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ScienceDirect
Pharmacology, Biochemistry and Behavior 85 (2006) 722–727
PHARMACOLOGY BIOCHEMISTRY AND BEHAVIOR
www.elsevier.com/locate/pharmbiochembeh

Beta-adrenergic-mediated inhibition of feeding by mercaptoacetate in food-deprived rats

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Received 9 August 2006; received in revised form 16 October 2006; accepted 2 November 2006
Available online 15 December 2006

Abstract

This study investigated the effect of intraperitoneal (IP) injections of the fatty acid oxidation (FAO) inhibitor mercaptoacetate (MA, 45.6 mg/kg) on feeding in food-deprived rats. As previously, MA significantly stimulated feeding in ad libitum-fed rats. MA, however, reduced feeding in 18 and 36 h-fasted rats despite apparently antagonizing the fasting-induced increase in hepatic FAO. To test whether this anorectic effect involves β-adrenergic stimulation, 36 h-fasted rats were IP injected with the nonspecific β-adrenergic receptor antagonist propranolol (PROP, 0.5 mg/kg) just before MA injection. PROP attenuated MA's feeding-inhibitory effect, suggesting that MA anorexia is at least partially mediated by β-adrenergic stimulation. Finally, we evaluated the role of subdiaphragmatic vagal afferent fibers in MA's feeding-inhibitory effect by testing the ability of MA to inhibit food intake in fasted rats after subdiaphragmatic vagal deafferentation (SDA). MA inhibited feeding similarly in SDA rats and sham-operated rats. These data demonstrate that subdiaphragmatic vagal afferents are not necessary for the feeding-inhibitory effect of peripheral MA. These results suggest that the FAO inhibitor MA elicits a feeding-inhibitory effect in fasted rats that is mediated by a different mechanism than its feeding-stimulatory effect.

Keywords: Satiety; Fatty acid oxidation; Liver; Food intake

1. Introduction

Considerable evidence supports the hypothesis that the organism can monitor ongoing levels of fuel oxidation and that this information plays a role in the control of food intake (Friedman, 1995; Langhans, 1996; Langhans and Scharrer, 1992; Leonhardt and Langhans, 2004). For example, peripheral administration of mercaptoacetate (MA) or other inhibitors of fatty acid oxidation (FAO) stimulates feeding in many species, including humans (Friedman and Tordoff, 1986; Horn et al., 2004; Kahler et al., 1999; Scharrer and Langhans, 1986). Although the exact mechanisms are not fully understood, there is strong evidence that MA's feeding-stimulatory signal arises from peripheral changes in FAO, presumably hepatic FAO, and is relayed to the brain via vagal afferent fibers (Langhans and Scharrer, 1987a; Ritter and Taylor, 1989, 1990). The feeding-stimulatory effect of MA seems to depend on current levels of FAO because MA stimulates feeding more in rats maintained on medium- or high-fat diets than on low-fat diets (Scharrer and Langhans, 1986; Singer-Koegler et al., 1996).

In the present study we attempted to increase hepatic FAO in low-fat fed rats by food deprivation. Because fasted rats depend on FAO to meet the energy needs of most peripheral tissues, we hypothesized that fasting would enhance the role of hepatic FAO in the control of feeding by hepatic FAO and thus increase the feeding-stimulatory effect of MA. To our surprise, blockade of FAO with MA in fasted rats decreased rather than stimulated feeding. As MA was reported to increase plasma epinephrine levels in food-deprived but not in ad libitum-fed rats (Van Dijk et al., 1995), we next tested whether β-adrenergic stimulation was involved in MA's feeding-inhibitory effect by using the nonspecific β-adrenergic receptor antagonist propranolol (PROP). The results indicated that the feeding-inhibitory effect of MA, but not its feeding-stimulatory effect, is partially mediated by β-adrenergic receptors. Finally, tests in rats with subdiaphragmatic vagal deafferentation (SDA) indicated that

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0091-3057/$ - see front matter © 2006 Elsevier Inc. All rights reserved.
doi:10.1016/j.pbb.2006.11.002

MA's feeding-inhibitory effect in fasted rats does not require intact vagal afferents. Given that vagal afferent fibers are necessary for MA's feeding-stimulatory effect (Langhans and Scharrer, 1987a; Ritter and Taylor, 1989, 1990), this further dissociates MA's feeding-stimulatory effect in ad libitum-fed rats from its feeding-inhibitory effect in fasted rats.

