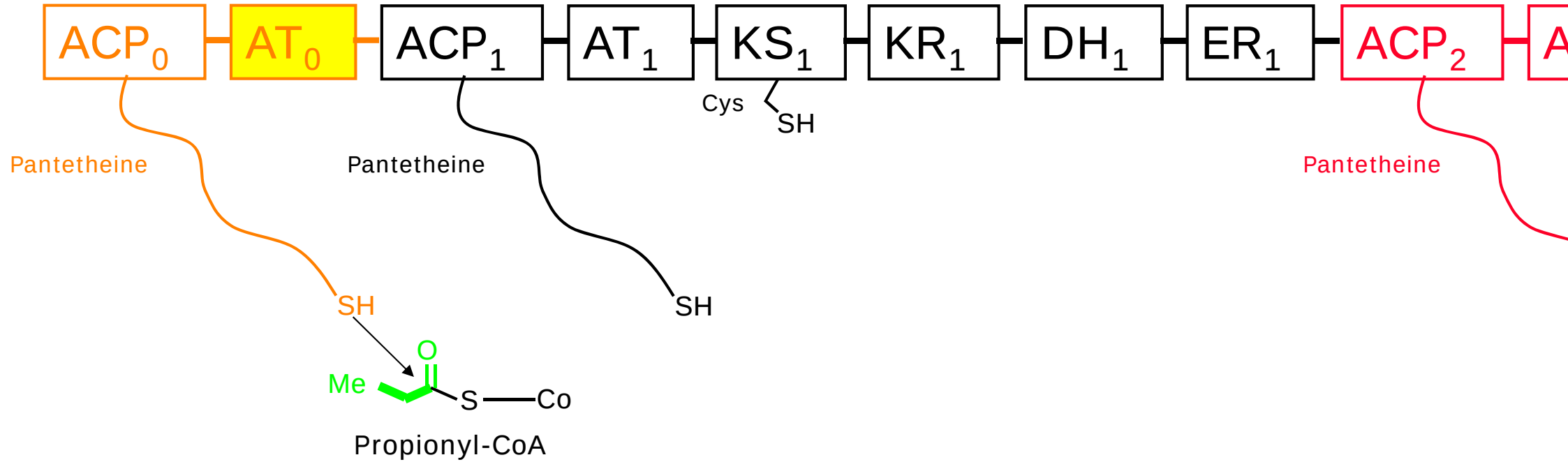

**KS** = keto synthase; **AT** = acetyl transferase; **MT** = malonyl transferase;
**KR** = keto reductase; **DH** = dehydratase; **ER** = enoyl reductase; **TE** = thioesterase; **ACP** = acyl carrier protein

# POLYKETIDE BIOSYNTHESIS [Type I – (modular)]



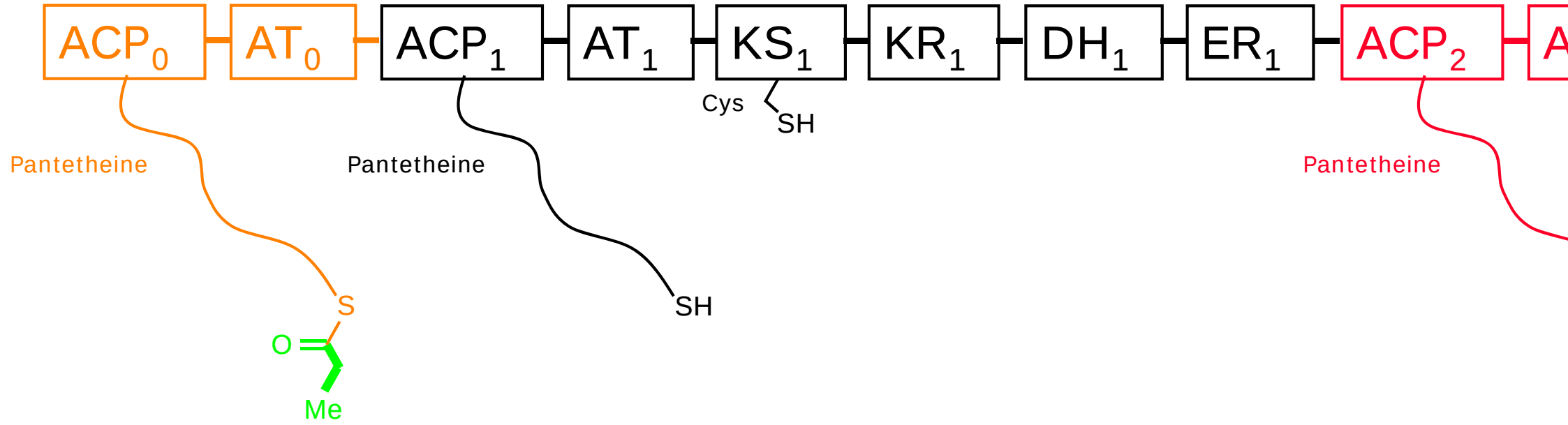

*NB*. the following sequence of slides has also been adapted from: http://www.courses.fas.harvard.edu/%7echem27/

