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Review

Regulation of food intake by oleoylethanolamide

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Abstract. Oleoylethanolamide (OEA), the naturally occurring amide of ethanolamine and oleic acid, is an endogenous lipid that modulates feeding, body weight and lipid metabolism by binding with high affinity to the ligand-activated transcription factor, peroxisome proliferator-activated receptor-alpha (PPAR-α). In the present article, we describe the biochemical pathways responsible for the initiation and termination of OEA signaling, and outline the pharmacological properties of this compound in relation to its ability to activate PPAR-α. Finally, we discuss the possible role of OEA as a peripheral satiety hormone.

Key words. Oleoylethanolamide; palmitoylethanolamide; peroxisome proliferator-activated receptor-alpha (PPAR-α); lipid; feeding; satiety; energy balance; metabolism.

Introduction

The fatty-acid ethanolamides (FAEs) are a family of naturally occurring lipids that are present in both plant and animal tissues [1,2]. Their physiological significance was first recognized nearly 50 years ago, with the discovery of the antiallergic effects of palmitoylethanolamide [1] (PEA, fig. 1), the amide of palmitic acid and ethanolamine. Nevertheless, these compounds received little attention until another member of the FAE family, arachidonoylethanolamide (anandamide, fig. 1), was found to serve as an endogenous ligand for cannabinoid receptors [3, 4], the G-protein-coupled receptors targeted by Δ^9-tetrahydrocannabinol in marijuana [5, 6]. The important roles played by anandamide in neural transmission (reviewed in [7]) renewed interest in the FAE family of lipids, leading in turn to the discovery of a number of biological effects exerted by these biomolecules, which are not mediated by cannabinoid receptors (reviewed in [8]).

In the present article, we review the roles of FAEs, and in particular oleoylethanolamide (OEA, fig. 1), in the regulation of energy balance. After a brief outline of the biochemical routes responsible for the formation and deactivation of OEA, we describe the pharmacological effects of this compound on feeding and body weight, as well as its ability to act as a high-affinity agonist for the ligand-activated transcription factor, peroxisome-proliferator activated receptor-alpha (PPAR-α). We conclude by discussing the possible physiological function of OEA as an intestinal satiety hormone.

OEA synthesis

Unlike classical neurotransmitters and hormones, which are stored in and released from secretory vesicles, OEA and other FAEs are produced through on-demand synthe-

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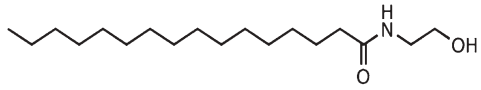
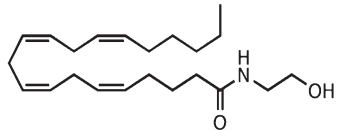
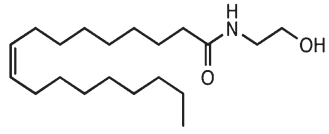


Palmitoylethanolamide (PEA)



Arachidonoylethanolamide (AEA)



Oleoylethanolamide (OEA)

Figure 1. Structures of various fatty-acid ethanolamides: oleoylethanolamide (OEA), palmitoylethanolamide (PEA) and anandamide (arachidonoylethanolamide, AEA).

