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Incentive motivation in humans is modulated by GLP-1 and hunger depending on insulin sensitivity

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Die in dieser Arbeit angegebenen Experimente sind nach entsprechender Anleitung durch Frau Dr. med. Ruth Hanßen von mir mit Unterstützung durch Mitarbeiter der AG Translational Neurocircuitry des Max-Planck- Instituts für Stoffwechselforschung in Köln durchgeführt worden. Insbesondere sind hier Tamara Sitnikow, Dr. Kerstin Albus, Elke Bannemer und Patrick Weyer zu nennen.

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List of abbreviations

ARC	Arcuate nucleus
BMI	Body Mass Index
CKK	Cholecystokinin
DA	Dopamine
DAT	Dopamine Transporter
DMH	Dorsomedial Hypothalamus
DPP-IV	Dipeptidyl-peptidase
EMA	European Medicines Agency
Ex4	Exendin-4
FDA	U.S. Food and Drug Administration
GLP-1	Glucagon-Like-Peptide-1
GLP-1R	GLP-1 receptors
HOMA	Homeostatic Model Assessment
HOMA-IR	Homeostatic Model Assessment of Insulin Resistance
IR	Insulin receptors
LH	Lateral Hypothalamus
NAc	Nucleus Accumbens
NPY	neuropeptide Y
NTS	Nucleus tractus solitarius
PD	Parkinson Disease
PVN	Paraventricular nucleus
PYY	peptide YY
T2DM	Type 2 Diabetes Mellitus
TH	Tyrosine Hydroxylase
VTA	Ventral Tegmental Area
WHO	World Health Organisation

Zusammenfassung

Im täglichen Leben muss unser Gehirn ständig externe Hinweise - wie den Belohnungswert einer potenziellen Nahrungsquelle - mit internen Signalen - wie Hungergefühlen - integrieren, um unser Nahrungsaufnahmeverhalten zu steuern und zu entscheiden, welche Belohnungen es wert sind, sich dafür anzustrengen. Daher müssen die Stoffwechselsignale aus der Peripherie unser Gehirn erreichen und mit den Schaltkreisen interagieren, die unsere Motivation, sich für bestimmte Nahrungsbelohnungen anzustrengen, regulieren. Die Anreizmotivation beschreibt die Prozesse, die den Belohnungswert von externen Reizen in körperliche Anstrengung umsetzen, um die Belohnung zu erhalten. Das dopaminerge Mittelhirn und die dopaminergen mesoaccumbens-Projektionen kodieren die Menge und die Wahrscheinlichkeit von Belohnungen, die motiviertes Verhalten auslösen. Das dopaminerge Mittelhirn und seine Projektionen besitzen zudem viele metabolische Rezeptoren, u.a. für Insulin und GLP-1, welche Informationen über den peripheren Stoffwechselzustand vermitteln. In Tierstudien wurde der modulatorische Effekt von Signalen des metabolischen Zustands auf motiviertes Verhalten bereits untersucht. Beim Menschen fehlen jedoch noch Informationen über die zustandsabhängige Modulation von motiviertem Verhalten und dessen Beitrag zur Adipositas. Ziel dieser Studie war es, den Einfluss von Stoffwechselsignalen, insbesondere von GLP-1, Insulinsensitivität sowie Hungerempfinden, auf die Regulation von motivierten Verhalten beim Menschen zu untersuchen.

Wir führten eine randomisierte, Placebo-kontrollierte, Crossover Studie mit 21 normalgewichtigen (Body-Mass-Index, BMI, < 25 kg/m^2) und 16 adipösen (BMI > 30 kg/m^2) freiwilligen Teilnehmer*innen durch, die an zwei separaten Testtagen entweder Liraglutid als GLP-1-Analogon oder Placebo erhielten. Die Anreizmotivation wurde mit Hilfe eines Computerexperiments erfasst, bei dem die Teilnehmer*innen mittels eines Handgriffes körperliche Anstrengung aufbringen mussten, um unterschiedliche Mengen an Nahrung- und Geldbelohnung zu gewinnen. Das Hungerempfinden wurde mit Hilfe von visuellen Analogskalen gemessen; Insulin, Glukose und die systemische Insulinresistenz, die mit dem Homeostasis Model

Assessment of Insulin Resistance (HOMA-IR) bestimmt wurde, wurden in einer Blutentnahme vor der Aufgabe quantifiziert.

Unsere Ergebnisse zeigten, dass die Anreizmotivation bei normalgewichtigen Menschen mit zunehmendem Hunger ansteigt ($F(1,42) = 5.31$, $p = 0.026$, $\beta = 0.19$), während Hunger bei adipösen Teilnehmer*innen keinen Einfluss auf die Motivation hatte ($F(1,62) = 1.93$, $p = 0.17$, $\beta = -0.12$). Dabei war der Effekt von Hunger auf die Motivation von der peripheren Insulinsensitivität der Teilnehmer*innen abhängig (Zweifachinteraktion, $F(1, 35) = 6.23$, $p = 0.017$, $\beta = -0.281$). Bei Menschen mit höherer Insulinsensitivität erhöhte Hunger die Motivation, während eine schlechtere Insulinsensitivität den motivierenden Effekt von Hunger verringerte. Bemerkenswert ist, dass dies sowohl für Essens- als auch für Geldbelohnungen galt. Die GLP-1-Analogapplikation schwächte den Effekt von Hunger auf die Motivation in Abhängigkeit von der Insulinsensitivität ab (Dreifachinteraktion, $F(1, 127) = 5.11$, $p = 0.026$); d.h. die GLP-1-Analogapplikation normalisierte das motivierte Verhalten der insulinresistenten Teilnehmer, so dass kein Unterschied mehr zwischen insulinresistenten und insulinsensitiven Teilnehmer*innen festgestellt werden konnte.

Um diese Verhaltenseffekte zu erklären, schlagen wir ein Modell der zugrundeliegenden neuronalen Prozesse vor. Da die Anreizmotivation in unserer Studie nicht von der Art der Belohnung beeinflusst wurde, deuten die Ergebnisse darauf hin, dass die Motivation sehr basal auf der Ebene des dopaminergen Mittelhirns statt auf kortikaler Ebene metabolisch reguliert wird. Weiterhin stützen unsere Ergebnisse die bestehende Annahme, dass Insulinsensitivität ein besserer Prädiktor für veränderte Dopaminsignalwege ist als der BMI.

Zusammenfassend berichten wir hier über einen differentiellen Effekt von Hunger auf die Motivation für Nahrungs- und Nicht-Nahrungsbelohnungen in Abhängigkeit von der Insulinsensitivität. Wir zeigen weiter, dass GLP-1 eine regulatorische Rolle bei adaptivem, motiviertem Verhalten beim Menschen spielt und dysregulierte Prozesse des dopaminergen Mittelhirns und damit das motivationale Verhalten bei insulinresistenten Menschen wiederherstellen kann.

Abstract

In everyday life, our brain constantly needs to integrate external cues – such as the rewarding value of a potential food reward – with internal signals – such as hunger feelings – in order to decide which rewards are worth spending effort for and to guide our food intake behaviour. Hence, metabolic signals from the periphery need to reach our brain and interact with the circuitries controlling our motivation to spend effort in order to obtain certain food rewards. Incentive motivation refers to the processes that translate the rewarding value of external cues into physical effort to obtain the reward. The dopaminergic midbrain and dopaminergic mesoaccumbens projections encode the amount and the probability of rewards, that initiate motivated behaviour. The dopaminergic midbrain and its projections are also particularly sensitive to the metabolic state, that is reflected by peripheral signals such as insulin and GLP-1. Animal studies have already assessed the modulatory effect of metabolic state signaling on motivated behaviour. However, in humans, we still lack information about state-dependent modulation of reward-related motivated behaviour and its contribution to obesity. The aim of this study was to assess the impact of metabolic signals, notably of GLP-1, insulin sensitivity and hunger, on the regulation of reward-related motivated behaviour in humans.

In a randomized, placebo-controlled, crossover study, 21 lean (body mass index, BMI, < 25 kg/m^2) and 16 obese (BMI > 30 kg/m^2) participants received either liraglutide as GLP-1 analogue or placebo on two separate testing days. Incentive motivation was measured using a computer experiment in which participants were required to exert physical effort using a handgrip in order to win different amounts of food and monetary reward. Hunger levels were measured using visual analogue scales; insulin, glucose and systemic insulin resistance as assessed by the homeostasis model assessment of insulin resistance (HOMA-IR) were quantified in a blood draw before the task.

Our results revealed that incentive motivation increases with hunger in lean humans ($F(1,42) = 5.31$, $p = 0.026$, $\beta = 0.19$), whereas hunger did not affect motivation in obese participants ($F(1,62) = 1.93$, $p = 0.17$, $\beta = -0.12$). We further showed that the effect of hunger on motivation depended on the peripheral insulin sensitivity of the

participants (two way interaction, $F(1, 35) = 6.23$, $p = 0.017$, $\beta = -0.281$). In humans with higher insulin sensitivity, hunger increased motivation, while poorer insulin sensitivity decreased the motivational effect of hunger. Notably, this holds true for both food and monetary reward. GLP-1 analogue application blunted the insulin-sensitivity-dependent effect of hunger on motivation (three-way interaction, $F(1, 127) = 5.11$, $p = 0.026$); i.e., GLP-1 analogue application normalized motivated behaviour of insulin resistant participants so that no difference between insulin resistant and insulin sensitive participants could be detected any more.

In order to explain these behavioural results, we suggest a model of the underlying neural processes. As incentive motivation was not affected by the type of reward in our study, we suggest motivation to be regulated very basally on the level of the dopaminergic midbrain rather than on a cortical level. We further provide support for the existing thesis that insulin sensitivity is a better predictor of altered DA signaling than the BMI.

In summary, we here report a differential effect of hunger on motivation to obtain food and non-food reward depending on insulin sensitivity. We further demonstrate that GLP-1 plays a regulatory role in adaptive, motivated behaviour in humans, and is able to restore dysregulated processes of the dopaminergic midbrain and hence motivational behaviour in insulin resistant humans.

1 Introduction

Emerging as one of the most critical public health issues in the global world, obesity accounts for a significant number of medical conditions and is responsible for relentlessly rising expenditures in the health care system. The WHO reported in 2008, that the prevalence of obesity had approximately doubled between 1980 and 2008[1]. The arising costs not only result from expenditures for the treatment of obesity itself but also from expenditures for the treatment of multiple obesity associated comorbidities. These comorbidities will promote increasing rates of morbidity and mortality in the western world, if the rising prevalence of obesity is not halted[2].

