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REVIEW

Combinatorial pathway disruption is a powerful approach to delineate metabolic impacts of endocrine disruptors

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The prevalence of metabolic diseases, such as obesity, diabetes, metabolic syndrome and chronic liver diseases among others, has been rising for several years. Epidemiology and mechanistic (*in vivo*, *in vitro* and *in silico*) toxicology have recently provided compelling evidence implicating the chemical environment in the pathogenesis of these diseases. In this review, we will describe the biological processes that contribute to the development of metabolic diseases targeted by metabolic disruptors, and will propose an integrated pathophysiological vision of their effects on several organs. With regard to these pathomechanisms, we will discuss the needs, and the stakes of evolving the testing and assessment of endocrine disruptors to improve the prevention and management of metabolic diseases that have become a global epidemic since the end of last century.

Keywords: appetite; bisphenol; dioxin; inflammation; insulin resistance; microbiota; perfluorinated compounds; phthalate; TBT

Abbreviations

BPA, bisphenol A; DDE, dichlorodiphenyldichloroethylene; DDT, dichlorodiphenyltrichloroethane; EDs or EDCs, endocrine disrupting compounds; HCB, hexachlorobenzene; MDCs, metabolism disrupting chemicals; PBDE, polybrominated diphenyl ether; PCBs, polychlorinated biphenyls; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonic acid; TBT, tributyl tin.

Introduction to the history of obesogenic endocrine disrupting chemicals

Since the middle of the twentieth century, the prevalence of obesity, diabetes and non-alcoholic fatty liver disease (NAFLD) has dramatically increased in developed countries. Now the rise is even faster in the low- and medium-income countries. In 2016, about 2 billion adults aged 18 or older and 340 million children and adolescents were overweight or obese. In parallel, the number of people with diabetes quadrupled from 1980 to 2014. NAFLD, which concerns around 25% of adults, is worsened by obesity, insulin resistance and type 2 diabetes [1]. Thus, these pathologies are related to each other and have common primary causative factors such as unbalanced diets, malnutrition, and lack of physical exercise or sedentary lifestyle. Besides psychosocial and physical factors, the environment, in particular the chemical environment, is suspected of contributing to the spread of this worldwide epidemic.

In 2006, Grun and Blumberg were the first to propose that endocrine disruptors (EDs), which are chemicals affecting endocrine systems and provoke deleterious effects, could induce or increase weight gain. They coined the name obesogens to describe these substances [2]. Obesogens have multiple modes of action: they can lead to an excessive development of the adipose tissue, increased inflammation that leads to abnormal adipocytes (e.g. resistant to insulin), increase in the production of pro-inflammatory cytokines, triacylglycerols storage, a decrease in fat consumption that leads to dysfunction of other organs, such as the liver and the pancreas, and of the inter-organ communication. These EDs have also been termed 'metabolism disrupting chemicals' (MDCs) [3] and there is converging evidence confirming their role in the development of metabolic diseases. At this stage, obesogens and MDCs overlap considerably and either term is suitable.

Among 1000 chemicals with presumed endocrine effects, epidemiological data indicate that exposure to some chemicals classified as persistent organic pollutants (POPs) by the Stockholm Convention alters metabolic functions and increases obesity. They include polychlorinated biphenyls (PCBs), polybrominated diphenyl ether (PBDE), perfluorooctane sulfonic acid (PFOS) and perfluorooctanoic acid (PFOA) (present in electrical appliances, paper, textiles or kitchenware), pesticides such as dichlorodiphenyl-trichloroethane (DDT) (or its metabolite dichlorodiphenyldichloroethylene, DDE) and hexachlorobenzene (HCB). Forbidden or restricted, these pesticides or chemicals used for their insulating, flame retardant or

anti-adhesive properties, are lipophilic and accumulate in fat mass of living organisms because of their resistance to biodegradation, except for PFAS. Alternatively, and more recently, many non-persistent pollutants present in everyday consumer products, used as plasticisers, food/cosmetic additives and preservatives (monosodium glutamate, phthalates and parabens) but also heavy metals (cadmium and arsenic) released from industrial, agricultural products and tobacco, are also suspected of favouring either increased weight mass or disturbed carbohydrate and lipid homeostasis. Thus, new and more appropriate terms, MDCs or obesogens, are now used to name the EDs that induce metabolic disorders and promote the development of obesity, type 2 diabetes and fatty liver diseases. In this review, we will describe the mechanisms targeted by the substances in key organs, that is, adipose tissue, liver, pancreas, gut and brain, which can alter inter-organ communication and lead to the development of metabolic diseases.

Epidemiological evidence for the metabolic effects of MDCs

The occurrence of metabolic diseases at the population level is influenced by exposure to several families of toxicants as shown by several epidemiological studies: evidence for inorganic compounds such as metals (arsenic, mercury and cadmium) remains limited [4,5] despite some trends of association (arsenic, mercury and dyslipidaemias [6-8]), while evidence of presumed risk for organic compounds is more conclusive. For persistent molecules such as organochlorine pesticides [beta-hexachlorocyclohexane (β -HCH) and heptachlor epoxide], associations are suggested with type 2 diabetes and metabolic syndrome [9-12].

There is growing evidence for a causal link to metabolic disorders concerning perfluorinated compounds (e.g. used to produce Teflon) such as PFOA and PFOS, which are highly persistent molecules (ecosystems and organisms): in young children exposed *in utero*, an increased risk of obesity, dyslipidaemia and hypertension is observed [13-15]. Exposure of adolescents or adults is associated with an increased risk of obesity, dyslipidaemia and type 2 diabetes in adults [16-18]. Alternatives to these perfluorinated compounds (GenX, Gi-PFESA) do not appear to be safer either, according to recent studies [19].

A large number of observations have also been carried out on compounds that are not persistent by nature but to which we are regularly exposed through our lifestyles: thus, among bisphenols (used in the plastics and packaging industries), a major focus on bisphenol

A (BPA) over the past several years has suggested that exposure to BPA, both during the perinatal period and in adulthood, increases the risk of metabolic syndrome, in particular, obesity and insulin resistance (one of the components of type 2 diabetes) [20–23]. Recent experimental studies suggest that BPA substituents (notably bisphenol S and F) are not safe alternatives [24].

For phthalates, which are also produced by the plastics industry, obesity appears to be one of the most common pathologies due to exposure during early life (*in utero*, childhood) or during adulthood [20,25,26]. In adults, disorders of glucose metabolism and hypertension are also identified with yet unexplained dimorphic effects [27].

Exposure to polycyclic aromatic hydrocarbons (PAHs, compounds resulting from combustion and cooking processes that are important air pollutants and food contaminants) is also associated with an increased risk of obesity in children and adults [28]. In adults, an increased risk of diabetes and hypertension is observed in association with environmental PAH exposure [29,30].

Experimental evidence for metabolic effects of MDCs

Numerous studies have been conducted on organs involved in the development of metabolic diseases including adipose tissue, liver, intestine, pancreas and specific systems or processes such as the microbiome, the central nervous system (regarding the control of appetite) or epigenetic mechanisms. We will now describe the most recent effects of EDs on these specific systems below.

Alteration of adipocyte functions: adipogenesis, adipokine secretion, insulin response

White adipose tissue (WAT) has a central role in metabolic homeostasis and its enlargement *via* hyperplasia and hypertrophy of adipocytes, is an essential adaptive response to caloric excess. This tissue is well studied in toxicology for several reasons: it is a target organ of EDs that can affect its development and functions; it is also a storage organ of POPs that can be released during lipolysis or on a long-term basis [31]. Formation of adipocytes or adipogenesis includes commitment of stem cells into the adipocyte lineage, proliferation of preadipocytes (the precursor adipocytic cells) and their differentiation into adipocytes [32]. Adipogenesis requires a sequential cooperation of transcription factors among which CCAAT-enhancer-binding protein- α (C/EBP α) and peroxisome

proliferator-activated receptor- γ (PPAR γ) play a crucial role. They activate target genes such as lipoprotein lipase (LPL), performing the lipolysis of circulating triacylglycerol-rich lipoproteins, CD36, a fatty acid (FA) cell-membrane transporter, fatty acid-binding protein 4 (FABP4), which binds and translocates intracellular FA, and perilipins that stabilise oil globules, all of them contributing to the formation of the lipid droplet of functional adipocytes that also secrete adipokines like leptin [33]. When the storage capacity of WAT is exceeded, this process is altered and leads to ectopic fat depots (in visceral WAT, liver and muscles) characterised by hypertrophic, inflammatory and insulin-resistant adipocytes [34].

Among the *in vitro* models used to assess the mechanisms of action of MDCs, 3T3-L1 and OP9 cell lines and C3H10-T1/2 mesenchymal stem cells (MSCs) are the most common murine models. The use of human models such as the Simpson–Golabi–Behmel syndrome (SGBS) preadipocytes and human adipose-derived stem cells is less common because of their more limited availability and their high cost. Rodents and zebrafish are essential *in vivo* models to highlight obesogenic and metabolic effects resulting from direct or *in utero* exposure to MDCs.

Organotin compounds such as tributyltin (TBT) represent an example of an environmental obesogen linked to abnormal adipocyte functions as shown by *in vivo* (rodents and zebrafish) and *in vitro* experiments. In different models, pre- or post-natal exposure to TBT increases body weight, fat mass, number of inflammatory cells in WAT, glycaemia and insulinaemia, while it decreases muscle mass [35–37]. Some of these effects persisted into adulthood and the next generations.

Similarly, BPA or its structural analogues (bisphenols AF, B, E, F and S) and halogenated derivatives [tetrabromobisphenol A (TBBPA) and tetrachlorobisphenol-A], phthalates [e.g. diethylhexyl phthalate (DEHP)], parabens, PFOS, PFOA and organochlorine pesticides such as DDT/DDE and HCB are associated with an increased adipogenesis and subsequently to an increased body weight and other disorders such as inflammation, hyperglycaemia, dyslipidaemia, glucose intolerance and insulin resistance [38–43]. For some chemicals such as tetrachlorodibenzo-*p*-dioxin (TCDD), PCB153, a second hit (e.g. an exposure to a high-fat diet) is necessary to observe an obesogenic effect, hyperglycaemia, or glucose and insulin intolerance [44–46].

