

**UNIVERSIDADE FEDERAL DE CIÊNCIAS DA SAÚDE DE
PORTO ALEGRE – UFCSPA
CURSO DE PÓS-GRADUAÇÃO EM CIÊNCIAS DA SAÚDE**

Vanessa Feistauer

**Variantes em Genes Relacionados
aos Sistemas Dopaminérgico e
Serotoninérgico Versus Padrão De
Ingestão Alimentar e Parâmetros De
Adiposidade em Crianças.**

**Universidade Federal de Ciências da Saúde
de Porto Alegre**

**Porto Alegre
2015**

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Dissertação submetida ao Programa
de Pós-Graduação em Ciências da
Saúde da Fundação Universidade
Federal de Ciências da Saúde de
Porto Alegre como requisito para a
obtenção do grau de Mestre.

Orientadora: Dra.Silvana de Almeida
Co-orientadora: Dra.Vanessa Suñé Mattevi

**Porto Alegre
2015**

Catálogo na Publicação

Feistauer, Vanessa

Variantes em genes relacionados aos sistemas dopaminérgico e serotoninérgico versus padrão de ingestão alimentar e parâmetros de adiposidade em crianças /

Vanessa Feistauer. -- 2015.

109 f. : tab. ; 30 cm.

Dissertação (mestrado) -- Universidade Federal de Ciências da Saúde de Porto Alegre, Programa de Pós-Graduação em Ciências da Saúde, 2015.

Orientador(a): Silvana de Almeida ; coorientador(a): Vanessa Suñé Mattevi.

1. Obesidade. 2. Crianças. 3. Dopamina. 4. Serotonina. 5. Polimorfismo. I. Título.

AGRADECIMENTOS

À minha orientadora, Silvana de Almeida, por todo o carinho, incentivo e tempo a mim dedicados. Obrigada pelo aprendizado e oportunidades que me proporcionou, mas principalmente por estar sempre presente, tanto nas férias quanto na gravidez!

À minha co-orientadora, Vanessa Mattevi, por todo o conhecimento proporcionado.

À Márcia Regina Vítolo e ao Grupo de Pesquisa em Nutrição (NUPEN), pela dedicação e incrível trabalho com a amostra do estudo.

Aos meus pais, Luís e Susana, por todo o amor que recebi ao longo da minha vida. Pelo incentivo constante e por nunca me permitirem desistir. Amo vocês para sempre!

Ao meu irmão, Lucas, que me incentivou, distraiu e me permitiu passar por mais essa fase com muita alegria. Te amo muito!

À Grasi e Magda, pelos conselhos técnicos e ajuda no laboratório. Vocês são essenciais para o funcionamento do laboratório, além de torna-lo um ambiente agradável e muito receptivo.

A todos os meus familiares e amigos, pelos incentivos e momentos de descontração.

Às minhas colegas de laboratório que dividiram comigo seus conhecimentos e tornaram os dias no laboratório muito mais alegres.

Ao Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) pela bolsa de mestrado concedida em parceria com a Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA).

À Fundação de Amparo a Pesquisa do Estado do Rio Grande do Sul (FAPERGS) e ao CNPq pelo financiamento deste projeto.