The evolution of the obesity epidemic is considered as a neurobehavioural problem resulting from the combination of a vulnerable brain and today's modern food environment. The rising production of high-calorie, energy dense palatable food available at low prices and its easy accessibility in most of the developed countries poses a major challenge to maintain a healthy and balanced diet. Especially highly processed western diet style food has strong rewarding values[3,4]. As a result, the human brain is constantly in conflict to process information about the current metabolic status keeping energy levels balanced, but at the same time gets confronted with rewarding external food cues. In order to appropriately weight the rewarding value of a possible reward against the effort required to obtain it, our brain needs to constantly integrate bodily signals in the decision making for or against food intake. Thus, constant overeating might result from a lack of integration of physiological signals into food intake decisions, causing alterations in neural signaling of reward and effort estimations and hence motivation. It is, therefore, of paramount importance to investigate how the central nervous system integrates information about the current metabolic state with external cues causing a promotion or reduction of motivational drive for food intake. Understanding the mechanisms of the motivation to eat beyond physiologic needs might reveal targets for the treatment of obesity.

This introduction will capture the definition and epidemiology of obesity, a short delineation of motivational behaviour, its encoding and aberrations in obesity as well as an approach to metabolic signaling of homeostatic needs and its dysregulations in obesity.

1.1 Definition of obesity

Obesity results from an excess of accumulated fat mass increasing the risk for negative consequences on health state[1]. The most commonly used measure to define obesity is the Body Mass Index (BMI). The BMI is calculated by dividing body weight in kilograms by height in meters squared (kg/m^2)[5]. Thus, the BMI defines obesity by the magnitude of body weight.

Published by the WHO, the BMI allows for a classification of adults as underweight, normal weight, overweight and obese.

Table 1: Body Mass Index (BMI) Classification

Classification	BMI [kg/m^2]
Underweight	< 18.5
Normal weight	18.5 – 24.9
Overweight	25.0 – 29.9
Obesity	
Obesity grade I	30 – 34.9
Obesity grade II	35 – 39.9
Obesity grade III	> 40

Although the BMI does not take into account the amount of adipose tissue and its distribution in the human body, it correlates well with the amount of body fat and positively correlates with an increase of the prevalence of hypertension, dyslipidemia and diabetes mellitus, all being comorbidities which are commonly associated with obesity[6,7].

1.1.1 Epidemiology of obesity worldwide and in Germany

Worldwide obesity rates have been rising dramatically since 1975. According to the WHO, in 2016 nearly 2 billion adults were counted overweight, of which more than 650 million were classified as obese[1].

In Germany, more than half of the population are overweight. In 1998 data from the German National Health Interview and Examination Survey (GNHIES98) showed that

52% of women and 67% of men in Germany were overweight (BMI > 25 kg/m^2)[8]. 22,5% of women and 18,9% of men were classified as obese (BMI > 30kg/m^2)[8].

In 2013 the DEGS-1 study revealed data collected in Germany from 2008 until 2011 showing that, compared to data from 1998, the prevalence of overweight remained on average the same[9]. However, the prevalence of obesity had increased compared to 1998. As such, 23.9% of women and 23.3% of men were considered obese (BMI > 30 kg/m^2)[9]. Prevalence for obesity grade I was found to be 15.9% for men and 18.1% for women. For obesity grade II it was 5.2% for men and 3.9% for women. Finally, 2.8% of men and 1.2% of women were classified in obesity grade III[9]. Furthermore, the prevalence of overweight in young men had increased considerably in Germany: 35.3% of 18-29 year old men were classified as overweight and in the cohort of 30 to 39 years old men, more than half of the cohort was classified as overweight (62.4%)[9].This development is worrying, as overweight young individuals tend to become obese over time and are consequently exposed to a higher risk of developing obesity-associated comorbidities[10].

1.2 Motivational behaviour and its alterations in obesity

External cues may comprise strong motivational signals, such as the palatable taste of a chocolate bar[3,11,12]. The incentive value of such a reward, however, also needs to be weight against the effort required to obtain the reward[13,14]. Hence, every-day decisions in favor of or against food intake are based on cost-benefit analyses weighing the potential food reward against the cost of spending effort in order to obtain it. E.g., is it worth the effort to go to the grocery store to get a piece of cake? For cake with high rewarding qualities, we might even go to the store further away, while for cake with low rewarding qualities, we might not leave the house.

The incentive theory of motivation[15] states that motivated behaviour is primarily affected by anticipated rewards and reinforcement. Incentive motivation, therefore, refers to the processes that translate expected reward into effort spending[16]. These processes include the integration of information about the perceived subjective reward magnitude, which is critical for determining effort exertion and initiating motivated behaviour[14,17].

1.2.1 Neural encoding of reward and motivation

Reward behaviour and motivation are encoded by the mesostriatal dopaminergic (DA) circuitry, which two main components for encoding of motivation are the ventral tegmental area (VTA) and the nucleus accumbens (NAc)[18-20]. Dopaminergic neurons projecting from the VTA to the NAc transmit DA and DA signaling has been shown to affect reward and motivational behaviour[21,22]. Several studies have shown that a lower dopaminergic tone results in lower effort spending and motivation[23,24].

Parkinson's disease (PD) for example, a disease associated with dopamine deficiency at striatal receptors, has been associated with motivational deficits. In pharmacological studies carried out in patients with PD, it could be shown that patients tested on their dopaminergic medication, showed increased willingness to exert force for reward compared to patients with Parkinson's disease off their usual dopaminergic treatment[25].

Two modes haven been described, in which transmission of DA from the VTA to the NAc occurs[26]. The tonic mode implies a steady, baseline level of DA in downstream regions of the VTA enabling normal neural function. In the phasic mode, DA neurons sharply increase their firing rate leading to a prominent change in DA concentrations in the NAc[27,28]. Phasic DA transmission from the VTA can be initialized not only by perceiving reinforcing and rewarding stimuli but also when processing non-reward related, aversive and alerting signals[28-30].

How motivational behaviour towards reward-related stimuli is modulated will depend on various components, such as wanting, liking and learning[4,31]. Findings suggest two types of dopamine neurons, one encoding motivational value, the other encoding motivational salience, modulating motivational behaviour[32]. In relation to reward-related learning, the role of dopamine has been specified. It has been found, that dopamine neurons are involved in encoding reward-related prediction errors, meaning the difference between the reward received and the reward predicted[33,34]. Schultz et al. (1997) found an immediate increase in phasic DA activity, when an unexpected food reward occurred. Inhibition of phasic DA activity occurred, when the reward was smaller than predicted and little or no DA activity occurred, when the reward size was predictable in advance[33].

To summarize, motivational behaviour is modulated by a complex interaction of several factors affecting dopaminergic activity in the midbrain and further investigations are needed on the motivational aspects of DA signaling in the brain.

1.2.2 Assessment of incentive motivation and its alterations in obesity

Several paradigms have been established to measure and objectivize incentive motivation.

One often used paradigm is a button press scenario, in which participants need to adapt their number of button presses per time in order to win a presented reward. In a study conducted by Epstein et al. (2007), effort spending as measured by button presses revealed that obese compared to lean participants had a higher willingness to spend effort for highly palatable food[35]. Similarly, Giesen et al. (2010) investigated, if obese compared to lean participants would invest more effort in button pressing to earn high-calorie snacks[36]. They could demonstrate, that obese and overweight individuals were more willing to spend effort to obtain high caloric food reward compared to lean individuals[36].

Another effort-based paradigm was recently designed, which measures motivation to obtain presented rewards by assessing hand-grip force[37]. The higher the force a participant applies, the more the participant can win.

In a behavioural study, Mathar et al. (2015) used a paradigm in which physical effort spending in humans was measured using a hand-grip[38]. Participants had to squeeze the grip while a picture of the reward was presented on a computer screen. Interestingly, they found out, that obese compared to lean participants were less motivated to spend effort, especially for food rewards. Notably, in the above mentioned studies, the homeostatic state of the participants was not included in the analysis. Given the paucity and heterogeneity of studies on incentive motivation in obesity, further research is needed to explore the motivational behaviour of obese individuals including measures of their internal state.

1.2.3 Alterations of the dopaminergic midbrain in obesity

Relating to the altered motivational behaviour in obese humans, the dopaminergic midbrain and its projections have been shown to be altered in obesity[39,40]. In mice, findings show that a long-term consumption of a high-fat diet leads to a decrease in

dopaminergic activity in the NAc[41]. Rats fed a high-fat diet showed attenuated dopamine turnover in the NAc[42].

Among the five subtypes of known DA receptors, D2 receptors are particularly important in the role of food intake[43,44]. In obese humans brain PET imaging studies have revealed a decreased dopamine D2 receptor availability[40] and specifically, human as well as animal studies have revealed a decreased binding potential of the dopamine D2 receptor in obesity[40,45].

Besides reduced DA activity and reduced availability of D2 receptors, the density of the dopamine transporters (DAT), which are important for regulating the transmission of dopamine, was found to be reduced in the striatum of obese people[46,47]. Furthermore, a high-fat diet in mice lead to a decrease of the expression of Tyrosine Hydroxylase (TH), the rate-limiting enzyme regulating dopamine in the brain[48,49]. Findings in obese humans support rodent data, as the expression of TH gene in the midbrain was significantly downregulated in obese compared to lean humans in a post-mortem study[46].

All these findings of altered dopaminergic signalling in the midbrain in obesity notably affecting the main components encoding reward and motivation, suggest altered reward-related motivational behaviour in obesity. It has, therefore, been suggested, that exceeding food intake in obese humans strives to compensate for the insufficient reward circuitry activation[50].

1.3 Metabolic signalling of homeostatic needs

Neuroendocrine hormones regulate food intake by signaling homeostatic needs to the brain[51]. Besides low blood sugar levels, the hormones leptin, insulin, ghrelin as well as GLP-1, cholecystokinin (CKK), neuropeptide Y (NPY) and peptide YY (PYY) have been identified to play a role as appetite regulating signals[52-54].

For this work, the focus lies on metabolic signaling by GLP-1 and insulin and their actions in the mesostriatal reward system.

1.3.1 GLP-1 and its peripheral effects

Specifically, one potent mediator of satiety signals is Glucagon like peptide 1 (GLP-1)[55]. GLP-1, an incretine hormone, is synthetized and secreted by enteroendocrine L-cells of the epithelium mainly situated in the distal part of the small intestine and the colon.