# POLYKETIDE BIOSYNTHESIS

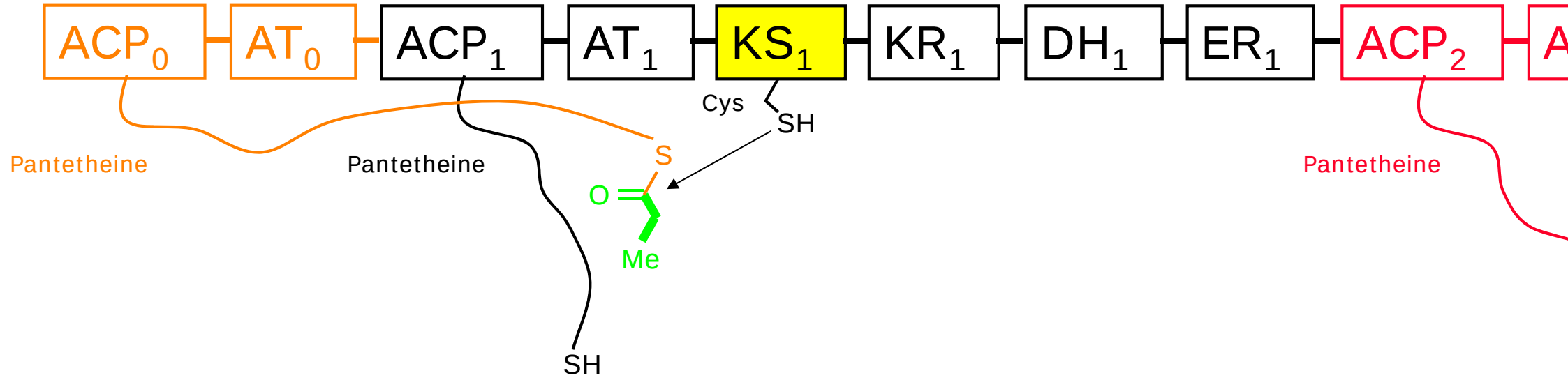



- $AT_0$ loads starting group (propionyl) onto $ACP_0$

# POLYKETIDE BIOSYNTHESIS

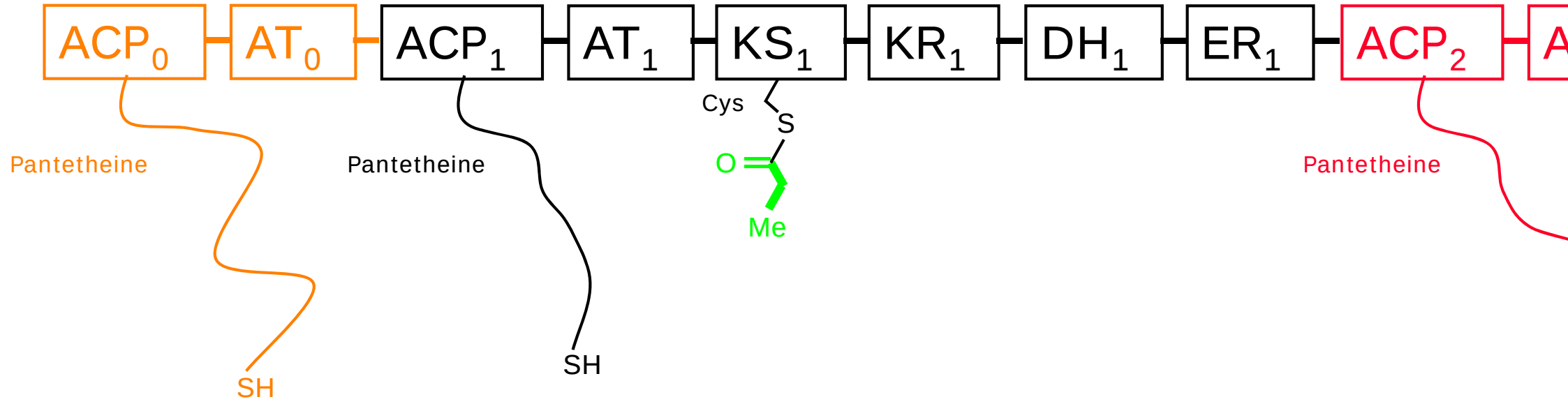


# POLYKETIDE BIOSYNTHESIS

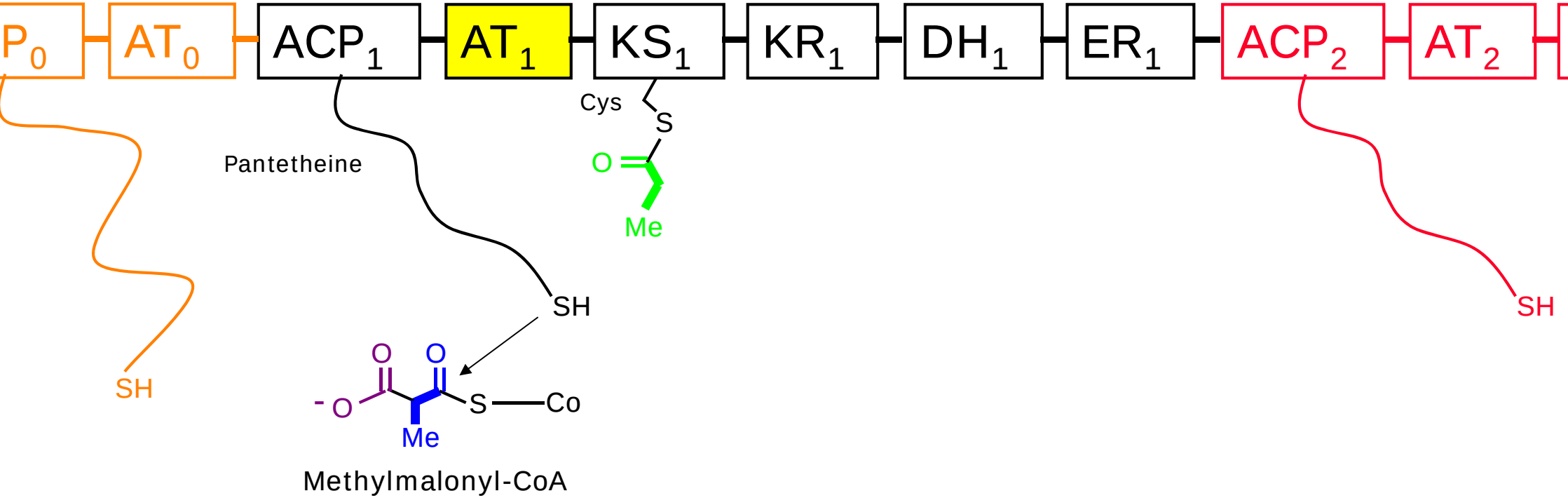



- $KS_1$ catalyzes translocation to module 1

# POLYKETIDE BIOSYNTHESIS

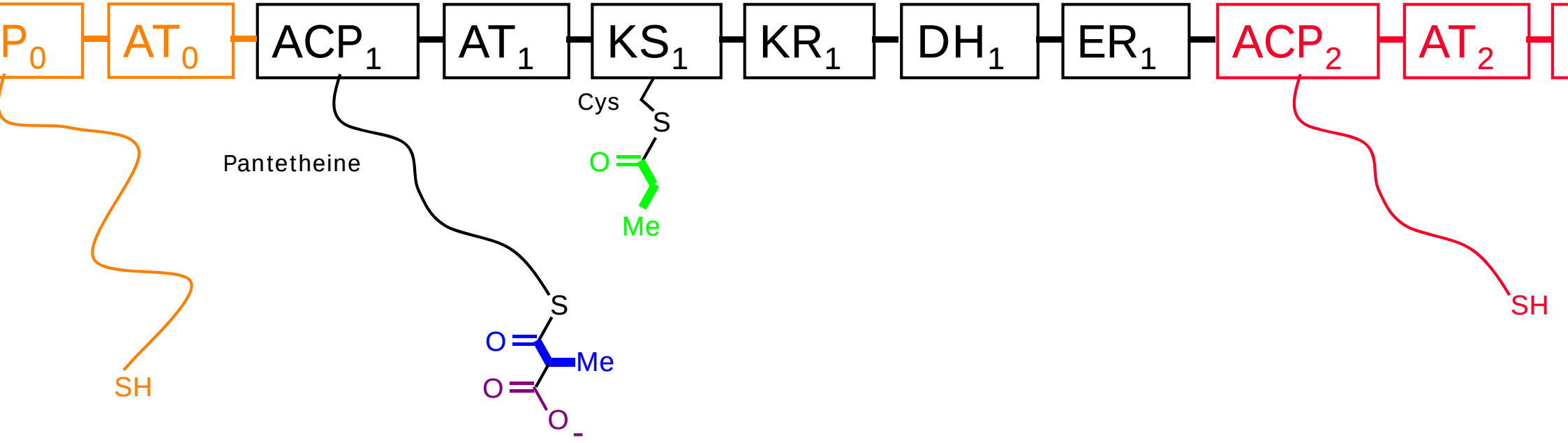


