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计算毒理学在内分泌干扰物筛选上的应用和展望

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摘要: 内分泌干扰物通过干扰内分泌系统导致多种疾病,如生殖疾病、肥胖症甚至癌症。然而,面对环境中大量潜在的内分泌干扰物,传统的体外、体内评估方法由于成本高、耗时长等问题,难以实现内分泌干扰物的高通量筛查。计算毒理学逐渐发展成为被美国环保局(Environmental Protection Agency, EPA)、经济合作与发展组织(Organization for Economic Co-operation and Development, OECD)等机构所推荐的内分泌干扰物筛选与预测方法。本文综述了计算毒理学在内分泌干扰物筛选上的进展,主要包括分子对接和分子动力学模拟的应用,并对有害结局路径(adverse outcome pathway, AOP)的方法进行介绍和展望。

关键词: 计算毒理学;内分泌干扰;分子对接;分子动力学模拟;AOP
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Application and Prospect of Computational Toxicology in Screening of Endocrine Disrupting Chemicals

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Abstract: Endocrine disrupting chemicals (EDCs) cause a variety of diseases, such as reproductive diseases, obesity and even cancer, by interfering with the endocrine system. However, in the face of a large number of potential endocrine disruptors in the environment, traditional *in vitro* and *in vivo* assays are difficult to achieve high throughput screening of endocrine disruptors due to their high cost and time consuming. Computational toxicology has been recommended as the screening and predicting method by the US Environmental Protection Agency (EPA), the Organization for Economic Co-operation and Development (OECD) and so on. Here, we discuss the application of computational toxicology methods, particularly molecular docking, molecular dynamics simulations and the developing adverse outcome pathway (AOP), in guiding the screening of EDCs.

Keywords: computational toxicology; endocrine disrupting; molecular docking; molecular dynamics; AOP

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1　前言(Introduction)

1.1　内分泌干扰效应与内分泌干扰物

内分泌系统指由一系列腺体分泌激素进入内循环系统运输并直接作用于目标器官的系统。内分泌系统的信号由激素传递,激素有不同的化学结构,主要包括 3 种:类花生酸类(eicosanoids)、甾体类(steroids)和氨基酸衍生物(胺类、肽链和蛋白质)。激素通过与目标细胞的特定受体蛋白结合,激活信号转导通路,达到调节细胞功能的作用,调节着生物体几乎所有生物学过程。已有报道表明,一些化学物质如双酚 A(bisphenol A, BPA)、多环芳烃(polycyclic aromatic hydrocarbons, PAHs)及一系列杀虫剂等的暴露会干扰内分泌系统并产生有害影响,这类物质被称为内分泌干扰物(endocrine disrupting chemicals, EDCs)。内分泌干扰物的暴露会增加生殖疾病、心肺疾病、免疫系统疾病和神经系统疾病的风险,甚至导致肿瘤和癌症的发生[1]。据 Attina 等[2]的模型估计,2010 年欧盟国家内分泌干扰物导致的疾病花费占国内生产总值(GDP)的 1.28%,为 2 170 亿美元,而美国的达到 3 400 亿美元,占 GDP 的 2.33%,内分泌干扰物筛选的研究迫在眉睫。

1.2　内分泌干扰物的实验检测手段

检测内分泌干扰物的实验手段包括体外(*in vitro*)和体内(*in vivo*)实验。体外实验包括细胞增殖实验(cell proliferation assays)[3]、报告基因实验(reporter gene assays)[4]、酵母双杂交实验(yeast two-hybrid assays)[5]、结合实验(binding assays)[6]等。体内实验多采用哺乳动物[7-8]、鸟类[9-10]、鱼类[11-12]、两栖类[13-14]等动物。过去十几年来,体外实验被合理开发应用到各种高通量测试筛选方法中,以应对数量巨大的潜在危害化合物,以及动物体内测试的巨大开销和伦理问题[15]。由美国环保局(Environmental Protection Agency, EPA)、国立卫生研究院(National Institutes of Health, NIH)和食品与药品管理局(Food and Drug Administration, FDA)等跨部门合作的 21 世纪毒理学(Tox21)项目,采用高通量筛选技术测试约 10 000 种环境化合物和药物的毒性,其中内分泌干扰是重要方面,关于雌激素干扰效应及信号通路的研究结果已于 2014 年发布[16]。