RESUMO

A obesidade infantil é considerada um problema de saúde pública, principalmente em países desenvolvidos e em desenvolvimento. O aumento do consumo de alimentos ricos em gordura e açúcares, associado à redução da atividade física, causam um desbalanço energético que leva ao acúmulo excessivo de gordura e, conseqüentemente, à obesidade. Esses alimentos ricos em gordura e carboidratos são altamente palatáveis, e estimulam uma fome hedônica, ou seja, uma fome relacionada ao prazer de comer, e não relacionada às necessidades calóricas. Os sistemas dopaminérgico e serotoninérgico estão diretamente relacionados ao prazer, apetite e motivação para comer. Entretanto, os indivíduos podem apresentar diferentes níveis de sensação de recompensa, através da ingestão alimentar, o que pode conferir diferenças no padrão da ingesta. Portanto, a variabilidade genética dos sistemas dopaminérgico e serotoninérgico, pode estar relacionada às diferentes respostas frente aos alimentos entre indivíduos expostos aos mesmos fatores ambientais. O objetivo do estudo é investigar a associação de variantes nos sistemas dopaminérgico e serotoninérgico com parâmetros de adiposidade e padrão de ingestão alimentar em crianças. O presente estudo é uma coorte prospectiva de 427 crianças que participaram de um estudo randomizado iniciado ao nascimento, na cidade de Porto Alegre. O estado nutricional das crianças, mensurado por dados antropométricos e dietéticos, foi coletado aos 12 meses e aos 2-3 anos de idade. O DNA (Ácido Desoxirribonucleico) extraído de células da mucosa bucal foi analisado para os seguintes polimorfismos através de PCR (Reação em Cadeia da Polimerase) em tempo real, utilizando sondas de hidrólise (Sistema TaqMan®): rs4680 (*COMT*); rs1800497 (*DRD2*); rs1048953 (*SLC6A3*); rs2071877, rs2312054 e rs12391221 (*SLC6A14*); e rs3813928 (*HTR2C*). O polimorfismo 5-HTTLPR (*SLC6A4*) foi analisado por PCR convencional seguido de eletroforese em gel de agarose. O genótipo A1/A1 do gene *DRD2/ANKK1* foi associado com maior espessura de dobras cutâneas subescapular em Z-score aos 12 meses ($p=0,022$) e em 2-3 anos ($p=0,020$) e maior IMC Z-score em 2-3 anos ($p=0,009$), quando comparados aos portadores do alelo A2. Aos 12 meses, as meninas homozigotas G/G do polimorfismo rs3813928 (*5-HTR2C*) apresentaram maior ingesta diária média de energia ($p=0,007$), de carboidratos ($p=0,019$), e de

lipídios ($p=0,005$); e maior espessura da dobra cutânea tricipital em Z-score ($p=0,034$) do que portadores do alelo A. Portanto, pressupõe-se que crianças homozigotas para o alelo A1 (*DRD2/ANKK1*) e meninas homozigotas G/G (*5-HTR2C*), apresentam menor densidade de receptores de dopamina e de serotonina, respectivamente, o que estimula a ingestão de alimentos palatáveis para prolongar a sensação de recompensa. Os demais polimorfismos não foram associados com padrões de ingestão alimentar e parâmetros de adiposidade na amostra estudada. No entanto, o acompanhamento desse estudo continua, e pode elucidar a contribuição genética para a obesidade ao longo dos anos.

Palavras-chave: Obesidade. Crianças. Dopamina. Serotonina. Polimorfismo.

ABSTRACT

Childhood obesity is considered a public health problem, especially in developed and developing countries. The increased consumption of high fat and high sugar foods, combined with reduced physical activity, cause an energy imbalance that leads to excessive fat accumulation and therefore leading to obesity. These high fat and high sugar foods are highly palatable and stimulate an hedonic hunger, that is, a hunger related to the pleasure of eating, and not related to caloric needs. The dopaminergic and serotonergic systems are directly related to pleasure, appetite and motivation to eat. However, individuals may have different levels of reward, which can grant different patterns of food intake. Therefore, the genetic variability of the dopaminergic and serotonergic systems may be related to different responses to food among individuals exposed to the same environmental factors. The objective of the study is to investigate the association of variants in dopaminergic and serotonergic systems with adiposity parameters and patterns of food intake in children. This study is a prospective cohort of 427 children who participated in a randomized study initiated at birth, in the city of Porto Alegre. The nutritional status of children, measured by anthropometric and dietary data was collected at 12 months and at 2-3 years of age. DNA (deoxyribonucleic acid) extracted from oral mucosa cells was analyzed for the following polymorphisms by PCR (Polymerase Chain Reaction) in real time using hydrolysis probes (TaqMan© System): rs4680 (*COMT*); rs1800497 (*DRD2*); rs1048953 (*SLC6A3*); rs2071877, rs2312054 and rs12391221 (*SLC6A14*); and rs3813928 (*HTR2C*). The 5-HTTLPR polymorphism (*SLC6A4*) was analyzed by conventional PCR followed by agarose gel electrophoresis. The A1/A1 genotype of *DRD2/ANKK1* gene was associated with increased subscapular skinfold thickness Z-score at 12 months ($p=0.022$) and 2-3 years ($p=0.020$) and BMI Z-score at 2-3 years ($p=0.009$) when compared to carriers of the allele A2. At 12 months, homozygous G/G girls of the rs3813928 polymorphism (*5-HTR2C*) presented higher average daily energy ($p=0.007$), carbohydrates ($p=0.019$) and lipid ($p=.005$) intake; and higher tricipital skinfold thickness Z-score ($p=0.034$) than carriers of the A allele. Therefore, it is presuppose that children homozygous for the A1 allele (*DRD2/ANKK1*) and girls homozygous G/G (*5-HTR2C*) have lower density of dopamine and serotonin receptors, respectively, which stimulates the intake of

palatable food to prolong the feeling of reward. Other polymorphisms were not associated with food intake patterns and adiposity parameters in the sample. However, the follow-up of this study continues, and may elucidate the genetic contribution to obesity over the years.

