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·综　述·

植物咖啡酸-*O*-甲基转移酶的研究进展

李元玉[1]，陈万生[1,2,3]，肖莹[2]

1 福建中医药大学 药学院，福建 福州 350122
2 上海中医药大学 中药资源与生物技术中心，上海 201203
3 海军军医大学附属长征医院医学保障中心，上海 200003

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摘　要：单木质素醇 (H 型、G 型和 S 型) 是构成植物木质素和木脂素的基本单元，其组成的不同直接决定木质素和木脂素的化学多样性和生物活性差异。咖啡酸-*O*-甲基转移酶 (caffeic acid *O*-methyltransferase, COMT) 可催化苯丙素类化合物羟基上氧原子的甲基化，在不同类型单木质素醇的构成中起决定作用，是木质素和木脂素生物合成途径的关键酶。2010 年的相关综述主要对 COMT 的基因特征和在木质素生物合成中的调控作用作了介绍，文中聚焦了近十多年来 COMT 的最新研究进展，从基因特征、表达特征、结构特征和调控作用几个方面进行全面综述，并对 COMT 的研究和应用前景进行展望。

关键词：咖啡酸-*O*-甲基转移酶；木质素；木脂素；单木质素醇

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Corresponding authors: XIAO Ying. Tel: +86-21-51322403; E-mail: xiaoyingtcm@shutcm.edu.cn
CHEN Wansheng. Tel: +86-21-51322404; E-mail: chenwansheng@shutcm.edu.cn
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Advances in plant caffeic acid-*O*-methyltransferase

LI Yuanyu[1], CHEN Wansheng[1,2,3], XIAO Ying[2]

1 School of Pharmacy, Fujian University of Traditional Chinese Medicine, Fuzhou 350122, Fujian, China
2 Research and Development Center of Chinese Medicine Resources and Biotechnology, Shanghai University of Traditional Chinese Medicine, Shanghai 201203, China
3 Medical Guarantee Center, Changzheng Hospital, Naval Medical University, Shanghai 200003, China

Abstract: Monolignin alcohols (type H, type G and type S) are the basic units of lignin and lignans in plants, and their composition differences directly determine the chemical diversity and biological activity of lignin and lignans. Caffeic acid *O*-methyltransferase (COMT) catalyzes the methylation of oxygen atoms on the hydroxyl groups of phenylpropanoids, playing a critical role in the composition of different types of monolignin alcohols, and thus acting as a key enzyme involved in the biosynthesis pathway of lignin and lignans. A previous review published in 2010 mainly introduced the gene characteristics of COMT and its regulatory role in lignin biosynthesis. This article summarized the latest research progress of COMT in the past decade, including the gene characteristics, expression characteristics, structural characteristics of COMT and its regulatory effects, and prospected future research and application of COMT.

Keywords: caffeic acid *O*-methyltransferase; lignin; lignans; monolignin alcohol

木质素是由苯丙烷代谢途径合成的 3 种单木质素醇通过化学键聚合而成的高分子化合物，其主要单体为对-香豆醇、松柏醇和芥子醇，主要存在于植物次生壁中，其含量仅次于纤维素。根据单体组成的不同，木质素可分为对-羟基苯基木质素 (p-hydroxyphenyl lignin, H 木质素)、愈创木基木质素 (guaiacyl lignin, G 木质素) 和紫丁香基木质素 (syringyl lignin, S 木质素)[1]。不同植物中木质素的组成不同，双子叶植物主要以 G 和 S 木质素为主；单子叶植物中，3 种类型木质素均有，但 H 木质素含量相对较少；而蕨类与裸子植物主要以 G 木质素为主[2-3]。木质素是一类复杂的酚类聚合物，木质素为植物细胞壁提供结构支持，能够增强细胞壁和茎秆的强度，对植物的生长发育具有重要作用[4-5]；同时木质素作为重要的物理抗菌物质，在植物体内构建了一道抵御有害生物的重要屏障，起到抑制病原菌扩散的作用[6-7]。

需要指出的是，3 种单木质素醇不同组合之间发生氧化二聚化生成木脂素，其种类多样，大多数以游离状存在，少数与糖结合，以苷的形式存在[8]。木脂素类化合物具有多种药理作用，如抗病毒、抗肿瘤、保肝、抗炎、抗菌等，包括板蓝根抗病毒活性成分落叶松脂素[9-11]，具有抗炎、抗氧化、保肝作用的五味子甲素[12-13]，抗肿瘤药物依托泊苷的先导化合物鬼臼毒素等[14-15]。此外，木脂素还参与植物对逆境胁迫的适应及病虫害防御调节[1]。