2. Materials and methods

2.1. Animals

Male Sprague–Dawley rats (Charles River, Sulzfeld, Germany) were housed individually in stainless-steel drawer cages with grated floors. The colony room was maintained at 22±2 °C and 60% atmospheric humidity, with a 12:12 h light dark cycle (lights out: 10.00 am). Rats had free access to water and ground low-fat (4.5%) chow (No. 3433, Provimi Kliba AG, Kaiseraugst, Switzerland), unless noted otherwise. Before the experiments, rats were adapted to diet and housing conditions for 2 weeks and were handled and weighed every morning. All protocols were approved by the Veterinary Office of the Canton of Zurich.

2.2. Drugs

MA (sodium thioglycolate, Sigma, Buchs, Switzerland, 45.6 mg/kg body weight, BW) was intraperitoneally (IP) injected in 2.7 ml/kg sterile water. MA is a potent inhibitor of acyl-CoA dehydrogenases and therefore inhibits mitochondrial β-oxidation (Bauche et al., 1981, 1983). The dose used inhibited FAO and increased feeding in ad libitum-fed rats (Scharrer and Langhans, 1986). Control injections were 2.7 ml/kg 0.15 M saline (SAL). PROP (DL-propranolol hydrochloride, Sigma, 0.5 mg/kg BW) was injected IP in 1 ml/kg BW 0.15 M SAL. The dose of PROP attenuated epinephrine's feeding-inhibitory effect in rats (De la Cruz et al., 1990). Control injections were 1 ml/kg BW 0.15 M SAL. Drug doses were calculated as salt and solutions were freshly prepared on the day of use.

2.3. Test protocols

2.3.1. MA and food intake in fasted rats

Fifty eight rats (BW: 534±4 g, mean±standard error of mean (SEM)) were used to test the effect of fasting on MA-induced feeding. Rats were randomly assigned to 6 groups of 9–10 rats each that were subjected to three pre-test fasting conditions, ad libitum, 18 h fast, and 36 h fast, and received IP injections of either MA or control SAL at lights off. Food was offered 0.5 h later, and intake was measured by weighing food containers and any spillage (±0.1 g) after 0.5 and 6 h.

2.3.2. Metabolic effects of MA after fasting

Four weeks later, the same rats (BW: 587±6 g) were regrouped and treated as described above, except they were not fed after injections. Rats were anesthetized with ether 1 h after injections, 3 ml blood samples were taken by heart puncture, and core body temperature was recorded to the nearest 0.1 °C using a quick response probe inserted about 7 cm into the rectum. Blood samples were chilled, mixed with 1.8 mg/ml EDTA and centrifuged for 8 min at 6000 ×*g* at 4 °C, and plasma was stored at −20 °C until assayed. Plasma glucose, free fatty acids (FFA), and β-hydroxybutyrate (BHB) were determined by standard colorimetric and enzymatic methods adapted for the Cobas Mira auto analyzer (Hoffman LaRoche, Basel, Switzerland).

2.3.3. Influence of PROP

Sixty new rats (BW: 497±4 g) were used to test the influence of PROP on MA's feeding effect in 36 h-food-deprived rats. Rats were distributed into 4 groups roughly matched for body weight and 36 h-food deprived. At lights off, PROP or its control was IP injected and immediately thereafter MA or its control was IP injected. Rats were offered food 0.5 h later, and intake was measured after 0.5 and 6 h. Four weeks later, the same rats (BW: 548±5 g) were treated as described above, but not fasted before testing.

2.3.4. Role of vagal afferents

The effect of MA on food intake in 36 h-fasted rats was tested in rats with SDA (n=13, BW: 477±9 g) or sham deafferentation (SHAM, n=13, BW: 481±7 g). On the test days, 36 h-food-deprived rats received a single IP injection of MA or SAL at lights off according to a crossover design, with 5 days of ad libitum feeding between test days, and food intake was measured as previously. The rats used here had previously been used in another experiment (Brandt et al., 2007).

SDA consisted of a transection of the left dorsal vagal rootlets at the brain stem and transection of the dorsal esophageal trunk of the vagus, resulting in complete subdiaphragmatic deafferentation, while sparing about half of the abdominal vagal efferents (Norgren and Smith, 1994; Schwartz et al., 1997). The SHAM procedure consisted of similarly opening the skull and abdomen to expose the vagal rootlets and abdominal vagus, but not further manipulating them. Rats were anesthetized by IP injection of a mixture of 80 mg/kg ketamine (Narketan, Vetóquinol AG Bern, Switzerland), 4 mg/kg xylazine (Rompun, Bayer, Leverkusen, Germany), and 0.05 mg/kg acepromazine (Prequillan, Arovet AG, Zollikon, Switzerland) in 1.1 ml/kg SAL. Supplemental injections of ketamine were given as required. Body temperature was maintained at 37–38 °C throughout surgery with a 39 °C water pad. A combination of trimethoprim and sulphadoxine (80 µl/rat Borgal 7.5%, Intervet, Boxmeer, The Netherlands) was injected subcutaneously immediately after surgery for infection prophylaxis, and 5 mg/kg carprofen (Rimadyl, E. Gräub AG, Bern, Switzerland) was injected subcutaneously after surgery and on each of the following two days for analgesia.