At the molecular level, several receptors have been involved in the effects of most MDCs: three xenobiotic receptors mediate such signalling including constitutive androstane receptor (CAR) and pregnane X receptor (PXR) whose activation by mono(2-thylhexyl)

phthalate (MEHP) and monoisononyl phthalate [47] can explain the ability of these phthalates to regulate the expression of genes involved in glyceroneogenesis and triacylglycerol metabolism [48]. *In vitro* data concerning TCDD and dioxin-like PCBs that activate the aryl hydrocarbon receptor (AhR) (a bHLH/PAS transcription factor), argue in favour of their anti-adipogenic and pro-inflammatory effects [46,49,50,51] (see above the necessity to have a second type of hit to observe an obesogenic effect).

Interestingly, receptors which were not traditional xenobiotic receptors are also involved in MDC effects: for example, BPA and its substitutes (AF, F and S), parabens (ethyl-, propyl-, butyl- and benzyl-parabens), PFOS and PFOA, act *via* the binding or upregulation of PPAR γ and C/EBP α , which increases expression of adipogenic genes such as FABP4, LPL, leptin and perilipin [42,52,53,54]. Some pollutants display multiple targets: obesogenic effects of TBT are related to its ability to activate several nuclear receptors (NRs) such as retinoid X receptor-alpha (RXR α) and PPAR γ [55], which increase the adipogenic commitment of MSCs, stimulate adipocyte proliferation and differentiation, and induce lipid uptake by adipocytes [3]. Similarly, phthalates and their metabolites can interact with different PPARs as agonists, and the androgen receptor (AR) as antagonists, and elicit endocrine-disrupting effects possibly contributing to obesity [56,57]. Several environmental compounds may activate the PPAR γ of different vertebrate species similarly [57]. In addition, oestrogen receptor-alpha (ER α) activation and/or AR inhibition by bisphenols, DDT/DDE and parabens [58,59] and glucocorticoid receptor (GR) activation by parabens [53] are also observed. Thus, MDCs by promoting the formation of hypertrophic and inflammatory adipocytes and counteracting the effects of endogenous ligands of several nuclear receptors (NRs), impair WAT functions. However, several signalling mechanisms appear to contribute to these effects.

Alteration of the hepatic functions: glucido-lipidic metabolism, secretion of hepatokines, response to insulin

During the last decades, the impact of EDs and/or MDCs on the liver and their consequences on metabolic disease outcomes have been largely supported. Most of them lead to NAFLD which are further linked to obesity, metabolic syndrome, insulin resistance (IR) and diabetes [1,60]. Thereafter, some examples of the effects of MDCs on mechanisms involved in the development of liver diseases from steatosis (lipid accumulation as droplets in hepatocytes) to non-

alcoholic steatohepatitis (NASH, characterised by liver cell death and inflammation) are presented through four non-exhaustive parts: (a) disruption of lipid homeostasis, (b) disruption of carbohydrate metabolism, (c) alteration of insulin responses and (d) inflammation and secretion of hepatokines.

With regard to the (a) disruption of lipid homeostasis, the first step of NAFLD is the accumulation of FA in the hepatocytes, mainly stored in the form of triglycerides; this can be driven by multiple processes: increased FA uptake, increased *de novo* lipogenesis (DNL), decreased mitochondrial FA oxidation (mFAO) and decreased FA export. These processes are highly regulated by nuclear receptors and other xenoreceptors, including PPARs, liver X receptors (LXR α), CAR, PXR, AhR and/or by other transcription factors, such as sterol regulatory element binding protein 1c (SREBP1c). Each of them has been shown to be altered by EDs [1,61]. A good example of such perturbations leading to liver steatosis could be the effect of an exposure to PCB156 which activates the AhR in mice while decreasing the expression of PPAR β/δ , a nuclear receptor which promotes FA oxidation [62]. Both PCB156 effects may then contribute to: an increased expression of CD36 (a plasma membrane transporter involved in FA uptake), and of SREBP1c (a transcription factor which regulates the expression of DNL-associated and cholesterol metabolism genes), a decreased expression of carnitine palmitoyl-transferase 1B (CPT1B) (a regulatory enzyme involved in mitochondrial FA entrance) and of apolipoprotein C2 (APOC2) (the co-factor activator of LPL), and ATP-binding cassette subfamily A member 1, ABCA1, involved in cellular cholesterol efflux [63]. Accumulation of lipids and disruption of expression of genes involved in the control of lipid metabolism can be similarly observed in liver cells exposed to perfluorinated compounds, such as PFOS [64] or in mice exposed to low doses of BPA (5 and 50 $\mu\text{g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$) increases the liver expression of lipogenic enzymes (Acc, Fasn, Scd1) and transcription factors which regulate lipogenesis (LXR, SREBP1c, ChREBP or carbohydrate responsive element binding protein). This effect is not observed at high doses (5000 $\mu\text{g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$) [65].

Regarding the (b) disruption of carbohydrate metabolism, it has been shown that several EDs bind CAR in hepatocytes. CAR is a nuclear receptor which binds xenobiotics and endobiotics, including retinoic acids and steroids, and it forms dimers with its partner, RXR α . In addition to xenobiotic metabolism and transport regulation, CAR regulates several liver functions, including, carbohydrate and lipid metabolism,

through a transcriptional activation associated with histone acetylation and a transcriptional repression when competing on enhancers with other regulatory factors such as HNF4 α , PPAR α or FXR [66]. CAR has been described as a repressor of gluconeogenesis: it binds to the transcriptional regulator FoxO1 and thus induces transactivation of several gluconeogenic genes including cytoplasmic phosphoenolpyruvate carboxykinase (PEPCK) and glucose 6-phosphatase (G6Pase) [1]. Phthalates and perfluorinated compounds have been described as CAR agonists [DEHP, Dibutyl phthalate (DBP) or PFOS] or antagonists (PFOA) [67–69]. Bisphenols have been described as either agonists (BPA) or antagonists (TBBPA). BPA acts on CAR as an inverse agonist, that is, its binding to the receptor exerts an opposite effect compared with an agonist [70]. In that case, BPA might favour gluconeogenesis and increase glycaemia. Moreover, BPA and its analogues can decrease glucokinase expression leading to an impairment of glucose sensing and glucose intolerance [71]. Glucose metabolism is also highly linked to insulin regulation and response, and EDs are also well-known to interfere with this pathway, as discussed in the next section.

Concerning the (c) alteration of insulin responses, the secretion of insulin, the main blood glucose-regulating hormone, is induced by increased blood glucose, upon the body shifting from a fasted to a fed state, and promotes liver glycogenesis and lipogenesis, while inhibiting liver gluconeogenesis and glucose secretion [72,73]. EDs are known to interfere with insulin response in the liver and other peripheral organs (muscles and AT) [74,75]. An example of direct effects of EDs on the liver is the disruption of insulin signalling by BPA, which reduces the level of insulin receptor and glycogenesis in the liver of rats [76]. It should be noted that reduction of insulin response by EDs in muscles or adipose tissues, leading respectively to hyperglycaemia or hyperlipidaemia, also promote both NAFLD and other metabolic diseases [74,75].

Finally, about (d) inflammation and secretion of hepatokines, it is worth noting that cytokines and pro-inflammatory proteins produced by the liver are crucial players in NAFLD progression to non-alcoholic steatohepatitis (NASH). NASH is a pathophysiological state in which liver steatosis is combined with inflammation and sometimes fibrosis; it can eventually lead to cirrhosis or hepatocellular carcinoma. Cytokine expression and secretion are regulated by EDs [1] such as PFOA in mice [77] or BPA that increases IL-8 and TNF- α secretion in HepG2 cells [78]. EDs such as BPA, PFOS and TCDD mediate liver inflammatory responses not only through pro-inflammatory

cytokines secretion but also by the activation and the polarisation of Kupffer cells (KCs) into pro-inflammatory M1-phenotype [79], the infiltration of immune cells in the liver and the activation of quiescent hepatic stellate cells (HSCs) into myofibroblast-like cells [80]. Furthermore, it has been shown in mice that polychlorinated biphenyls (PCBs) could increase the expression of several secreted proteins, known as hepatokines (Fgf21, Igf1 and betatrophin), thus promoting NAFLD progression and pancreatic alteration favouring diabetes [81].

Impaired pancreatic functions

Pancreatic β -cell dysfunction is known to be the hallmark of type 2 diabetes but also a major contributing factor in the aetiology of other metabolic diseases like obesity or metabolic syndrome. Experimental evidence has revealed that certain EDs may display direct effects on pancreatic β -cells leading to several adverse outcomes including oxidative stress, mitochondrial damage, cell apoptosis, altered electrical activity, impaired Ca²⁺ signalling and insulin secretory defects. Here we give a brief overview of the currently known EDs affecting β -cells and of the mechanisms of action of POPs and non-POPs.

Current experimental evidence supports that BPA can disrupt glucose metabolism by affecting pancreatic β -cell physiology. Acute low doses of BPA have been reported to alter the expression and activity of the main ion channels implicated in the coupling of glucose metabolism to insulin secretion. In particular, BPA was found to promote the closure of the K_{ATP} channel, increase the frequency of [Ca²⁺]_i oscillations, as well as to reduce Na⁺ and K⁺ currents in mouse and human pancreatic β -cells. Furthermore, BPA significantly disturbed the expression of genes encoding important Na⁺ and K⁺ subunits. These changes ultimately led to increased glucose-stimulated insulin secretion (GSIS) through the activation of the extranuclear oestrogen receptor beta (ER β) [82,83]. Longer exposures to BPA upregulated insulin content in an oestrogen receptor alpha (ER α)-mediated manner [84]. BPA-treated animals also manifested an excessive insulin secretory response leading to hyperinsulinaemia and insulin resistance [85,86].

A number of studies have also examined the impact of a direct DEHP exposure on pancreatic β -cells and, although limited to immortalised β -cell lines, they all indicate that DEHP can promote increased apoptosis [87–90]; various mechanisms have been proposed to explain this effect including oxidative stress [87–90], DNA damage [90], decline of antioxidant protection

[89], activation of ER (endoplasmic reticulum) stress responses [89] or interaction between oxidative stress and autophagy [88]. DEHP exposure has also been shown to suppress GSIS [87,89], which could be partially explained by the DEHP-induced β -cell loss.

Among POPs, studies performed in the mouse pancreatic β -cell line β -TC6 have revealed that PFOS may differently impact pancreatic β -cell function depending on time of exposure. Thus, acute PFOS treatment for 1 h resulted in augmented intracellular $[Ca^{2+}]_i$ and, consequently, insulin release, in a mechanism depending on G protein coupled receptor (GPR40) activation [91,92]. This was confirmed *in vivo* [91]. In contrast, longer exposure to PFOS (48 h) promoted decreased mitochondrial membrane potential, ATP production, Ca^{2+} influx, as well as insulin secretion, *via* downregulation of SIRT1-UCP2 pathway [93]. Decreased ATP levels and GSIS were also reported in *ex vivo* treated mouse islets together with a significant reduction on Pdk1-AktmTOR pathway expression levels [94].