作为构成植物木质素和木脂素的基本单元，不同类型单木质素醇的组成直接决定木质素和木脂素的化学多样性和生物活性差异。咖啡酸-*O*-甲基转移酶 (caffeic acid *O*-methyltransferase, COMT) 可催化咖啡酸、5-羟基松柏醛和 5-羟基松柏醇甲基化分别生成阿魏酸、芥子醛和芥

子醇，参与了芥子醇 (S 单元) 和松柏醇 (G 单元) 合成的甲基化反应，对不同类型单木质素醇的组成起决定作用，是调控木质素和木脂素生物合成的核心关键酶[16]。本文对 COMT 的基因特征、表达特征和调控作用进行综述，并结合笔者团队的工作对 COMT 的研究和应用前景进行展望。

1 COMT 的基因特征

咖啡酸-*O*-甲基转移酶 (caffeic acid *O*-methyltransferase, COMT)，主要是以腺苷蛋氨酸 (S-adenosyl methionine, SAM) 为甲基供体，可催化多种底物的甲基化，归属于不需要 Mg^{2+}的植物Ⅱ型 *O*-甲基转移酶 (*O*-methyltransferase, OMT)，主要定位于细胞骨架和细胞质中[17-18]。

1.1 序列特征

目前已经有许多植物的 *COMT* 基因被报道，如表 1 中列举出几个具有明确功能的 *COMT* 基因。Southern 杂交表明，*COMT* 为多基因家族，其基因的长度范围为 1 000–2 000 bp，其酶的氨基酸数目大致在 300–400 之间，COMT 蛋白大小在 40 kDa 左右[19]。COMT 蛋白氨基酸序列从 N 端到 C 端，存在氧甲基转移酶共有的 5 个序列保守区，分别为：A: LVDGGGxG；B: GINFDLPHV；C: EHVGGDMF；D: NGKVI；E: GGKERT[20-21]。5 个保守区共包含 36 个氨基酸残基，其中靠近 C 端的 3 个保守区富含甘氨酸残基，保守区 A 和 D 被认为是 SAM 和金属离子 Mg^{2+}的结合部位[18,22]，这些区域在大多数植物的 COMT 中具有高度保守性[19] (图 1)。

对不同植物 COMT 同源分析发现，两种单子叶植物或两种双子叶植物之间具有很高的同源性，例如，板栗与垂枝桦 (*Betula pendula* Roth.) 和白桦的同源性均在 90%以上，且与桦木科植物物种亲缘关系较近[22]；甘蔗与玉米的同源性为 91.2%，而甘蔗与其他双子叶植物同源性则为 59.3%–65.5%[19,23]；葡萄 (*Vitis vinifera* L.) *Vv*COMT 与番茄和拟南芥 COMT 同源性都在 80%以上[24]。这些特征为植物中 *COMT* 基因的识别奠定了基础。从 NCBI 获取 48 个不同植物来源的 COMT 蛋白序列进行系统进化分析，发现总共分为两大分支 (Ⅰ和Ⅱ)，其中Ⅱ包含数量较多，继续分化为多个小分支。该进化树显示，大部分物种来源相同或相近的 COMT 被分为同一分支，提示它们可能具有相近的功能，例如：灯笼椒的两个 COMT (GenBank 登录号：AAG43822.1、AAC78475.1) 在分支Ⅰ中；来源于伞形科的峨参 (GenBank 登录号：BAO79380.1) 和大阿米芹 (GenBank 登录号：AAR24097.1) 同属于分支Ⅱ中的小分支②，两者同源性高达 88%；小麦 (GenBank 登录号：AAP23942.1)、黑麦草 (GenBank 登录号：AAD10253.1) 和高粱 (GenBank 登录号：AAL57301.1) 等禾本科植物的 COMT 均在分支Ⅱ的小分支②中 (图 2)。

表 1 不同植物来源 COMT 蛋白信息

Table 1 COMT from various plant sources

Sequence name	GenBank Accession No.	Plant source
*At*COMT	AAM10127.1	*Arabidopsis thaliana* (L.) Heynh.
*Nt*COMT	AAL91506.1	*Nicotiana tabacum* L.
*Os*COMT	ABB90678.1	*Oryza sativa* L.
*Lp*COMT	AAD10253.1	*Lolium perenne* L.
*Ii*COMT	AAZ95246.1	*Isatis indigotica* Fort.
*Hv*COMT	BAC54275.1	*Hordeum vulgare* L.
*Zm*COMT	AAB03364.1	*Zea mays* L.
*So*COMT	CAA13175.1	*Saccharum officinarum*
*Ms*COMT	ACY06328.1	*Medicago sativa* L.
*Bep*COMT	AGG91492.1	*Betula platyphylla* Suk.
*Cm*COMT	AOT85273.1	*Castanea mollissima* BL.
*Pt*COMT	AFZ78575.1	*Populus tomentosa* Carrière