现阶段评估一个化合物的内分泌干扰作用往往需要体内和体外实验的结合。美国 EPA 内分泌干扰物筛选项目(Endocrine Disruptor Screening Program, EDSP)于 2012 年开展了 EDSP21 项目,采用 2 个级别的筛选测试手段(Tier 1 和 Tier 2)评价化合物的内分泌干扰活性。其中级别 1 测试包括 5 种体外实验和 5 种体内实验,级别 2 则是更深层次或多代体内测试。经过级别 1 测试具有干扰活性的化合物进入级别 2 测试,评估其内分泌干扰效应。目前,超过 1 800 种化合物内分泌相关活性的高通量筛选数据(主要为包括雌激素受体 ER、雄激素受体 AR 结合和转录激活的级别 1 体外筛查数据)已可在 EDSP21 Dashboard 网站(http://actor. epa. gov/edsp21/)获取。

1.3　内分泌干扰物的模拟预测手段

体外和体内实验手段虽然能完整评价化合物的内分泌干扰效应,但是其成本高、耗时长,难以对全球现有超过 126 000 000 种化合物参考(http://www.cas.org)进行逐一筛选。因此,亟需发展化学品内分泌干扰效应筛选的计算毒理学(computational toxicology)方法[17]。计算毒理学方法指通过综合体内、体外实验和计算机模拟等不同来源的数据,开发数学或计算机模型,以更好理解或预测化合物干扰效应的方法[18]。近年来,计算毒理学得到越来越多的关注,美国 EPA 于 2005 年成立了国家计算毒理学中心(National Center for Computational Toxicology, NCCT),致力于开发新的评估化合物安全性的方法,即计算毒理方法;经济合作与发展组织(Organization for Economic Co-operation and Development, OECD)于 2008 年开发的计算毒理软件 OECD QSAR Toolbox 如今也进入 3.4 版本,并得到各国政府、化学工业的接受和使用。

定量结构-效应关系(quantitative structure-activity relationship, QSAR)是最早开发和发展的计算毒理学方法,将代表化合物结构、物理、化学性质的分子描述符(molecular descriptors)与特定效应终点或有害结局(adverse outcome, AO)建立联系,达到预测目的,在不同尺度的内分泌干扰效应,如核受体结合[19-20]、转录激活[21-22]、器官和个体有害结局[23-24]等,都得到广泛运用。我国陈景文教授、张爱茜教授、高士祥教授、王连生教授和于红霞教授等的团队在内分泌干扰物的 QSAR 研究上都做了大量工作[25-29],比如 Li 等[25]计算了 517 种有机化合物的 705 个分子描述符,并选取其中的 13 个分子描述符建立雌激素效应的 QSAR 模型,发现有机分子的雌激素活性主要与分子尺寸、形状特征、电负性和范德华体积等相关。如今,QSAR 已经发展成较为成熟的计算毒

理学方法,得到 OECD 等组织的认可,基于 QSAR 开发的毒性预测软件,如 TEST、ECOSAR、OncoLogic 等,也得到广泛应用。然而,QSAR 往往忽略干扰物的效应机制,采用的分子描述符往往也没有直接或明确的药理学或生物学意义[30]。

激素分子与体内调节相关生理功能的大分子,如受体蛋白等之间的相互关系在内分泌系统信号传递中具有重要作用。因此对干扰物与受体作用关系的研究是内分泌干扰物筛选的重要研究手段,很多体外实验都是以干扰物与受体作用关系为对象研究化合物的内分泌干扰效应的[31-32],而分子对接(molecular docking)和分子动力学(molecular dynamics, MD)模拟方法作为基于干扰物与受体作用关系的计算毒理学研究方法也得到越来越多的应用。在此基础上,为了更加深入地理解内分泌干扰效应作用机制,OECD、美国 EPA 等组织开展了开发有害结局路径(adverse outcome pathway, AOP)的项目,将极大促进基于效应机制的计算毒理学的发展。因此,本文将对分子对接、MD 模拟和 AOP 等基于效应机制的计算毒理学方法在内分泌干扰物筛选上的应用进行综述。