As it was the case for PFOS, TBT effects on pancreatic β -cells was largely dependent on the length of treatment. Acute exposure (4 h) significantly increased $[Ca^{2+}]_i$ and insulin secretion. These effects were attenuated in the presence of the oestrogen receptor antagonist ICI 182780, the antioxidant *N*-acetylcysteine, and the specific PKC inhibitor Ro32-0432, suggesting that both oestrogen-related and ROS/PKC signalling pathways were involved. This was confirmed in the β -cell line Rin-m5F but also in mouse and human islets treated *ex vivo* [95]. TBT exposure for 24 h led to oxidative stress and consequent activation of JNK pathway resulting in increased apoptosis and decreased GSIS in Rin-m5F β -cells and mouse isolated islets [96].

Among dioxins, TCDD is the best known to be harmful for pancreatic β -cells. Studies in mouse and human islets have demonstrated that TCDD may impair insulin release through an AhR-dependent pathway, as CYP1A1 expression and activity were found to be elevated [97]. In addition, TCDD has been reported to promote increased apoptosis through ERK1/2 and JNK signalling pathways [98]. Both increased apoptosis and decreased GSIS were related to ultrastructural alterations such as increased mitophagy and mitochondrial swelling in mouse and human islets [99].

Impaired intestinal and microbiota functions

While the impact of EDs on the liver: adipose axis has been well studied through their roles on metabolic diseases like NAFLD, diabetes or obesity [100–102], their effect on the intestine still needs to be investigated.

Notwithstanding, the gastrointestinal tract constitutes a prime target of many pharmaceuticals and xenobiotics that can affect human health and disease [103].

The gut microbiota is an organ, capable of producing a large number of biologically active molecules that influence the physiological functions of the host whose secretions in turn influence the composition and function of the microbiota.

Indeed, there is mounting evidence that the gut microbiome, through crosstalk with the gut–liver and gut–brain axis, could mediate the outcome of ED chemical exposure. This could lead to reproductive and mental disorders as well as to metabolic diseases by altering hormone regulation of food intake, appetite and satiety [104]. Even if the mechanisms behind this dysbiosis are still not clarified, the impact of EDs on intestinal microbiota may be responsible for obesity even in young children [105]. EDs influence the gut-microbiome dialogue and can lead to dysbiosis. The microbiota can also be responsible for the metabolism of food contaminants (dechlorination of DDT to DDD). Finally, a sexual dimorphism in the composition of microbiota could play a role in sexual predisposition to diseases [106], including metabolic syndrome (NAFLD) [107].

Some studies have identified putative mechanisms leading to metabolic pathologies: indeed, in different animal models (mice, zebrafish and dogs), BPA is able to induce changes in the microbiota, both in terms of its composition and its function: BPA (a) favours Proteobacteria populations (as in case of exposure to a high-fat diet) or CKC4 [108,109] and disfavors the phylum Bacteroides, Flexispiraphyla, Oscillospira and Ruminococcaceae [110,111], (b) changes the blood metabolome (increased plasma bicarbonate concentrations in relation to Bacteroides disruptions), [110], (c) feminises the microbiota [112]. Other EDCs have been studied such as phthalates, diethylphthalate (DEP), methylparaben (MP) and triclosan (as well as their mixture) but they had significantly different population effects with a relative increase in Bacteroides and decrease in Firmicutes in female rats [113]. In contrast, MEHP causes a relative increase of Firmicutes and a reduction in Verrucomicrobia in mice, whereas DEP causes a decrease in Firmicutes [114]. The importance of pattern and chemical nature of molecules is confirmed with pesticides: carbemazine, a reprotoxic fungicide, and DDE and β -HCH, insecticidal organochlorines, reduce microbiota diversity by decreasing bacteroids, and favouring firmicutes [115,116] while pentachlorophenol (PCP), a herbicide and insecticide, favours bacteroids at the expense of firmicutes [117]. These studies raise the question of

antagonistic and potentiating effects of mixtures of molecules especially since some persistent molecules (such as 2,3,7,8-tetrachlorodibenzofuran, PCB126, TCDD and DDE) are also responsible for these imbalances, for some in relation to metabolic diseases [116,118,119]. In particular, phosphatidylcholine, phosphatidylserine, phosphatidylethanolamine and triacylglycerols have been identified as key metabolites affected by DDE treatment, the levels of which are strongly correlated with altered microbiota composition [119]. At this stage it is not completely clear whether the effects on microbiota account for the MDC metabolic effects or whether the impacts on metabolism can account for the observed imbalance in microbial species.

Several studies attempted to address these mechanisms. Male mice treated with BPA showed, in addition to the bacterial composition alterations seen above, an accumulation of hepatic lipids. The suspected mechanism is that the decrease in the diversity of the gut microbiota leads to increased permeability related to elevated levels of endotoxins, and an increase in liver inflammation (IL-1 β and 6, TNF- α) that promotes steatosis [120]. In BPA-treated rats, intestinal imbalance (decrease in faecal bifidobacterial elements) is associated with impaired glucose tolerance that subsequently leads to the development of obesity, type 2 diabetes (through insulin resistance and inflammation of peripheral tissues such as adipose tissue) [121].

TCDD, a ubiquitous POP, could alter the composition of the microbiota in male rats and thus disrupt the enterohepatic cycle leading to a significant decrease in faecal bile acids, and thus an increase in intestinal transit time and intestinal permeability [122]. One of the modes of action of dioxins is through their binding to the AhR; however, this receptor also binds tryptophan metabolites that can be produced by the microbiota, and an alteration of this metabolic capacity is associated with the metabolic syndrome, in mice and humans. Dioxins such as TCDD could hijack 'Trp metabolites – AhR' signalling; indeed, restoration of AhR signalling through the use of natural agonists, attenuates both intestinal permeability and metabolic syndrome in mice [122–124].

Finally, the microbiota produces short-chain FA (SCFAs) such as propionate, an anti-inflammatory molecule that may reduce the development of hepatic steatosis by reducing the transcription of several *de novo* lipogenesis enzymes [125]. Butyrate potentiates the secretion of pituitary GH and its stimulatory factor, ghrelin [126] which increases food intake. Yet it has been shown that chronic exposure to chlorpyrifos

(an organophosphate insecticide) can reduce the production of SCFA and lactate.

The intestine itself plays a central role in fat homeostasis by regulating intestinal lipid transport. Furthermore, several relevant molecular actors are expressed in the intestine. For instance, PXR is a ligand-dependent transcription factor that is activated by numerous endogenous hormones, dietary steroids, pharmaceutical agents, and xenobiotics and regulates the expression of genes required for xenobiotic metabolism in the liver and intestine, including Phase I–III proteins [127]. Whereas the role of PXR in xenobiotic metabolism has been well established, its role in mediating the pathophysiological effects of ED chemicals in humans and animals has been less investigated although recent studies reported the role of PXR in dyslipidaemia and atherosclerosis. The principal role of PXR in the intestine is to maintain barrier function and reduce inflammation, as well as to regulate intestinal transcription of metabolic enzymes [100]. The dietary xenobiotics resulting from PXR-regulated intestinal absorption could impact the physiological function of the liver and adipose tissue and promote steatosis and obesity (among others) [100]. Recently, Kim et al. [128] showed the role of PXR in physiopathology of NAFLD, obesity and inflammation linked to gut microbiome dysbiosis using PXR-knockout mice. The activation of intestinal PXR is responsible for transcriptional activity of key metabolic enzymes such as the CYP3A4 which is the major expressed P450 in intestinal enterocytes, with levels uncorrelated to those of liver. It is involved in the metabolism of endogenous compounds like cholesterol [129] and contributes to the first-pass metabolism of drugs [130]. PXR may link hypercholesterolaemia and exposure to ED chemicals as it is involved in both cholesterol and xenobiotics metabolism [131]. Thus, the disruption of intestinal PXR and/or CYP3A4 expression could lead to serious metabolic diseases through the deregulation of signalling pathways implicated in intestinal barrier function, inflammation, xenobiotics detoxification and endogenous metabolism.

Alteration of appetite and thermogenesis

Endocrine disruptors can influence food intake and thermogenesis at multiple levels and thus contribute to an increased risk of obesity [132,133].

Obesogenic molecules are likely to influence the function of the hypothalamus, an area of the brain responsible for controlling eating behaviour. Similarly, at the peripheral level, they influence the secretion and production of adipokines by the adipose tissue.

Specifically, perinatal exposure to BPA in rats influences pre- and post-synaptic connections at the hypothalamic level, increasing food consumption by stimulating compulsive eating behaviours, which ultimately leads to obesity [134]. A recent study shows that exposure to BPA and a brominated derivative, TBBPA at concentrations commonly found in the environment (between 20 and 500 $\mu\text{g}\cdot\text{L}^{-1}$) alters the behaviour of adult male zebrafish. The latter consumes a greater amount of food, which leads to obesity and hepatic steatosis. BPA and its derivative bind the cannabinoid receptor type 1 (CB1), a G protein-coupled receptor expressed in the peripheral and central nervous system. Selective antagonists of this CB1 receptor are used for weight reduction. Binding of BPA or TBBPA may promote food intake and contribute to the development of metabolic diseases [135].

Obesogens also control the levels of adipokine production and secretion that influence food intake; for example, in humans, serum BPA levels are associated with increased body weight and serum leptin and ghrelin levels [136]. In mice, at the level of adipocytes, BPA promotes the production of leptin transcripts (using the 3T3-L1 cell line), which leads to the hypothesis of an increased production of this hormone, which could therefore influence food intake [137]. This is also observed *in vivo* in mice treated with methyl-paraben and DEHP which increase serum leptin levels; the latter (DEHP) also decreases adiponectin levels [138,139].

Regarding thermogenesis, recent studies deciphered the mechanisms of communication between organs which govern the metabolic switches and potentially, how pollutants may affect this communication [140]. Indeed, cold-induced thermogenesis leads to the release of free fatty acids from white adipose tissue, which are taken up by the liver, which in turn produces a large amount of acylcarnitines in an HNF4 α -dependent process. These molecules reach the brown adipose tissue (BAT) but neither the liver nor the white adipose tissue, for which the capture is blocked [141]. A recent study showed that chlorpyrifos impairs mitochondrial respiration in BAT in mice at very low concentrations (1 μM). Rearing temperature is a key element of this experiment since at thermoneutrality, subjected to a high-fat diet, mice treated with chlorpyrifos develop obesity, NAFLD and insulin resistance. Thus chlorpyrifos, seems to inhibit diet-induced thermogenesis and activation of BAT [142].