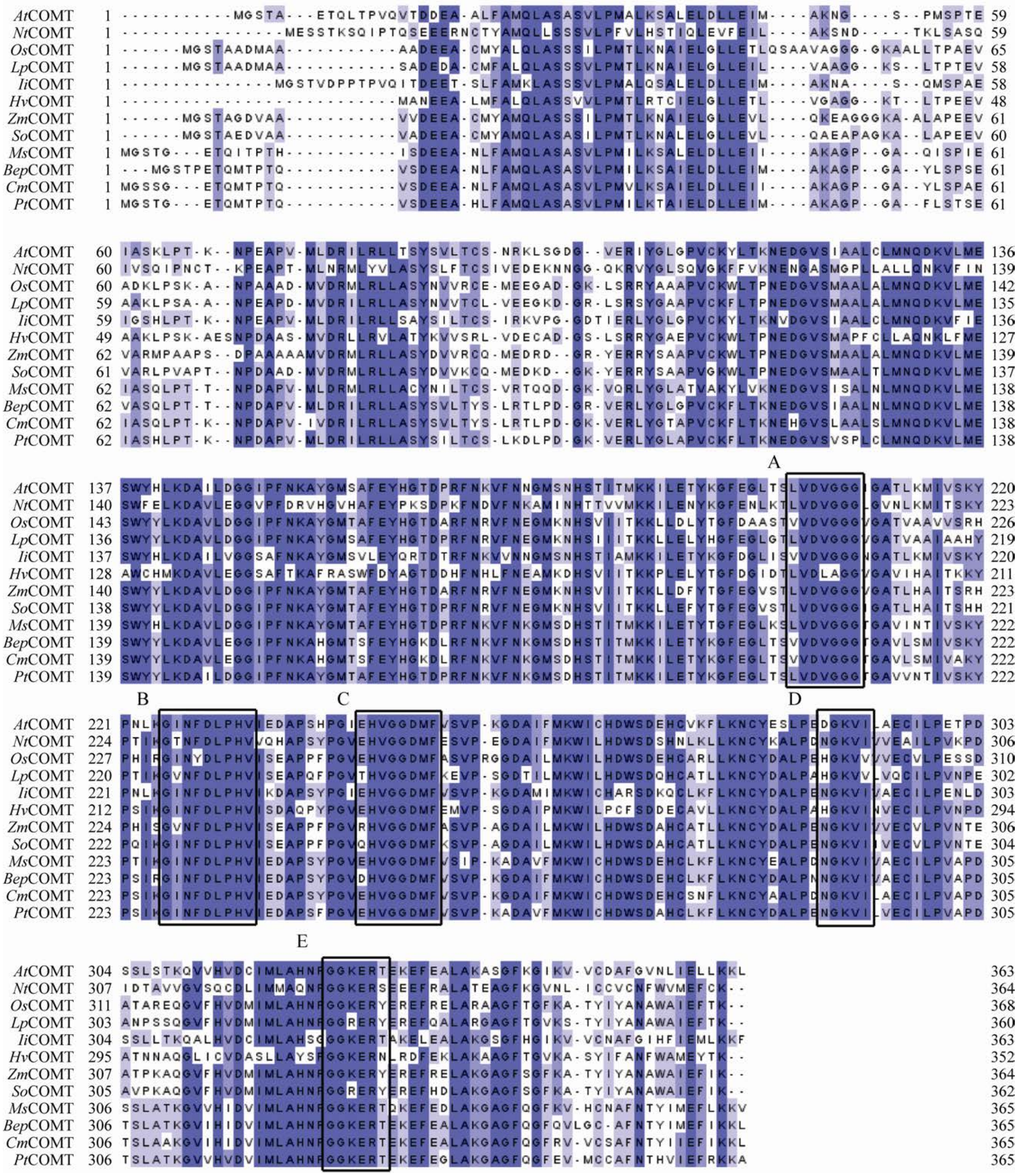

图 1　不同植物来源 COMT 氨基酸序列比对　A：LVDGGGxG；B：GINFDLPHV；C：EHVGGDMF；D：NGKVI；E：GGKERT

Figure 1　Alignment of COMT amino acid sequences from different plant sources. (A) LVDGGGxG. (B) GINFDLPHV. (C) EHVGGDMF. (D) NGKVI. (E) GGKERT.