After the experiment, SDA was verified functionally by testing the feeding response to IP injection of 4 µg/kg BW CCK-8 in 12 h-food-deprived rats. In SHAM rats the mean± standard deviation inhibition of intake was 80±21% (31–97%). Therefore, any SDA rat whose reduction of 0.5 h food intake after CCK-8 was >30% control was considered an incomplete lesion. All SDA rats met this criterion. Vagotomy was also verified histologically, by verifying the absence of retrograde

transport of IP injected fluorogold (Fluorochrome, Denver, CO, USA) to the dorsal motor nucleus (DMX) as previously described (Arnold et al., 2006). After preparation of 40 μm sections of the medulla, an observer blind to the behavioral data counted the number of fluorogold-labeled neurons in the left and right DMX in all sections that included the area postrema. This region of the brainstem is richly innervated by both motor and sensory fibers from all three subdiaphragmatic vagal branches (Norgren and Smith, 1988). The inclusion criterion for SDA rats, based on that of Phillips et al. (2000), was that the number of labeled cells in the right (disconnected) DMX be <3% of the number in the left (intact) DMX. Three SDA rats failed this criterion, and their data were not further analyzed. The completeness of the vagal rhizotomies was judged visually at surgery. Our previous data indicate that additional histological verification of vagal rhizotomies detects <5% additional incomplete surgeries (Arnold et al., 2006).

2.4. Statistical analyses

In order to maximize statistical power, and because our hypothesis involved testing a small number of pre-assigned comparisons, including complex comparisons, data were analyzed using planned comparisons tested with the sequentially rejective Bonferroni–Holm procedure (Holm, 1979). Two-factor ANOVAs, with drug treatments and pre-test fasting conditions as between-subject factors, were performed to generate an experiment-wide error term. Six planned comparisons were done to test MA's effect after different pre-test fasting conditions: the MA versus SAL contrasts in each of the three fasting groups and the three contrasts of MA's effects between fasting groups (i.e., MA's effect in 0 h-fasted rats versus in 18 h-fasted rats, MA's effect in 0 h-fasted rats versus in 36 h-fasted rats, and MA's effect in 18 h-fasted rats versus in 36 h-fasted rats). Four comparisons were done to test PROP's ability to alter MA's feeding effect: SAL versus MA; SAL versus MA/PROP; MA versus MA/PROP and the SAL–MA difference versus the PROP–MA/PROP difference. Data obtained from SHAM and SDA rats were analyzed with two-factor ANOVA, with surgery as between-subject factor and drug treatment as within-subject factor. The three planned comparisons tested here were MA versus SAL within each surgical group and the difference in MA's effect between SHAM and SDA rats. Differences were considered significant when $P<0.05$. Data are reported as mean±SEM. When the Bonferroni–Holm test revealed a significant difference (experiment-wide $P<0.05$), the t statistic is given. The standard error of the difference (SED, (2 [mean square error]/n)$^{1/2}$; n=mean # observation per point) is reported as a measure of experiment-wide error variability (McNicol, 1999).

3. Results

3.1. MA and food intake in fasted rats

MA's effect on food intake changed with the duration of food deprivation prior to administration (Fig. 1). In ad libitum-fed rats, MA increased 0.5 h food intake ($t(52)=4.53$; SED=0.6 g) but, in contrast, it reduced 0.5 h food intake in both 18 and 36 h-fasted rats ($t(52)=3.23$ and 6.74, respectively). The reduction was significantly larger in 36 h-fasted rats (4.2 g versus 2.0 g, $t(52)=3.52$). After 6 h, MA had only one significant effect, a reduction in food intake in 36 h-deprived rats ($t(52)=4.96$, SED=1.5 g, Fig. 1), and even after 24 h this reduction was still present (data not shown).