Epigenetic effects of EDs

Several EDs are associated with epigenetic modifications (modifications of DNA methylation, post-

translational histones modifications and microRNA expression), possibly transgenerational: diethylstilbestrol (DES) represents a classical example: DES, a nonsteroidal oestrogen commonly prescribed during pregnancy between 1947 and 1971, is a potent ED whose prenatal exposure in animals causes developmental defects of the reproductive system; several epidemiological and animal studies suggest that prenatal exposure to this EDC is linked to obesity. More recently, it was shown that exposure to three EDs that are constituents of certain plastics, BPA, DEHP and DBP, to female rats (F0) during gestation induced in the F3 generation (which has never been in contact with these EDCs), changes in DNA methylation at genes associated with obesity, with increased susceptibility to this pathology in both males and females [143].

This is also true for POPs such as methoxychlor [144], DDT [145,146] or TBT, an organotin antifouling agent, historically used to control the appearance of organisms on the hulls of ships. TBT is indeed a prominent example of obesogens as it has been documented to have obesogenic effects in animals [2,37,147]. TBT is a potent environmental ligand of RXR and promote adipogenesis and alter lipid homeostasis *via* RXR-dependent pathways [148]. TBT promotes, in different generations (F1 and F3), an increase in perigonadal fat deposits but without any change in body weight [149]. However, a susceptibility to weight gain is observed specifically in males of the next generation (F4) [150]. Changes in DNA methylation are also observed for example in the leptin gene [151]. For DDT, a 2019 study suggests by isolating adipocytes, that this insecticide has a sexual dimorphic effect in terms of DNA methylation modification [152].

Inorganic pollutants exerting endocrine disrupting effects such as cadmium (in a mixture associated with mercury), are also incriminated by studies conducted in rats exposed over several generations, to metabolic alterations (glucose intolerance and increased abdominal fat deposition) up to the F4 generation [153].

Multi-organ alterations contributing to the development of metabolic diseases: an integrative view

Several types of pollutants have been epidemiologically associated with the development of metabolic diseases in young children, adolescents or adults (sometimes due to perinatal exposure) and several inter-organ mechanisms of action can be identified for these molecules.

The most common mechanism of action is the stimulation of inflammatory processes that can impact, for example, the function of adipose tissue (particularly in terms of insulin resistance) or that of the liver; inflammation being an essential component of NASH. Metabolic processes can also be directly affected: for example, the membrane FA transporter CD36 is regulated by several transcriptional factors in certain tissues (such as C/EBP α for AT), some of which are also nuclear receptors (PPAR γ) or xenobiotic receptors (AhR). CD36 allows the import of FA into cells, particularly at the adipocyte and hepatic levels, contributing respectively to adipogenesis and NAFLD formation. Another example is SREBP1c, a transcription factor that is synthesised as a precursor anchored to the nuclear membrane and the endoplasmic reticulum. Its expression is increased by insulin, which induces, after cleavage of the precursor, translocation of the mature protein to the nucleus leading to the regulation of genes involved in lipogenesis and glucose metabolism. Several pollutants increase the expression of CD36 or SREBP1c such as TCDD [154,155], PCB156 [62], PFOS [64], BPA [156] or atmospheric particles [157].

Some pollutants such as BPA have the capacity to target several key organs in metabolic regulation; their mode of action can thus be understood at the scale of the organism and of the inter-organ communication. Indeed, BPA displays a variety of effects in different organs that concur in leading to metabolic disruption. (a) BPA acts as an endocrine disruptor by impacting the production and secretion of insulin by the pancreas in an ER-dependent manner. While BPA augments insulin content through an ER α /ERK pathway, ER β modulates the expression and activity of ion channels in pancreatic β -cells leading to an increase in glucose-stimulated insulin release. Overall, this excessive secretory response may be a contributing factor in the long-term development of insulin resistance [82–86]. (b) At the hepatic level, there are multiple targets of BPA: it behaves as a reverse agonist of CAR [70], a receptor suppressing gluconeogenesis; as a consequence, BPA potentially contributes to hyperglycaemia but it also reduces the level of insulin receptor contributing to insulin resistance [76], which could be enhanced by inflammatory cytokine production [78] and polarisation of KCs to a pro-inflammatory phenotype. (c) BPA promotes in AT, increased adipogenesis associated with increased body weight but also inflammation that contributes to insulin resistance and hyperglycaemia [38–40]. BPA and its substitutes (AF, F and S), act by modulating the activity of PPAR γ ; it should be noted that, in this tissue, the effects of the BPA

substitute, BPS, are even more powerful and include a robust induction of adipogenic genes such as FABP4, LPL, leptin and perilipin [52].

Beyond the liver–pancreas–adipose tissue network, other tissues are impacted by BPA, such as the intestine, in particular the microbiota. The diversity of the gut microbiota appears to be reduced in favour of endotoxin-producing bacteria leading to increased permeability of the gut barrier, increased inflammation of the liver, and consequently insulin resistance and steatosis [120]. BPA would also act at the central level (hypothalamus) by increasing compulsive eating behaviours [134] and peripheral (AT) by increasing leptin secretion [137]. In addition to all the direct effects mentioned, this would result in a dietary energy imbalance (Fig. 1).

Conclusion and future perspectives

A major step forward in toxicology has been an increasing attention to combinations of exposure pathways, of stressors and of mechanisms of action. In this review, we did not discuss either aggregated exposure pathways or mixture effects, which are highly relevant but have been covered elsewhere [158,159].

The importance of taking into account multi-organs interaction needs to be addressed and we thus have highlighted that combining the disrupting effects that a given environmental chemical has on different organs is key to providing a better understanding of toxicity at the organism level. The combinatorial approach whereby organ-level pathway disruption is integrated into a global organism-level toxicity is particularly well suited for metabolic disruption. Indeed, it is well known that bodily metabolism is influenced by endocrine and signalling pathways at play in multiple organs and that exhibit considerable crosstalk: the gut–liver–pancreas–adipose–muscle–brain network governs organism metabolism and therefore it is particularly important to assess the effects of chemicals on these organs and to attempt to integrate them. This is what we have tried to do here for BPA.

The Adverse Outcome Pathway Network (AOPN) provides a particularly suitable framework to address combinatorial pathway disruption effects. By integrating different mechanisms of action at the molecular, cellular, organ and organism levels, it allows researchers to describe how combinations of multiple organ- or cellular-level pathways disruptions contribute to adverse outcomes. AOPN makes it possible to better determine the critical events leading to metabolic outcomes and possibly to identify effect markers for epidemiological or toxicological studies. Such markers

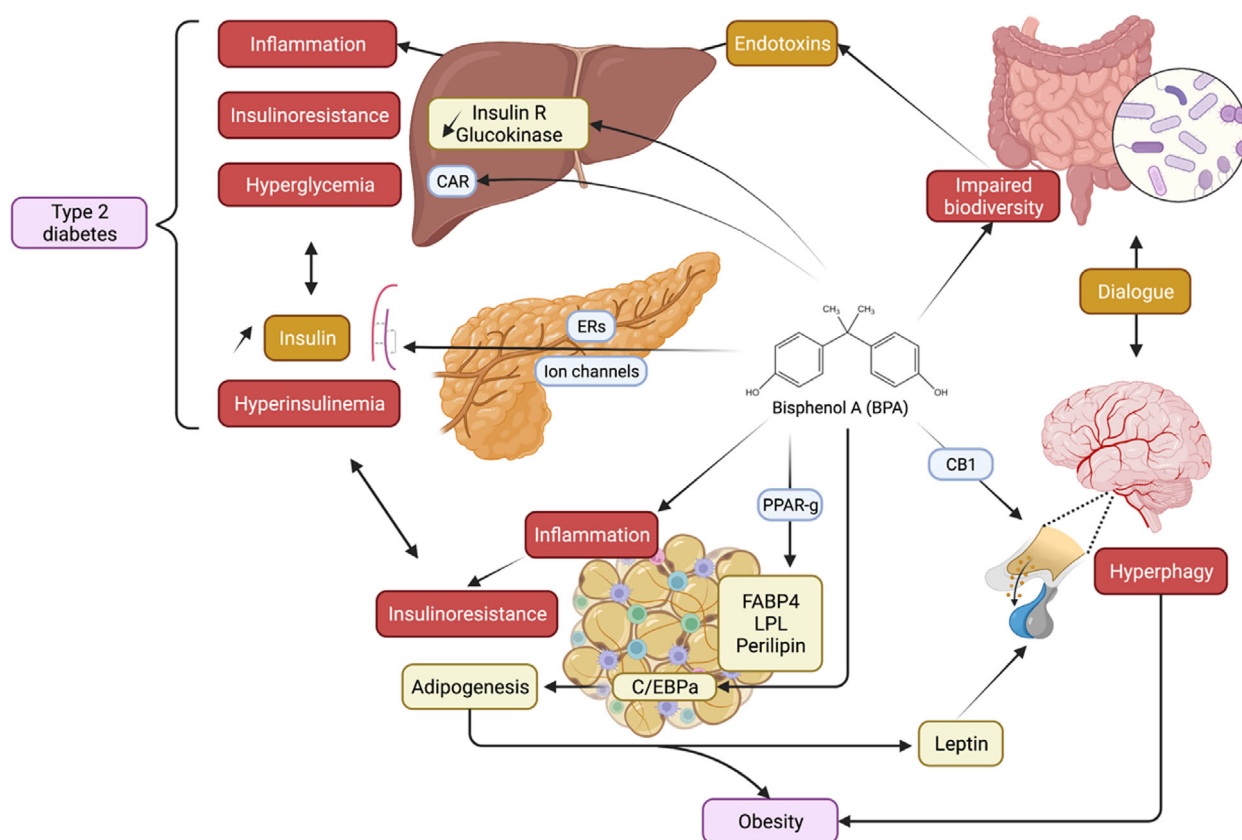


Fig. 1. An integrated view of the modes of action of bisphenol A (BPA) on metabolic parameters. In blue, the molecular targets of BPA; in yellow, indirect targets of BPA. In red, impaired physiological functions. In purple, the adverse outcomes (created with BioRender.com).

could also be useful to rapidly verify the safety of chemical substituents and contribute to a safe by design approach to the development of new chemicals.