# POLYKETIDE BIOSYNTHESIS

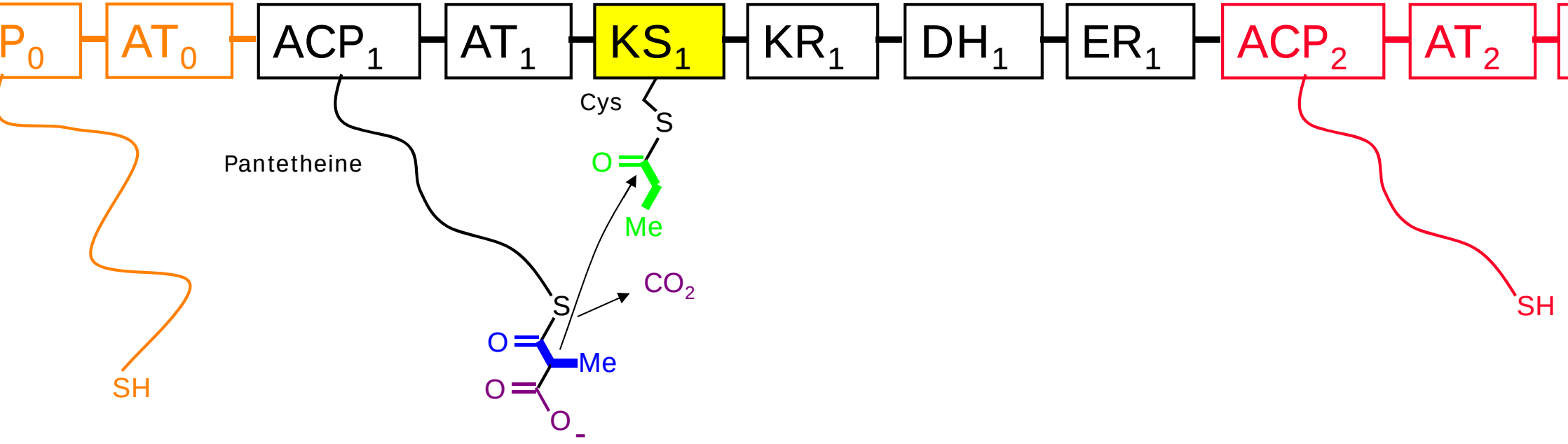



- $AT_1$ loads methylmalonyl group onto $ACP_1$

# POLYKETIDE BIOSYNTHESIS

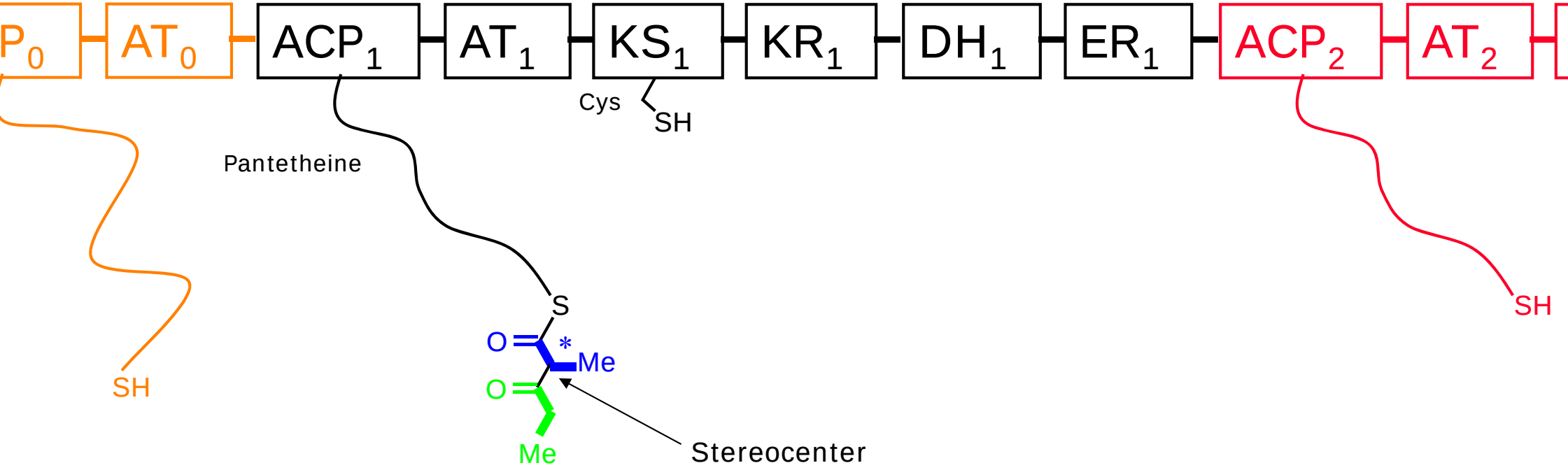


# POLYKETIDE BIOSYNTHESIS

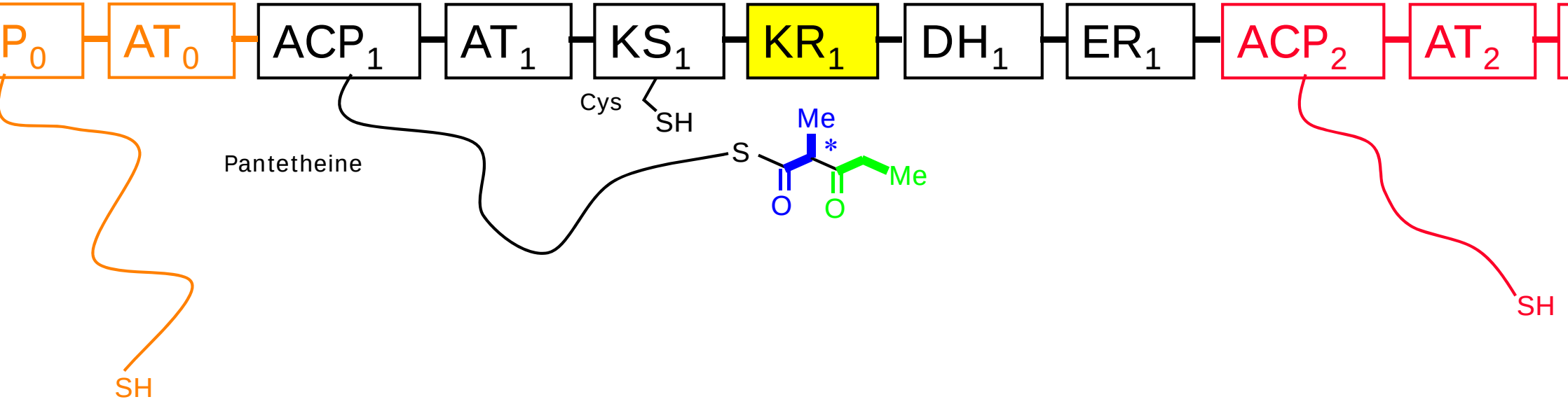



- $KS_1$ catalyzes Claisen condensation

# POLYKETIDE BIOSYNTHESIS

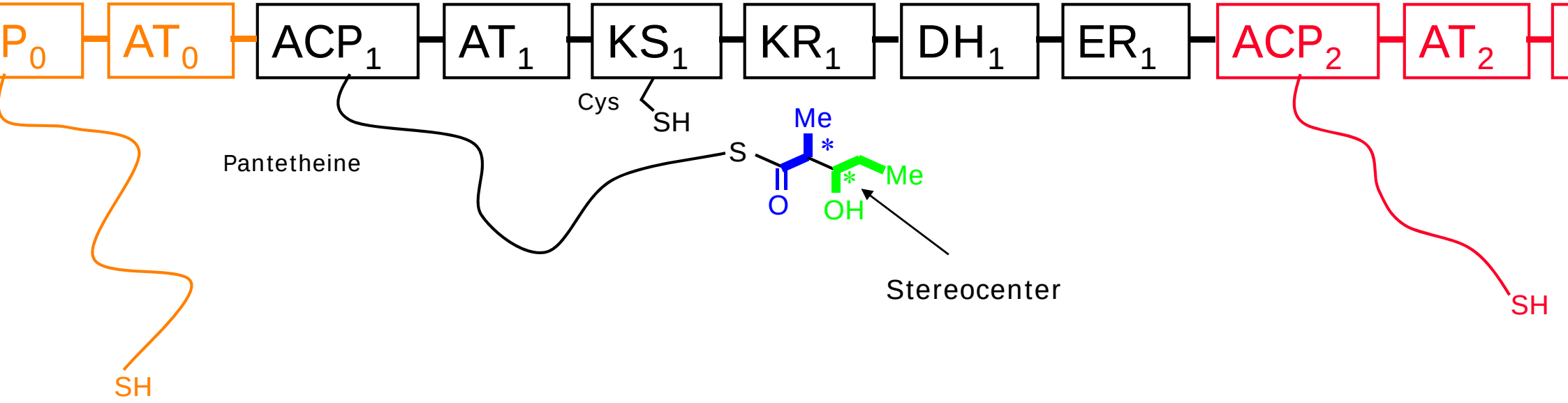