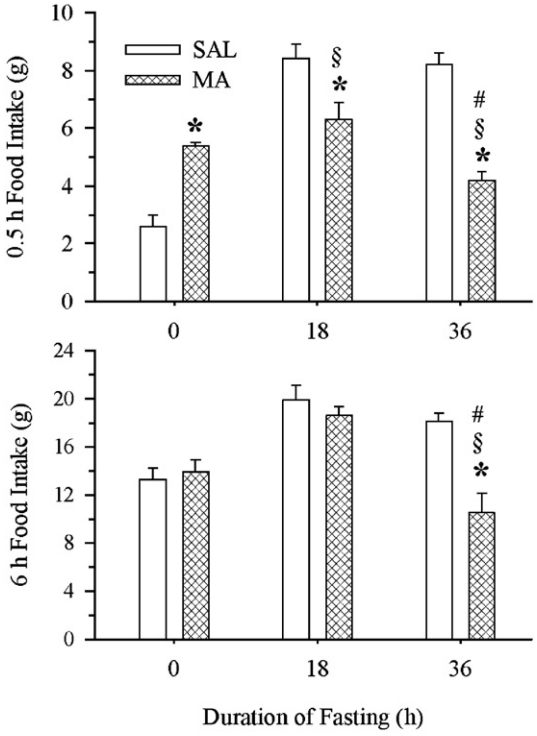

Fig. 1. Mercaptoacetate (MA, 45.6 mg/kg IP) stimulates feeding in ad libitum-fed rats and decreases feeding in 18 and 36 h-food-deprived rats. Each bar represents the mean±SEM of 9–10 rats. *Significantly different from the corresponding control saline rats (SAL). [§]MA's effect in fasted rats is significantly different from MA's effect in ad libitum-fed rats. [#]MA reduced feeding more in 36 h-fasted rats than in 18 h-fasted rats.

3.2. Metabolic effects of MA after fasting

The metabolic effects of MA depended on pre-test feeding conditions (Table 1). MA did not reduce plasma BHB levels in ad libitum-fed rats, but it did significantly reverse the fasting-induced increases in plasma BHB levels in 18 h ($t(52)=11.66$, SED=0.05 mmol/l) and 36 h ($t(52)=21.49$)-fasted rats. Conversely, MA treatment increased plasma FFA in both 18 h ($t(52)=7.84$, SED=0.07 mmol/l) and 36 h ($t(52)=9.47$)-fasted rats, but not in ad libitum-fed rats. The increase in plasma FFA following MA was larger in 18 h ($t(52)=6.02$) and 36 h ($t(52)=7.65$)-fasted rats compared to ad libitum-fed rats, but similar between 18 and 36 h-fasted rats. MA increased plasma glucose levels significantly in ad libitum-fed rats ($t(52)=3.69$, SED=0.51 mmol/l), but not in fasted rats. MA did not decrease body temperature significantly in ad libitum-fed rats, but it did in 18 h ($t(52)=2.71$, SED=0.2 °C) and 36 h ($t(52)=7.73$)-food-deprived rats. The body temperature

Table 1
Effects of mercaptoacetate (MA, 45.6 mg/kg) injected IP after 0, 18, and 36 h fasting on plasma metabolites and body temperature

Fasting	0 h		18 h		36 h	
Drug	SAL	MA	SAL	MA	SAL	MA
FFA, mmol/l	0.39±0.03	0.53±0.06	0.58±0.04	1.17±0.09*,§	0.53±0.03	1.23±0.04*,§
BHB, mmol/l	0.23±0.04	0.24±0.02	0.71±0.04	0.08±0.02*,§	1.21±0.07	0.06±0.01*,§
GLC, mmol/l	8.4±0.4	10.3±0.3*	7.9±0.4	7.3±0.4§	7.1±0.3	6.3±0.5§
TEM, °C	37.8±0.1	37.6±0.1	37.9±0.1	37.4±0.1*	38.2±0.1	36.9±0.1*,§,#

Data are mean±SEM for 9–10 rats/group. Samples were obtained 1 h after MA injection, as described. SAL, saline control; FFA, free fatty acids; BHB, β-hydroxybutyrate; GLC, glucose; TEM, temperature.
*Significantly different from the corresponding SAL value.
§MA's effect in fasted rats significantly different from MA's effect in ad libitum-fed rats.
#MA's effect in 36 h-fasted rats significantly different from MA's effect in 18 h fasted rats.

reduction in 36 h-fasted rats (1.3 °C) was larger than that in ad libitum-fed rats (0.2 °C, $t(52)=6.09$) or 18 h-fasted rats (0.5 °C, $t(52)=5.01$).

3.3. Influence of PROP

MA's feeding-inhibitory effect in 36 h-fasted rats was attenuated by PROP pretreatment (Fig. 2). MA reduced 0.5 h-food intake in PROP ($t(55)=4.61$; SED=0.6 g) and SAL pretreated rats ($t(55)=7.52$). The reduction in food intake, however, was significantly smaller in the PROP pretreated rats (3.0 g versus 4.3 g, $t(55)=2.3$) and MA/PROP rats ate significantly more than MA rats ($t(55)=2.9$). After 6 h, MA significantly inhibited food intake only in SAL ($t(55)=5.87$; SED=1.5 g), but not in PROP pretreated rats, and the reduction in food intake in SAL pretreated rats was significantly larger than in PROP pretreated rats (9.0 g versus 2.5 g, $t(55)=4.25$).