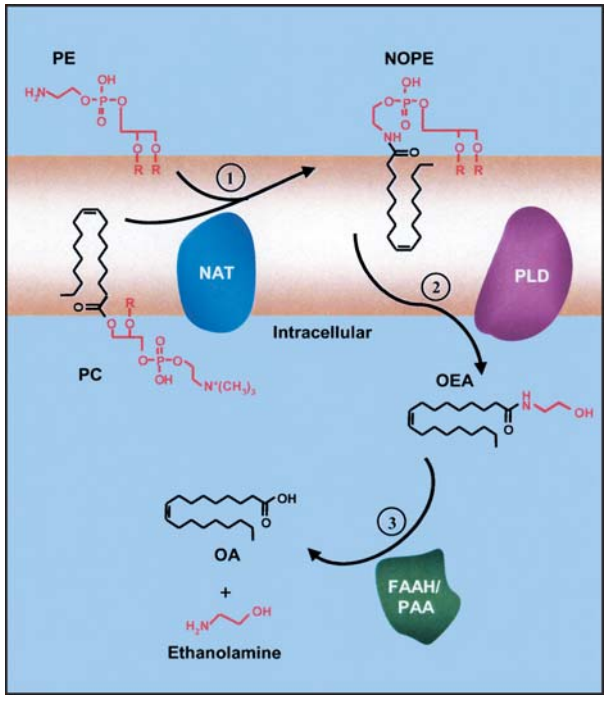


Figure 2. Fatty-acid ethanolamide biosynthesis and deactivation, as exemplified by OEA. (1) A calcium-dependent N-acyltransferase activity catalyzes the transfer of oleic acid from the *sn*-1 position of phosphatidylcholine (PC) to the free amino group of phosphatidylethanolamine (PE) to form N-oleoyl-phosphatidylethanolamine (NOPE). (2) NOPE is cleaved by NAPE-specific phospholipase D (PLD) to release OEA. (3) OEA is hydrolyzed by fatty-acid amide hydrolase (FAAH) or PEA-preferring acid amidase (PAA) to yield oleic acid and ethanolamine.

sis from a phospholipid precursor found within the membrane lipid bilayer. In mammalian tissues, two concerted biochemical reactions mediate this process (Fig. 2). The first consists in the transfer of a fatty-acid residue from the *sn*-1 position of phosphatidylcholine to the free amine of phosphatidylethanolamine (PE). This reaction is catalyzed by a calcium and cyclic AMP-regulated N-acyltransferase (NAT) activity, which remains to be molecularly cloned, and yields a diverse family of N-acyl-phosphatidylethanolamines (NAPEs, fig. 2). The second reaction is catalyzed by NAPE-specific phospholipase D (PLD), which cleaves NAPEs to produce FAEs [9, 10]. The first NAPE-specific PLD was recently identified [11]. This lipid hydrolase shares little sequence homology with other members of the PLD family and recognizes multiple NAPE species, producing OEA along with other FAEs (fig. 2) [11].

An additional mechanism of FAE biosynthesis has been proposed, which involves the hydrolysis of NAPE to N-acyl-lyso-PE (lyso-NAPE), catalyzed by a secretory phospholipase A_2 ($sPLA_2$), followed by the cleavage of lyso-NAPE to FAEs, catalyzed by a lysophospholipase D (lyso-PLD) [12]. The relative contribution of the NAT/PLD and $sPLA_2$/lyso-PLD pathways to OEA formation is unknown at present.

OEA deactivation

In mammalian tissues, OEA is primarily eliminated through enzymatic hydrolysis to oleic acid and ethanolamine. Two enzymes have been identified which may catalyze this reaction: fatty-acid amide hydrolase (FAAH) [13, 14] and PEA-preferring acid amidase (PAA) [15].

FAAH is a membrane-bound intracellular serine hydrolase that catalyzes the hydrolysis of all FAEs, including OEA (fig. 2). It is present in all mammalian tissues, but is particularly abundant in brain and liver [16]. Although most investigations have focused on the roles of FAAH in anandamide degradation, OEA is also an excellent substrate for this enzyme [17–19]. In fact, mice lacking the *faah* gene have dramatically reduced OEA hydrolysis and increased OEA levels in brain and liver tissues [20, 21].

Unlike FAAH, PAA activity is most abundant in the rodent lung, spleen and small intestine [15]. In the presence of detergent this activity displays a marked preference for PEA as a substrate; however, when detergents are omitted from the assay PAA recognizes all FAEs, suggesting that it may play a broad role in the deactivation of these compounds by intact cells [15].