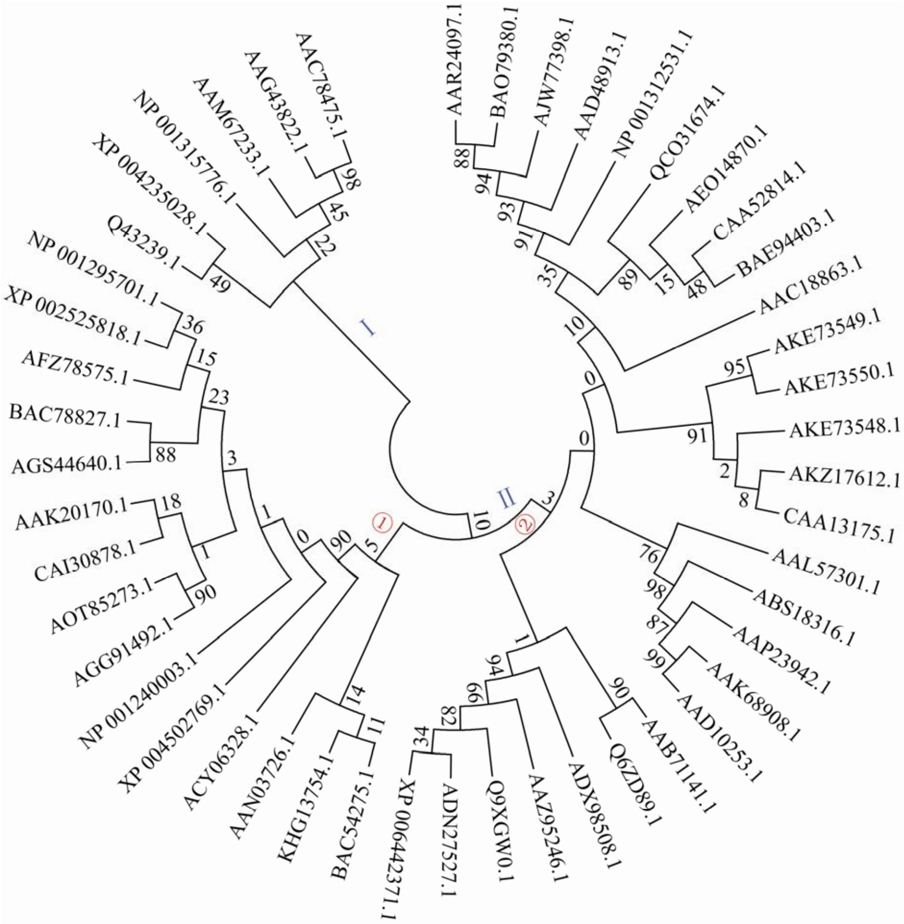

图 2　多种来源 COMT 的系统进化树

Figure 2　Phylogenetic tree of COMT from various sources. AAR24097.1: *Ammi majus* L.; BAO79380.1: *Anthriscus sylvestris* (L.) Hoffm. Gen.; AJW77398.1: *Angelica sinensis* (Oliv.) Diels; AAD48913.1: *Liquidambar styraciflua* L.; NP_001312531.1: *Nicotiana tabacum* L.; QCO31674.1: *Peucedanum praeruptorum* Dunn; AEO14870.1: *Salvia miltiorrhiza* Bge.; CAA52814.1: *Eucalyptus robusta* Smith; BAE94403.1: *Ipomoea nil* (L.) Roth; AAC18863.1: *Mesembryanthemum crystallinum* L.; AKE73549.1: *Miscanthus sinensis* Anderss.; AKE73550.1: *Miscanthus floridulus* (Lab.) Warb. ex Schum. et Laut.; AKE73548.1: *Triarrhena sacchariflora* (Maxim.) Nakai; AKZ17612.1: *Ligusticum chuanxiong* Hort.; CAA13175.1: *Saccharum officinarum* L.; AAL57301.1: *Sorghum bicolor* (L.) Moench; ABS18316.1: *Bambusa oldhamii* Munro; AAP23942.1: *Triticum aestivum* L.; AAK68908.1: *Lolium arundinaceum* (Schreb.) DARBYSH.; AAD10253.1: *Lolium perenne* L.; AAB71141.1: *Clarkia elegans* Douglas; Q6ZD89.1: *Oryza sativa* L.; ADX98508.1: *Panicum virgatum* L.; AAZ95246.1: *Isatis indigotica* Fortune.; Q9XGW0.1: *Ocimum basilicum* L.; ADN27527.1: *Camellia sinensis var. Assamica* (Mast.) Kitam; XP_006442371.1: *Citrus maxima* (Burm) Merr.; BAC54275.1: *Hordeum vulgare* L.; KHG13754.1: *Gossypium arboreum*; AAN03726.1: *Coffea canephora* Pierre ex Froehn.; ACY06328.1: *Medicago sativa* L.; XP_004502769.1: *Cicer arietinum* Linn.; NP_001240003.1: *Glycine max* (Linn.) Merr.; AGG91492.1: *Betula platyphylla* Suk.; AOT85273.1: *Castanea mollissima* Bl.; CAI30878.1: *Picea abies* (L.) Karst.; AAK20170.1: *Catharanthus roseus* (L.) G. Don; AGS44640.1: *Pyrus bretschneideri* Rehd.; BAC78827.1: *Rosa chinensis* Jacq.; AFZ78575.1: *Populus tomentosa* Carrière; XP_002525818.1: *Ricinus communis* L.; NP_001295701.1: *Jatropha curcas* L.; Q43239.1: *Zinnia elegans* Jacq.; XP_004235028.1: *Lycopersicon esculentum* Miller; NP_001315776.1: *Malus pumila* Mill.; AAM67233.1: *Arabidopsis thaliana* (L.) Heynh.; AAG43822.1, AAC78475.1: *Capsicum chinense* L..