2 分子对接及其应用(Molecular docking and its applications)

分子对接是预测配体与受体结合成稳定复合体时配体所处的最佳位置和方向的方法[33]。内分泌系统中主要的受体蛋白(图 1)是包括雌激素受体(estrogen receptor, ER)、雄激素受体(androgen receptor, AR)、甲状腺激素受体(thyroid hormone receptor, TR)、糖皮质激素受体(glucocorticoid receptor, GR)等在内的核受体(nuclear receptor, NR),它们都受激素调节并控制大量基因的表达[34]。随着晶体学和生物化学技术的发展,越来越多核受体的晶体结构被解析出来(图 1)[35-39],这些晶体结构都可以通过 Protein Data Bank 网站(http://www.rcsb.org/pdb/home/home.do)获得,使采用对接和 MD 模拟等方法筛选内分泌干扰物成为可能[40]。但已有的多为人类的受体,对于其他物种,往往需要通过同源建模(homology modeling)构建受体结构[41]。

分子对接的使用有助于加深对配体受体相互作用机制的理解。Nose 等[42]用对接的方法从 14 个酚类物质中筛选出 4-(1-adamantyl)phenol 为拟雌激素物质,经验证确实具有很强的雌激素活性。对对接结果的分析发现,与雌激素类似,4-(1-adamantyl)phenol 的羟基也能与 ERα 中 Glu353 和 Arg394 氨基酸形成氢键。D' Ursi 等[43]采用柔性对接的方法探索了内分泌干扰物与 ER、孕酮受体(progesterone receptor, PR)和 AR 的相互作用,发现这些内分泌干扰物与受体的相互作用主要取决于化合物与配体结合腔(ligand binding cavity, LBC)中多个氨基酸残基之间的疏水性作用,对于亲脂性内分泌干扰物,它们有能力适应甾体受体的疏水性 LBC,并呈现非特异

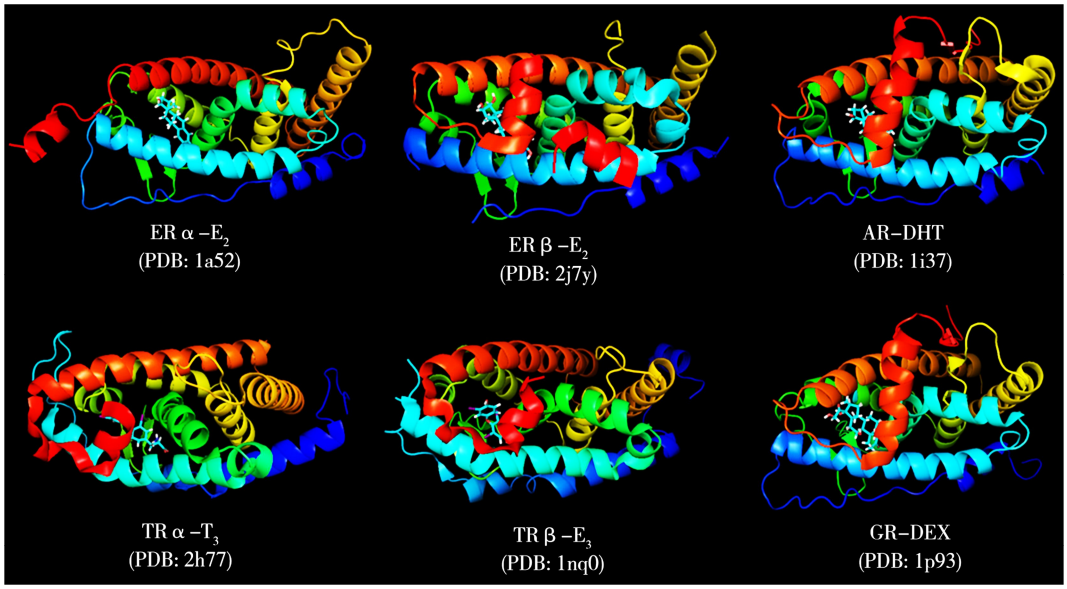

图 1 部分已解析的核受体结构[35-39]

Fig. 1 Some of the structures of nuclear receptors (NRs) that have been refined[35-39]