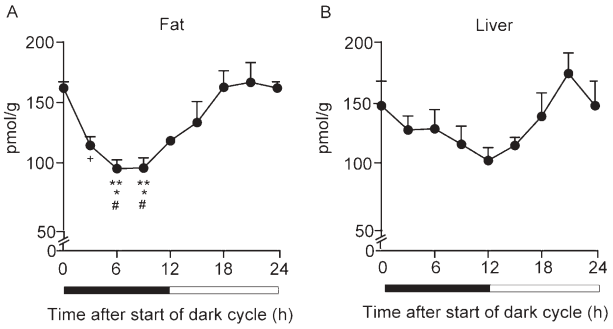


Figure 3. Diurnal fluctuations of OEA in rat white adipose and liver tissue. OEA levels in (*A*) epididymal fat or (*B*) liver from free-feeding male Wistar rats. OEA was measured by isotope-dilution high-performance liquid chromatography/mass spectrometry (HPLC/MS) [71] at 3-h intervals following the onset of the dark phase (17:00–05:00 h, closed bars). *, $p < 0.05$: 0 vs. 6h, 0 vs. 9 h; **, $p < 0.01$: 6 vs. 18 h, 9 vs. 18 h; #, $p < 0.05$: 6 vs. 21 h, 9 vs. 21 h; +, $p < 0.05$: 3 vs. 18 h. No significant differences were detected in liver tissue. ANOVA (analysis of variance) followed by Bonferroni's multiple comparison test (n = 4–5).

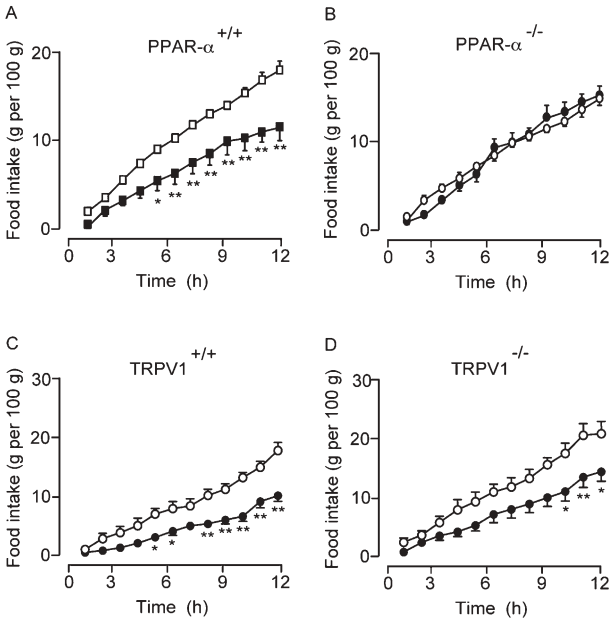


Figure 4. OEA inhibits feeding. (*A–B*), Effects of vehicle (70% DMSO/30% saline, 1 ml kg^{-1}) OEA (10 mg kg^{-1}, i.p, administered 15 min before the onset of dark) on food intake in (*A*) wild-type and (*B*) PPAR-α-null mice (n = 5–11). Reproduced from [27]. (*C–D*), Effects of vehicle or OEA (10 mg kg^{-1}, i.p) on food intake in (*C*) wild-type and (*D*) TRPV1-null mice (n = 7–10). ANOVA followed by a Dunnett's test. Significantly different from vehicle: *, $p < 0.05$; **, $p < 0.01$.

Regulation of OEA levels

In primary cultures of rat brain neurons, the synthesis of OEA and its precursor NAPE is stimulated by a variety of pharmacological treatments that elevate intracellular calcium levels. These include membrane-depolarizing agents such as kainate (a glutamate receptor agonist) and calcium ionophores such as ionomycin [4, 10, 22–24]. In addition, OEA formation may be elicited in cortical neurons by coactivation of N-methyl-D-aspartate (NMDA)-type glutamate receptors and cholinergic muscarinic receptors [24]. However, the physiological stimuli that control brain OEA levels in live animals remain unexplored.

In the duodenum and jejunum of rats and mice, OEA levels change in response to nutrient status: they are lower in food-deprived than free-feeding animals, and return to baseline values upon refeeding [25]. These alterations are restricted to the upper intestine – a structure intimately involved in the control of feeding behavior [26] – and are accompanied by parallel changes in NAT activity within the upper region of the intestine, which is suggestive of a stimulatory effect of feeding on OEA synthesis [25].

OEA levels in the rodent small intestine also display diurnal fluctuations. They are higher during the daytime, when animals are satiated, and lower during the night, when they are awake and actively feeding [27]. A parallel diurnal cycle is present in rat white adipose tissue, but not in the liver (fig. 3). The molecular mechanisms that control OEA turnover in response to feeding and diurnal cycles have not been characterized.