# POLYKETIDE BIOSYNTHESIS

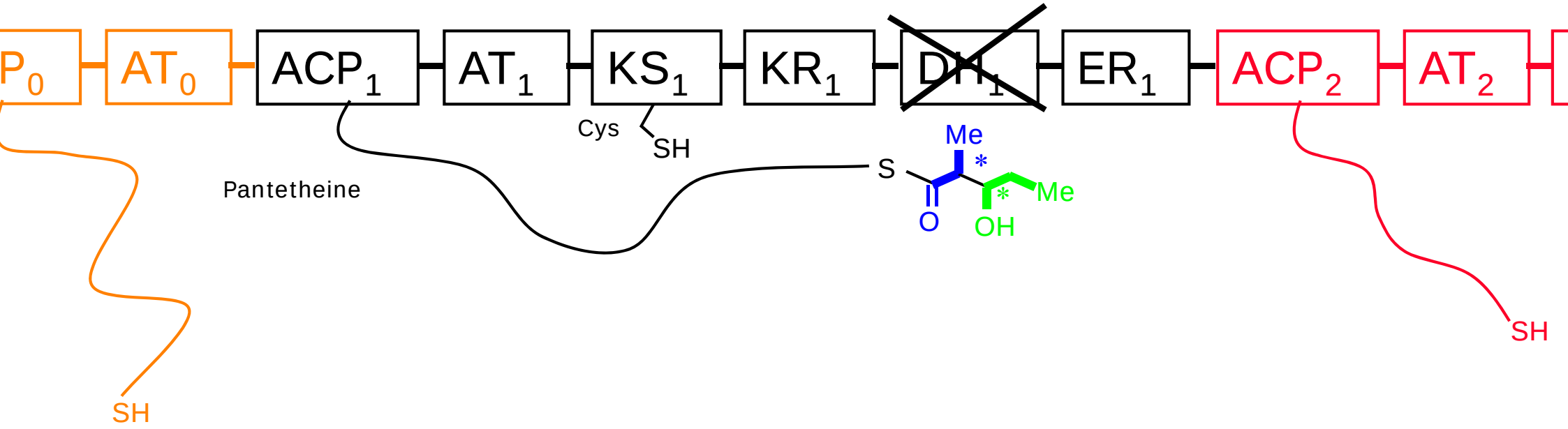



- $KR_1$ catalyzes reduction of ketone

# POLYKETIDE BIOSYNTHESIS

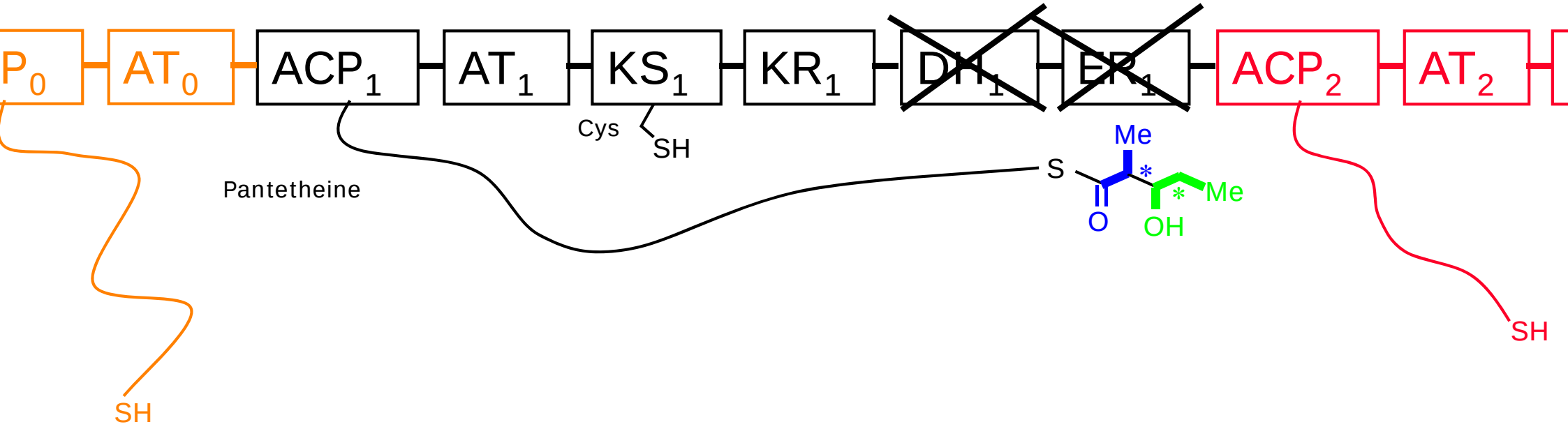


# POLYKETIDE BIOSYNTHESIS



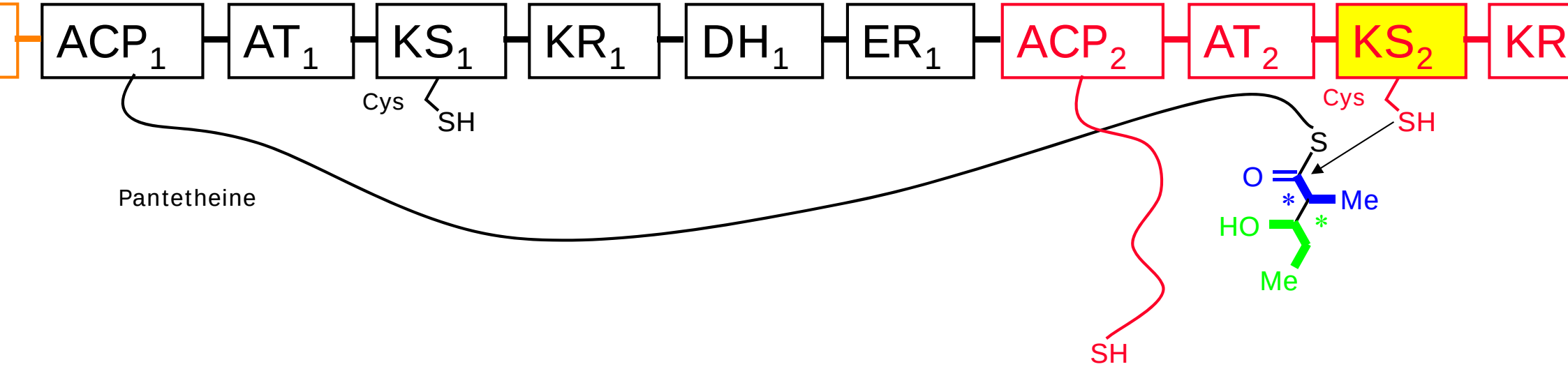

- no $DH_1$ activity

# POLYKETIDE BIOSYNTHESIS



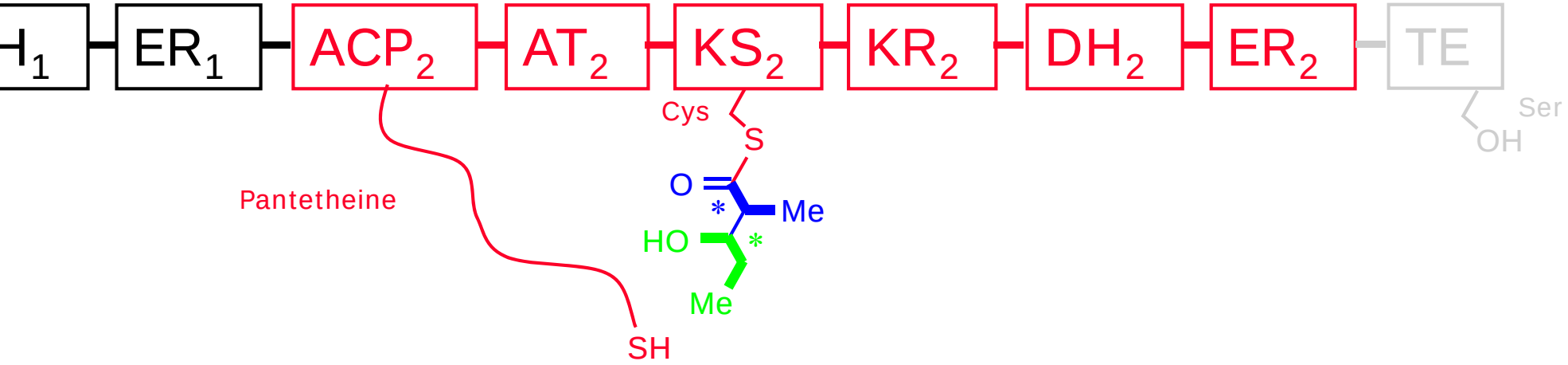

- no $ER_1$ activity