In ad libitum-fed rats the increase in 0.5 h food intake after MA was not significantly affected by PROP pretreatment (data not shown). Comparable to the results of Experiment 3.1 (Fig. 1), MA did not increase 6 h food intake (data not shown).

3.4. Role of vagal afferents

MA inhibited 0.5 h food intake similarly in SHAM ($t(24)=4.19$, SED=0.6 g) and SDA rats ($t(24)=3.77$, Fig. 3). After 6 h, MA again inhibited feeding in both SHAM ($t(24)=3.72$, SED=

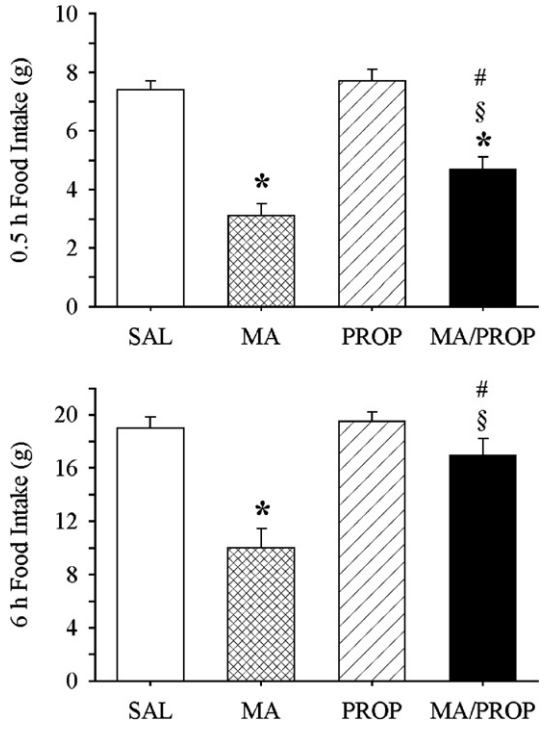

Fig. 2. Propranolol pretreatment (PROP, 0.5 mg/kg IP) attenuated the feeding-inhibitory effect of mercaptoacetate (MA, 45.6 mg/kg IP) in 36 h-food-deprived rats. Each bar represents the mean±SEM of 14–15 rats. *Significantly lower than control saline rats (SAL). §Rats pretreated with PROP (MA/PROP) ate more than MA rats. #SAL–MA is significantly different from PROP–MA/PROP.

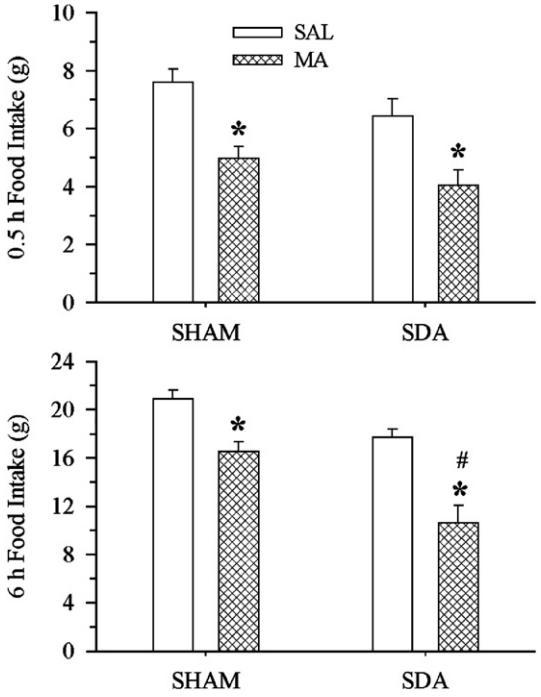

Fig. 3. Mercaptoacetate (MA, 45.6 mg/kg IP) reduced feeding in 36 h-food-deprived sham-operated controls (SHAM, $n=13$) and in rats with subdiaphragmatic vagal deafferentation (SDA, $n=13$). *Significantly lower than corresponding control saline rats (SAL). #MA reduced feeding more in SDA than SHAM rats.

1.2 g) and SDA ($t(24)=5.99$) rats, but the reduction was significantly larger in SDA rats (7.1 g vs. 4.4 g, $t(24)=2.29$).