OEA regulates feeding behavior

The finding that intestinal OEA levels are elevated in the post-absorptive state suggests that this lipid amide may contribute to the regulation of feeding behavior. In support of this possibility, administration of OEA to rats or mice was found to produce a dose and time-dependent inhibition of food intake [25, 27–30] (fig. 4a). This effect is both structurally and behaviorally selective: close structural analogues of OEA are either less potent than OEA at reducing feeding (e.g. PEA) or are completely ineffective (e.g. anandamide or oleic acid) [25]. In fact, anandamide stimulates feeding in partially satiated animals via activation of cannabinoid receptors located both in the brain and peripheral tissues [31, 32]. Importantly, OEA is inactive in a rat model of conditioned taste aversion, does not cause anxiety-like behaviors in the elevated plus-maze test and does not change plasma corticosterone levels, indicating that its anorexiant effects cannot be accounted for by malaise anxiety or stress [25, 33]. As discussed later, the ability of OEA to reduce feeding may be predominantly, if not exclusively, ascribed to an enhanced state of satiety.

At doses higher than those required to produce hypophagia, OEA causes moderate hypolocomotion [25]. This effect is unlikely to contribute to the anorexi-

ant properties of OEA, however, not only for its clear dose separation, but also because OEA has no effect on water intake or need-induced sodium appetite [25, 33].

OEA is an endogenous PPAR-α agonist

Although the structural and behavioral specificity of OEA and other FAEs suggest that these biomolecules may interact with a selective receptor site [25, 34], the identity of this putative receptor has long remained elusive. The fact that OEA resembles lipid compounds that activate the PPAR family of nuclear receptors, such as non-esterified fatty acids [35], prompted us to explore the possibility that this molecule might target one of these ligand-activated transcription factors.

PPAR receptors, first identified in 1992 [36], are a family of ligand-activated transcription factors that comprises three known isoforms, PPAR-α, PPAR-δ (also called PPAR-β) and PPAR-γ. PPAR-α receptors, the molecular target of the fibrate class of antihyperlipidemic drugs, induce a variety of transcriptional changes that result in increased fatty-acid catabolism, reduced blood lipid levels and lowered body-weight gain (reviewed in [35, 37]). They also regulate inflammation through transcriptional and non-transcriptional actions [38]. Like PPAR-α, PPAR-δ stimulates adipose tissue utilization and protects animals from diet-induced obesity [39]. In addition, activation of this receptor modulates muscle function by enhancing the transformation of type II to type I fibers and stimulating mitochondrial biogenesis [40]. In contrast to PPAR-α and PPAR-δ, PPAR-γ activation facilitates fat storage, promotes adipocyte maturation and improves insulin sensitivity (reviewed in [41]).

OEA activates PPAR-α in standard transactivation assays with a half-maximal concentration (EC_{50}) of 120 nM and PPAR-δ with an EC_{50} of 1.1 μM, whereas it does not engage PPAR-γ or retinoid-X receptor – an obligatory partner in PPAR-activated transcription [27]. As illustrated in table 1, the potency of OEA to activate PPAR-α exceeds that of other natural ligands and even rivals that of many synthetic agonists. Activation of PPAR-α by OEA is structurally selective, since related FAEs – including stearoylethanolamide (18:0), myristoylethanolamide (14:0) and anandamide (20:4) – have no effect on this receptor [27]. Consistent with its ability to activate PPAR-α, saturation binding experiments show that OEA associates with the purified ligand-binding domain (LBD) of PPAR-α with a dissociation constant (K_D) of approximately 40 nM [27]. Furthermore, in competitive binding studies, OEA displaces radiolabeled OEA from the PPAR-α LBD with a half-maximal inhibitory concentration (IC_{50}) of 120 nM [27]. These results indicate that OEA is a high-affinity agonist of PPAR-α.

Table 1. Potencies of various natural and synthetic PPAR-α agonists in vitro.

Compound	EC_{50} (μM)	
OEA	0.120	[27]
GW7647	0.006	[72]
WY-14,643	0.650	[37]
Oleic acid	10	[27]
EPA	10	[73]
Linoleic acid	7	[45]
Clofibrate	50	[37]
Fenofibrate	30	[72]
8(S)-HETE	0.2	[74]

EPA, eicosapentaenoic acid; 8(S)-HETE, 8(S)-hydroxyeicosatetraenoic acid. Results are from references listed in brackets.

Table 2. Effects of OEA on meal pattern in free-feeding Wistar rats.