While integrated multi-organ toxicity can obviously be tested *in vivo*, it is also possible to address these integrated multi-organ mechanisms by combining several *in vitro* and *in silico* assays. This requires a mechanistic understanding of ED effects at the organ level. While significant advances in our understanding of the modes of action and effects of EDs on the metabolic physiopathology of organisms have been made in recent years, there is still a need to pursue this research to enhance such knowledge at the molecular, cellular and tissue levels, particularly in order to develop mechanism-based assays, which could be implemented at the regulatory level and thereby fill existing gaps in hazard and risk assessment of chemicals. In this perspective, it is important to design these assays so that they could be used to efficiently establish causal and possibly quantitative links between mechanisms and adverse effects by taking advantage of the Adverse Outcome Pathways. The development of species-specific *in silico*, *in vitro* and *in vivo* models and their integration into an

efficient strategy to monitor human health must precede their regulatory recognition and use.

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References

- 1 Cano R, Pérez JL, Dávila LA, Ortega Á, Gómez Y, Valero-Cedeño NJ, et al. Role of endocrine-disrupting chemicals in the pathogenesis of non-alcoholic fatty

- liver disease: a comprehensive review. *Int J Mol Sci*. 2021;**22**:4807.
- 2 Grün F, Blumberg B. Environmental obesogens: organotins and endocrine disruption via nuclear receptor signaling. *Endocrinology*. 2006;**147**:S50–5.
 - 3 Heindel JJ, Blumberg B, Cave M, Machtinger R, Mantovani A, Mendez MA, et al. Metabolism disrupting chemicals and metabolic disorders. *Reprod Toxicol*. 2017;**68**:3–33.
 - 4 Bulka CM, Persky VW, Daviglus ML, Durazo-Arvizu RA, Argos M. Multiple metal exposures and metabolic syndrome: a cross-sectional analysis of the National Health and Nutrition Examination Survey 2011–2014. *Environ Res*. 2019;**168**:397–405.
 - 5 Xu P, Liu A, Li F, Tinkov AA, Liu L, Zhou J-C. Associations between metabolic syndrome and four heavy metals: a systematic review and meta-analysis. *Environ Pollut*. 2021;**273**:116480.
 - 6 Planchart A, Green A, Hoyo C, Mattingly CJ. Heavy metal exposure and metabolic syndrome: evidence from human and model system studies. *Curr Environ Health Rep*. 2018;**5**:110–24.
 - 7 Roy C, Tremblay P-Y, Ayotte P. Is mercury exposure causing diabetes, metabolic syndrome and insulin resistance? A systematic review of the literature. *Environ Res*. 2017;**156**:747–60.
 - 8 Tinkov AA, Ajsuvakova OP, Skalnaya MG, Popova EV, Sinitskii AI, Nemereshina ON, et al. Mercury and metabolic syndrome: a review of experimental and clinical observations. *Biomol*. 2015;**28**:231–54.
 - 9 Kim S-K, Park S, Chang S-J, Kim S-K, Song JS, Kim H-R, et al. Pesticides as a risk factor for metabolic syndrome: population-based longitudinal study in Korea. *Mol Cell Toxicol*. 2019;**15**:431–41.
 - 10 Park S-K, Son H-K, Lee S-K, Kang J-H, Chang Y-S, Jacobs DR, et al. Relationship between serum concentrations of organochlorine pesticides and metabolic syndrome among non-diabetic adults. *J Prev Med Public Health*. 2010;**43**:1.
 - 11 Rosenbaum PF, Weinstock RS, Silverstone AE, Sjödin A, Pavuk M. Metabolic syndrome is associated with exposure to organochlorine pesticides in Anniston, AL, United States. *Environ Int*. 2017;**108**:11–21.
 - 12 Mustieles V, Fernández MF, Martín-Olmedo P, González-Alzaga B, Fontalba-Navas A, Hauser R, et al. Human adipose tissue levels of persistent organic pollutants and metabolic syndrome components: combining a cross-sectional with a 10-year longitudinal study using a multi-pollutant approach. *Environ Int*. 2017;**104**:48–57.
 - 13 Mondal D, Lopez-Espinosa M-J, Armstrong B, Stein CR, Fletcher T. Relationships of perfluorooctanoate and perfluorooctane sulfonate serum concentrations between mother–child pairs in a population with perfluorooctanoate exposure from drinking water. *Environ Health Perspect*. 2012;**120**:752–7.
 - 14 Braun JM, Chen A, Romano ME, Calafat AM, Webster GM, Yolton K, et al. Prenatal perfluoroalkyl substance exposure and child adiposity at 8 years of age: the HOME study: prenatal PFAS exposure and child adiposity. *Obesity*. 2016;**24**:231–7.
 - 15 Halldorsson TI, Rytter D, Haug LS, Bech BH, Danielsen I, Becher G, et al. Prenatal exposure to perfluorooctanoate and risk of overweight at 20 years of age: a prospective cohort study. *Environ Health Perspect*. 2012;**120**:668–73.
 - 16 Averina M, Brox J, Huber S, Furberg A-S. Exposure to perfluoroalkyl substances (PFAS) and dyslipidemia, hypertension and obesity in adolescents. The fit futures study. *Environ Res*. 2021;**195**:110740.
 - 17 Qi W, Clark JM, Timme-Laragy AR, Park Y. Per- and polyfluoroalkyl substances and obesity, type 2 diabetes and non-alcoholic fatty liver disease: a review of epidemiologic findings. *Toxicol Environ Chem*. 2020;**102**:1–36.
 - 18 Liu G, Zhang B, Hu Y, Rood J, Liang L, Qi L, et al. Associations of perfluoroalkyl substances with blood lipids and apolipoproteins in lipoprotein subspecies: the POUNDS-lost study. *Environ Health*. 2020;**19**:5.
 - 19 Mi X, Yang Y-Q, Zeeshan M, Wang Z-B, Zeng X-Y, Zhou Y, et al. Serum levels of per- and polyfluoroalkyl substances alternatives and blood pressure by sex status: isomers of C8 health project in China. *Chemosphere*. 2020;**261**:127691.
 - 20 Stojanoska MM, Milosevic N, Milic N, Abenavoli L. The influence of phthalates and bisphenol A on the obesity development and glucose metabolism disorders. *Endocrine*. 2017;**55**:666–81.
 - 21 Teppala S, Madhavan S, Shankar A. Bisphenol A and metabolic syndrome: results from NHANES. *Int J Endocrinol*. 2012;**2012**:1–5.
 - 22 Kim KY, Lee E, Kim Y. The association between bisphenol A exposure and obesity in children—a systematic review with meta-analysis. *Int J Environ Res Public Health*. 2019;**16**:2521.
 - 23 Provisiero D, Pivonello C, Muscogiuri G, Negri M, de Angelis C, Simeoli C, et al. Influence of bisphenol A on type 2 diabetes mellitus. *Int J Environ Res Public Health*. 2016;**13**:989.
 - 24 Rochester JR, Bolden AL. Bisphenol S and F: a systematic review and comparison of the hormonal activity of bisphenol A substitutes. *Environ Health Perspect*. 2015;**123**:643–50.
 - 25 Golestanzadeh M, Riahi R, Kelishadi R. Association of exposure to phthalates with cardiometabolic risk factors in children and adolescents: a systematic review and meta-analysis. *Environ Sci Pollut Res*. 2019;**26**:35670–86.

- 26 Radke EG, Galizia A, Thayer KA, Cooper GS. Phthalate exposure and metabolic effects: a systematic review of the human epidemiological evidence. *Environ Int.* 2019;**132**:104768.
- 27 James-Todd TM, Huang T, Seely EW, Saxena AR. The association between phthalates and metabolic syndrome: the National Health and Nutrition Examination Survey 2001–2010. *Environ Health.* 2016;**15**:52.
- 28 Poursafa P, Dadvand P, Amin MM, Hajizadeh Y, Ebrahimpour K, Mansourian M, et al. Association of polycyclic aromatic hydrocarbons with cardiometabolic risk factors and obesity in children. *Environ Int.* 2018;**118**:203–10.
- 29 Zhang B, Pan B, Zhao X, Fu Y, Li X, Yang A, et al. The interaction effects of smoking and polycyclic aromatic hydrocarbons exposure on the prevalence of metabolic syndrome in coke oven workers. *Chemosphere.* 2020;**247**:125880.
- 30 Hu H, Kan H, Kearney GD, Xu X. Associations between exposure to polycyclic aromatic hydrocarbons and glucose homeostasis as well as metabolic syndrome in nondiabetic adults. *Sci Total Environ.* 2015;**505**:56–64.
- 31 La Merrill M, Emond C, Kim MJ, Antignac J-P, Le Bizec B, Clément K, et al. Toxicological function of adipose tissue: focus on persistent organic pollutants. *Environ Health Perspect.* 2013;**121**:162–9.
- 32 Audano M, Pedretti S, Caruso D, Crestani M, De Fabiani E, Mitro N. Regulatory mechanisms of the early phase of white adipocyte differentiation: an overview. *Cell Mol Life Sci.* 2022;**79**:139.
- 33 Costa V, Gallo MA, Letizia F, Aprile M, Casamassimi A, Ciccodicola A. PPAR γ : gene expression regulation and next-generation sequencing for unsolved issues. *PPAR Res.* 2010;**2010**:409168.
- 34 Longo M, Zatterale F, Naderi J, Parrillo L, Formisano P, Raciti GA, et al. Adipose tissue dysfunction as determinant of obesity-associated metabolic complications. *Int J Mol Sci.* 2019;**20**:E2358.
- 35 Chamorro-García R, Sahu M, Abbey RJ, Laude J, Pham N, Blumberg B. Transgenerational inheritance of increased fat depot size, stem cell reprogramming, and hepatic steatosis elicited by prenatal exposure to the obesogen tributyltin in mice. *Environ Health Perspect.* 2013;**121**:359–66.
- 36 Li B, Guo J, Xi Z, Xu J, Zuo Z, Wang C. Tributyltin in male mice disrupts glucose homeostasis as well as recovery after exposure: mechanism analysis. *Arch Toxicol.* 2017;**91**:3261–9.
- 37 Bertuloso BD, Podratz PL, Merlo E, de Araújo JFP, Lima LCF, de Miguel EC, et al. Tributyltin chloride leads to adiposity and impairs metabolic functions in the rat liver and pancreas. *Toxicol Lett.* 2015;**235**:45–59.
- 38 Yang M, Chen M, Wang J, Xu M, Sun J, Ding L, et al. Bisphenol A promotes adiposity and inflammation in a nonmonotonic dose-response way in 5-week-old male and female C57BL/6J mice fed a low-calorie diet. *Endocrinology.* 2016;**157**:2333–45.
- 39 Aaseth J, Javorac D, Djordjevic AB, Bulat Z, Skalny AV, Zaitseva IP, et al. The role of persistent organic pollutants in obesity: a review of laboratory and epidemiological studies. *Toxics.* 2022;**10**:65.
- 40 Jia Y, Liu T, Zhou L, Zhu J, Wu J, Sun D, et al. Effects of di-(2-ethylhexyl) phthalate on lipid metabolism by the JAK/STAT pathway in rats. *Int J Environ Res Public Health.* 2016;**13**:E1085.
- 41 Cohen IC, Cohenour ER, Harnett KG, Schuh SM. BPA, BPAF and TMBPF alter adipogenesis and fat accumulation in human mesenchymal stem cells, with implications for obesity. *Int J Mol Sci.* 2021;**22**:5363.
- 42 Reina-Pérez I, Olivas-Martínez A, Mustieles V, Ruiz-Ojeda FJ, Molina-Molina JM, Olea N, et al. Bisphenol F and bisphenol S promote lipid accumulation and adipogenesis in human adipose-derived stem cells. *Food Chem Toxicol.* 2021;**152**:112216.
- 43 Ramskov Tetzlaff CN, Svingen T, Vinggaard AM, Rosenmai AK, Taxvig C. Bisphenols B, E, F, and S and 4-cumylphenol induce lipid accumulation in mouse adipocytes similarly to bisphenol A. *Environ Toxicol.* 2020;**35**:543–52.
- 44 Brulport A, Le Corre L, Chagnon M-C. Chronic exposure of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) induces an obesogenic effect in C57BL/6J mice fed a high fat diet. *Toxicology.* 2017;**390**:43–52.
- 45 Wahlang B, Falkner KC, Gregory B, Ansert D, Young D, Conklin DJ, et al. Polychlorinated biphenyl 153 is a diet-dependent obesogen that worsens nonalcoholic fatty liver disease in male C57BL/6J mice. *J Nutr Biochem.* 2013;**24**:1587–95.
- 46 Wu H, Yu W, Meng F, Mi J, Peng J, Liu J, et al. Polychlorinated biphenyls-153 induces metabolic dysfunction through activation of ROS/NF- κ B signaling via downregulation of HNF1b. *Redox Biol.* 2017;**12**:300–10.
- 47 Laurenzana EM, Coslo DM, Vigilar MV, Roman AM, Omiecinski CJ. Activation of the constitutive androstane receptor by monophthalates. *Chem Res Toxicol.* 2016;**29**:1651–61.
- 48 Ellero-Simatos S, Claus SP, Benelli C, Forest C, Letourneur F, Cagnard N, et al. Combined transcriptomic-(1)H NMR metabolomic study reveals that monoethylhexyl phthalate stimulates adipogenesis and glyceroneogenesis in human adipocytes. *J Proteome Res.* 2011;**10**:5493–502.
- 49 Gadupudi G, Gourronc FA, Ludewig G, Robertson LW, Klingelutz AJ. PCB126 inhibits adipogenesis of human preadipocytes. *Toxicol In Vitro.* 2015;**29**:132–41.