L-cells can also be found in the proximal part of the small intestine. Furthermore, neurons in the nucleus tractus solitarius (NTS) synthetize GLP-1 as well[56,57]. Overall, GLP-1 is one of the main factors of the gut-brain axis affecting food intake[58].

The hormone is postprandially released into the bloodstream as its biological active form GLP-1(7-36) amid or GLP-1(7-37) amid, especially after the intake of carbohydrate-rich and fatty food[59].

GLP-1 acts on the GLP-1 receptor (GLP-1R) localized in different organs and tissues such as the gut, the lungs, the brain and the pancreatic beta-cells[60-62].

Its main effect as incretine hormone is the insulinotropic effect as GLP-1 potentiates biosynthesis and secretion of insulin upon meal ingestion. If blood glucose levels are low, no GLP-1 stimulated insulin secretion occurs[63].

The second important action of GLP-1 is the inhibition of glucagon secretion. Again, this varies with the amount of plasma glucose concentration: low and hypoglycemic glucose concentrations do not prevent counter-regulation of glucagon secretion[64]. If hyperglycemia occurs, secretion of glucagon is mostly inhibited. Furthermore, GLP-1 decreases gastric emptying[65], lowers the synthesis of gastric acid and has a supporting effect on the neogenesis of pancreatic beta-cells[66].

Endogenous GLP-1 is quickly degraded by the enzyme dipeptidyl peptidase IV (DPP-IV), which is the reason for the very short plasma half-life of about 2 minutes[56,67]. Gutzwiller, Göke et al. (1999) identified GLP-1 as an important factor affecting food intake in 1999, as intravenous infusion of GLP-1 was found to reduce food consumption and increased satiety[58].

1.3.2 Central effects of GLP-1 and its effect on motivational behaviour

Besides its peripheral actions, GLP-1 is able to act in the central nervous system. GLP-1R in the brain have been found to be located in areas implicated in the control of food intake such as in the lateral hypothalamus (LH)[68], the paraventricular nucleus of the hypothalamus (PVN), the dorsomedial hypothalamus (DMH), the arcuate nucleus (ARC) and the supraoptic nucleus but most importantly in areas of the reward system: the VTA and the NAc[69]. GLP-1 presumably affects the brain via two pathways: by passing the blood brain barrier and indirectly via its receptors on the vagal nerve, signaling metabolic state to the NTS. As the endogenous half-life of the hormone is

under 2 minutes, it remains unclear which amount of the gut secreted hormone reaches the brain via the blood brain barrier[12]. However, binding to enteric vagal afferents signalling metabolic state to the NTS, might activate GLP-1 producing neurons within the NTS modulating hypothalamic and limbic areas, affecting food intake[70].

Interventional studies have repeatedly demonstrated the role of GLP-1 in food intake regulation. The LH represents an important regulatory centre of homeostatic feeding and administration of the GLP-1 agonist exendin-4 (Ex4) in the LH has been shown to effectively decrease food-intake in rats[71]. An experiment carried out by Alhadeff AL et al. (2012) showed that a significant reduction especially in highly palatable food intake in rats resulted, after injection of the GLP-1R agonist Ex4 in the VTA and the NAc, the two main components in the dopaminergic midbrain encoding reward and motivation. Further, by direct administration of GLP-1R agonist Ex4 in in the dopaminergic midbrain notably the VTA and the NAc, a reduction in motivated behaviour could be observed in rodents[72]. These findings suggest that binding of GLP-1 to GLP-1R in the dopaminergic midbrain modulates motivational behaviour. The role of GLP-1 in the regulation of human motivation is, however, unknown.

1.3.3 Impairments of GLP-1 signaling in obesity

Obesity is associated with metabolic impairments and as such, it has been found that obese compared to lean individuals show reduced GLP-1 signaling[73,74]. In obese humans, intake of carbohydrates leads to a decrease of GLP-1. However, the mechanisms contributing to this effect are still unclear[74]. Moreover, the secretion of GLP-1 from the L-cells is positively modulated by the orexigenic hormone Ghrelin[75] and it could be demonstrated that Ghrelin levels are reduced in obesity, possibly leading to decreased GLP-1 release[76]. In view on central effects of GLP-1, results from studies conducted with diet induced obese mice and obese humans suggest a central resistance to hormones affecting food intake such as GLP-1[77].

However, there are also studies that revealed no difference between post-prandial GLP-1 release in obese compared to lean humans[78] and studies that showed increased basal GLP-1 levels in obese compared to lean[79]. Taken together, these findings show inconsistent results concerning alterations of GLP-1 signaling in obese and evidence

how impairments of GLP-1 signaling might affect incentive motivation in obesity is still lacking.

1.3.4 Insulin and its peripheral effects

Besides GLP-1, insulin plays an important role as appetite regulating signal. Insulin, a hydrophilic peptide hormone is produced by pancreatic beta-cells and glucose-dependently released into the bloodstream after food intake[80]. Insulin acts via tyrosine kinase receptors at insulinotropic organs (especially skeletal muscle and fatty tissue) by transporting glucose into the cells and thus lowering blood glucose levels[81]. Furthermore, insulin reduces gluconeogenesis and glycogenolysis in high-glucose states after food intake[82]. Moreover, insulin has a growth-stimulating effect[83].

1.3.5 Central effects of insulin and its effects on motivational behaviour

Insulin receptors (IR) are also found in the central nervous system, specifically on dopaminergic neurons originating from the VTA[84]. As dopaminergic midbrain signalling has been implicated in mediating motivational behaviour in response to rewarding cues such as food reward, the presence of IR in the dopaminergic midbrain suggests, that insulin modulates motivational and reward-related behaviour[85,86]. This is further supported by the fact that insulin regulates the reuptake of dopamine by inducing the expression of dopamine reuptake transporter[87]. It has been shown that central insulin has an important role in regulating food intake and affects weight control, as mice with a neuron-specific disruption of the IR gene in the brain developed obesity and showed increased food intake[88]. Furthermore, intranasally applicated insulin acts on dopamine neurons in the midbrain which supports the assumption, that insulin might modulate motivational behaviour[85].

1.3.6 Impairments of insulin signalling in obesity

Obesity is strongly associated with insulin resistance[89]. Insulin resistance can be assessed by the Homeostatic Model Assessment (HOMA), a method for assessing beta-cell function and insulin resistance (IR).

The homeostatis model assessment of insulin resistance (HOMA-IR) is calculated as [90]:

fasting serum glucose in mg/dl × fasting serum insulin in mU/l)/405

The lower the calculated HOMA-IR, the higher the degree of insulin sensitivity. In insulin resistant humans, the action of insulin is diminished. The proposed cut-off value for the diagnosis of insulin resistance is a HOMA-IR equal or greater than 2.5[90,91]. As such, not only peripheral insulin resistance, but also central insulin resistance has been reported in obesity[92]. This is important, as insulin receptors have been found in brain regions involved in the reward processing. Hence, alterations in insulin sensitivity might affect motivational behaviour. Insulin resistant mice, for example, have a decreased synthesis of tyrosine hydroxylase, a rate-limiting enzyme in dopamine production[93]. In mice, in which insulin receptors in tyrosine hydroxylase expressing dopaminergic cells were inactivated, body weight was shown to increase due to increased food intake[94].

These results support the thesis that insulin resistance in dopaminergic cells might increase dopaminergic turnover and thus have a significant influence on motivational behaviour. Still, further investigation is needed to understand the exact mechanisms how peripheral insulin resistance affects insulin signalling in the central nervous system, and, therefore, has an impact on food intake and motivated behaviour in humans.

1.4 Impact of metabolic state on motivation

So far it has been demonstrated, that reward-related motivational behaviour is encoded by dopaminergic neurons in the mesostriatal DA system and that functional dopaminergic signalling between the VTA and the NAc is mandatory for the processing of reward-related associations.

Furthermore, the dopaminergic midbrain, specifically the VTA and the NAc, express many receptors that process information of hormones signaling metabolic state from the periphery, i.e. GLP-1 and insulin, suggesting these signals have a modulatory role in motivational behaviour.

To a great part, hormones are involved in signalling homeostatic needs to the brain in order to maintain a balanced energy homeostasis. However, the subjective rewarding value of an external presented cue and, therefore, the willingness to spend effort to obtain it, is also significantly dependent on the current metabolic state in humans[95].

In animals, hunger represents one of the greatest drives regarding motivational behaviour[96]. In human studies the rewarding value of food has been shown to be more desirable in hungry persons in contrast to sated persons[95,97]. The lateral hypothalamus (LH) plays an essential role in regulating energy homeostasis and food intake[98]. This area is closely interacting with the dopaminergic projections of the VTA and has been suggested to affect motivational behaviour in view of food and reward[99]. In rodents, activation of GABAergic LH neurons projecting to the VTA resulted in increased motivational behaviour towards food reward, measured by an increase in lever-pressing for food reward[100].

Considering obesity, the mesostriatal DA system has been shown to be altered in obese compared to lean humans, suggesting altered reward-related motivational behaviour in obese humans contributing to and promoting overeating. Besides alterations of the dopaminergic midbrain, obesity is associated with metabolic impairments affecting hormones signalling the metabolic state such as GLP-1 and insulin.

1.5 Hypothesis

We, therefore, hypothesized that internal state signals, such as hunger, as well as GLP-1 and insulin sensitivity modulate reward-related motivational behaviour in humans.

The aim of this randomized, placebo-controlled and crossover study was to assess the modulatory role of GLP-1 in reward-related motivated behaviour of humans with varying insulin sensitivity and hunger states[101].

To investigate reward-related motivational behaviour, we asked participants to undergo an adapted behavioural paradigm on the computer, first presented by Pessiglione et al. (2007) and further refined by Le Bouc et al. (2016)[37,102], in a fasted state. In this task, different amounts of food and monetary reward could be earned by investing effort in squeezing a handgrip.

Before subjects performed the task, hunger was assessed using visual analogue scale and blood samples were taken to measure glucose and insulin.

The effect of GLP-1 was assessed using a liraglutide intervention in comparison to a placebo condition.

The present study was approved by the Ethics Committee of the University Hospital of Cologne (Cologne, Germany; No. 16-251).