Parameter	*OEA (5 mg/kg)*	*OEA (10mg/kg)*	*OEA (20mg/kg)*
Latency	↑↑	↑↑↑	↑↑↑↑
First meal size	—	—	—
PMI after 1st meal	—	—	—
$PMI_{ave.}$	—	—	—
Average meal size	—	—	—
Meal frequency	—	↓	↓

Doses are in mg/kg, i.p. Arrows symbolize a 100% change from vehicle alone, half-arrows indicate a 50% change. PMI, post meal interval. Data are adapted from [57].

Does PPAR-α activation contribute to the hypophagic actions of OEA? Experiments in genetically modified mice show that OEA does not reduce feeding in animals lacking a functional PPAR-α gene (fig. 4b). Underscoring the selectivity of this defect, PPAR-α-deficient mice retain the ability to respond to other appetite suppressants, such as *d*-fenfluramine and cholecystokinin [27]. The possibility that OEA causes hypophagia by activating PPAR-α is supported by two additional findings. First, potent PPAR-α agonists, such as GW7647 and Wy-14643, inhibit food intake [27]. Interestingly, fibrate drugs, which are widely used in the clinic as antihyperlipidemic agents, but are weak PPAR-α agonists [42], have no anorexiant effect [27]. Second, administration of OEA or synthetic PPAR-α agonists modifies expression of PPAR-α-regulated genes in the small intestine and other tissues. Genes modulated by OEA include, along with PPAR-α itself, fatty-acid translocase (FAT/CD36) and fatty-acid transport protein (FATP) [27], which encode for proteins involved in intestinal lipid transport (table 3). This result is interesting because it suggests that OEA may facilitate the absorption of lipid nutrients, an effect functionally analogous to that of cholecystokinin stimulation of pancreas and gall bladder secretion (reviewed in [43]).

Other members of the PPAR receptor family do not appear to directly modulate feeding behavior, as potent activators of PPAR-δ (GW501516) and PPAR-γ (ciglitazone) have

Table 3. Effects of OEA (10 mg/kg, i.p.) on gene expression in various mouse tissues.

Gene	Jejunum	Doudenum	Ileum	Liver
PPAR-α	↑↑↑	↑↑↑	—	↑↑↑↑
PPAR-δ	—	—	—	N/A
PPAR-γ	—	—	—	N/A
FAT/CD36	↑↑↑	↑↑↑↑	—	↑↑
FATP	↑↑	↑↑	—	N/A
FABP	—	—	—	↑↑
iNOS	↓	N/A	N/A	N/A

Arrows symbolize a 100% change from vehicle alone, half-arrows indicate a 50% change. FAT, fatty-acid translocase; FATP, fatty-acid transport protein 1; FABP, fatty acid-binding protein; iNOS, inducible nitric-oxide synthase; N/A, tissue not analyzed. Data are adapted from [27].

no effect on food intake in mice [27]. These findings indicate that the anorexiant effects of OEA are selectively mediated through PPAR-α activation, and demonstrate a role for this nuclear receptor in the regulation of feeding.

The fatty-acid sensor hypothesis, revisited

PPAR-α is generally regarded as a metabolic sensor of diet-derived fatty acids [44, 45]. According to this view, PPAR-α may be activated by the heightened levels of non-esterified fatty acids reached in tissues after a meal. Though elegant, this hypothesis does not satisfactorily explain all of the available data. Fatty acids absorbed are rapidly modified by cells through conversion to coenzyme A (CoA) esters, and their metabolic products (phospholipids, glycerol esters, cholesterol esters) do not activate PPAR-α. Furthermore, if fatty acids directly interact with PPAR-α, they would be expected to exert anorexiant effects similar to those of OEA [27]. In contrast, on a molar basis, fatty acids appear to be weaker than other nutrients at causing such effects [46, 47]. Even further, if the role of PPAR-α is to sense fatty-acid flux into cells, then a high-fat diet should trigger PPAR-α activation in tissues containing this receptor. In contrast, rats fed either normal chow or a high-fat diet display identical levels of two downstream targets of PPAR-α, acyl-CoA-dehydrogenase (ACD) – the rate-limiting enzyme of peroxisomal β-oxidation – and acyl-coA-oxidase (AOX) – a mitochondrial β-oxidation enzyme – indicating that an elevated fatty-acid load in the liver does not alter PPAR-α function [48]. Similarly, dietary supplementation of fat to humans fails to increase fat utilization or stimulate energy balance up to 24 h following their ingestion [49, 50]. Finally, many tissues that do not experience fatty-acid fluxes or have limited capacity to synthesize fatty acids contain PPAR-α. For example, PPAR-α is present at significant levels in select regions of the rat brain [51], where there are no significant post-prandial fluxes of free fatty acids. Unlike fatty acids, OEA binds to and activates PPAR-α at low nanomolar concentrations, and its levels in small intestinal tissue are not only tightly controlled by feeding, but can also reach values that are sufficient to fully activate PPAR-α [27]. Moreover, OEA is present in all tissues that express PPAR-α, including the brain, though the regulation of its biosynthesis in many of these tissues is still unknown. It appears, therefore, that OEA signaling may complement in important ways non-esterified fatty acids in their role as PPAR-α activators, particularly in cells where fatty-acid buffering is most effective.