性结合模式。

由于内分泌干扰物往往能同时作用于不同受体,研究干扰物与不同受体的相互作用能预测潜在的效应终点。Yuriev 等[45]针对人体 14 种 NR,选取了 18 个完整、可靠的晶体结构建立对接模型,通过对接得到化合物与各个 NR 的结合能,判断对哪些受体更敏感,从而判断化合物潜在的内分泌干扰效应[44],这种方法被称为反向对接法。于红霞教授团队最近也采用反向对接开发了一个针对 39 个人类 NR 的程序 SPEN,该程序经过 10 种化合物的验证具有良好的表现[46]。

分子对接可以与 QSAR 结合,构建多维 QSAR 模型。Vedani 等[47]采用柔性对接和 QSAR 结合的方法建立了预测 ER 结合能力的 6 维 QSAR 模型,并用于对 106 种内分泌干扰物的筛选,结果 r^2 达到 0.885,表明具有很好的筛选能力。Vedani 团队将这种方法拓展到 AR、TR、GR 等 11 个核受体,并建立了基于网站的预测平台 VirtualToxLab[48]。分子对接在药物开发领域也产生了一些新方法,如基于分子碎片的对接技术正在快速发展,虽然在内分泌干扰物筛选上还没有得到应用,但是不失为提高对接结果精确度的选择[49]。然而分子对接在很大程度上仍受受体柔性的制约,而对受体赋予过多的柔性会导致对计算的要求呈指数增加并且变得不切实际[50]。

在实际应用中分子对接受受体结构和配体种类的影响较大,针对相同种类、数量较少的化合物时,分子对接能得到较好的预测效果,对接的结合能可以很好预测化合物与受体的结合效力[51];而针对大量具有不同结构的化合物时,预测效果一般,甚至比普通的 QSAR 模型差[52]。因此,选择合适、可靠的受体模型是分子对接模型构建的重点。针对不同种类的化合物提供不同对接模型是可行的解决方法;也可以借助分子动力学模拟,产生多个受体模型进行对接[53]。

3　分子动力学模拟及其应用(Molecular dynamics simulations and the applications)

随着计算机技术的发展和计算能力的提升,分子动力学模拟逐渐成为研究生物大分子作用的标准方法[54]。分子动力学模拟是研究原子和分子物理运动的计算机模拟方法,所有分子和原子在给定的时间范围内相互作用,形成一个动态变化的系统,以此研究生物分子之间的相互作用[55]。

MD 模拟有助于探索干扰物作用下受体蛋白及配体本身的构象变化。Li 等[56]用 MD 模拟研究了内分泌干扰物的雌激素干扰效应,发现干扰物与 ER 在 2 ns 的模拟下都能达到稳定状态,并且干扰效力更强的化合物能与 ER 的 His524 氨基酸稳定形成氢键。采用 MD 模拟还能发现蛋白质的关键结构及活性产生的关键变化[57]。Wang 等[58]通过对 AR 骨架变构情况的比较发现 12 号螺旋(Helix 12, H12)在 MD 模拟过程中具有最显著的位置变化,认为 H12 的位置变化是抗雄激素活性产生的关键。Wang 等[58]还发现 H12 在 10 ns 模拟时间内达到平衡是抗雄性活性产生的重要特征,且稳定时间与活性强弱呈负相关。

干扰物与受体蛋白 LBC 的结合情况仍然是 MD 研究的重点。Martínez 等[59]通过 MD 模拟发现了配体逃离 TR-LBC 的 3 种可能途径。有学者进一步用操纵分子动力学(steered molecular dynamics, SMD)模拟探究配体从各逃离途径逃离配体结合腔的难易程度[60-62]。Martínez 等[61]发现配体逃离 TR 的最佳途径是位于 H1、H2 和 H3 处的通道 3,而且当配体亲水部分能与受体外部的水分子接触时逃离过程会变得更轻松。另一方面,有研究表明,干扰物的诱导能使受体 H12 的位置发生变化,而其稳定位置正好挡住配体,使配体无法从 LBC 中逃离,受体形成的这种结构称为“老鼠夹(mousetrap)”结构[56, 63]。