2 Publication

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Brief Communication

MOLECULAR METABOLISM

GLP-1 and hunger modulate incentive motivation depending on insulin sensitivity in humans

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ABSTRACT

Objective: To regulate food intake, our brain constantly integrates external cues, such as the incentive value of a potential food reward, with internal state signals, such as hunger feelings. Incentive motivation refers to the processes that translate an expected reward into the effort spent to obtain the reward; the magnitude and probability of a reward involved in prompting motivated behaviour are encoded by the dopaminergic (DA) midbrain and its mesoaccumbens DA projections. This type of reward circuity is particularly sensitive to the metabolic state signalled by peripheral mediators, such as insulin or glucagon-like peptide 1 (GLP-1). While in rodents the modulatory effect of metabolic state signals on motivated behaviour is well documented, evidence of state-dependent modulation and the role of incentive motivation underlying overeating in humans is lacking.

Methods: In a randomised, placebo-controlled, crossover design, 21 lean (body mass index [BMI] < 25 kg/m^2) and 16 obese (BMI[3] 30 kg/m^2) volunteer participants received either liraglutide as a GLP-1 analogue or placebo on two separate testing days. Incentive motivation was measured using a behavioural task in which participants were required to exert physical effort using a handgrip to win different amounts of food and monetary rewards. Hunger levels were measured using visual analogue scales; insulin, glucose, and systemic insulin resistance as assessed by the homeostasis model assessment of insulin resistance (HOMA-IR) were quantified at baseline.

Results: In this report, we demonstrate that incentive motivation increases with hunger in lean humans ($F_{(1,42)} = 5.31$, $p = 0.026$, $\beta = 0.19$) independently of incentive type (food and non-food reward). This effect of hunger is not evident in obese humans ($F_{(1,62)} = 1.93$, $p = 0.17$, $\beta = -0.12$). Motivational drive related to hunger is affected by peripheral insulin sensitivity (two-way interaction, $F_{(1,\ 35)} = 6.23$, $p = 0.017$, $\beta = -0.281$). In humans with higher insulin sensitivity, hunger increases motivation, while poorer insulin sensitivity dampens the motivational effect of hunger. The GLP-1 analogue application blunts the interaction effect of hunger on motivation depending on insulin sensitivity (three-way interaction, $F_{(1,\ 127)} = 5.11$, $p = 0.026$); no difference in motivated behaviour could be found between humans with normal or impaired insulin sensitivity under GLP-1 administration.

Conclusion: We report a differential effect of hunger on motivation depending on insulin sensitivity. We further revealed the modulatory role of GLP-1 in adaptive, motivated behaviour in humans and its interaction with peripheral insulin sensitivity and hunger. Our results suggest that GLP-1 might restore dysregulated processes of midbrain DA function and hence motivational behaviour in insulin-resistant humans.

Keywords Glucagon-like peptide-1; Insulin sensitivity; Regulation of motivational behaviour; Obesity; Hunger

1. INTRODUCTION

The growing obesity epidemic represents one of the greatest health challenges of the 21st century, leading to increased risk of severe comorbidities such as cardiovascular disease or cancer [1,2]. While continuous excessive food intake has long been identified as one of the leading causes promoting obesity, the physiological mechanisms driving food intake behaviour and overeating remain poorly understood, especially in humans.

Our daily behaviours are driven by basic needs often without us noticing. Hunger is one of these basic behavioural drivers, as food serves as the energetic foundation for all biological processes. To

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regulate the body's need for food and the subsequent internal sensation of hunger, the brain has developed precise physiological and behavioural mechanisms to keep the body operating at optimal levels; to ensure this physiological homeostasis and adapt behavioural responses, our brain constantly integrates information about the metabolic state with external environmental cues [3–5]. Although there has been much investigation on the basic homeostatic mechanisms of hunger and the behavioural consequences thereof [6,7], significant gaps remain in understanding how metabolic signals prompt and external cues incentivise the behavioural aspect of the hunger response.

External cues comprise strong motivational signals, such as the incentive value of an expected reward, but also the effort required to obtain the reward [8–10]. Hence, everyday decisions in favour of or against food intake are based on cost-benefit analyses weighing the potential food reward against the cost of spending effort to obtain it. The incentive theory of motivation [11,12] suggests that behaviour is primarily motivated by anticipated rewards and reinforcement. Thus, behavioural drive and hence incentivised motivation refers to the processes that translate expected reward into effort spent [13]. These processes include forming a subjective representation of the potential reward magnitude, which determines effort exertion [14] and is critical for initiating motivated behaviour [9,15]. Notably, the subjective reward magnitude depends markedly on the internal state of the organism [10]; a food reward is regarded as more valuable in a hungry than in a sated state [16].

The magnitude and probability of a reward [9,17] is encoded by dopamine (DA) neurons in the ventral tegmental area (VTA) and its mesoaccumbens DA projections. By promoting the formation of cue–reward associations, VTA DA neurons play a central role in mediating motivated behaviours [18]. DA terminals in the nucleus accumbens (NAc) specifically respond to reward-predictive cues [19,20]. In fact, the activity of VTA DA neurons and NAc DA levels have the capacity to prompt reward-directed action initiation and effort exertion [21,22]. In human pharmacological intervention studies, a lower dopaminergic tone was shown to result in lower effort spending and motivation [8,23,24].

Related to overeating, alterations in the mesoaccumbens DA pathway have been consistently linked to obesity in animal studies [25–27]. In obese humans, alterations in the fronto-mesostriatal DA circuitry have been generally related to an impaired reward system [28,29]. Reduced binding potential of striatal dopamine receptors has been hypothesised to be associated with a heightened striatal dopaminergic tone, leading to an imbalance between anticipation and consumption of food reward [30–35]. While it was reproducibly shown that obese vs lean humans show greater neural activation in reward-related regions anticipating rewarding stimuli, neural activation in response to obtained food rewards decreases [36–39]; however, findings of the reinforcing capacity and its link to incentive motivation and effort spending underlying obesity portray a heterogeneous picture [40,41].

These studies, however, may rest on an incomplete assumption about modulatory aspects of midbrain DA function and body mass index (BMI) as decisive variables to nuanced facets of motivated behaviour. While being strongly implicated in incentive motivation, VTA DA neuron firing and mesoaccumbens DA pathways are also particularly sensitive to nutritional value [16,27], post-ingestive effects of food [42,43], and metabolic state signalled by neuropeptides and peripheral peptidergic mediators [44–47]. These bodily signals may bias our food intake behaviour more than just external cues per se; in fact, they may also shape the incentive cue value, mediating its reinforcement efficiency depending on the metabolic state.

Notably, VTA DA neurons express many receptors that respond to peripheral peptides that signal metabolic status [48]; the glucagon-like peptide 1 (GLP-1) receptor is a particularly prominent example [49,50] that has also been used as target for the development of drug therapies aimed at curbing overeating [51,52]. While GLP-1 acts primarily on pancreatic islets to enhance glucose-induced insulin secretion, it can induce metabolic actions to maintain glucose homeostasis by interacting with its receptors expressed on neurons and cells in the enteric and central nervous system [53,54].

Related to the mesoaccumbens DA pathway, rodent studies demonstrated that activation of GLP-1 receptors in the VTA by endogenous GLP-1 specifically reduces the excitatory synaptic strength of DA neurons that project to the NAc [55], decreasing the reinforcing efficiency of appetitive cues and adapting motivated behaviour [51,56–58].

In line with the work in rodents, GLP-1 analogues were reliably shown to lead to reduced food intake and to induce weight loss in obese humans [59–61]. However, while in rodents the modulatory effect of GLP-1 on DA neurocircuitry and motivational behaviour is well documented [56,62], evidence of a modulatory role of GLP-1 affecting motivational behaviour in humans is lacking.

To this end, the present randomised, placebo-controlled, and crossover study assessed the modulatory role of GLP-1 in motivated behaviour of lean (BMI $<$ 25 kg/m^2) in comparison to obese (BMI^3 30 kg/m^2) humans. To account for the hunger state, subjective hunger ratings were assessed. To consider the metabolic state and particularly the physiological role of GLP-1 in maintenance of glucose homeostasis, fasted insulin and glucose levels were acquired. As a readout for incentivised motivated behaviour, we adapted a classic behavioural paradigm of effort spending that was first suggested by Pessiglione et al. [63] and further refined by Le Bouc et al. [23]. In this task, different amounts of possible reward are used as incentives. Volunteer participants were required to exert physical effort (force) on a handgrip to win different amounts of food and non-food rewards (money).

Interestingly, although we detected a differential effect of hunger on incentive motivation between lean and obese humans ($F_{(1, 137)} = 3.98$, $p = 0.048$), we could not find a GLP-1-interaction with BMI. Based on the physiological role of GLP-1 in the regulation of insulin secretion and prior evidence that insulin sensitivity modulates excitatory input of VTA DA neurons [64] as well as mesostriatal functional connectivity [65], we predicted that GLP-1 interactions with motivational functions of DA might change with BMI depending on peripheral insulin sensitivity. Hence, we analysed the effect of insulin sensitivity on incentive motivation with peripheral insulin sensitivity being assessed by the homeostasis model assessment of insulin resistance (HOMA-IR) [66,67]. It is important to note that none of the studies' participants suffered from diabetes.

2. MATERIALS AND METHODS

2.1. Participants

Twenty-five subjects with normal weight (BMI: 22.42 $\pm$ 0.22 kg/m2) and 25 obese subjects (BMI: 35.61 $\pm$ 0.87 kg/m2) were recruited from the pre-existing database of volunteers maintained at the Max Planck Institute of Metabolism Research based on a power analysis assuming an a error = 0.05, power = 0.95, and a small-to medium-effect size of Cohen's d = 0.35 (equivalent to f = 0.175). This power analysis was performed for a mixed-effects model targeting the three-way interaction of group (lean vs obese), intervention (GLP-1 vs placebo), and hunger level, yielding a total sample size of N = 46.

All the participants were non-smokers between the ages of 20–40 years with no history of neurological, psychiatric, metabolic, or eating disorders. In the course of the analysis, 13 subjects had to be excluded due to low engagement in the task: 7 subjects were excluded as they reported wanting of the monetary or food reward lower than 3 (of 10) points, 3 participants had not invested sufficient effort in the calibration so they engaged constantly to press with >75% of their maximum force for every single trial, and for 3 participants their preferred amount of food reward was not clearly identifiable. Hence, 21 lean (BMI: 22.56 ± 0.38 kg/m2, age: 26.87 ± 1.4 years, 9 female) and 16 obese (BMI: 35.32 ± 1.36 kg/m2, age: 27.20 ± 1.3 years, 9 female) subjects were included for further data analysis (Table 1).