- 50 Mullerova D, Pesta M, Dvorakova J, Cedikova M, Kulda V, Dvorak P, et al. Polychlorinated biphenyl 153 in lipid medium modulates differentiation of human adipocytes. *Physiol Res*. 2017;**66**:653–62.
- 51 Kim MJ, Pelloux V, Guyot E, Tordjman J, Bui L-C, Chevallier A, et al. Inflammatory pathway genes belong to major targets of persistent organic pollutants in adipose cells. *Environ Health Perspect*. 2012;**120**:508–14.
- 52 Ahmed S, Atlas E. Bisphenol S- and bisphenol A-induced adipogenesis of murine preadipocytes occurs through direct peroxisome proliferator-activated receptor gamma activation. *Int J Obes (Lond)*. 2016;**40**:1566–73.
- 53 Hu P, Chen X, Whitener RJ, Boder ET, Jones JO, Porollo A, et al. Effects of parabens on adipocyte differentiation. *Toxicol Sci*. 2013;**131**:56–70.
- 54 Liu S, Yang R, Yin N, Wang Y-L, Faiola F. Environmental and human relevant PFOS and PFOA doses alter human mesenchymal stem cell self-renewal, adipogenesis and osteogenesis. *Ecotoxicol Environ Saf*. 2019;**169**:564–72.
- 55 Shoucri BM, Hung VT, Chamorro-García R, Shioda T, Blumberg B. Retinoid X receptor activation during adipogenesis of female mesenchymal stem cells programs a dysfunctional adipocyte. *Endocrinology*. 2018;**159**:2863–83.
- 56 Kim SH, Park MJ. Phthalate exposure and childhood obesity. *Ann Pediatr Endocrinol Metab*. 2014;**19**:69–75.
- 57 Fang M, Webster TF, Ferguson PL, Stapleton HM. Characterizing the peroxisome proliferator-activated receptor (PPAR γ) ligand binding potential of several major flame retardants, their metabolites, and chemical mixtures in house dust. *Environ Health Perspect*. 2015;**123**:166–72.
- 58 Amir S, Shah STA, Mamoulakis C, Docea AO, Kalantzi O-I, Zachariou A, et al. Endocrine disruptors acting on estrogen and androgen pathways cause reproductive disorders through multiple mechanisms: a review. *Int J Environ Res Public Health*. 2021;**18**:1464.
- 59 TTB V, Jeung E-B. An evaluation of estrogenic activity of parabens using uterine calbindin-d9k gene in an immature rat model. *Toxicol Sci*. 2009;**112**:68–77.
- 60 Foulds CE, Treviño LS, York B, Walker CL. Endocrine-disrupting chemicals and fatty liver disease. *Nat Rev Endocrinol*. 2017;**13**:445–57.
- 61 Deierlein AL, Rock S, Park S. Persistent endocrine-disrupting chemicals and fatty liver disease. *Curr Environ Health Rep*. 2017;**4**:439–49.
- 62 Chen N, Shan Q, Qi Y, Liu W, Tan X, Gu J. Transcriptome analysis in normal human liver cells exposed to 2, 3, 3', 4, 4', 5-hexachlorobiphenyl (PCB 156). *Chemosphere*. 2020;**239**:124747.
- 63 Shan Q, Li H, Chen N, Qu F, Guo J. Understanding the multiple effects of PCBs on lipid metabolism. *Diabetes Metab Syndr Obes*. 2020;**13**:3691–702.
- 64 Liang X, Xie G, Wu X, Su M, Yang B. Effect of prenatal PFOS exposure on liver cell function in neonatal mice. *Environ Sci Pollut Res Int*. 2019;**26**:18240–6.
- 65 Marmugi A, Ducheix S, Lasserre F, Polizzi A, Paris A, Priymenko N, et al. Low doses of bisphenol A induce gene expression related to lipid synthesis and trigger triglyceride accumulation in adult mouse liver. *Hepatology*. 2012;**55**:395–407.
- 66 Küblbeck J, Niskanen J, Honkakoski P. Metabolism-disrupting chemicals and the constitutive androstane receptor CAR. *Cells*. 2020;**9**:E2306.
- 67 Wyde ME, Kirwan SE, Zhang F, Laughter A, Hoffman HB, Bartolucci-Page E, et al. Di-n-butyl phthalate activates constitutive androstane receptor and pregnane X receptor and enhances the expression of steroid-metabolizing enzymes in the liver of rat fetuses. *Toxicol Sci*. 2005;**86**:281–90.
- 68 Eveillard A, Mselli-Lakhal L, Mogha A, Lasserre F, Polizzi A, Pascussi J-M, et al. Di-(2-ethylhexyl)-phthalate (DEHP) activates the constitutive androstane receptor (CAR): a novel signalling pathway sensitive to phthalates. *Biochem Pharmacol*. 2009;**77**:1735–46.
- 69 Bjork JA, Butenhoff JL, Wallace KB. Multiplicity of nuclear receptor activation by PFOA and PFOS in primary human and rodent hepatocytes. *Toxicology*. 2011;**288**:8–17.
- 70 Kojima H, Takeuchi S, Sanoh S, Okuda K, Kitamura S, Uramaru N, et al. Profiling of bisphenol A and eight its analogues on transcriptional activity via human nuclear receptors. *Toxicology*. 2019;**413**:48–55.
- 71 Yue S, Yu J, Kong Y, Chen H, Mao M, Ji C, et al. Metabolomic modulations of HepG2 cells exposed to bisphenol analogues. *Environ Int*. 2019;**129**:59–67.
- 72 Kubota T, Kubota N, Kadowaki T. Imbalanced insulin actions in obesity and type 2 diabetes: key mouse models of insulin signaling pathway. *Cell Metab*. 2017;**25**:797–810.
- 73 Mu W, Cheng X-F, Liu Y, Lv Q-Z, Liu G-L, Zhang J-G, et al. Potential nexus of non-alcoholic fatty liver disease and type 2 diabetes mellitus: insulin resistance between hepatic and peripheral tissues. *Front Pharmacol*. 2018;**9**:1566.
- 74 Rotondo E, Chiarelli F. Endocrine-disrupting chemicals and insulin resistance in children. *Biomedicine*. 2020;**8**:E137.
- 75 Sargis RM. The hijacking of cellular signaling and the diabetes epidemic: mechanisms of environmental disruption of insulin action and glucose homeostasis. *Diabetes Metab J*. 2014;**38**:13–24.
- 76 Jayashree S, Indumathi D, Akilavalli N, Sathish S, Selvaraj J, Balasubramanian K. Effect of bisphenol-A