1.2 催化功能与蛋白结构

COMT 是植物苯丙素生物合成途径的关键酶，其底物范围广泛，催化苯丙素类化合物羟基上氧原子的甲基化，不仅是甲基化单体酚的3′位置，同时还协助 CCoAOMT 甲基化单体酚的 5′位置[25]。Nakatsubo 等[26]对拟南芥中注释出的 *COMT* 基因 (*At5g54160*) 进行了功能研究，发现其能够催化多种苯丙素类化合物和黄酮类化合物，包括咖啡酸、咖啡醇、5-羟基阿魏酸、5-羟基松柏醛、5-羟基松柏醇、槲皮素和木犀草素等。近几年对 COMT 的深入研究发现，除了催化苯丙素和类黄酮类化合物的甲基化，某些 COMT 还具有类似褪黑素生物合成过程中 *N*-乙酰-5-羟色胺甲基转移酶的作用，不仅能够催化 *N*-乙酰-5-羟色胺生成褪黑素，还能够催化 5-羟色胺生成 5-氧甲基色胺[27-29] (图 3)。

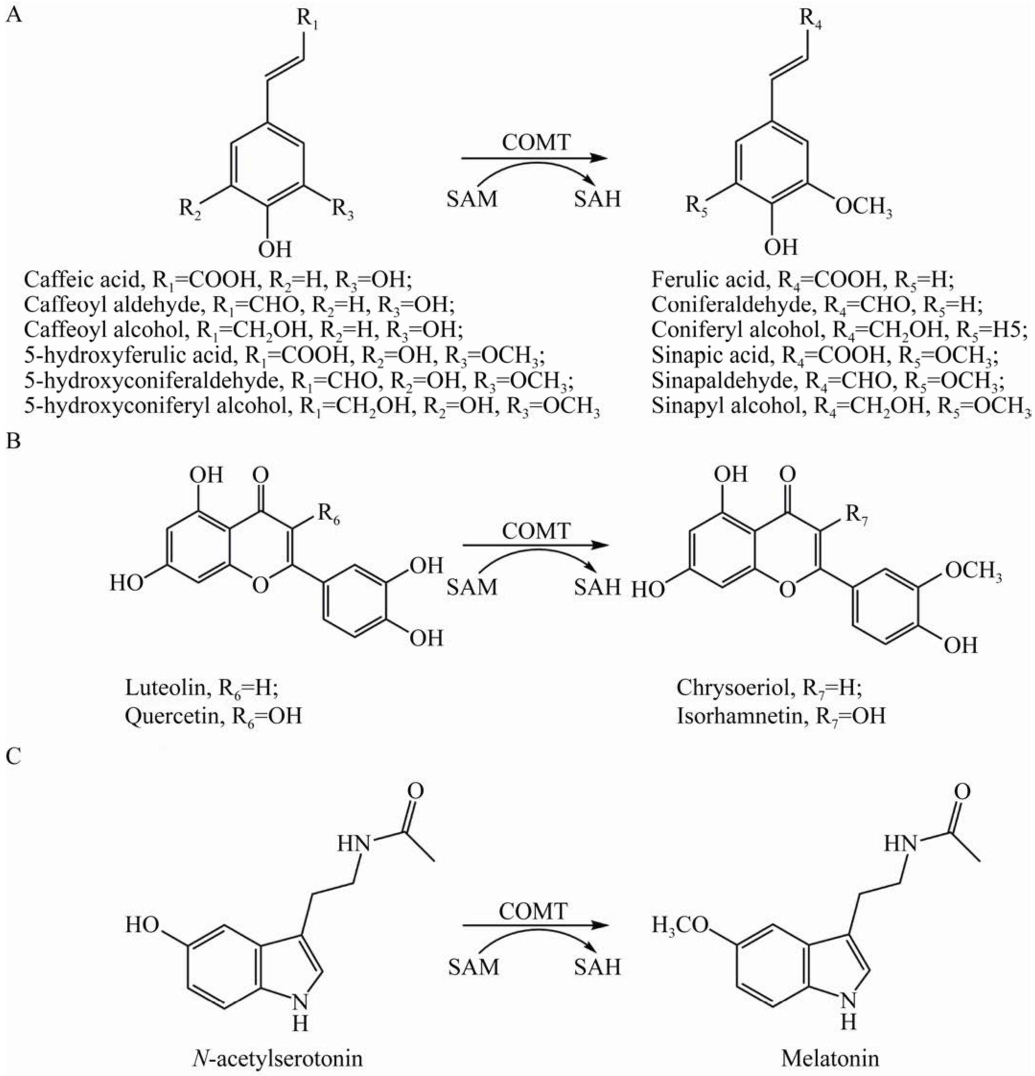

图 3 COMT 催化不同底物的甲基化反应 A：催化苯丙素类化合物；B：催化黄酮类化合物；C：催化乙酰-5-羟色胺

Figure 3 COMT catalyzes the methylation reaction of different substrates. (A) Catalysis of phenylpropanoid compounds. (B) Catalysis of flavonoids. (C) Catalysis of *N*-acetyl-5-hydroxytryptamine.

COMT 属于 SAM 依赖性植物Ⅰ型家族氧甲基转移酶，这类酶的长度通常约为 360 个氨基酸残基，主要由 α-螺旋和 β-折叠构成，具有 3 个重要的结构域，分别是咖啡酸结合域、S-腺苷甲硫氨酸结合域以及在同源二聚化中起作用的辅助 N 端结构域[30-34]。2010 年，Louie 等[35]对黑麦草的 COMT (*Lp*OMT1) 的蛋白晶体结构进行了解析，其属于典型的植物Ⅱ型 OMTs，多肽链折叠形成独特的二聚体和 SAM/SAH 结合域，在 2 个结构域之间夹有一个 α 螺旋层。两个紧密缠绕的对称同源二聚体结构域位于中央核心位置，SAM 结构域位于外围，两个单体的活性位点裂缝位于 SAM 双链结合域和相邻 α 螺旋层之间的界面上，是酚基结合位点，处于两个 α 螺旋构成的口袋中。