Keywords: Obesity. Children. Dopamine. Serotonin. Polymorphism.

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1 INTRODUÇÃO

1.1 OBESIDADE INFANTIL

A obesidade é definida pela Organização Mundial da Saúde (OMS), como o acúmulo excessivo ou anormal de gordura que confere riscos à saúde. A prevalência global de sobrepeso (incluindo obesidade) em pré-escolares (<5 anos) aumentou de 4,2% em 1990 para 6,7% em 2010 (1). Estima-se que até 2020 essa prevalência atinja a marca de 9,1%, ou aproximadamente 60 milhões de crianças (1). Países desenvolvidos e em desenvolvimento tiveram um padrão similar no aumento da prevalência de obesidade em pré-escolares (1). Apesar da prevalência de crianças obesas em países desenvolvidos (11,7%) ser o dobro do que em países em desenvolvimento (6,1%), é nesse último em que o aumento relativo nos últimos anos foi maior (65% nos países em desenvolvimento, contra 48% nos países desenvolvidos) (1). Os países com maior índice de obesos, de 1980 a 2008 são China e Estados Unidos, seguidos por Brasil e México (2). A prevalência estimada de obesidade infantil no Brasil é de 21,1% em meninas e 23,1% em meninos (2). As principais consequências da obesidade são doenças cardiovasculares, resistência à insulina e síndrome metabólica, diabetes mellitus tipo dois, marcadores de inflamação elevados, distúrbios musculoesqueléticos, além de distúrbios psicológicos associados (3). Crianças obesas são mais prováveis de se tornarem adultos obesos e possuem maior risco de desenvolvimento de doenças coronárias e demais doenças associadas, que diminuem a expectativa de vida (4-6).

Os fatores determinantes da obesidade infantil são, principalmente: nutrição inadequada, com elevado consumo calórico; atividade física reduzida; urbanização e residência em cidades metropolitanas; fatores socioculturais, como a falta de conhecimento dos pais sobre nutrição; idade e sexo; programas de refeições escolares; e causas endócrinas e/ou genéticas (3, 7, 8).

1.1.1 Obesidade x Ingestão alimentar

A disponibilidade de alimentos altamente palatáveis e calóricos somados a baixos níveis de atividade física causam um desbalanço energético, diretamente associado à epidemia de obesidade (9, 10). Esses ambientes obesogênicos acabam

influenciando na escolha de alimentos pouco nutritivos e encorajando o consumo além do necessário e, assim, tornam a população cada vez mais obesa (9-11). O *neuromarketing* em crianças é muito rentável para as empresas e, considerando os níveis de exposição de crianças a aparelhos eletrônicos além das técnicas persuasivas utilizadas, levam as crianças a um consumo exagerado e desnecessário de alimentos com alto teor de açúcar e gordura (10). Insinuações que evocam memórias e imagens de alimentos ao longo do dia estimulam uma fome hedônica mesmo quando já há a sensação de saciedade (10).

Após uma dieta composta apenas de leite materno, crianças, como onívoras, precisam ingerir uma variedade de alimentos. Essa transição de dieta deve ocorrer ao fim do primeiro ano de vida, entretanto, os novos alimentos nem sempre são prontamente aceitos, uma vez que na fase pré-escolar, a criança prefere geralmente alimentos mais palatáveis, com alta concentração de carboidratos, gorduras, açúcares e sal (12). Ao proporcionar à criança algumas comidas e sabores e não outros, a mesma adquire preferências, principalmente devido a três fatores: a percepção sensorial proporcionada pelo alimento, a consequência pós-ingestão e o contexto social (12, 13). Essas preferências podem ser saudáveis ou não, o que pode levar à obesidade. Algumas consequências fisiológicas podem influenciar também na dieta. Se ao provar um alimento novo, seguem-se sensações positivas, como saciedade, uma preferência pelo mesmo se forma (14). As preferências alimentares são, por isso, muito particulares, e podem ser diferentes até mesmo entre membros da mesma família (15). Em um determinado ambiente, dois indivíduos podem apresentar comportamentos e estados psicológicos diferentes frente a estímulos como, por exemplo, o alimento (9).