Vanilloid receptors do not contribute to OEA hypophagia

OEA modulates agonist-evoked vanilloid type-1 receptor (TRPV1) currents in *Xenopus* oocytes expressing the receptor [52], raising the possibility that OEA might interact with TRPV1 to regulate satiety. TRPV1 receptors are temperature and acid-sensitive cation channels, which are activated pharmacologically by a diversity of lipid compounds, including anandamide and lipoxygenase metabolites of arachidonic acid [53]. To examine whether TRPV1 receptors participate in the anorexiant actions of OEA, we used mice deficient in the TRPV1 gene (TRPV1$^{-/-}$). Using previously established protocols [27], we administered OEA [10 mg-kg^{-1}, intraperitoneally (i.p.)] 30 min prior to the inception of the dark phase and measured feeding for the following 12 h, using an automated monitoring system. The results of these experiments show that disruption of the TRPV1 gene does not affect OEA-induced hypophagia (fig. 4c, d), indicating that TRPV1 activation does not play a significant role in the anorexiant effects of this lipid amide.

OEA induces satiety

To explore the behavioral mechanism by which OEA inhibits feeding, we examined how this compound affects normal feeding patterns in rats. Rats feed during the night, in a series of episodes (meals) that are separated by intervals of variable duration (fig. 5). Anorexiant drugs and feeding-regulating hormones act by modifying different aspects of this patterned behavior. For example, the serotonergic anorexiant *d*-fenfluramine (4 mg-kg^{-1}, i.p.) prolongs the time preceding the first meal (latency of feeding onset) and shortens meal size [54, 55]. In contrast, the peptide hormone cholecystokinin selectively reduces meal size [56]. OEA appears to operate through a distinct mechanism: when administered systemically to free-feeding rats or mice before dark, OEA increases feeding latency and decreases meal frequency, but has no effect on meal size [57] (table 2). Moreover, synthetic PPAR-α

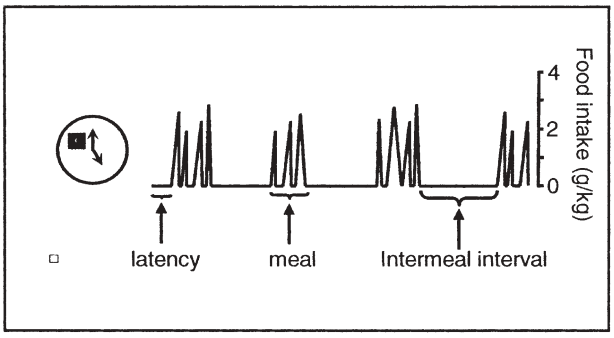


Figure 5. Feeding in rodents starts at the onset of dark and consists of multiple meals, each consisting of several eating bouts. Hormones and anorexiant drugs modify different aspects of this patterned behavior; they can prolong the time preceding the first meal or prolong the interval between meals (i.e. induce satiety); or shorten the duration and size of a meal (i.e. induce satiation). OEA selectively prolongs the latency of feeding onset [57].

agonists have very similar effects [27]. An economical interpretation of these findings is that OEA activation of PPAR-α induces a state of satiety (tonic inhibition over eating) rather than one of satiation (phasic termination of eating, resulting from the act of food ingestion).