- on insulin signal transduction and glucose oxidation in liver of adult male albino rat. *Environ Toxicol Pharmacol.* 2013;**35**:300–10.
- 77 Yang B, Zou W, Hu Z, Liu F, Zhou L, Yang S, et al. Involvement of oxidative stress and inflammation in liver injury caused by perfluorooctanoic acid exposure in mice. *Biomed Res Int.* 2014;**2014**:409837.
 - 78 Huc L, Lemarié A, Guéraud F, Héliers-Toussaint C. Low concentrations of bisphenol A induce lipid accumulation mediated by the production of reactive oxygen species in the mitochondria of HepG2 cells. *Toxicol In Vitro.* 2012;**26**:709–17.
 - 79 Han R, Zhang F, Wan C, Liu L, Zhong Q, Ding W. Effect of perfluorooctane sulfonate-induced Kupffer cell activation on hepatocyte proliferation through the NF- κ B/TNF- α /IL-6-dependent pathway. *Chemosphere.* 2018;**200**:283–94.
 - 80 Han M, Liu X, Liu S, Su G, Fan X, Chen J, et al. 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) induces hepatic stellate cell (HSC) activation and liver fibrosis in C57BL6 mouse via activating Akt and NF- κ B signaling pathways. *Toxicol Lett.* 2017;**273**:10–9.
 - 81 Shi H, Jan J, Hardesty JE, Falkner KC, Prough RA, Balamurugan AN, et al. Polychlorinated biphenyl exposures differentially regulate hepatic metabolism and pancreatic function: implications for nonalcoholic steatohepatitis and diabetes. *Toxicol Appl Pharmacol.* 2019;**363**:22–33.
 - 82 Soriano S, Alonso-Magdalena P, García-Arévalo M, Novials A, Muhammed SJ, Salehi A, et al. Rapid insulinotropic action of low doses of bisphenol-A on mouse and human islets of Langerhans: role of estrogen receptor β . *PLoS One.* 2012;**7**:e31109.
 - 83 Martinez-Pinna J, Marroqui L, Hmadcha A, Lopez-Beas J, Soriano S, Villar-Pazos S, et al. Oestrogen receptor β mediates the actions of bisphenol-A on ion channel expression in mouse pancreatic beta cells. *Diabetologia.* 2019;**62**:1667–80.
 - 84 Alonso-Magdalena P, Roperio AB, Carrera MP, Cederroth CR, Baquie M, Gauthier BR, et al. Pancreatic insulin content regulation by the estrogen receptor ER α . *PLoS One.* 2008;**3**:e2069.
 - 85 Alonso-Magdalena P, Morimoto S, Ripoll C, Fuentes E, Nadal A. The estrogenic effect of bisphenol A disrupts pancreatic beta-cell function in vivo and induces insulin resistance. *Environ Health Perspect.* 2006;**114**:106–12.
 - 86 Alonso-Magdalena P, Vieira E, Soriano S, Menes L, Burks D, Quesada I, et al. Bisphenol A exposure during pregnancy disrupts glucose homeostasis in mothers and adult male offspring. *Environ Health Perspect.* 2010;**118**:1243–50.
 - 87 Li L, Wang F, Zhang J, Wang K, De X, Li L, et al. Typical phthalic acid esters induce apoptosis by regulating the PI3K/Akt/Bcl-2 signaling pathway in rat insulinoma cells. *Ecotoxicol Environ Saf.* 2021;**208**:111461.
 - 88 Li J, Zheng L, Wang X, Yao K, Shi L, Sun X, et al. Taurine protects INS-1 cells from apoptosis induced by Di(2-ethylhexyl) phthalate via reducing oxidative stress and autophagy. *Toxicol Mech Methods.* 2019;**29**:445–56.
 - 89 Sun X, Lin Y, Huang Q, Shi J, Qiu L, Kang M, et al. Di(2-ethylhexyl) phthalate-induced apoptosis in rat INS-1 cells is dependent on activation of endoplasmic reticulum stress and suppression of antioxidant protection. *J Cell Mol Med.* 2015;**19**:581–94.
 - 90 She Y, Jiang L, Zheng L, Zuo H, Chen M, Sun X, et al. The role of oxidative stress in DNA damage in pancreatic β cells induced by di-(2-ethylhexyl) phthalate. *Chem Biol Interact.* 2017;**265**:8–15.
 - 91 Qin W-P, Cao L-Y, Li C-H, Guo L-H, Colbourne J, Ren X-M. Perfluoroalkyl substances stimulate insulin secretion by islet β cells via G protein-coupled receptor 40. *Environ Sci Technol.* 2020;**54**:3428–36.
 - 92 Zhang L, Duan X, Sun W, Sun H. Perfluorooctane sulfonate acute exposure stimulates insulin secretion via GPR40 pathway. *Sci Total Environ.* 2020;**726**:138498.
 - 93 Duan X, Sun W, Sun H, Zhang L. Perfluorooctane sulfonate continual exposure impairs glucose-stimulated insulin secretion via SIRT1-induced upregulation of UCP2 expression. *Environ Pollut.* 2021;**278**:116840.
 - 94 Wan HT, Cheung LY, Chan TF, Li M, Lai KP, Wong CKC. Characterization of PFOS toxicity on in-vivo and ex-vivo mouse pancreatic islets. *Environ Pollut.* 2021;**289**:117857.
 - 95 Chen Y-W, Lan K-C, Tsai J-R, Weng T-I, Yang C-Y, Liu S-H. Tributyltin exposure at noncytotoxic doses dysregulates pancreatic β -cell function in vitro and in vivo. *Arch Toxicol.* 2017;**91**:3135–44.
 - 96 Huang C-F, Yang C-Y, Tsai J-R, Wu C-T, Liu S-H, Lan K-C. Low-dose tributyltin exposure induces an oxidative stress-triggered JNK-related pancreatic β -cell apoptosis and a reversible hypoinsulinemic hyperglycemia in mice. *Sci Rep.* 2018;**8**:5734.
 - 97 Ibrahim M, MacFarlane EM, Matteo G, Hoyeck MP, Rick KRC, Farokhi S, et al. Functional cytochrome P450 1A enzymes are induced in mouse and human islets following pollutant exposure. *Diabetologia.* 2020;**63**:162–78.
 - 98 Martino L, Novelli M, Masini M, Chimenti D, Piaggi S, Masiello P, et al. Dehydroascorbate protection against dioxin-induced toxicity in the beta-cell line INS-1E. *Toxicol Lett.* 2009;**189**:27–34.
 - 99 Novelli M, Beffy P, Masini M, Vantaggiato C, Martino L, Marselli L, et al. Selective beta-cell toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin on isolated pancreatic islets. *Chemosphere.* 2021;**265**:129103.

- 100 Cave MC, Clair HB, Hardesty JE, Falkner KC, Feng W, Clark BJ, et al. Nuclear receptors and nonalcoholic fatty liver disease. *Biochim Biophys Acta*. 2016;**1859**:1083–99.
- 101 Vanni R, Bussuan RM, Rombaldi RL, Arbex AK. Endocrine disruptors and the induction of insulin resistance. *Curr Diabetes Rev*. 2021;**17**:e102220187107.
- 102 Darbre PD. Endocrine disruptors and obesity. *Curr Obes Rep*. 2017;**6**:18–27.
- 103 Wilson ID, Nicholson JK. Gut microbiome interactions with drug metabolism, efficacy, and toxicity. *Transl Res*. 2017;**179**:204–22.
- 104 Hampl R, Stárka L. Endocrine disruptors and gut microbiome interactions. *Physiol Res*. 2020;**69**:S211–23.
- 105 Sarni ROS, Kochi C, Suano-Souza FI. Childhood obesity: an ecological perspective. *J Pediatr (Rio J)*. 2022;**98**(Suppl 1):S38–46.
- 106 Santos-Marcos JA, Haro C, Vega-Rojas A, Alcalá-Díaz JF, Molina-Abril H, Leon-Acuña A, et al. Sex differences in the gut microbiota as potential determinants of gender predisposition to disease. *Mol Nutr Food Res*. 2019;**63**:e1800870.
- 107 Tripathi A, Debelius J, Brenner DA, Karin M, Loomba R, Schnabl B, et al. The gut-liver axis and the intersection with the microbiome. *Nat Rev Gastroenterol Hepatol*. 2018;**15**:397–411.
- 108 Liu Y, Yao Y, Li H, Qiao F, Wu J, Du Z, et al. Influence of endogenous and exogenous estrogenic endocrine on intestinal microbiota in zebrafish. *PLoS One*. 2016;**11**:e0163895.
- 109 Lui JB, McGinn LS, Chen Z. Gut microbiota amplifies host-intrinsic conversion from the CD8 T cell lineage to CD4 T cells for induction of mucosal immune tolerance. *Gut Microbes*. 2016;**7**:40–7.
- 110 Koestel ZL, Backus RC, Tsuruta K, Spollen WG, Johnson SA, Javurek AB, et al. Bisphenol A (BPA) in the serum of pet dogs following short-term consumption of canned dog food and potential health consequences of exposure to BPA. *Sci Total Environ*. 2017;**579**:1804–14.
- 111 Reddivari L, Veeramachaneni DNR, Walters WA, Lozupone C, Palmer J, Hewage MKK, et al. Perinatal bisphenol A exposure induces chronic inflammation in rabbit offspring via modulation of gut bacteria and their metabolites. *mSystems*. 2017;**2**:e00093-17.
- 112 Javurek AB, Spollen WG, Johnson SA, Bivens NJ, Bromert KH, Givan SA, et al. Effects of exposure to bisphenol A and ethinyl estradiol on the gut microbiota of parents and their offspring in a rodent model. *Gut Microbes*. 2016;**7**:471–85.
- 113 Hu J, Raikhel V, Gopalakrishnan K, Fernandez-Hernandez H, Lambertini L, Manservigi F, et al. Effect of postnatal low-dose exposure to environmental chemicals on the gut microbiome in a rodent model. *Microbiome*. 2016;**4**:26.
- 114 Wang C, Yue S, Hao Z, Ren G, Lu D, Zhang Q, et al. Pubertal exposure to the endocrine disruptor mono-2-ethylhexyl ester at body burden level caused cholesterol imbalance in mice. *Environ Pollut*. 2019;**244**:657–66.
- 115 Jin Y, Zeng Z, Wu Y, Zhang S, Fu Z. Oral exposure of mice to carbendazim induces hepatic lipid metabolism disorder and gut microbiota dysbiosis. *Toxicol Sci*. 2015;**147**:116–26.
- 116 Zhang L, Nichols RG, Correll J, Murray IA, Tanaka N, Smith PB, et al. Persistent organic pollutants modify gut microbiota–host metabolic homeostasis in mice through aryl hydrocarbon receptor activation. *Environ Health Perspect*. 2015;**123**:679–88.
- 117 Kan H, Zhao F, Zhang X-X, Ren H, Gao S. Correlations of gut microbial community shift with hepatic damage and growth inhibition of *Carassius auratus* induced by pentachlorophenol exposure. *Environ Sci Technol*. 2015;**49**:11894–902.
- 118 Chi Y, Lin Y, Lu Y, Huang Q, Ye G, Dong S. Gut microbiota dysbiosis correlates with a low-dose PCB126-induced dyslipidemia and non-alcoholic fatty liver disease. *Sci Total Environ*. 2019;**653**:274–82.
- 119 Liang Y, Liu D, Zhan J, Luo M, Han J, Wang P, et al. New insight into the mechanism of POP-induced obesity: evidence from DDE-altered microbiota. *Chemosphere*. 2020;**244**:125123.
- 120 Feng P, Ye Z, Kakade A, Virk A, Li X, Liu P. A review on gut remediation of selected environmental contaminants: possible roles of probiotics and gut microbiota. *Nutrients*. 2018;**11**:22.
- 121 Malaisé Y, Menard S, Cartier C, Gaultier E, Lasserre F, Lencina C, et al. Gut dysbiosis and impairment of immune system homeostasis in perinatally-exposed mice to bisphenol A precede obese phenotype development. *Sci Rep*. 2017;**7**:14472.
- 122 Fader KA, Nault R, Zhang C, Kumagai K, Harkema JR, Zacharewski TR. 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD)-elicited effects on bile acid homeostasis: alterations in biosynthesis, enterohepatic circulation, and microbial metabolism. *Sci Rep*. 2017;**7**:5921.
- 123 Natividad JM, Agus A, Planchais J, Lamas B, Jarry AC, Martin R, et al. Impaired aryl hydrocarbon receptor ligand production by the gut microbiota is a key factor in metabolic syndrome. *Cell Metab*. 2018;**28**:737–49.e4.
- 124 Fader KA, Nault R, Doskey CM, Fling RR, Zacharewski TR. 2,3,7,8-Tetrachlorodibenzo-p-dioxin abolishes circadian regulation of hepatic metabolic activity in mice. *Sci Rep*. 2019;**9**:6514.
- 125 Tilg H, Cani PD, Mayer EA. Gut microbiome and liver diseases. *Gut*. 2016;**65**:2035–44.
- 126 Weger BD, Rawashdeh O, Gachon F. At the intersection of microbiota and circadian clock: are