4. Discussion

This study compared the effects of IP MA injection in ad libitum-fed and in 18 and 36 h-food-deprived rats. The novel findings are 1) that fasting reverses the feeding-stimulatory effect of peripheral MA, 2) that the feeding-inhibitory effect of MA in food-deprived rats is at least partially mediated by β-adrenergic stimulation, and 3) that, unlike the feeding-stimulatory effect of peripheral MA, its feeding-inhibitory effect does not require intact vagal afferents. The recruitment of a β-adrenergic feeding-inhibitory mechanism in fasted rats prevented this design from providing a test of our original hypothesis, that the increase in fatty acid mobilization during fasting would increase the control of food intake by hepatic FAO and, consequently, increase the feeding-stimulatory effect of MA.

That MA stimulates feeding in non-deprived rats has been reported frequently (Langhans and Scharrer, 1987b; Scharrer and Langhans, 1986; Singer-Koegler et al., 1996) and seems to be due to inhibition of hepatic FAO (Leonhardt and Langhans, 2004; Scharrer and Langhans, 1986). That MA did not detectably decrease plasma BHB in ad libitum-fed rats here is presumably due to the fact that these rats consumed a carbohydrate-rich meal just before injections, which rapidly inhibits ketogenesis, thus rendering BHB an inaccurate indicator of hepatic FAO (McGarry and Foster, 1971; Surina-Baumgartner et al., 1996). MA did appear to effectively antagonize hepatic FAO in 18 or 36 h-fasted rats, as the plasma BHB concentrations were decreased in these groups after MA. Nevertheless, the inhibition of hepatic FAO by MA was not sufficient to stimulate feeding under these conditions. Rather, MA inhibited feeding in food-deprived rats. These data suggest that MA can elicit a feeding-inhibitory effect in food-deprived rats that does not depend on hepatic FAO and that this effect can antagonize its feeding-stimulatory effect.

A likely candidate for the feeding-inhibitory effect of MA is activation of the sympathetic nervous system. This is because the nonspecific β-adrenergic receptor antagonist PROP significantly attenuated MA's feeding-inhibitory effect in 36 h-food-deprived rats without significantly affecting MA's feeding-stimulatory effect in non-deprived rats. Whether PROP acted in the periphery, the brain, or both to attenuate MA's feeding-inhibitory effect is uncertain. PROP can penetrate the blood brain barrier (Pardridge et al., 1983), and several lines of evidence suggest that activation of β-adrenergic sites in the hypothalamus can inhibit feeding in rats (Borsini et al., 1982; Goldman et al., 1971; Leibowitz, 1970). Alternatively, MA may have inhibited feeding in fasted rats in part via a peripheral β-adrenergic mechanism (Tordoff et al., 1982). A role of the sympathetic nervous system is also consistent with the report that MA induced an increase in plasma epinephrine levels in food-deprived rats but not in ad libitum-fed rats (Van Dijk et al., 1995). Epinephrine released from the adrenal glands is one of the principal effectors of the sympathetic nervous system, and peripheral epinephrine administration potently inhibits feeding (Russek et al., 1967, 1987), at least in part through stimulation of β-adrenergic receptors (De la Cruz et al., 1990; Langhans et al., 1985). Furthermore, peripheral administration of the β-adrenergic agonist salbutamol reversed MA's feeding-stimulatory effect in non-deprived rats (Nisoli et al., 1996).

The cause of the increased β-adrenergic activity in MA-treated fasted rats is not clear. Hypothermia, increased plasma FFA levels, and disturbances in glucose metabolism may have contributed (Benthem et al., 2000; Grekin et al., 1995; Kozyreva et al., 1999; Paolisso et al., 2000; Van Dijk et al., 1995). Our data suggest that increased plasma FFA levels and the decreased body temperature are the strongest candidates, as these parameters were changed significantly by MA treatment in both 18 and 36 h-fasted rats, at least at our sampling point 1 h after injection. An increase in plasma FFA levels could of course also be the result, rather than the cause, of β-adrenergic stimulation by MA. Alternatively, MA may have had some other, unidentified aversive effect in fasted rats, such as impairment of motor capacity, that contributed to its feeding-inhibitory effect. This possibility needs further investigation.

Finally, we demonstrated that SDA, the most selective method available to completely lesion abdominal vagal afferents without severely compromising vagal motor function, failed to attenuate MA's feeding-inhibitory effect. Thus, intact vagal afferents are not required for the feeding-inhibitory effect of MA in fasted rats. In line with this conclusion, Langhans and Scharrer (1987a) previously reported that the feeding-inhibitory effect of high doses of MA in non-deprived rats did not depend on the hepatic branch of the vagus. These data dissociate the feeding-inhibitory effect of MA from its feeding-stimulatory effect, which numerous studies indicate depends on vagal afferent signaling (for review see Langhans, 1996; Leonhardt and Langhans, 2004). Indeed, the observation that MA reduced 6 h food intake more in SDA rats than in intact rats may reflect the elimination of the feeding-stimulatory action after SDA. Two other findings are also consistent with this reasoning. First, the report that the feeding-inhibitory effect of epinephrine does not depend on hepatic vagal afferents (MacIsaac and Geary, 1985) is consistent with our conclusion that MA's feeding-inhibitory is mediated in part by activation of the sympathetic nervous system. Second, capsaicin treatment, which lesions vagal and non-vagal non-myelinated afferents, blocked the feeding-stimulatory effect of MA (68.4 mg/kg) without blocking the conditioned taste aversion elicited by the same dose of MA (Singer et al., 1999), again suggesting that the feeding-stimulatory, but not the feeding-inhibitory effect of MA depends on vagal afferent signaling.