After analysing the effects of groups stratified by BMI (normal vs obese), we examined how incentive motivation may change with systemic insulin resistance as assessed by the homeostatic model of insulin resistance (HOMA-IR) [66; 67]. For this purpose, the HOMA-IR of each participant was calculated as (fasting serum glucose in mg/dl × fasting serum insulin in mU/l)/405, with lower values indicating a higher degree of insulin sensitivity. Only the HOMA-IR of the placebo day was calculated, as GLP-1 analogues may increase insulin secretion and alter the HOMA-IR. The HOMA-IR was used as a continuous variable later in the analysis. The final subject selection (N = 37) allowed for a power of 0.91 for the three-way interaction of the HOMA-IR, hunger, and intervention within the used mixed-effects model, with an effect size of partial h2 = 0.03 (equivalent to f = 0.176) and an a error = 0.05.

All the subjects provided written informed consent to participate in the experiment, which was approved by the local ethics committee of the Medical Faculty of the University of Cologne (Cologne, Germany; No. 16–251).

2.2. Study design

The study was conducted in a single blinded, placebo-controlled, randomised, cross-over design. Each volunteer participated on two testing days lasting a maximum of 2 h each. Both testing days were separated by at least one week to allow for a sufficient wash-out period [68]. The order of the intervention (GLP-1 vs placebo) was counter-balanced (Figure 1A).

The evening prior to each testing day, the participants received an agonistic GLP-1 analogue (see as follows) or an equal volume of saline solution and a standardised dinner with equal kcal amounts per individual (see Supplemental Material). The next morning, the participants arrived fasted at the institute at 8 a.m. and their BMI was measured using a Seca mBCA 515 (medical Body Composition Analyser). As this study was part of a larger experiment, all the participants not only underwent the behavioural task detailed as follows but also an fMRI task that was related to a different study question (sensory learning) and that is reported elsewhere. The order of the behavioural task and fMRI task was counterbalanced.

Table 1 – Participants' characteristics.

Parameter	Mean ± SEM
N (lean)	37 (21)
Age (years)	26.49 ± 0.9
lean	26.87 ± 1.4
obese	27.20 ± 1.3
BMI (kg/m^2)	27.68 ± 1.22
lean	22.56 ± 0.19
obese	35.32 ± 0.66

Note: BMI = body mass index, SEM = standard error of the mean.

Before each task, hunger levels were assessed via visual analogue scales and blood drawn to measure insulin and glucose. After the behavioural task, individual liking and wanting of the reward types (money and food) and the participants' compliance were evaluated in a short debriefing.

2.3. GLP-1 analogue

A subcutaneous injection of 0.6 mg of liraglutide (Novo Nordisk) was used as an agonistic GLP-1 analogue. As the maximum plasma concentration of liraglutide is reached approximately 11–13 h after injection [68], liraglutide was administered the evening prior to the testing day between 7 and 8 p.m. to ensure sufficient blood plasma levels at the start of the testing day. As a placebo condition, an equal volume of saline solution was injected subcutaneously.

2.4. Incentive motivation task

Incentive motivation was assessed by measuring effort spending for external cues in a behavioural paradigm (Figure 1B). Two incentive types (food and money) could be earned by squeezing a handgrip device (hand dynamometer HD-BTA, Vernier). The task was programmed in MATLAB (version 2014b, MathWorks) using the Psychophysics Toolbox (version 3.0.11) [69,70] and a toolbox dedicated to enabling communication between MATLAB and the device (https://github.com/lionel-rigoux/vernier-toolbox).

Prior to the task, the participants performed a calibration in which they were instructed to squeeze the handgrip three times in a sequence as hard as possible. For each participant, the highest force reached, $F_{max} = \max(F)$, was scaled to define the subject's individual maximum voluntary contraction C_{max} :

$$C_{max} = 1.05 * F_{max}.$$

Each participant's individual maximum contraction C_{max} was used as a reference point related to the force of each trial in the task. Note, F_{max} was scaled up to ensure that C_{max} was always higher than the maximum force in each trial during the task. However, if the subjects' maximum force in one trial during the incentive motivation task exceeded the initially calibrated C_{max}, the maximum force exerted during this trial was used to recalculate the maximum voluntary contraction in subsequent analyses.

Following the calibration, printed instructions were provided to each participant. The subjects were informed that the percentage of exerted maximum force related to how much of the presented stimulus they could win (e.g. if a subject squeezed with 80% of their maximum voluntary contraction, they could win 80% of the displayed food or monetary stimulus). Before starting the task, a short training session of 4 test trials (2 food and 2 money trials) with randomly assigned stimulus amounts was provided.

In total, the task comprised 128 trials with a total duration of 25 min. The trials were divided into 4 blocks displaying monetary stimuli and 4 blocks with food stimuli. Hence, any block consisted of 16 trials in which varying stimulus amounts were presented. Food and monetary blocks alternated. Each trial consisted of two phases: a response phase and feedback phase.

During the response phase, one of four monetary amounts (0.6 €, 1.2 €, 1.8 €, or 2.4 €) or one of four food amounts (1/4, 2/4, 3/4, or 4/4 of a bread roll with cheese) was displayed as an incentive on the left-hand side of the screen. While the food or monetary stimulus was displayed, the participants exerted force on the handgrip. Feedback of the performance was directly provided by an orange bar ascending in

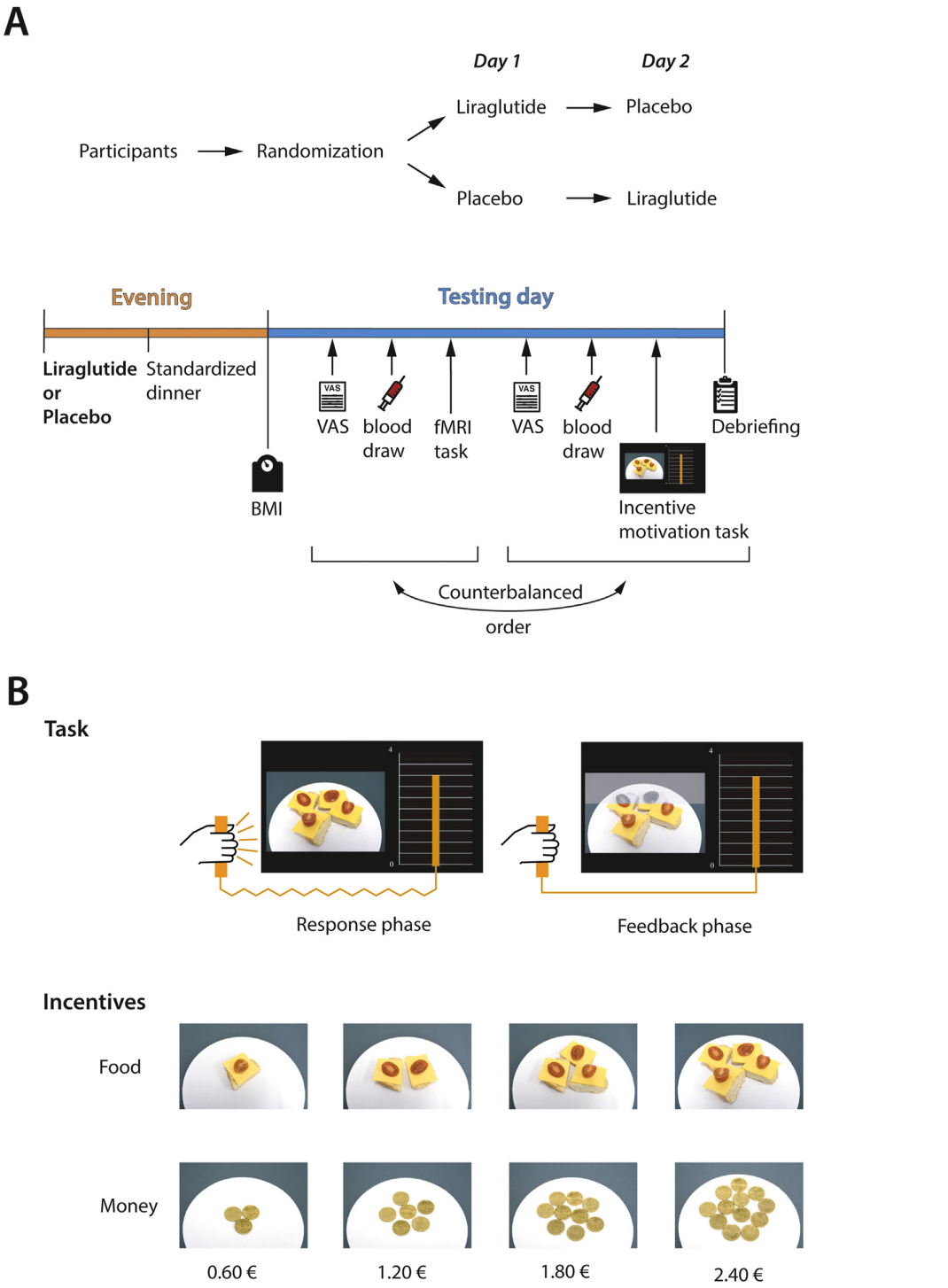

Figure 1: Study design and behavioural task. A. In this within-subject design, the subjects received either placebo or liraglutide and a standardised dinner the evening prior to each testing day. After fasting overnight, BMI, hunger rating, and a blood draw (glucose and insulin levels) were assessed the next morning followed by the behavioural task with a short debriefing. B. After a fixation cross, the subjects were shown either a monetary cue (0.6 €, 1.2 €, 1.8 €, or 2.4 €) or a food cue (1/4, 2/4, 3/4, or 4/4 of a bread roll with cheese) presented on a white plate, for which the participants exerted force to win the presented cues. Online feedback on the force produced was provided by an orange bar ascending with increasing force exertion. During the following feedback phase, the participants could relax their arm, and direct feedback about the amount of the presented reward that the subjects would have won was displayed by a colour change in the cue image. The different levels of incentives (food and money) are also depicted in detail. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

height in proportion to the exerted force. Each stimulus amount was displayed 4 times per block. The response phase lasted for 3 s.
During the following feedback phase, the participants could relax their arm, and direct feedback on the amount of the presented reward the participant would have won was displayed (for example, if the participant exerted 80% of his/her maximum voluntary contraction, 80% of the displayed stimulus changed in colour representing the amount won). The feedback phase lasted for 3.5 s.
The subjects were informed that one food trial and one monetary trial were chosen at random at the end of the experiment and that they were granted the reward won in this trial. To provide a supplementary stimulus to motivate the subjects for greater effort exertion, one plate with the food stimulus and one with the monetary stimulus were placed within viewing range of each subject during the task.