Estudos com animais mostram que a estimulação de áreas específicas do cérebro (relacionadas à dopamina) pode induzir uma ingestão compulsiva, bem como a sensação de recompensa (16, 17). Outro estudo em que ratos possuíam livre acesso a uma dieta constituída de alimentos palatáveis com elevado índice de gordura levou a um comportamento semelhante ao de viciados em drogas de abuso, além de redução na concentração de receptores de dopamina estriatais (18). Essa redução no nível de receptores estimula o consumo compulsivo para igualar os níveis iniciais de dopamina (18). Em humanos, a concentração de receptores de

dopamina D2 em indivíduos obesos e em dependentes de cocaína apresenta um padrão muito semelhante (10, 19). A exposição excessiva a drogas de abuso diminui a concentração desses receptores no cérebro e, com isso, o consumo torna-se mais frequente (10, 19). Da mesma forma, o consumo constante de alimentos ricos em açúcar e gorduras pode causar dependência, semelhante à dependência de drogas de abuso (10, 19).

Ingesta excessiva e aumento de peso em animais também foram observados quando há diminuição nos níveis de serotonina no cérebro e deficiência no transportador de serotonina (5-HTT) (20-23). Agonistas da serotonina ou inibidores da recaptação de serotonina diminuem a ingestão de alimentos, aumentando a perda de peso (20, 24). Estudos em humanos demonstram que alterações na neurotransmissão serotoninérgica estão associadas a desordens na ingesta (20, 25, 26). A inibição da expressão do transportador de serotonina através de metilação da região promotora também foi relacionada com peso e índice de massa corporal elevados em um estudo com gêmeos monozigóticos (20).

A sensação de recompensa, no entanto, é maior em alguns indivíduos do que em outros, por isso há diferença na ingestão alimentar (27). Por conseguinte, a variabilidade genética dos sistemas dopaminérgico e serotoninérgico, pode estar relacionada às diferentes respostas entre indivíduos expostos aos mesmos fatores ambientais.

1.2 SISTEMA DOPAMINÉRGICO

Responsável por diversas funções fisiológicas (como atividade locomotora, cognição, emoção, recompensa, sono e alimentação), a dopamina é a principal catecolamina no cérebro de mamíferos. A síntese de dopamina se inicia com a conversão de tirosina à dopa (etapa limitante da velocidade) e descarboxilação à dopamina. Após sua liberação nos terminais nervosos, esse neurotransmissor é amplamente recaptado por um transportador específico da dopamina (DAT) e metabolizada pelas enzimas monoaminooxidase (MAO) e catecol-O-metiltransferase (COMT). Sua ação ocorre após ligação a seus receptores presentes nas células-alvo, os ativadores (classe D1) ou os inibidores (classe D2) da adenilciclase (28, 29).

Os receptores da classe D1 (subtipos D1 e D5) aumentam o monofosfato cíclico de adenosina (AMPC) intracelular quando ativados, enquanto que os receptores da classe D2 (subtipos D2, D3 e D4) o inibem (28).

1.2.1 Catecol-O-metiltransferase (COMT)

Distribuída em todos os órgãos, a principal função da enzima COMT é degradar catecolaminas, como a epinefrina, norepinefrina e dopamina, essencialmente no córtex pré-frontal (30, 31). O gene *COMT* está localizado no do cromossomo 22q11 e codifica duas proteínas: COMT solúvel, encontrada em tecidos periféricos; e COMT ligada à membrana, expressa predominantemente no sistema nervoso central (32, 33). Variantes genéticas modificam a atividade dessa enzima e, dessa forma, podem alterar alguns processos neurocomportamentais relacionados com a dopamina (34). Uma dessas variantes é o polimorfismo de sequência única (SNP, *Single Nucleotide Polimorphysm*) *Val158Met* (rs4680) no quarto éxon, no qual ocorre uma troca dos aminoácidos valina (Val, alelo G) para metionina (Met, alelo A) na posição 158 da proteína, alterando sua termoestabilidade (35). Indivíduos Val/Val apresentam maior atividade da enzima, se comparados com indivíduos Met/Met, e os heterozigotos apresentam atividade enzimática intermediária (35). Portanto, indivíduos Met/Met possuem níveis elevados de dopamina no córtex pré-frontal, enquanto os portadores de Val apresentam níveis menores desse neurotransmissor. Por esse motivo, o polimorfismo *Val158Met* é candidato nos estudos de desordens associadas à dopamina, como esquizofrenia, doença de Parkinson e até mesmo obesidade (36).