# POLYKETIDE BIOSYNTHESIS



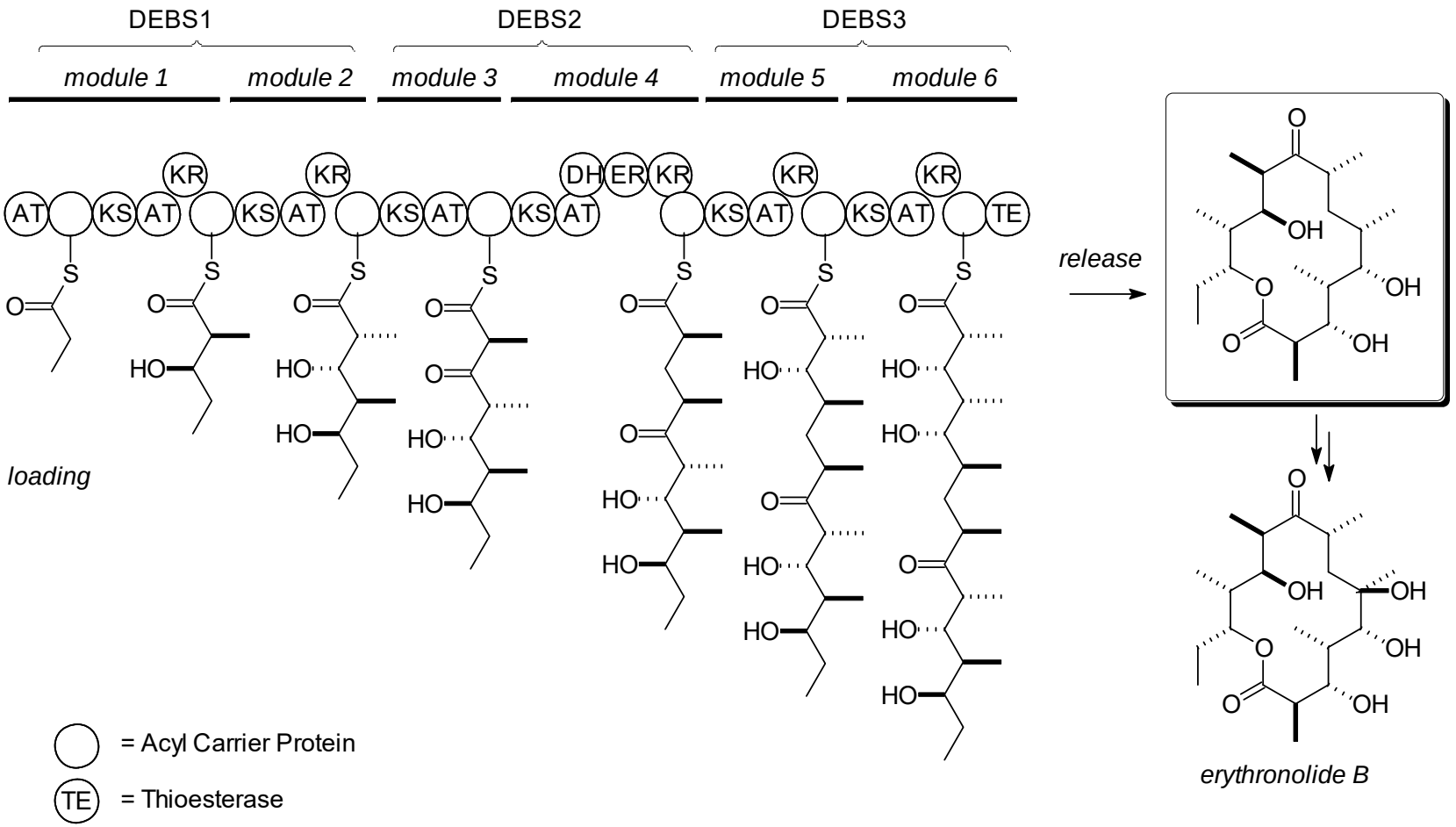

- $KS_2$ catalyzes translocation to module 2

# POLYKETIDE BIOSYNTHESIS



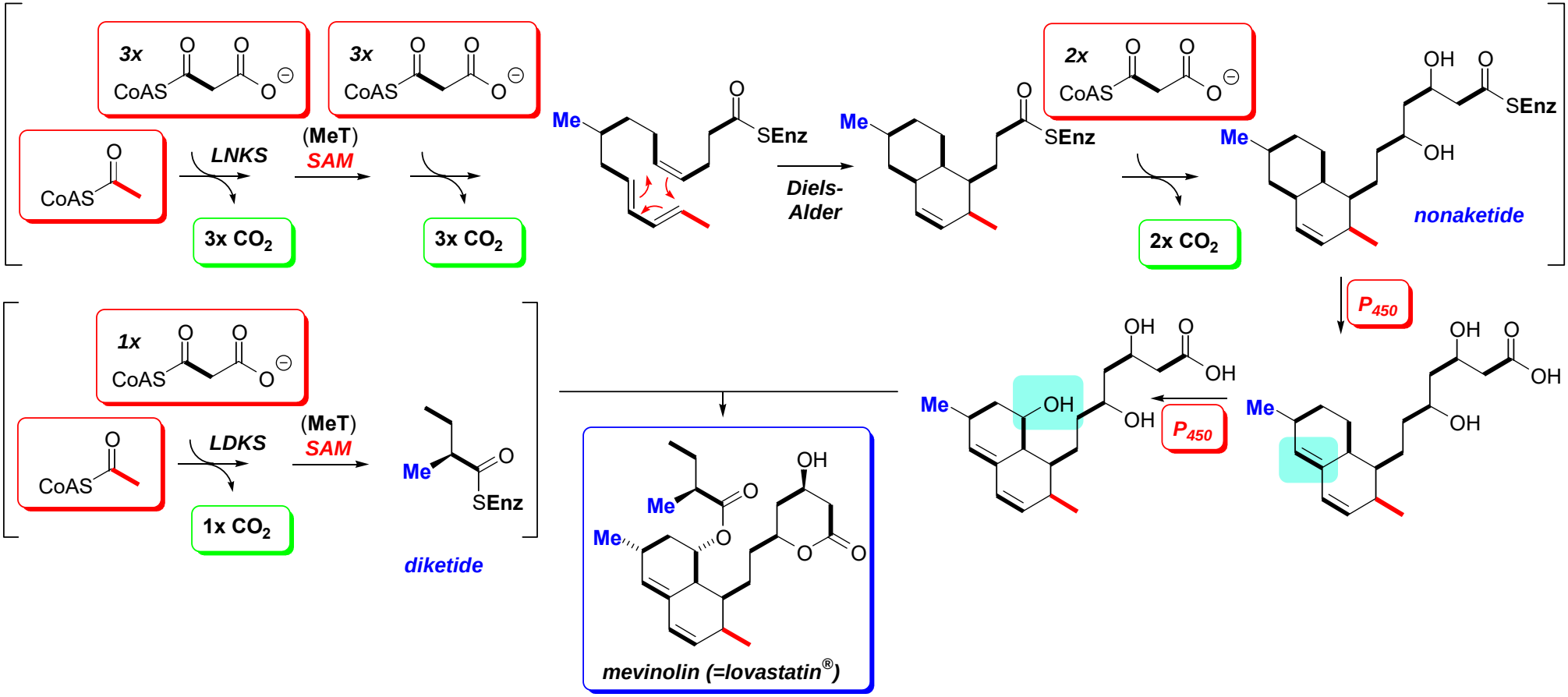
# Biosynthesis of Erythromycin – *Type I(modular) PKS*

- ***6-deoxyerthronolide*** is a precursor to ***erythromycin A*** – ***bacterial*** antibiotic (*Streptomyces erythreus*):
  - ***propionate*** based ***heptaketide***; 3 multifunctional polypeptides (DEBS1, DEBS2 & DEBS3, all ~350 kDa)
  - Katz *et al. Science* **1991**, *252*, 675 (**DOI**); Staunton, Leadley *et al. Science* **1995**, *268*, 1487 (**DOI**); Khosla *et al. J. Am. Chem. Soc.* **1995**, 9105 (**DOI**); ***review:*** Staunton & Weissman *Nat. Prod. Rep.* **2001**, *18*, 380 (**DOI**)