OEA engages vagal sensory fibers

Several feeding-controlled signals of peripheral origin, such as cholecystokinin, act by engaging vagal sensory fibers that converge on the nucleus of the solitary tract (NST) in the brainstem. From the NST, higher-order brain regions are recruited to process this information. Pivotal in this regard is the paraventricular nucleus of the hypothalamus (PVH), which integrates central and peripheral satiety signals and orchestrates autonomic responses by adjusting the balance between energy intake and energy expenditure (reviewed in [58]).

Several lines of evidence suggest that OEA acts in a similar manner to produce satiety. First, OEA fails to reduce feeding in rats in which the subdiaphragmatic vagus nerve has been severed [27] (but, for potentially contrasting results see [30]). Second, rats deprived of peripheral sensory fibers by treatment with the neurotoxin capsaicin fail to respond to OEA, Wy-14643 or the vagal-dependent peptide cholecystokinin, but retain their ability to respond to centrally acting anorexiant drugs, such as the serotonergic agonist CP-93129 [25]. Third, although potent when administered peripherally, OEA is ineffective after injection into the rat brain ventricles [25]. Fourth, *c-fos* in situ hybridization experiments show that systemically administered OEA selectively activates the NST, PVH and supraoptic nucleus [25]. The neuromodulatory systems recruited by OEA remain unclear, but peptides such as neuropeptide Y, cocaine and amphetamine-regulated transcript (CART), arginine-vasopressin and oxytocin are likely to be involved.

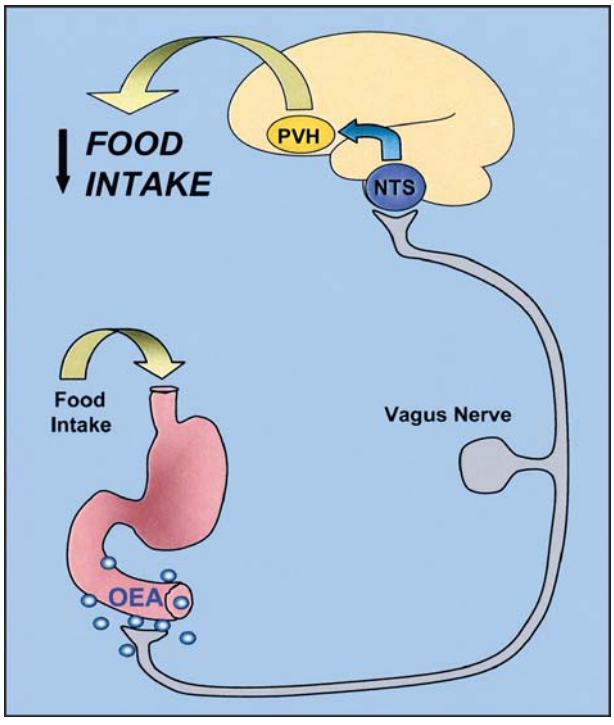


Figure 6. Regulation of feeding by intestinal OEA. According to this hypothetical model, OEA accumulated in the small intestine in response to feeding activates intestinal PPAR-α receptors, which engage vagal sensory fibers. This leads in turn to the recruitment of the nucleus of the solitary tract (NST) in the brainstem and the paraventricular nucleus of the hypothalamus (PVH), ultimately causing induction of satiety.

The findings discussed above support the model illustrated in figure 6. According to this hypothetical model, OEA is produced in the small intestine in response to feeding, reaching local concentrations that are sufficient to activate PPAR-α. Acting through an unknown mechanism, these receptors may in turn activate vagal sensory afferents leading to the subsequent recruitment of the NST, PVH and other higher brain structures involved in the control of energy balance.

A role for intestinal PPAR-α receptors

How does PPAR-α activation mediate the anorexiant effects of OEA? This important question has not been addressed yet, but the fact that feeding regulates OEA levels in the duodenum and jejunum [25,27] implies that this lipid signal may act on PPAR-α localized within cells of the small intestine. In keeping with this possibility, we found that OEA content in the rat small intestine rises from 132 ± 18 pmol-g^{-1} in the fasting state to 329 ±

150 pmol-g^{-1} in the post-ingestive state [25]. The latter value is approximately twofold higher than the EC_{50} (half maximal response) of OEA for PPAR-α (120 nM), suggesting that post-prandial levels of endogenous OEA can fully activate this receptor [27].