Estudos nutrigenéticos relacionaram o genótipo Val/Val do polimorfismo *Val158Met* com elevado índice de massa corporal (IMC), maior circunferência da cintura (37, 38). Um estudo prévio do presente grupo demonstrou que crianças portadoras do alelo Val ingeriam em média maior quantidade de alimentos ricos em lipídios do que as homozigotas Met/Met (39).

1.2.2 Receptores de dopamina D2 (DRD2)

Os receptores de dopamina pertencem à família de receptores acoplados à proteína-G. Quando a dopamina extracelular liga-se a seus receptores, uma resposta celular é desencadeada, ou seja, pode-se aumentar ou diminuir os níveis de AMPc intracelular. A ação do DRD2 quando ativado pela dopamina é inibitória (40, 41). Esse receptor é um dos mais abundantes do sistema nervoso central, e essencial na coordenação motora, atividade locomotora, motivação ou aversão e recompensa (41, 42).

O gene *DRD2/ANKK1* possui o SNP *TaqIA* (rs1800497), localizado a 10kb do gene *DRD2*, no éxon 8 de um gene adjacente, denominado *ANKK1* (*ankyrin repeat and kinase domain containing 1*) (43). Esse polimorfismo apresenta dois alelos, o alelo A1 (T) e o alelo A2 (C). Estudos indicam que o alelo A1 é responsável pela diminuição de 30 a 40% na densidade de receptores D2 (principalmente nas partes ventrais dos núcleos caudado e putâmen), aumentando a ingesta de alimentos palatáveis (43-47). A ingestão repetitiva de alimentos doces e gordurosos resulta em uma regulação negativa dos receptores pós-sinápticos D2, diminuindo sua sensibilidade (44). Um estudo de imagens através de *PET-scans* (*positron emission tomography*) mostra que indivíduos obesos apresentam baixa concentração de receptores de dopamina D2 estriatais, como um mecanismo de regulação negativa devido a elevados níveis de dopamina, indicando que a redução destes receptores estaria associada a um comportamento de dependência, também observado em usuários de drogas ilícitas (48). Estudos associaram o polimorfismo *TaqIA* com obesidade, IMC elevados e aumento da ingesta, além de desordens psiquiátricas, como a esquizofrenia (49-55).

1.2.3 Transportador de Dopamina (DAT)

O transportador de dopamina é uma proteína localizada na membrana plasmática do neurônio pré-sináptico, e sua função é transportar ativamente a dopamina livre na fenda sináptica para o citoplasma. Pertencente à família de transportadores SLC6 (SLC6A3), dependentes de Na^+ e Cl^- , e regula os níveis de dopamina e influencia a ingestão alimentar (56-59).

O gene *SLC6A3* está localizado no cromossomo 5p15.3 e contém 15 éxons e 14 íntrons (60, 61). Estudos com animais demonstram que ratos obesos apresentam diminuição na densidade de DAT na superfície celular dos neurônios estriatais ou na expressão gênica (62-66). Diversos polimorfismos foram descritos nesse gene, entre eles o rs1048953 localizado no íntron 3, caracterizado por uma troca de bases C>T (60). Em um estudo de nosso laboratório, pode-se observar que crianças (3-4 anos de idade) homozigotas para o alelo T possuíam ingestão média diária de energia maior do que portadores do alelo C (homozigotos e heterozigotos). Além disso, medidas antropométricas elevadas também foram associadas com esse polimorfismo (67).