2.5. Hunger and liking ratings

To control for differences in hunger states between testing days, we instructed the participants to rate their hunger prior to the task on each testing day using a visual analogue scale as previously described [71]. In brief, on a 100 mm visual analogue scale (0 mm = "sehr hungrig (very hungry)" and 100 mm = "gar nicht hungrig (not hungry at all)"), the subjects were asked to mark the point that most accurately represented their perception of their current hunger state.
Likewise, to explore the individual incentive value of cues, liking of the items (separately for money and food) was rated on a 100 mm vertical scale with "Mag ich gar nicht (not liking the item at all)" at the lower anchor point and "Mag ich sehr (liking the item a lot)" at the upper anchor point [72].

2.6. Insulin levels

As GLP-1 analogues are reported to increase insulin secretion [73], we monitored insulin levels to control for insulin effects at the onset of the behavioural task. This monitoring was achieved by a blood draw directly before starting the task and measuring the insulin level within. Glucose levels were assessed from the same blood draw.

2.7. Statistical analysis

Statistical analysis was performed in MATLAB (version 2014b, MathWorks) and R (version 4.0.0) [74] using the lmerTest package (version 3.1–2) [75]. GraphPad Prism (version 8.0) was used to visualise the results. Statistical significance was reported at a level of $p < .05$.
The analysis of the acquired data followed a two-level approach. On a first (subject) level, a general linear model was used to assess the interaction of incentive and force exerted for food or monetary reward separately. Note, as not all the participants experienced the same amount of food as rewarding, incentive levels were recalibrated based on the amount the subjects individually preferred (for details, see Supplemental Material). Both incentive types were analysed separately to include the type of incentive for the subsequent second-level analysis. Hence, the following statistical models were applied to the data:

$$\text{force} \sim \beta_0^m + \beta_1^m \text{. level of incentive}$$

for money as an incentive type and

$$\text{force} \sim \beta_0^f + \beta_1^f \text{. level of incentive}$$

for food as an incentive type, where the coefficients indexed with m related to monetary and those indexed with f related to food incentives. The regression coefficients β_1 represent the individual motivation to spend physical effort for incentives. That is, a low β_1 indicates that a participant did not increase their effort spending much with increasing incentive and thus revealed a low incentive motivation.
A further analysis was performed to test for differences in force exertion between the two types of incentives. In this study, a mixed-effects model was established including "type of incentive" (money/food), "level of incentive," and their interaction as independent variables (fixed effects) and subject as the random effect. Post hoc comparisons were calculated using Tukey's procedure facilitated by the lsmeans package (version 2.30-0; [76]) in R.
On the second (group) level, we assessed the effect of the intervention (GLP-1 vs placebo) on incentive motivation (β_1^m and β_1^f) with a mixed-effects model considering the independent variables "type of incentive" (money vs food), intervention (GLP-1 vs placebo), hunger, and group (lean vs obese) as fixed effects with the subject as a random effect. This design lent structure to examining the effects of the intervention and hunger while considering the BMI as a group-differentiating criterion. Thus, hunger was used as a continuous variable while group, intervention, and "type of incentive" were used as factorial variables. We also controlled for the individual baseline insulin level, liking of the respective incentive, and order of the testing days. To consider an effect of peripheral insulin sensitivity on motivation, we established a further mixed-effects model with the independent variable HOMA-IR instead of group (lean vs obese). Thus, HOMA-IR and hunger were used as continuous variables and intervention (GLP-1 vs placebo) as well as "type of incentive" (money vs food) as factorial variables:

$$\beta_1^m \text{ and } \beta_1^f \sim \textit{hunger} * \textit{HOMA} - \textit{IR} * \text{int}\textit{ervention} * \textit{incentive type} + \textit{insulin} + \textit{liking} + \textit{day} + (1|\textit{subject ID})$$

Given the complexity of our three-way interaction of intervention, hunger, and HOMA-IR ($F_{(1,\ 127)} = 5.11$, $p = 0.026$), we also analysed the effect of the intervention (GLP-1 or placebo) separately with mixed-effects models testing the effect of incentive, hunger, and HOMA-IR while controlling for insulin levels, liking of incentive, and measuring day.

3. RESULTS

To evaluate the modulatory role of GLP-1 in motivated behaviour, we conducted a placebo-controlled behavioural study employing an established effort spending task in lean and obese human participants. In this task, individuals were required to exert physical effort using a handgrip to win different amounts of reward and adapt their behaviour to changing external cues with different reward types. To examine implications of reinforcement, we incentivised the task with either a food or non-food reward; to consider the impact of internal state modulation, we assessed hunger levels; and we tested for the modulatory role of GLP-1 and its interaction with peripheral insulin sensitivity, hunger, and incentive on task outcome.

3.1. Effort exertion increased with increasing incentive value and liking

In addition to the suggestion that behaviour is motivated by internal drives (hunger), incentive theory suggests that external cues leverage reinforcement (incentives). Thus, in comparing behavioural performance on the task between different types of incentives (food vs money), we examined force exertion for the varying levels of food and monetary amounts (1/4 up to 4/4 of a bread roll with cheese or 0.60 € up to 2.4 €). In congruence with Pessiglione et al. (2007) revealing that individuals spend higher effort for higher reward value per se (the

main effect of level of incentive on force: $F_{(3,9435)} = 1886.73$, $p < 0.0001$; see Supplemental Material), we also found a significant two-way interaction revealing that force exertion increased more for increasing monetary rewards than for increasing food rewards ($F_{(3,9435)} = 65.42$, $p < 0.0001$, Figure 2A; for further details, see Supplemental Material).

Furthermore, as liking of a reward is essential for its incentive value, we controlled for the effect of liking on effort exertion, revealing that higher liking of the incentive linked to stronger motivational drive ($\beta = 0.016$, $t = 2.35$, $p = 0.02$; for further details, see Supplemental Material).

3.2. In lean humans, incentive motivation increased with increasing hunger levels

We assessed the modulation of effort exertion under GLP-1 depending on hunger level in lean and obese humans (lean BMI $<$ 25 kg/m^2, obese BMI ^{3} 30 kg/m^2). While the three-way interaction of intervention, hunger, and group was borderline to significant ($F_{(1,\ 116)} = 3.68$, $p = 0.057$; Supplemental Table 4), a significant two-way interaction of group and hunger ($F_{(1,\ 137)} = 3.98$, $p = 0.048$) was detected. Given the complexity of such an interaction, however, and to ascertain how hunger affects incentive motivation differently in lean and obese humans, we analysed both groups separately showing that in the lean

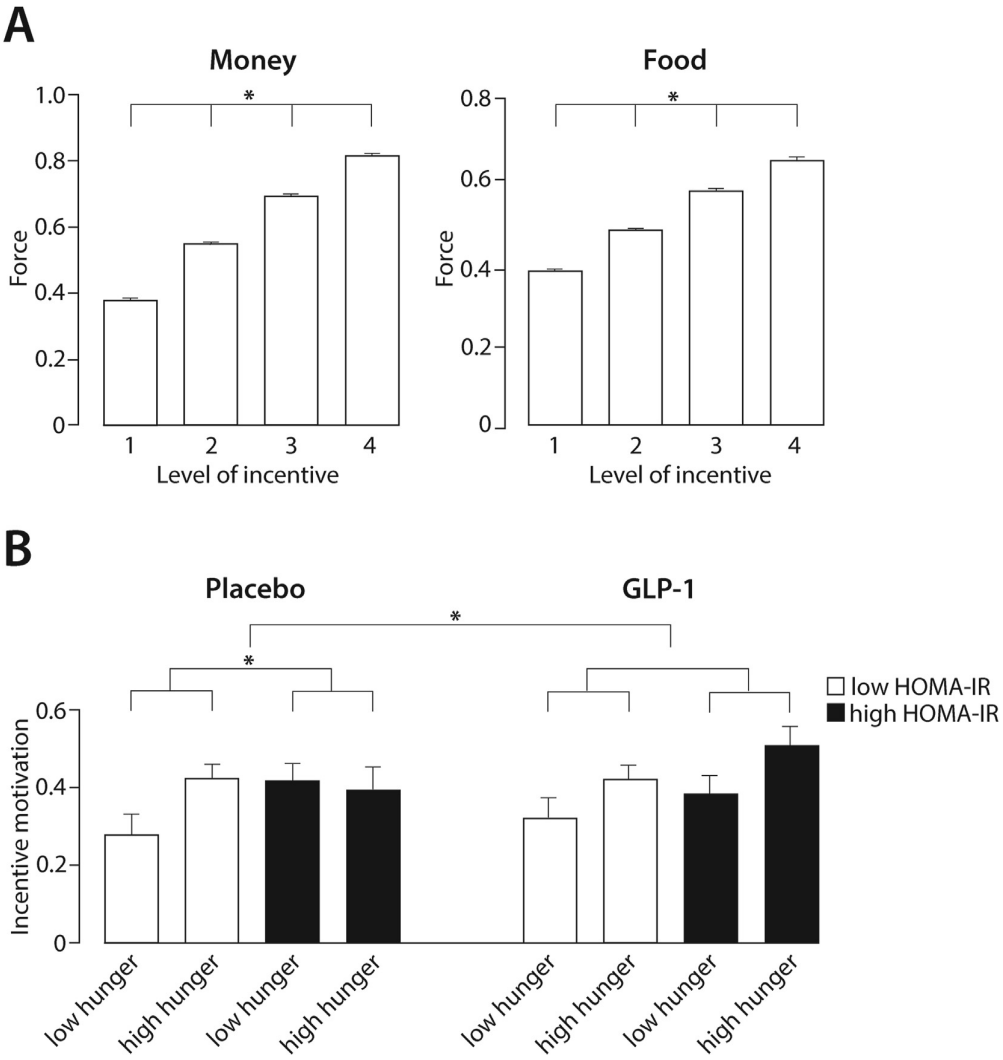

Figure 2: A. Force exertion for different levels of incentive. The force exerted by all of the participants for monetary and food rewards is shown as mean ± SEM. The exerted force increased with higher amounts of incentives. B. GLP-1 modulated incentive motivation depending on insulin sensitivity and hunger. The modulatory effect of GLP-1 was tested in a three-way interaction of hunger HOMA-IR intervention ($F_{(1,\ 120)} = 5.11$, $p = 0.026$). In the placebo condition, in the individuals with a low HOMA-IR, increasing hunger levels promoted incentive motivation; in turn, in those with a high HOMA-IR, higher hunger levels did not lead to an increase in incentive motivation ($F_{(1,\ 35)} = 6.23$, $p = 0.017$, $\beta = -0.281$). Under the GLP-1 intervention, no significant difference between the participants with low HOMA-IR and high HOMA-IR could be detected (two-way interaction, $F_{(1,\ 34)} = 0.13$, $p = 0.72$). In the analysis, hunger and HOMA-IR were used as continuous variables. For illustration purposes, HOMA-IR is depicted as a categorical variable distinguishing between the participants with a low HOMA-IR and high HOMA-IR using a median split (median = 1.68) shown as mean with SEM.

subjects, incentive motivation increased with increasing hunger levels ($F_{(1,\ 42)} = 5.31$, $p = 0.026$, $\beta = 0.19$; Supplemental Table 5). In the obese group, however, no significant effect of hunger on incentive motivation was detectable ($F_{(1,\ 62)} = 1.93$, $p = 0.17$, $\beta = -0.12$; Supplemental Table 6).