- sexual dimorphism and growth hormones the missing link to pathology?: circadian clock and microbiota: potential effect on growth hormone and sexual development. *Bioessays*. 2019;**41**:e1900059.
- 127 Zhou C, Verma S, Blumberg B. The steroid and xenobiotic receptor (SXR), beyond xenobiotic metabolism. *Nucl Recept Signal*. 2009;**7**:e001.
 - 128 Kim S, Choi S, Dutta M, Asubonteng JO, Polunas M, Goedken M, et al. Pregnane X receptor exacerbates nonalcoholic fatty liver disease accompanied by obesity- and inflammation-prone gut microbiome signature. *Biochem Pharmacol*. 2021;**193**:114698.
 - 129 Shin K-H, Choi MH, Lim KS, Yu K-S, Jang I-J, Cho J-Y. Evaluation of endogenous metabolic markers of hepatic CYP3A activity using metabolic profiling and midazolam clearance. *Clin Pharmacol Ther*. 2013;**94**:601–9.
 - 130 Zanger UM, Schwab M. Cytochrome P450 enzymes in drug metabolism: regulation of gene expression, enzyme activities, and impact of genetic variation. *Pharmacol Ther*. 2013;**138**:103–41.
 - 131 Sui Y, Helsley RN, Park S-H, Song X, Liu Z, Zhou C. Intestinal pregnane X receptor links xenobiotic exposure and hypercholesterolemia. *Mol Endocrinol*. 2015;**29**:765–76.
 - 132 Grün F, Blumberg B. Minireview: the case for obesogens. *Mol Endocrinol*. 2009;**23**:1127–34.
 - 133 Gluckman PD, Hanson MA. Developmental origins of disease paradigm: a mechanistic and evolutionary perspective. *Pediatr Res*. 2004;**56**:311–7.
 - 134 MacKay H, Patterson ZR, Abizaid A. Perinatal exposure to low-dose bisphenol-A disrupts the structural and functional development of the hypothalamic feeding circuitry. *Endocrinology*. 2017;**158**:768–77.
 - 135 Tian S, Yan S, Meng Z, Huang S, Sun W, Jia M, et al. New insights into bisphenols induced obesity in zebrafish (*Danio rerio*): Activation of cannabinoid receptor CB1. *Journal of Hazardous Materials*. 2021;**418**:126100.
 - 136 Rönn M, Lind L, Örborg J, Kullberg J, Söderberg S, Larsson A, et al. Bisphenol A is related to circulating levels of adiponectin, leptin and ghrelin, but not to fat mass or fat distribution in humans. *Chemosphere*. 2014;**112**:42–8.
 - 137 Ariemma F, D'Esposito V, Liguoro D, Oriente F, Cabaro S, Liotti A, et al. Low-dose bisphenol-A impairs adipogenesis and generates dysfunctional 3T3-L1 adipocytes. *PLoS One*. 2016;**11**:e0150762.
 - 138 Schmidt J-S, Schaedlich K, Fiandanese N, Pocar P, Fischer B. Effects of di(2-ethylhexyl) phthalate (DEHP) on female fertility and adipogenesis in C3H/N mice. *Environ Health Perspect*. 2012;**120**:1123–9.
 - 139 Hao C. Perinatal exposure to diethyl-hexyl-phthalate induces obesity in mice. *Front Biosci*. 2013;**5**:725–33.
 - 140 Fougerat A, Schoiswohl G, Polizzi A, Régner M, Wagner C, Smati S, et al. ATGL-dependent white adipose tissue lipolysis controls hepatocyte PPAR α activity. *Cell Rep*. 2022;**39**:110910.
 - 141 Simcox J, Geoghegan G, Maschek JA, Bensard CL, Pasquali M, Miao R, et al. Global analysis of plasma lipids identifies liver-derived acylcarnitines as a fuel source for brown fat thermogenesis. *Cell Metab*. 2017;**26**:509–22.e6.
 - 142 Wang B, Tsakiridis EE, Zhang S, Llanos A, Desjardins EM, Yabut JM, et al. The pesticide chlorpyrifos promotes obesity by inhibiting diet-induced thermogenesis in brown adipose tissue. *Nat Commun*. 2021;**12**:5163.
 - 143 Manikkam M, Tracey R, Guerrero-Bosagna C, Skinner MK. Plastics derived endocrine disruptors (BPA, DEHP and DBP) induce epigenetic transgenerational inheritance of obesity, reproductive disease and sperm epimutations. *PLoS One*. 2013;**8**:e55387.
 - 144 Manikkam M, Haque MM, Guerrero-Bosagna C, Nilsson EE, Skinner MK. Pesticide methoxychlor promotes the epigenetic transgenerational inheritance of adult-onset disease through the female germline. *PLoS One*. 2014;**9**:e102091.
 - 145 King SE, McBirney M, Beck D, Sadler-Riggelman I, Nilsson E, Skinner MK. Sperm epimutation biomarkers of obesity and pathologies following DDT induced epigenetic transgenerational inheritance of disease. *Environ Epigenet*. 2019;**5**:dvz008.
 - 146 Skinner MK, Manikkam M, Tracey R, Guerrero-Bosagna C, Haque M, Nilsson EE. Ancestral dichlorodiphenyltrichloroethane (DDT) exposure promotes epigenetic transgenerational inheritance of obesity. *BMC Med*. 2013;**11**:228.
 - 147 Grün F, Blumberg B. Perturbed nuclear receptor signaling by environmental obesogens as emerging factors in the obesity crisis. *Rev Endocr Metab Disord*. 2007;**8**:161–71.
 - 148 le Maire A, Grimaldi M, Roecklin D, Dagnino S, Vivat-Hannah V, Balaguer P, et al. Activation of RXR-PPAR heterodimers by organotin environmental endocrine disruptors. *EMBO Rep*. 2009;**10**:367–73.
 - 149 Grün F, Watanabe H, Zamanian Z, Maeda L, Arima K, Cubacha R, et al. Endocrine-disrupting organotin compounds are potent inducers of adipogenesis in vertebrates. *Mol Endocrinol*. 2006;**20**:2141–55.
 - 150 Chamorro-Garcia R, Diaz-Castillo C, Shoucri BM, Käch H, Leavitt R, Shioda T, et al. Ancestral perinatal obesogen exposure results in a transgenerational thrifty phenotype in mice. *Nat Commun*. 2017;**8**:2012.
 - 151 Diaz-Castillo C, Chamorro-Garcia R, Shioda T, Blumberg B. Transgenerational self-reconstruction of disrupted chromatin organization after exposure to an environmental stressor in mice. *Sci Rep*. 2019;**9**:13057.

- 152 Qian Y, Liao J, Suen AHC, Lee AWT, Chung HS, Tang NLS, et al. Comparative analysis of single-cell parallel sequencing approaches in oocyte application. *Int J Biochem Cell Biol.* 2019;**107**:1–5.
- 153 Camsari C, Folger JK, Rajput SK, McGee D, Latham KE, Smith GW. Transgenerational effects of periconception heavy metal administration on adipose weight and glucose homeostasis in mice at maturity. *Toxicol Sci.* 2019;**168**:610–9.
- 154 Forgacs AL, Kent MN, Makley MK, Mets B, DelRaso N, Jahns GL, et al. Comparative metabolomic and genomic analyses of TCDD-elicited metabolic disruption in mouse and rat liver. *Toxicol Sci.* 2012;**125**:41–55.
- 155 Lee JH, Wada T, Febbraio M, He J, Matsubara T, Lee MJ, et al. A novel role for the dioxin receptor in fatty acid metabolism and hepatic steatosis. *Gastroenterology.* 2010;**139**:653–63.
- 156 Sui Y, Park S-H, Helsley RN, Sunkara M, Gonzalez FJ, Morris AJ, et al. Bisphenol A increases atherosclerosis in pregnane X receptor-humanized ApoE deficient mice. *J Am Heart Assoc.* 2014;**3**:e000492.
- 157 Ding S, Yuan C, Si B, Wang M, Da S, Bai L, et al. Combined effects of ambient particulate matter exposure and a high-fat diet on oxidative stress and steatohepatitis in mice. *PLoS One.* 2019;**14**:e0214680.
- 158 Drakvik E, Altenburger R, Aoki Y, Backhaus T, Bahadori T, Barouki R, et al. Statement on advancing the assessment of chemical mixtures and their risks for human health and the environment. *Environ Int.* 2020;**134**:105267.
- 159 Teeguarden JG, Tan Y-M, Edwards SW, Leonard JA, Anderson KA, Corley RA, et al. Completing the link between exposure science and toxicology for improved environmental health decision making: the aggregate exposure pathway framework. *Environ Sci Technol.* 2016;**50**:4579–86.