热力学计算也是 MD 模拟的常用分析方法。采用 MM/PBSA 或 MM/GBSA(molecular mechanics with Poisson-Boltzmann or generalized Born and surface area)方法计算配体-受体结合自由能 $\Delta G_{binding}$ 可用于预测配体与受体间的结合效力。van Lipzig 等[64]将计算得到的雌激素干扰物与 ER 的结合自由能和实验测得的结合效力比较,发现两者的相关系数达到 0.94。结合自由能还能区分干扰物对受体不同亚型的选择性[65-66]。Martínez 等[65]分别计算了配体 Triac 与 TRα 和 TRβ 相互作用的结合自由能,发现 Triac 与 TRα 的结合自由能显著低于 TRβ,导致其对 TRα 具有高度选择性。

除了干扰物与核受体的结合,核受体与其他蛋白质的相互作用,如与共调节因子作用、二聚现象等[67-69],也是影响内分泌干扰效应产生的重要过程。研究表明 ER 的二聚作用大大抑制了 E_2 逃离 LBC[69]。于红霞教授团队[67]最近的 MD 研究也表明,共调节因子在化合物甲状腺激素干扰活性产生过程具有重要作用,抗甲状腺激素干扰物与 TR 结合能促进共抑制因子而不是共激活因子与 TR 结

合,从而导致抗性的产生。因此,考虑蛋白质受体与其他调剂因子的作用过程,对生物大分子间作用,如共调节因子结合、二聚作用和与 DNA 的结合等进行模拟,是 MD 模拟在内分泌干扰物筛选上的重要发展方向。另外,在 MD 模拟中采用量子力学/分子力学(QM/MM)耦合的方法,将配体部分用 QM 计算,其他部分用 MM 模拟的方法,有助于提高模拟的精确度,并有利于更深入探索配体受体之间的相互作用。陈景文教授团队[70]采用 QM/MM 的方法,探索了电中性和阴离子形态下酚类内分泌干扰物与甲状腺素运载蛋白(transthyretin, TTR)的结合,发现阴离子形态比电中性的酚类物质与 TTR 结合更强,认为离子形态的考虑是内分泌干扰物虚拟筛选过程不可忽视的机制。

4 AOP 的发展和展望(Development and prospect of AOP)

随着对效应机制理解的不断深化,AOP 概念逐渐发展起来。AOP 就是描绘从分子启动事件(MIE)的开始,由一系列关键事件(KE)和之间关系(KER)连接,到有害结局(AO)之间关系的框架[71],与 AOP 相关的各种概念如表 1 所示。内分泌干扰效应的产

表 1 与有害结局路径(AOP)相关概念的定义[71]

Table 1 Definition of concepts relevant to adverse outcome pathway (AOP)[71]

项目(Concept)		定义(Definition)
有害结局路径	AOP (adverse outcome pathway)	描绘现有知识中直接的分子启动事件(如外源干扰物与生物大分子相互作用)和与风险评估相关的特定生物层面的有害结局之间联系的概念性框架。 A conceptual construct that portrays existing knowledge concerning the linkage between a direct molecular initiating event (e.g., a molecular interaction between a xeno-biotic and a specific biomolecule) and an adverse outcome at a biological level of organization relevant to risk assessment.
作用方式	MOA (mode of action)	导致有害影响的,生物学上可信的系列关键事件。指一系列关键事件和过程,从媒介与细胞的作用开始,经过器官、组织上可观察的变化,最终导致有害影响。作用方式与作用机理(mechanism of action)不同,作用机理更强调细节的理解和事件的描述。 A biologically plausible series of key events leading to an adverse effect. A sequence of Key Events and processes, starting with interaction of an agent with a cell, proceeding through operational and anatomical changes, and resulting in an adverse effect. Mode of action is contrasted with "mechanism of action," which implies a more detailed understanding and description of events.
关键事件	KE (key event)	指可实际观察的步骤或标志,是作用方式决定最终结局的必要元素(必要而不充分),关键事件是可测且可重复的。 An empirically observable step or its marker, which is a necessary element of the mode of action critical to the outcome (i.e., necessary, but not necessarily sufficient in its own right); key events are measurable and reproducible.
关键事件关系	KER (key event relationship)	成对关键事件之间的预见或因果关系。关键事件关系是推理或推测的单元,指从上游关键事件推理或推测下游关键事件所提供的科学可信的生物学依据,推理或推测的置信度由支撑证据的权重表示。 The predictive or causal linkages between a pair of KEs. KERs, in contrast, are a unit of inference or extrapolation. They are defined by the biological plausibility and evidence that provide a scientifically credible basis for inferring or predicting the state of a downstream KE based on the known state of an upstream KE and the confidence in that inference or prediction is defined by the weight of supporting evidence.
分子起始事件	MIE (molecular initiating event)	AOP 中第一个关键事件,指由外源干扰物与特定生物大分子相互作用导致的生物学干扰。 The first KE within an AOP representing the biologic perturbation resulting from a molecular interaction between a xenobiotic and a specific biomolecule.
有害结局	AO (adverse outcome)	AOP 后期的关键事件,指在管理层面会认为是有害的生物学干扰。有害结局特指个体或种群层面的变化。 Late stage KE in an AOP representing a biologic perturbation that would be considered adverse in a regulatory context. These typically occur at either the individual or population levels of organization.