3.3. Modulation of incentive motivation by GLP-1 differed depending on insulin sensitivity and hunger level

Based on the physiological role of GLP-1 in regulating insulin secretion [53] and evidence linking insulin signalling to motivational behaviour (see Introduction), we hypothesised that GLP-1 interactions with motivational functions might change depending on peripheral insulin sensitivity. Hence, we tested the three-way interaction of GLP-1, HOMA-IR, and hunger level ($F_{(1,\ 127)} = 5.11$, $p = 0.026$; Supplemental Table 7), which clearly indicated a modulation of incentive motivation by GLP-1 differing between the participants depending on their peripheral insulin sensitivity and hunger level. To ascertain this complex interaction, we analysed the GLP-1 and placebo condition separately (Figure 2B).

For the placebo condition, we found a significant interaction between hunger and insulin resistance ($F_{(1,\ 35)} = 6.23$, $p = 0.017$, $\beta = -0.281$), revealing that with increasing HOMA-IR, the positive effect of hunger on incentive motivation was reduced. Hence, in the subjects with a high peripheral insulin sensitivity, increasing hunger levels promoted incentive motivation; in turn, in those with poorer insulin sensitivity, higher hunger levels did not lead to an increase in incentive motivation (see Supplemental Table 8). In the GLP-1 intervention, no significant difference between the participants with good insulin sensitivity and poor insulin sensitivity could be detected (two-way interaction hunger: insulin sensitivity, $F_{(1,\ 34)} = 0.13$, $p = 0.72$; see Supplemental Table 9).

4. DISCUSSION

To regulate food intake, our brain constantly integrates internal state signals such as hunger with external cues, such as the incentive value of a potential food reward. To adapt behavioural responses, metabolic modulators from the periphery impact on brain circuitry to ensure physiological homeostasis. This study provides an analysis of the modulatory effect of hunger on motivated behaviour in humans by considering GLP-1 and insulin sensitivity as metabolic modulators as well as the role of the incentive value reflected by different external cues (money and food).

Regarding the role of external cues, we demonstrated that the level of liking of the presented cues determined their incentive value as higher liking rendered higher motivation. To this end, it is important to note that our findings suggest a role of metabolic modulation of motivated behaviour independent of incentive type (food or money). This finding underscores evidence from psychological literature suggesting that biologically based motivation can affect behaviours in unrelated domains that are irrelevant to the biological motive [77]. From a more neurobiologically centred perspective, modulation of motivational drive independent of incentive type might indicate neural encoding by basal subcortical circuits, as "cognitive" cortical representations of motivationally relevant cues would influence action selection by weighing the current physiological needs against the predicted consequences of responding to certain cues [78].

Addressing the role of the internal state and metabolic signals as modulators of motivation, we demonstrated that incentive motivation depends on the internal state in lean humans, as it increases with increasing hunger. In obese humans, however, hunger does not affect incentive motivation. This effect of hunger on motivational drive is modulated by the insulin sensitivity of the individual, as the degree of insulin resistance determines the magnitude of the effect of hunger on incentive motivation under placebo conditions. The higher the HOMA-IR index, the lower is the positive effect of hunger on incentive motivation (see Figure 2B). This effect of hunger revealed in our work was also observed in animal (rodent) studies showing that neurons in the lateral hypothalamus (LH) sense fasting (or sated) states and regulate motivation [45,79,80]; low glucose levels that occur in the fasting state activate glutamate and orexin co-expressing neurons in the LH, which project to and excite VTA DA neurons [81]. GABAergic LH-neurons project to the VTA [82], and their activation increases motivation for food [79]. As previously mentioned, our findings significantly extend the effect of hunger on incentive motivation and non-food rewards in humans, as the influence of hunger and insulin sensitivity applied for both types of incentives (food and money). This is in line with animal studies deciphering GABAergic LH inputs in the VTA that contribute to motivational salience in multiple contexts [83].

Our results also reveal that the effect of hunger on incentive motivation is modulated by the peripheral insulin sensitivity of the individual. In previous studies, we showed that systemic insulin sensitivity may have an impact on DA projections of the midbrain [65], which is in line with other recent human studies emphasising that peripheral insulin sensitivity is a better predictor of altered DA signalling than BMI [84–86]. Animal studies revealed altered DA clearance and synthesis in the VTA and DA terminals in the NAc due to insulin resistance [87,88], and insulin resistance has been associated with maladaptive eating and motivational behaviour [87,89,90]. However, the detailed neuronal mechanisms on how insulin sensitivity affects DA signalling within the midbrain and hence incentive motivation remain to be elucidated.

We further show that upon GLP-1 receptor agonist (liraglutide) treatment, the effects of hunger and insulin resistance on incentive motivation are blunted as no differential effect of hunger on motivation depending on insulin sensitivity could be detected. Thus, no difference between insulin-resistant and insulin-sensitive humans could be identified under GLP-1 treatment, indicating that GLP-1 normalises the effect of hunger and insulin sensitivity on motivation. While Figure 2B suggests that GLP-1 restores motivational drive in insulin-resistant humans to a non-insulin-resistant levels, it cannot be statistically differentiated if GLP-1 reinstated the effect of hunger on motivation in the insulin-resistant participants or blunted the effect of hunger on motivation in the insulin-sensitive subjects (or both; see Supplemental Tables 10 and 11). Considering that GLP-1 application has a stronger effect on peripheral insulin secretion in insulin-resistant humans than insulin-sensitive individuals [91], it seems reasonable to assume that the central effect of GLP-1 is equally stronger in insulin-resistant humans than insulin-sensitive individuals, indicating that improvement of insulin-resistant humans is likely.

Related to treatment with GLP-1 receptor agonists, while the peripherally administered agonist exendin-4 was revealed to bind to both astrocytes and neurons in the VTA in rodents [50], peripherally administered liraglutide has not yet been shown to enter the VTA or NAc but could be detected in the circumventricular organs, hypothalamus (the paraventricular nucleus, supraoptic nucleus, and supraoptic decussation but not the lateral hypothalamus) [92,93], and solitary nucleus [NTS, 93]. Hence, although not demonstrated, it can be hypothesised that peripheral liraglutide may also bind directly to GLP-1 receptors within the VTA and thus affects motivational behaviour. In rodent studies with normal-weight animals, GLP-1 receptor activation in the VTA was reported to reduce motivational behaviour [56,57]. In detail, phasic DA responses in the VTA to food-predictive cues could be

suppressed by the central administration of the GLP-1 receptor agonist exendin-4 [62]. However, it needs to be considered that these murine results are not fully comparable to our human results as hunger/fasting times were not included in the analysis.

An alternative and more likely access route might be via vagal afferents in the NTS, as peripheral liraglutide was shown to enter the NTS [93]. Peripherally administered liraglutide might bind to GLP-1 receptor-expressing glutamatergic and GABAergic neurons [93] as well as astrocytes [94,95] within the NTS, which regulate the GLP-1 producing neurons in the NTS. These GLP-1 producing neurons project to the VTA, suppressing activity of DA neurons in the mesoaccumbens pathway [55]. Comparing the influence of GLP-1 on incentive motivation in our human study with animal reports, similar behavioural results were found in normal-weight rodents with activation of GLP-1 receptors in the NTS reducing food and drug reward behaviour by targeting VTA DA neurons [96,97]. However, data on the effect of GLP-1 on motivational behaviour in insulin-resistant/obese rodents and the effect of hunger/fasting time are lacking.

Collectively, one reasonable mechanism underlying our behavioural findings is that GLP-1 receptor activation possibly in the NTS modulates incentive motivation through its action on midbrain DA neurons, which are regulated by hunger and insulin sensitivity.

Methodological caveats worth pointing out are first, that although it was controlled for peripheral insulin levels in our analyses, it needs to be considered that the effect attributed to GLP-1 on motivation could also be an overlapping effect of GLP-1 and insulin, as insulin is secreted in a GLP-1 and insulin sensitivity-dependent manner [91]. Second, although our behavioural results are greatly compatible with the aforementioned animal data on the underlying neuronal processes, the proposed molecular mechanisms explaining the observed behaviour remain speculative. Our approach will thus require further validation on a neural level.

In sum, we provide an assessment of the regulation of incentive motivation in humans by internal and metabolic state parameters as reflected by hunger, GLP-1, and insulin sensitivity, respectively. We propose an explanatory approach addressing the underlying neural mechanisms of the observed behaviour. Moreover, our results suggest a role of GLP-1 to restore dysregulated processes underlying motivational behaviour in obesity.

AUTHOR CONTRIBUTIONS

Ruth Hanssen: Conceptualisation, methodology, software, formal analysis, investigation, and writing.
Alina Chloé Kretschmer: Software, investigation, and formal analysis.
Lionel Rigoux: Methodology, software, and validation.
Kerstin Albus: Investigation and project administration.
Sharmili Edwin Thanarajah: Software and validation.
Tamara Sitnikow: Investigation.
Corina Melzer: Software and validation.
Oliver A. Cornely: Project administration and funding acquisition.
Jens C. Brüning: Funding acquisition, conceptualisation, and supervision.
Marc Tittgemeyer: Funding acquisition, conceptualisation, supervision, and writing.
All of the authors agreed on the final version of the manuscript.