通过分析 *Lp*OMT1 在不同构象 (开放和封闭) 以及不同形式 (脱辅基酶、全酶和与产物结合的三元复合物) 下的蛋白晶体结构，解析了 *Lp*OMT1 对醛类底物的偏好性的结构基础，产物结合复合物揭示了 COMT 催化基团与反应物分子的甲基转移后形成的酚配体结合位点。

作为一个底物杂泛性较高的酶，COMT 底物选择性的机制研究受到了极大的关注。来自紫花苜蓿 (*Medicago sativa* L.) 的 COMT (*Ms*COMT) 能够甲基化含有咖啡酰和 5-羟基阿魏酸的酸、醛和醇，且对醇和醛的催化活性显著强于游离酸。Zubieta 等[30]对 *Ms*COMT 与 S-腺苷-L-高半胱氨酸 (SAH)、阿魏酸以及 5-羟基松柏醛复合物的晶体结构进行解析，识别出了蛋白底物接触面上的活性位点残基，提示可以通过氨基酸定点诱变来改变底物的偏好性。对 *Ms*COMT 的 Leu136、Ala162、Met130 和 Asn131 等一系列关键氨基酸位点进行突变，发现其对醛和醇的结合能力显著加强，验证了其底物选择性的结构基础。Wang 等[36]发现，拟南芥 COMT (*At*COMT) 对底物咖啡酸和非天然底物 *N*-乙酰-5-羟色胺 (*N*-acetylserotonin, NAS) 催化活性具有显著差异，通过底物与酶之间的分子对接推测差异可能源于 NAS 较长的“尾巴”限制了其与 *At*COMT 的结合，以增强底物亲和力为目标进行蛋白理性突变，三重突变体 (C296F-Q310L-V314T) 将 *At*COMT 催化 NAS 生成褪黑素的活性提高了 9.5 倍。

通过 COMT 蛋白结构理性设计，还可以实现其对底物氧甲基化位置的改变。Zhang 等对 *At*COMT 的 4 个氨基酸残基进行突变，使其能特异催化单木质素醇的 4 位氧甲基化 (催化效率提高了 200 倍)，阻碍了后续木质素单体的聚合反应，将其转入拟南芥后，显著降低了木质素的含量[37]，将该结构改造后的 COMT 转入杨树，显著提高了杨树细胞壁转化为乙醇的效率[38]，该研究在培育可高效利用的生物质原料植物方面将有潜在的应用前景。