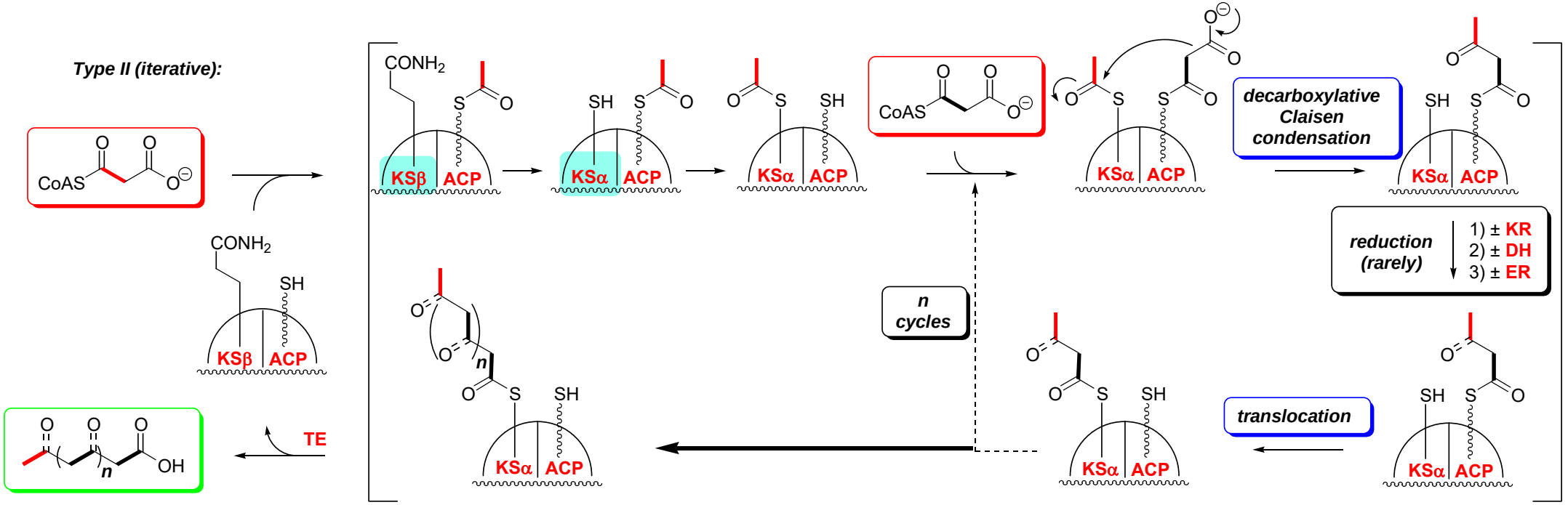
# Biosynthesis of Mevinolin – *Type I(iterative) PKS*

- ***mevinolin (=lovastatin®)*** – cholesterol lowering metabolite of filamentous ***fungus*** *Aspergillus terreus*
  - inhibits HMG-CoA → mevalonate (see next lecture) – rate-limiting step in biosynthesis of ***cholesterol***
  - ***acetate*** based polyketide composed of a diketide and nonaketide linked by an ester
  - 2 × Type I (iterative) PKSs: LNKS and LDKS...both contain ***MeT*** (***methyl transferase***) activities
  - Hutchinson *et al. Science* **1999**, *284*, 1368 (**DOI**)



# Type II PKSs – *Enzyme Clusters (Microbial)*

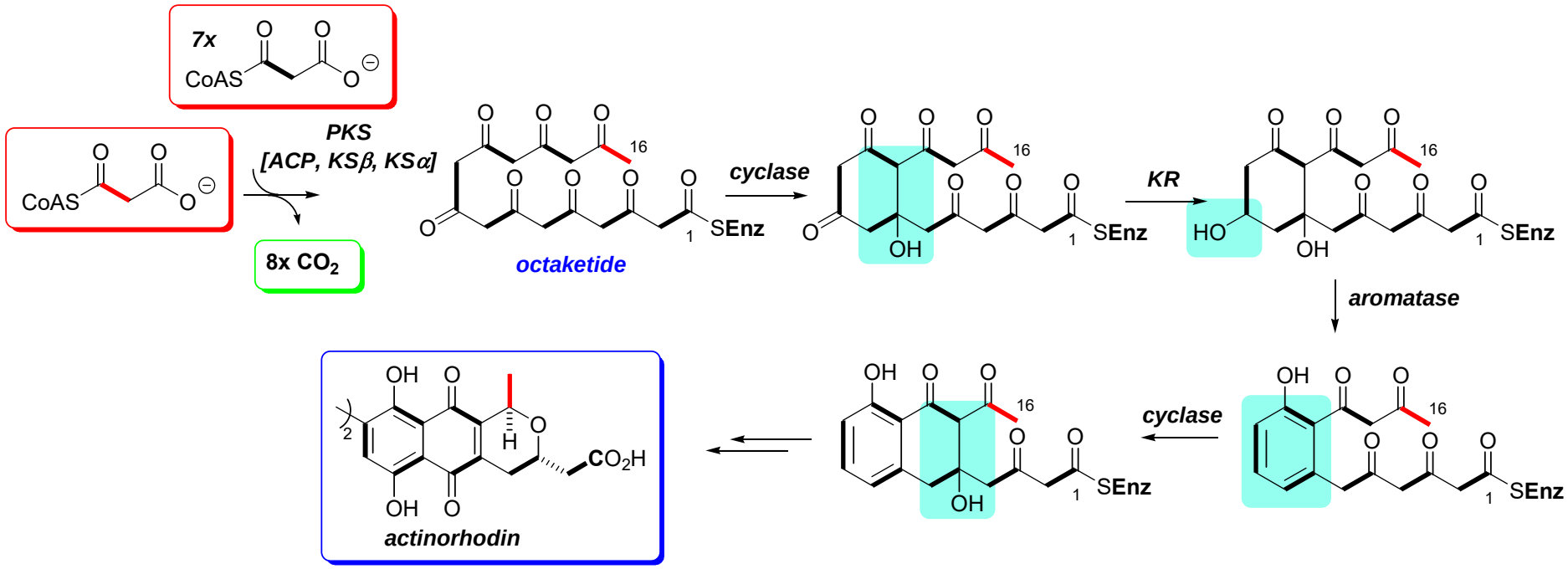
- ***Type II PKSs:*** single set of discrete, dissociable single-function proteins (ACP & 6× catalytic functions): ***ACP***, ***KS$_\alpha$***, ***KS$_\beta$***, [***KR***, ***DH***, ***ER***, & ***TE***] [*NB.* NO acetyl or malonyl transferases (AT, MT)]
  - ***iterative*** - each catalytic module *may* operate in each iteration (*cf.* FASs) (→ ***aromatics/polyphenols***)
- these clusters (generally) use ***malonate*** as BOTH ***starter*** & ***extender*** unit
- their ***ACP proteins*** are able to load malonate direct from malonyl CoA (no MT required)
  - the ***starter malonate*** is ***decarboxylated*** by '***ketosynthase' β (KS$_\beta$)*** to give ***S-acetyl-ACP***
  - the ***extender malonates*** undergo ***decarboxylative Claisen condensations*** by ***ketosynthase α (KS$_\alpha$)***
- these clusters rarely utilise ***KR***, ***DH*** or ***ER*** activities and produce 'true' polyketides:




**KS$_\beta$** = 'keto synthase β' (=decarboxylase!); **KS$_\alpha$** = 'keto synthase α' (=ketosynthase!); **KR** = keto reductase; **DH** = dehydratase; **ER** = enoyl reductase; **TE** = thioesterase; **ACP** = acyl carrier protein

# Biosynthesis of Actinorhodin – *Type II PKS*

- ***actinorhodin –*** octaketide ***bacterial antibiotic*** (*Streptomyces coelicolor*)
  - Hopwood *Chem. Rev.* **1997**, *97*, 2465 (**DOI**)

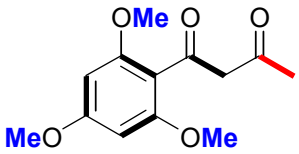



  - ***timing*** of ***1st cyclisation*** and mechanism of ***control of chain length*** uncertain
    - ***octaketide*** synthesis then cyclisation? (as shown above)
    - ***hexaketide*** synthesis then cyclisation then two further rounds of extension?
  - indications can sometimes be gleaned from ***biomimetic syntheses***...

# Scope of Structures - *Type II PKS*

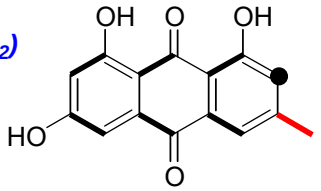
- ***microbial polyphenolic*** metabolites:

***pentaketides (5x $C_2$)***



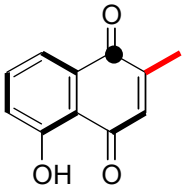

***eugenone***

***octaketides (8x $C_2$)***



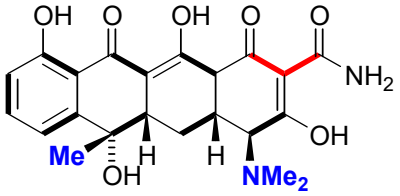

***emodin***

***hexaketides (6x $C_2$)***



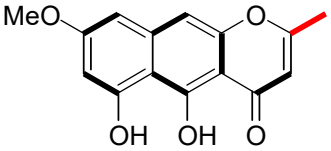

***plumbagin***

***nonaketides (9x $C_2$)***



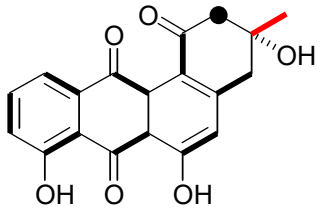

***tetracycline***

***heptaketides (7x $C_2$)***




***rubrofusarin***

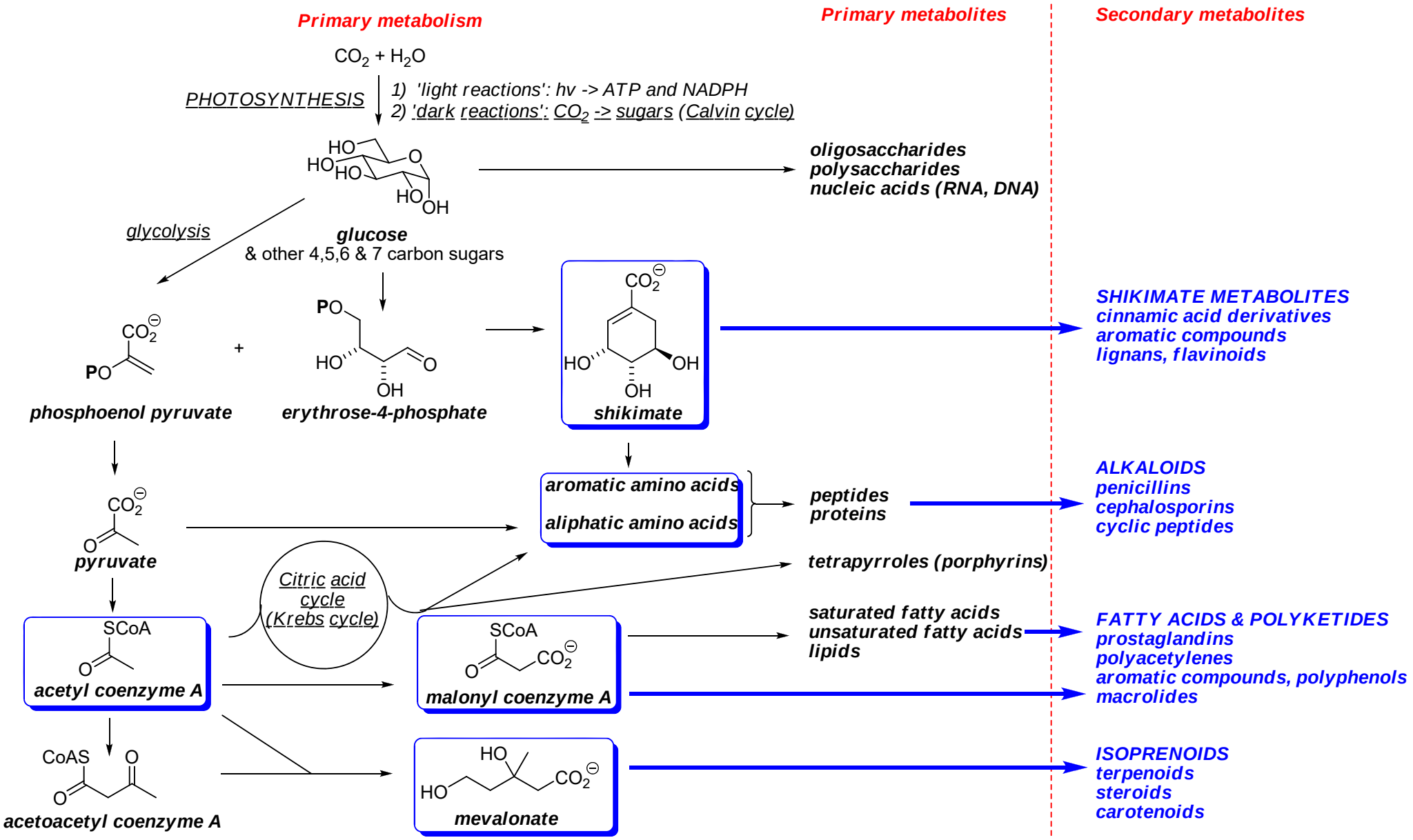
***decaketides (10x $C_2$)***




***rabelomycine***

- many display interesting biological activities...

# Primary Metabolism - *Overview*




For interesting animations' of *e.g.* photosynthesis see: **http://www.johnkyrk.com/index.html**