DATA AVAILABILITY STATEMENT

All the data will be made available upon request. The request necessitates that the purpose of data re-analysis is in line with the study's aims as approved by the ethics review board (see text) and the participants' consent. Furthermore, consent to data privacy must be assured by signing an agreement form accordingly.

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APPENDIX A. SUPPLEMENTARY DATA

Supplementary data to this article can be found online at https://doi.org/10.1016/j.molmet.2021.101163.

CONFLICT OF INTEREST

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3 Discussion

Obesity represents one of the biggest global health issues and is associated with altered reward-related behaviour. As such, alterations in mesolimbic DA midbrain signaling as well as dysregulated metabolic signaling of homeostatic needs in obesity have been related to overeating and to promote obesity. The reinforcing value of palatable food is able to promote motivated behaviour in humans, going beyond actual metabolic needs. Motivated behaviour towards food reward in the absence of an energy deficit can lead to excessive food intake and calorie intake, which can lead to obesity. The present study analyzed the modulatory role of internal state signals, such as hunger and metabolic signaling by GLP-1 and insulin sensitivity on motivated behaviour towards food and monetary reward in humans.

3.1 "Liking" and the reward value predict effort spending in humans

We found, that participants were more motivated to spend physical effort for rewards with higher value (e.g. greater amount of food or monetary reward) and rewards, which received higher ratings on the "liking" scale. This is in line with several studies investigating cost-benefit decision making in humans observing that the value and, therefore, the expected reward outcome represent an important motivational drive to exert physical effort[103]. Le Bouc et al. (2016) assessed motivational behaviour in humans in an incentive motivation task and found, that the force peak during squeezing a handgrip increased with higher levels of incentive in the task[102].

Evidence suggests, that subjective liking and wanting of a possible reward has the ability to surpass homeostatic needs and to be processed independent of the actual metabolic state[104]. The pleasure of experiencing the taste of highly palatable food, for example, might represent a strong reinforcer and motivate effort exertion even in the absence of homeostatic needs[105,106]. This is important, as this behaviour could further enhance overeating and promote obesity. Furthermore, subjective "liking" of a reward is believed to be processed independent of dopamine. Berrige et al. (2016) hypothesized, that "liking" as well as "wanting" would be decreased when dopamine is depleted, assuming, that "liking" is mediated by dopamine, as it is the case for "wanting". However, in their experiment carried out with rats, they revealed that

reactions of "liking" towards sweet taste did not change after depletion of dopamine[107]. Our work underscores their result, as liking did not differ between groups and conditions (lean subjects treated with placebo, lean subjects treated with GLP-1, obese subjects treated with placebo, obese subjects treated with GLP-1; see supplemental material of the manuscript) even though motivation, which is encoded by dopamine, differed between groups and conditions.

3.2 Motivational behaviour in lean and obese is not affected by the type of incentive

In our task, subjects could earn food as well as monetary rewards. In the analysis testing the effect of intervention, insulin sensitivity, type of incentive and hunger on incentive motivation, no significant effect of type of incentive was found to affect motivation. This is interesting, as other studies have proposed money to be a more desirable reward compared to food. In the study of Mathar et al. (2016), obese compared to lean participants more often chose to grip for monetary rewards compared to palatable snack reward[38]. However, in our analysis we considered the metabolic state of the participants allowing us to disentangle the different motivational states of both lean and obese humans. We further considered that participants possibly differed in their preferred food and monetary reward amount. As such, participants were interviewed about their preferred food reward amount after performing the task. In a second step, we analyzed for each participant, for which reward amount the highest force was exerted during the task. The mean force exerted for each cue level was calculated and the highest exerted mean force regarded as their preferred reward amount. We excluded participants, for whom there was a mismatch between the highest exerted force and statement about their preferred food reward. Based on the obtained information, the cue levels in the task were recalibrated, taking into account participants' preferred food reward. The same procedure was applied to monetary reward. Here, the maximum reward value was set as the preferred amount of reward.

Our findings suggest that metabolic modulation of reward-orientated behaviour is independent of the reward type and that the homeostatic needs promoting biologically based motivational behaviour affect also behaviours, which do not

correspond to the domains relating to the biological motive. This is in line with findings that have shown that people tend not only to acquire food but also non-food related rewards, when they are hungry[108]. Subcortical circuits have been discussed to be involved in the process of acquisition of reward, that does not satisfy the actual biological need. As no difference in motivated behaviour towards food and monetary reward could be observed in our analysis, our findings support the assumption, that motivation is regulated on a subcortical level, specifically in the dopaminergic midbrain, without cortical influences differentiating between different types of reward.

3.3 Insulin sensitivity modulates the effect of hunger on motivated behaviour

Considering the internal state affecting motivational behaviour, we hypothesized, that obese compared to lean participants would demonstrate altered reward-related motivational behaviour. As expected, reward-related motivated behaviour increased with higher hunger ratings in lean subjects. However, motivational behaviour in obese subjects was not affected by hunger. Our results revealed, that hunger affects motivational behaviour depending on the insulin sensitivity in humans. In the placebo condition, participants with low HOMA-IR reporting high hunger ratings showed higher motivation to spend effort than participants with lower hunger rating. However, in participants with high HOMA-IR the effect of hunger was blunted.

As we have seen previously, insulin is involved in the regulation of dopaminergic projections between the VTA and the NAc and it has been suggested that alterations in insulin signaling affect motivational behaviour. DA neurons in the VTA and the NAc express insulin receptors insulin reduces the concentration of DA in the VTA in rodents[109,110]. Consequently, intra-VTA injected insulin reduces the intake of palatable food in mice, once sated[109]. However, our results revealed, that individuals with high HOMA-IR showed increased motivated behaviour towards reward in a sated state. A possible mechanism explaining high motivation for reward in sated participants with high HOMA-IR, might be that peripheral insulin resistance is accompanied by central insulin resistance reducing the inhibitory effect of basal insulin levels on the dopaminergic tone in the midbrain. The ability of insulin to depress DA concentrations

in the VTA is believed to result from an increased uptake of DA via dopamine reuptake transporter[109] and it has recently been shown, that a high HOMA-IR, indicating peripheral insulin resistance, is associated with an insufficient reuptake of DA in humans[111]. Regarding acquisition of food reward, high motivation for reward in the sated state due to insulin resistance could further promote overeating and the development of obesity.

Additionally, our findings underscore the findings from recent human data suggesting that peripheral insulin sensitivity is a better predictor of altered DA signaling in humans than the BMI. Eckstand et al. (2017) could demonstrate that the HOMA-IR was a better predictor of the performance in a stop signal task than the BMI in obese humans with type II diabetes mellitus (T2DM)[112]. Regarding reward related motivated behaviour, a study conducted by Edwin et al. (2019) found, that intranasally administrated insulin affected functional connectivity of the dopaminergic midbrain depending on the insulin sensitivity assessed by the HOMA-IR and not depending on the BMI in humans[85]. Insulin sensitivity might not only better predict altered DA signaling but also maladaptive behaviours promoting obesity[113,114].

Overall, detailed neuronal mechanisms how insulin sensitivity influences DA signaling in the midbrain and thus modulates reward-related motivated behaviour in humans require further investigation.

3.4 GLP-1 normalizes the motivational effect of hunger in insulin resistant humans

Assessing motivational behaviour in subjects treated with GLP-1, we found no difference between subjects with good and poor insulin sensitivity. GLP-1R have been found to be expressed in the CNS, notably the NAc and the VTA. Experiments carried out with rodents found, that the activation of GLP-1R in the VTA was able to reduce reward-related motivated behaviour. In animal studies, a decreased motivation to obtain food was shown in mice that were treated with the direct GLP-1 agonist exendin-4. In a conditioned place preference test these mice avoided the place that was associated with food reward in form of chocolate pellets[115]. In the same study, the exendin-4 treated mice showed a decrease in motivation to obtain sucrose in a

progressive ratio (PR) operant-conditioning task[115]. Results from a similar experiment performed by Hsu T. et al (2015), support the findings, that activation of GLP-1 R results in a decrease of motivation to obtain sucrose pellets[116]. However, these investigations have only analysed the direct, central effect of GLP-1 on motivational behaviour for food reward and not the peripheral action. Furthermore, the hunger state or fasting duration of these animals was not considered so that these animal results can only be compared to our human results with certain restrictions.

Considering, how peripherally applied GLP-1 analogues affect the brain, in one study conducted by Hernandez et al. (2018) peripherally administered GLP-1R agonist Ex4 was able to pass the blood-brain-barrier and bind to receptors localized in the VTA in rats[117]. Even though Ex4 can cross the blood-brain barrier and act in the VTA, to date we lack evidence confirming that peripherally injected liraglutide also binds to receptors in the VTA or the NAc.

Peripherally injected liraglutide might reach the CNS by binding to vagal afferents in the NTS. In the NTS the GLP-1R agonist possibly binds on glutamatergic and GABAergic neurons that express GLP-1R[118]. These neurons act on regulating GLP-1 producing neurons within the NTS that project to the VTA and the NAc[119] resulting in a decrease of the activity of DA neurons[120]. In rodents with normal weight, the activation of GLP-1R located in the NTS was shown to reduce food and drug related incentive motivation[121]. Our study further analysed the effect of GLP-1R activation in insulin resistant individuals and suggests, that GLP-1 restores motivational behaviour in insulin resistant humans to the behaviour of insulin sensitive humans[101]. The mechanism by which GLP-1 may affect motivational behaviour in humans may be due to GLP-1 R activation in the NTS by acting on DA neurons in the dopaminergic midbrain, that are regulated by hunger and insulin sensitivity[101].

In 2009 the European Medicines Agency (EMA) approved long acting GLP-1 agonists such as exenatide and liraglutide for the treatment of type II diabetes mellitus. More recent, the U.S Food and Drug Administration (FDA) and EMA approved the treatment of obesity with liraglutide 3.0mg (Saxenda) as well as the treatment of overweight adults (BMI over 27) with comorbidities in 2015. Our work reveals one possible

mechanism contributing to the success of GLP-1 analogues as anti-obesity drugs apart from their central homeostatic effects.

Overall, we reveal how motivated behaviour is metabolically regulated by insulin sensitivity, GLP-1 and hunger in humans and that GLP-1 analogue application normalizes motivation in obese, insulin resistant humans. To further prevent the worldwide increase in the prevalence of obesity, it is of paramount importance to expand investigations on how neural and homeostatic pathways interact in the regulation of our behaviour to ameliorate treatment options in the field of obesity.

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5 Appendix

5.1 Supplementary material

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