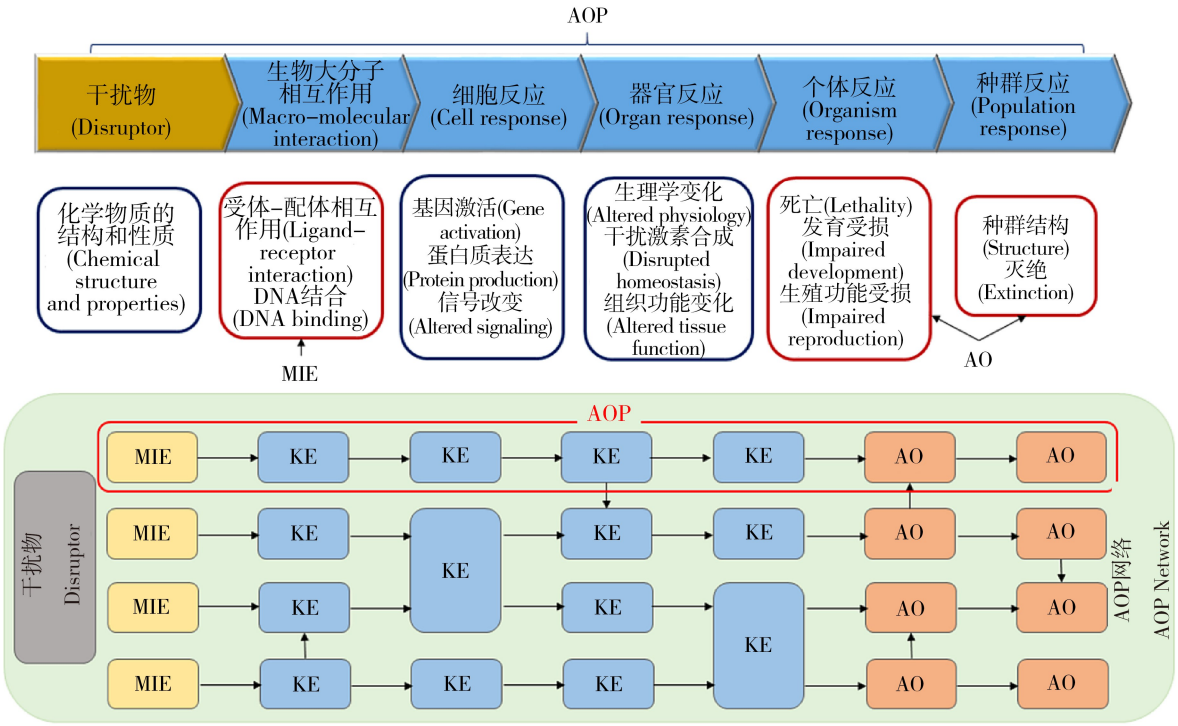

图 2　AOP 示意图

Fig. 2　Schematic of AOP

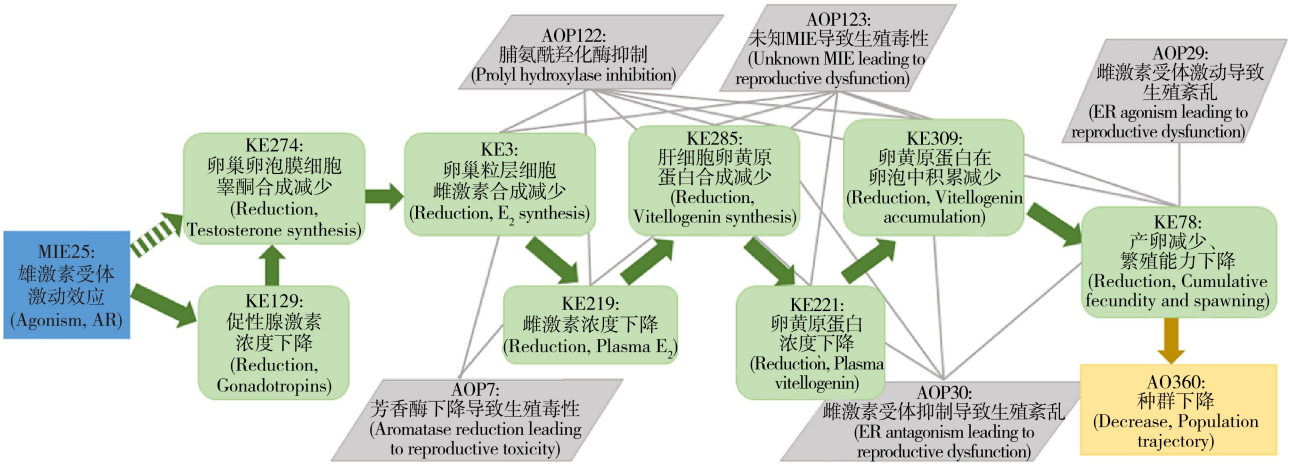

图 3　雄激素受体激动效应导致生殖紊乱的 AOP[73]

Fig. 3　AOP: Androgen receptor agonism leading to reproductive dysfunction[73]

生不只是干扰物与靶标相互作用,还包括生物大分子间、细胞层次、器官层次的变化(如图 2),因此,对 AOP 的研究有助于获得更加精确和透彻的预测效果。然而这种预测方法是建立在对干扰效应作用通路足够明晰的基础上,这也是目前面临的最大挑战[72]。随着有害结局路径知识库(Adverse Outcome Pathway Knowledge Base, AOP-KB: http://aopkb.org/)的建立,越来越多 AOP 被开发并在 AOP-KB 平台上共享,这将大大促进 AOP 在计算毒理学预测上的应用。

目前已有的 AOP 为 20 个,其中包括与雌激素、雄激素、甲状腺激素受体等相关的 5 个内分泌干扰 AOP。Villeneuve[73]开发并发表了有关雄激素受体激动效应导致生殖功能紊乱的 AOP(AOP23,如图 3 所示),干扰物激活雄激素受体(MIE),导致睾酮、雌

激素合成下降,血液雌激素浓度降低,进而肝脏卵黄原蛋白合成下降,血液中卵黄原蛋白浓度降低,卵母细胞吸收量减少,进而使产卵下降,最终导致生殖功能紊乱的 AO 产生。Villeneuve 等[74]还总结了 AOP 开发的五大原则:(1)AOP 不具有化合物特异性;(2)AOP 是模块化的,且构成要素可重复利用;(3)每个独立的 AOP 都由单一系列的 KE 和 KER 构成;(4)由具有相同 KE 和 KER 的 AOP 构成的网络(图 2)是预测真实世界情况的基础单元;(5)AOP 是可以随着新认识的形成而不断演化的。这些原则的考虑有助于对 AOP 的理解和应用。