In summary, our findings confirm that peripheral MA stimulates feeding in ad libitum-fed rats, but show that it inhibits feeding in 18 or 36 h-food-deprived rats, and reveal two dissociations between MA's feeding-stimulatory effect in non-deprived rats and its feeding-inhibitory effect: First, β-adrenergic mechanisms mediate at least part of the feeding-inhibitory effect of MA, but are apparently not involved in its feeding-stimulatory effect. Second, vagal afferents mediate the feeding-stimulatory effect, but not the feeding-inhibitory effect.

Acknowledgements

We are grateful to Dr. Lori Asarian, Anthony Moses and Sepp Torluccio for the technical assistance. This work was supported by U.S. N.I.H. Research grant DK 060735.

References

Arnold M, Mura A, Langhans W, Geary N. Gut vagal afferents are not necessary for the eating-stimulatory effect of intraperitoneally injected ghrelin in the rat. J Neurosci 2006;26:11052–60.

Bauche F, Sabourault D, Giudicelli Y, Nordmann J, Nordmann R. 2-Mercaptoacetate administration depresses the beta-oxidation pathway through an inhibition of long-chain acyl-CoA dehydrogenase activity. Biochem J 1981;196:803–9.

Bauche F, Sabourault D, Giudicelli Y, Nordmann J, Nordmann R. Inhibition in vitro of acyl-CoA dehydrogenases by 2-mercaptoacetate in rat liver mitochondria. Biochem J 1983;215:457–64.

Benthem L, Keizer K, Wiegman CH, de Boer SF, Strubbe JH, Steffens AB, et al. Excess portal venous long-chain fatty acids induce syndrome X via HPA axis and sympathetic activation. Am J Physiol 2000;279:E1286–93.

Borsini F, Bendotti C, Thurlby P, Samanin R. Evidence that systemically administered salbutamol reduces food intake in rats by acting on central beta-adrenergic sites. Life Sci 1982;30:905–11.

Brandt K, Arnold M, Geary N, Langhans W, Leonhardt M. Vagal afferents mediate the feeding response to mercaptoacetate but not to the beta (3) adrenergic receptor agonist CL 316,243. Neurosci Lett 2007;411:104–7.

De la Cruz F, Soto-Mora ML, Uriostegui T, Russek M. Adrenaline anorexia blocked by alpha and beta adrenergic antagonists in 24-h fasted rats. Behav Brain Res 1990;37:145–8.

Friedman MI. Control of energy-intake by energy-metabolism. Am J Clin Nutr 1995;62:S1096–100.

Friedman MI, Tordoff MG. Fatty-acid oxidation and glucose-utilization interact to control food-intake in rats. Am J Physiol 1986;251:R840–5.

Goldman HW, Lehr D, Friedman E. Antagonistic effects of alpha and beta-adrenergically coded hypothalamic neurones on consummatory behaviour in the rat. Nature 1971;231:453–5.

Grekin RJ, Vollmer AP, Sider RS. Pressor effects of portal venous oleate infusion — a proposed mechanism for obesity hypertension. Hypertension 1995;26:193–8.

Holm S. A simple sequentially rejective multiple test procedure. Scand J Statist 1979;6:65–70.

Horn CC, Ji H, Friedman MI. Etomoxir, a fatty acid oxidation inhibitor, increases food intake and reduces hepatic energy status in rats. Physiol Behav 2004;81:157–62.

Kahler A, Zimmermann M, Langhans W. Suppression of hepatic fatty acid oxidation and food intake in men. Nutrition 1999;15:819–28.

Kozyreva TV, Tyachenko EY, Kozaruk VP, Latysheva TV, Gilinsky MA. Effects of slow and rapid cooling on catecholamine concentration in arterial plasma and the skin. Am J Physiol 1999;276:R1668–72.

Langhans W. Role of the liver in the metabolic control of eating: what we know — and what we do not know. Neurosci Biobehav Rev 1996;20:145–53.