2 COMT 的表达特征

2.1 组织表达

COMT 基因在植物不同组织部位的表达不同，直接影响不同部位木质素构成的差异。例如：兴安落叶松 (*Larix gmelinii* (*Rupr*.) Kuzen.) 的 *COMT* 基因在根、茎、针叶中均有表达，但在茎中的表达量最高，这与茎中需要合成更多的木质素来维持机械化强度相对应[39]；砀山酥梨 (*Pyrus bretschneideri cv*. Dangshan Su) 的 *COMT* 基因，开花后 47 d 在果中部表达量最高，63 d 在近果核部表达量最高，且基因总表达量在开花后呈现先升高后降低的趋势，并在 63 d 达到最大值，这与其石细胞的发育规律有很大的相关性[40]；丹参 *COMT* 基因在茎中表达最丰富，其次是叶中，而根中表达量最低，这与其机械支持和保护功能相符[41]。

2.2 诱导表达

胁迫处理对植物中 *COMT* 的表达具有显著影响。UV-B 处理后，当归 (*Angelica sinensis* (Oliv.) Diels) 的根、叶柄、叶中 *COMT* (*AsCOMT*) 的表达水平显著提高[42]；聚乙二醇 6000 (PEG) 模拟干旱胁迫能提高番茄 (*Lycopersicon esculentum* Miller) *COMT1* (*SiCOMT1*) 表达量[43]；割胶处理后，橡胶树 (*Hevea brasiliensis* (*Willd. ex A. Juss*.) Muell. Arg.) 中的 *COMT* (*HbCOMT1*) 的表达水平快速上升[44]，这些研究结果均提示 *COMT* 可能参与了植物胁迫响应。

3 COMT 的调控作用

3.1 调控木质素的组成

COMT 作为木质素单体生物合成的关键酶，已成为通过植物次生代谢工程调控木质素含量和分布的重要靶点。运用正义或反义抑制、基因沉默、基因下调或者基因突变的手段调控 *COMT* 基因的表达，这些表达变化对大部分植物中木质素的含量和组成均有较大影响。例如，构建玉米突变体 (bm3) 抑制 *COMT* 表达，玉米中总木质素含量减少、S-木质素含量减少、S/G 木质素比例下降，并且玉米出现典型的棕色中脉叶表型[45]；高粱 (*Sorghum bicolor* (*L*.) Moench) *comt* 突变体 (bm12) 也出现跟玉米类似的现象，且高粱突变体的根系结构以及对水分限制的响应也发生了改变[46]；在烟草、大麦、杨树中，抑制 *COMT* 的表达，不仅总木质素含量和 S/G 比例均下降，还掺入了 5-羟基-G 木质素 (S 木质素的前体)[47-49]。但在一些植物中，如白桦 (*Betula platyphylla* Suk.)、拟南芥、颤杨 (*Quaking aspen*) 中，*COMT* 抑制表达对木质素总量影响不大，主要是 S/G 木质素比例下降[50-52]。表 2 列举了几种不同植物 *COMT* 被抑制后木质素成分的变化情况及其对植物的影响。

3.2 提高植物抗性

褪黑素参与植物的生长发育过程并且能够对各种非生物的胁迫做出响应，通过表达 *COMT* 来增加植物内褪黑素的含量，有助于提高植物抗性，对植物生长发育意义重大。Yang 等[18]过表达小麦中的 *COMT* 基因 (*TaCOMT*) 后，转基因植物中褪黑素含量和抗性均显著高于野生型。Zhang 等[53]过表达白颖薹草 (*Carex duriuscula* C. A. Mey. subsp. *rigescens* (Franch.) S. Y. Liang et Y. C. Tang) 中的 *CrCOMT* 基因，转基因植物在盐胁迫下表现出更强的生长和生理性能，如侧根数量、脯氨酸水平和叶绿素含量等均显著提高。孙莎莎等[43]过表达 *SlCOMT1* 提高了番茄内源褪黑素的含量和耐盐性，同时提高了干旱胁迫下番茄植株的光合作用和抗氧化能力，诱导了胁迫相关信号基因的表达，提高了番茄抗旱能力。

4 总结与展望

COMT 是植物苯丙素途径的关键酶，对不同类型单木质素醇 (H 型、G 型和 S 型) 的构成起重要调控作用，从而影响植物生长发育及抗逆活性。目前，通过改变 *COMT* 表达水平已经在烟草、杨树、苜蓿等植物中成功地改变了木质素含量和组分，对生产中降低造纸成本、减轻污染和改良饲草品质等方面均展现了潜在应用前景。另一方面，COMT 也是木脂素生物合成的关键酶，例如其催化咖啡醇发生氧甲基化反应生成松柏醇，松柏醇发生二聚化生成重要木脂素类成分松脂醇，松脂醇的一系列衍生物具有多种药理作用，是许多药用植物发挥药效的重要物质基础。