MIE 和 KE 都是 AOP 必不可少的组成部分。由于 MIE 直接与干扰物相互作用,由此开始整条通路的调节,并最终到达 AO 终点,干扰物的结构和性质与 MIE 之间的联系比其他任一节点和效应终点都强[75]。因此,正如 QSAR 和分子模拟所做的,大多数计算机预测模型都以 MIE 为研究对象。然而,MIE 与 AO 并不是直接的相互关系,它们之间至少存在一个 KE,并且不同的 MIE 都有可能导致相同的 AO 产生,形成 AOP 网络(图 2)[76]。以前面 Villeneuve[73]开发的雄激素受体激动效应导致生殖功能紊乱的 AOP 为例,血液雌激素浓度降低同时会与芳香酶活性降低导致生殖毒性的 AOP(AOP7)产生交联,通过另一条 AOP 造成生殖问题,多条通路的交联从而形成 AOP 网络(图 3)。干扰物与最终 AO 之间的网络关系,使针对单一靶标受体的预测方法具有更大的不确定性。

将 MIE、KE、AO 之间用一系列的数学模型联系起来[77],可建立定量 AOP(qAOP)模型。学者们相信,通过建立 qAOP 可以将体外实验得到的数据,如配体受体结合效力,作为 qAOP 的输入信息,通过系列数学模型的模拟计算预测潜在的内分泌干扰活性,甚至模拟剂量-效应关系和时间进程行为[71]。AOP-KB 平台上推出了 Effectopedia 模块,通过量化 KE 之间的关系,建立 qAOP。目前,Effectopedia 模块还处于发展阶段,但 Beta 版本的软件已发布(http://www.effectopedia.org/)。此外,还有其他系统生物学计算模拟软件,如 PK-Sim 和 MoBi 都具有很好的 qAOP 模型构建和模拟功能[78]。然而,qAOP 需建立于明确的作用机制之下,目前还处于发展阶段,其定量预测能力还有待实验验证。

既然可以利用体外实验的数据,通过 AOP 预测最终的干扰效应,那么计算预测方法与 AOP 的结合也将成为可能。事实上,AOP 概念正是来源于利用 QSAR、生物标记物(biomarker)和其他机制数据提高对毒理学的认识和预测化学品暴露的潜在有害影响的想法[64]。由于对接和 MD 模拟的靶标往往与 AOP 中的 MIE 或 KE 相对应,分子模拟与 AOP 结合进行模拟预测将成为 AOP 发展中的重要研究方向。然而,限于目前 AOP 仍处于起步阶段,还没有较为成功的 AOP 与模拟预测方法结合的案例。

5 总结和展望(Conclusions and prospect)

内分泌干扰物是导致多种疾病,如生殖疾病、肥胖症和与激素相关的癌症等的重要诱因,众多化合物都具有潜在的内分泌干扰效应,使内分泌干扰问题在化合物风险评估上显得尤为突出。然而,评估一个化合物的内分泌干扰效应需要耗费大量的成本,无法对成千上万种化学品进行逐一测试。计算毒理学大大简化了这一过程,其使用也逐渐受到认可。本文基于内分泌干扰的效应机制,介绍了分子对接、MD 模拟和 AOP 这 3 种计算毒理学方法及其在内分泌干扰物筛选上的应用。

分子对接与 QSAR 相比更有助于效应机制的理解,通过反向对接能预测化合物可能的内分泌干扰活性终点,还能与 QSAR 结合构建多维 QSAR 模型。MD 模拟有助于探索配体-受体相互作用关系及两者的变化,探索重要的结构变化,并借助热力学计算预测结合效力。AOP 将 MIE 与最终 AO 用一系列 KE 和 KER 连接,形成完整的、明晰的通路甚至网络,借助 AOP 和 AOP 网络将推动计算毒理学进入新的阶段。

内分泌干扰物主要通过与内分泌系统相关靶标的相互作用,正如 AOP 中的 MIE 和 KE 的激活,因此,分子对接、MD 模拟和 AOP 的结合将使计算毒理学更加面向效应机制。在内分泌干扰物的筛选中,将反向对接技术与 AOP 模拟结合,不仅能预测干扰物潜在的内分泌干扰敏感靶标,还能进一步通过 AOP 网络预测可能造成的有害结局。MD 模拟的发展使得通过模拟区分促进和抑制作用逐渐成为可能,甚至可以模拟生物大分子之间的相互作用,研究 MIE、KE 和 KE、KE 之间的关系。因此,将对接和分子动力学模拟技术运用到 AOP 和 AOP 网络中进行预测和筛选,将有助于计算毒理学在内分泌干扰物筛选上的发展与应用。

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