Langhans W, Scharrer E. Evidence for a vagally mediated satiety signal derived from hepatic fatty acid oxidation. J Auton Nerv Syst 1987a;18:13–8.

Langhans W, Scharrer E. Role of fatty-acid oxidation in control of meal pattern. Behav Neural Biol 1987b;47:7–16.

Langhans W, Scharrer E. Metabolic control of eating. World Rev Nutr Diet 1992;70:1–67.

Langhans W, Pantel K, Scharrer E. Dissociation of epinephrine's hyperglycemic and anorectic effect. Physiol Behav 1985;34:457–63.

Leibowitz SF. Hypothalamic beta-adrenergic "satiety" system antagonizes an alpha-adrenergic "hunger" system in the rat. Nature 1970;226:963–4.

Leonhardt M, Langhans W. Fatty acid oxidation and control of food intake. Physiol Behav 2004;83:645–51.

MacIsaac L, Geary N. Partial liver denervations dissociate the inhibitory effects of pancreatic glucagon and epinephrine on feeding. Physiol Behav 1985;35:233–7.

McGarry JD, Foster DW. The regulation of ketogenesis from octanoic acid. The role of the tricarboxylic acid cycle and fatty acid synthesis. J Biol Chem 1971;246:1149–59.

McNicol JW. Recommendations of the Editorial Board on use of statistics in papers submitted to JSFA. J Sci Food Agric 1999;79:152–6.

Nisoli E, Garosi V, Blundell JE, Carruba MO. Salbutamol antagonizes insulin- and sodium mercaptoacetate-induced but not 2-deoxy-D-glucose-induced hyperphagia. Pharmacol Biochem Behav 1996;54:409–13.

Norgren R, Smith GP. Central distribution of subdiaphragmatic vagal branches in the rat. J Comp Neurol 1988;273:207–23.

Norgren R, Smith GP. A method for selective section of vagal afferent or efferent axons in the rat. Am J Physiol 1994;267:R1136–41.

Paolisso G, Manzella D, Rizzo MR, Ragno E, Barbieri M, Varricchio G, et al. Elevated plasma fatty acid concentrations stimulate the cardiac autonomic nervous system in healthy subjects. Am J Clin Nutr 2000;72:723–30.

Pardridge WM, Sakiyama R, Fierer G. Transport of propranolol and lidocaine through the rat blood–brain barrier. Primary role of globulin-bound drug. J Clin Invest 1983;71:900–8.

Phillips RJ, Baronowsky EA, Powley TL. Regenerating vagal afferents reinnervate gastrointestinal tract smooth muscle of the rat. J Comp Neurol 2000;42:325–46.

Ritter S, Taylor JS. Capsaicin abolishes lipoprivic but not glucoprivic feeding in rats. Am J Physiol 1989;256:R1232–9.

Ritter S, Taylor JS. Vagal sensory neurons are required for lipoprivic but not glucoprivic feeding in rats. Am J Physiol 1990;258:R1395–401.

Russek M, Mogenson GJ, Stevenson JAF. Calorigenic hyperglycemic and anorexigenic effects of adrenaline and noradrenaline. Physiol Behav 1967;2:429–33.

Russek M, Vega C, Barrera J, Soto-Mora LM, Lanzagorta A, Racotta R. Anorexia elicited by different catecholamines in rats. Appetite 1987;9:119–26.

Scharrer E, Langhans W. Control of food intake by fatty acid oxidation. Am J Physiol 1986;250:R1003–6.

Schwartz GJ, Plata-Salamán CR, Langhans W. Subdiaphragmatic vagal deafferentation fails to block feeding-suppressive effects of LPS and IL-1b in rats. Am J Physiol 1997;273:R1193–8.

Singer LK, York DA, Berthoud HR, Bray GA. Conditioned taste aversion produced by inhibitors of fatty acid oxidation in rats. Physiol Behav 1999;68:175–9.

Singer-Koegler LK, Magluyan P, Ritter S. The effects of low-, medium-, and high-fat diets on 2-deoxy-D-glucose- and mercaptoacetate-induced feeding. Physiol Behav 1996;60:321–3.

Surina-Baumgartner DM, Arnold M, Moses A, Langhans W. Metabolic effects of a fat-and carbohydrate-rich meal in rats. Physiol Behav 1996;59:973–81.

Tordoff MG, Novin D, Russek M. Effects of hepatic denervation on the anorexic response to epinephrine, amphetamine, and lithium chloride: a behavioral identification of glucostatic afferents. J Comp Physiol Psychol 1982;96:361–75.

Van Dijk G, Scheurink A, Ritter S, Steffens A. Glucose-homeostasis and sympathoadrenal activity in mercaptoacetate-treated rats. Physiol Behav 1995;57:759–64.