The chain of molecular events bridging PPAR-α activation to vagal sensory fiber recruitment is unknown. It is possible that this message is conveyed, at least in part, through nitric oxide (NO) release. Enterocytes produce large amounts of this gaseous transmitter, which may act as a peripheral appetite-stimulating signal [59, 60]. Moreover, PPAR-α represses the expression of inducible NO synthase (iNOS), one of the enzymes responsible for intestinal NO production (table 3) [27, 61]. Thus, repression of iNOS expression by PPAR-α may contribute to the long-term satiating effects of OEA, which last for many hours after injection of the compound [27, 57].

Inception of satiety occurs within minutes of OEA administration [57], a time-course that cannot be accounted for by changes in gene transcription. This suggests that, in addition to its transcriptional actions, PPAR-α also engages non-transcriptional mechanisms, possibly analogous to those recruited by receptors for estrogen hormones and vitamin D. Such effects include activation of phosphoinositol-3-kinase [62], NOS [63] and guanylyl cyclase [64]. Whether PPAR-α initiates similar or different molecular events remains to be determined, but initial evidence suggests that PPAR-α agonists can rapidly transactivate epidermal growth factor receptor, extracellular regulated kinase and p38-MAP kinase in vitro [65].

Effects of OEA on body weight and fat utilization

In addition to its anorexiant actions, OEA also reduces body-weight gain when administered subchronically to lean [25] or obese rats [66] and mice [27], but not PPAR-α-null mutants [27]. These effects are accompanied by parallel changes in tissue triacylglycerols and serum lipids [27, 66]. Pair-feeding experiments suggest that OEA-induced hypophagia may be sufficient to suppress body-weight gain in lean rats [25]. The situation may be different, however, in high-fat-diet-induced obese rats. In these animals, pair-feeding has no effect on body mass, which implies that the weight-controlling actions of OEA might primarily be due to stimulation of energy expenditure [66]. In fact, OEA promotes the expression of PPAR-α-regulated genes that participate in fat catabolism such as FATP and FAT/CD36, or energy homeostasis, such as uncoupling protein-2 (UCP-2) (table 3) [27,66]. In addition to these transcriptional events, OEA promotes a rapid lipolytic response, which occurs within minutes of OEA administration and, therefore, is likely to be independent of genomic activation [66].

A physiological role for OEA in the regulation of satiety?

As discussed above, several lines of evidence suggest that OEA regulates feeding in a physiological manner. First, the increase in intestinal OEA levels that occurs in post-ingestive states is in accord with a role of this compound as a local satiety hormone. This possibility is reinforced by results showing that OEA levels are higher during the daytime (16:30 h: 289 ± 29 pmol-g^{-1}), when animals are satiated, and lower during the night (01:30 h: 138 ± 12 pmol-g^{-1}) when they feed [27]. Moreover, diurnal fluctuations of intestinal PPAR-α expression parallel changes in tissue OEA levels [27]. Thus, a plausible interpretation of these convergent results is that intestinal OEA may help maintain satiety in the post-ingestive state via activation of local PPAR-α (fig. 6). We cannot exclude, however, the possibility that PPAR-α receptors in other visceral organs, such as the liver, may also contribute to this response.

Conclusions and future directions

The discovery that OEA is an endogenous high-affinity PPAR-α agonist raises many questions, three of which appear to be particularly urgent. First, how is intestinal OEA synthesis regulated physiologically? OEA levels in the small intestine are increased post-prandially, but the physiological and molecular mechanisms or the cellular localization of this phenomenon are unclear. Second, what are the roles of OEA in other tissues, such as fat, liver or brain? The existence of diurnal OEA fluctuation in white adipose tissue (fig. 3) suggests the enticing possibility that this lipid amide may serve local modulatory functions, possibly complementary to those of signaling proteins, such as leptin and adiponectin [67]. This possibility remains, however, to be explored. Third, how do other non-cannabinoid FAEs act? Recent work has shown that PEA reduces inflammation in a PPAR-α-dependent manner [68]. Is this true also for other FAEs such as stearoylethanolamide, or do these molecules interact with additional members of the nuclear transcription factor family [69, 70]? As these questions are progressively unraveled, we may not only gain insight on the OEA signaling system, but also identify new potential drug targets for the treatment of appetite disorders and obesity.

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