表 **2** ***COMT*** 基因表达对植物中木质素的影响

Table 2 Effect of *COMT* gene expression on lignin in plants

Species	Method	Changes of content	Changes of composition	Plant phenotype	References
Panicum virgatum L.	Down-regulation/RNAi	Decrease	Decreased S-lignin; increased G-lignin; incorporated benzodioxane structure and 5-hydroxyguaiacyl unit	–	[54-55]
Zea mays L.	Mutant(bm3)/ antisense suppression	Decrease	Decreased S-lignin; decreased S/G lignin	Typical brown midrib leaf phenotype	[45,56]
Medicago sativa L.	Antisense suppression	Decrease	Decreased G-lignin and S-lignin	–	[57-58]
Hordeum vulgare L.	RNAi	Decrease	Reduced S/G lignin by about 50%, and increased 5-hydroxyguaiacyl units by 10–15 times higher than the normal level	–	[49]
Nicotiana tabacum L.	Antisense suppression	Decrease	Decreased S/G lignin，and increased accumulation of 5-hydroxyguaiacyl units	Light red color of the peeled stems	[48,59]
Saccharum officinarum	RNAi/multiple allelic mutations	Decrease	Decreased S/G lignin	Improved saccharification efficiency	[60-62]
Triticum aestivum L.	Antisense suppression	Slightly down	Sharply decreased S-lignin	Improved disease resistance and the mechanical strength of stalks	[63]
Miscanthus sinensis Anderss.	Down-regulation	Decrease	–	–	[64]
Brachypodium sylvaticum (Huds.) Beauv.	Gene silence	Unchanged	Decreased S-lignin and S/G lignin	Changed flowering time; increased number of stems and stem weight; improved saccharification efficiency	[65-66]
Populus tremula L×*Populus alba* L.	Gene silence	Decrease	Increased G-lignin; Decreased S/G lignin; Incorporated 5-hydroxyguaiacyl units	–	[47,67]
Lolium perenne L.	RNAi	Decrease	Decreased S/G lignin	–	[68]
Festuca elata Keng ex E. Alexeev	Cosuppression	Decrease	Decreased S/G lignin	–	[69]
Brassica napus L.	RNAi	Decrease	–	–	[70]
Sorghum bicolor (L.) Moench	Mutant(bm12)	Decrease	Decreased S/G lignin	Typical brown midrib leaf phenotype; changed root anatomy and response to water restriction	[46,71]
Quaking aspen	Down-regulation	Unchanged	Decreased S/G lignin	–	[50]
Arabidopsis thaliana (L.) Heynh.	Mutant	Decrease	Decreased S-lignin; increased G-lignin and 5-hydroxyguaiacyl units	–	[25,51,72]
Betula platyphylla Suk.	RNAi	Unchanged	Decreased S/G lignin	–	[52]
Eucalyptus urophylla S.T. Blake	Sense suppression	Increase	Decreased S/G lignin	–	[73]

对于 COMT 的研究，目前有几个方面相对缺乏：1) COMT 作为木脂素前体化合物松柏醇合成的关键酶之一，与木脂素之间的调控关系报道较少；2) COMT、木质素和木脂素三者之间的动态平衡规律尚缺乏系统的认识；3) COMT 作为一种杂泛性较广的酶，如何提高 COMT 的催化专一性，减少副产物，提高特定化合物的合成和积累；4) COMT 缺失或抑制表达对于植物生长发育影响的分子机制不明。本课题组前期研究提示菘蓝 (中药板蓝根的基源植物) COMT 的表达水平与板蓝根抗病毒活性木脂素的积累水平相互关联[74]，但目前通过调控 COMT 表达水平改变植物中特异木脂素类成分的积累的研究还相对薄弱，尚需进一步实验验证，笔者认为探究 COMT 对活性木脂素类成分的调控作用以提高其含量积累将成为重要研究方向。同时构建 COMT、木质素和木脂素之间的相互关联，实现木质素和木脂素的最佳动态平衡也应给予重点关注。

另外，植物 COMT 底物杂泛性较广，既能催化苯丙素类化合物，又能催化黄酮、*N*-乙酰-5-羟色胺等，其作为催化元件应用于合成生物学生产特定活性苯丙素类成分时存在代谢流分散、副产物较多等问题。尽管已经有报道通过 COMT 蛋白定点突变等提高底物专属性的相关报道，但是其底物特异性的分子机制尚未完全阐明，如何控制 COMT 驱动的目标成分的定向合成仍是目前研究的难点。本文建议通过构建催化性质差异的 COMT 差异蛋白库，借助结构生物学等技术手段全面解析其底物选择性的结构基础，并通过理性设计进行蛋白改造，从根本上提升底物专属性，为实现特定活性木脂素的定向生物合成提供可行方案。

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