1.3 SISTEMA SEROTONINÉRGICO

A serotonina, ou 5-hidroxitriptamina (5-HT) é um neurotransmissor pertencente ao grupo das aminas biogênicas, envolvido em vias relacionadas ao humor, comportamento, sono, fadiga, percepção da dor, termoregulação e apetite (68). A 5-HT é sintetizada a partir da descarboxilação do triptofano, um aminoácido aromático essencial obtido principalmente a partir da dieta (68). A síntese de serotonina depende da disponibilidade do triptofano e a atividade da enzima triptofano hidroxilase, que converte o triptofano em 5-hidroxitriptofano (5-HTP) e, em seguida, descarboxilado à 5-HT (68, 69). O triptofano, aminoácido obtido através da dieta, é transportado para o citosol por um carreador de solutos (transportador) pertencente à família 6 (*SLC6A14*) (70). Também pertencente à essa família (*SLC6A4*), o transportador de serotonina (5-HTT), um cotransportador dependente de Na^+/Cl^- , controla os níveis de serotonina na fenda sináptica através da recaptação desse neurotransmissor (69, 70). Estão localizados principalmente na membrana pré-sináptica, e sua disfunção tem sido associada à obesidade, à depressão e a transtornos de ansiedade, e, por esse motivo, são alvos de drogas inibidoras da recaptação da serotonina, como a fluoxetina (29, 69).

1.3.1 Transportador de aminoácidos

O gene *SLC6A14* codifica uma proteína de membrana que transporta aminoácidos neutros e catiônicos através da membrana plasmática (71). O triptofano, aminoácido essencial obtido através da dieta, compete com outros aminoácidos neutros para ser transportado pela barreira hematoencefálica, regulando, dessa forma, a síntese de serotonina (71-73). Encontrado no cromossomo Xq24, o *SLC6A14* possui os polimorfismos: 20649C>T (rs2071877) no íntron 12; 303A/T (rs2312054) no íntron 1; e 501C/A (rs12391221), no íntron 5, entre outros 494 descritos no banco de dados *dbSNP* do *National Center for Biotechnology Information* em 18 de dezembro de 2015.

Estudos com população de origem europeia relacionaram os polimorfismos 22510C/G e 303A/T com obesidade (74-76). Entretanto, ao nosso conhecimento, não há estudos relacionando o polimorfismo 501C/A com índices antropométricos e ingestão alimentar. Dados obtidos por nosso grupo indicam que não há associação entre os genótipos dos polimorfismos rs2312054 e rs12391221 e o consumo de alimentos de crianças de um ano e 3-4 anos de idade. Entretanto, aos 7-8 anos de idade, as medidas de consumo foram maiores nos portadores do alelo A, tanto no polimorfismo rs2312054 ($p=0,035$) quanto no rs12391221 ($p=0,017$) (77).

1.3.2 Transportador de serotonina (5-HTT)

O 5-HTT é um importante regulador da função da serotonina (36). Através da recaptação de 5-HT, esse transportador dependente de sódio regula os níveis extracelulares dessa catecolamina, inativando assim a transmissão serotoninérgica (36, 78).

O gene *SLC6A4* está localizado no cromossomo 17q11.2 e possui um polimorfismo do tipo VNTR na região promotora, denominado *5-HTTLPR*. Esse polimorfismo possui um alelo longo, com uma inserção de 44-pb, e um alelo curto, com uma deleção de 44-pb (79). Indivíduos com a versão curta desse polimorfismo possuem uma transcrição pouco eficiente do gene, o que reduz a recaptação de serotonina, e por isso tem sido relacionado com depressão, ansiedade, diabetes e obesidade (36, 79-81). Uma pesquisa prévia de nosso grupo buscou associação

entre o polimorfismo *5-HTTLPR* e a ingestão alimentar em crianças nas idades de um ano, 3-4 anos e 7-8 anos (82). Os dados indicam que há associação entre o alelo curto e o índice de massa corporal nas 3 idades analisadas ($p=0,031$, $p=0,017$, $p=0,007$, respectivamente) (82). Esse alelo também foi associado com aumento do consumo alimentar aos 3-4 anos ($p=0,033$) e com elevada soma das dobras cutâneas aos 3-4 e 7-8 anos ($p=0,021$ e $p=0,035$, respectivamente) (82).

1.3.3 Receptor de serotonina (5-HTR2C)

O receptor de serotonina (5-HTR2C) é um membro da superfamília de receptores com sete domínios, que sinaliza para o meio intracelular do neurônio pós-sináptico através da proteína G quando estimulado por hormônios, neurotransmissores ou ligantes farmacológicos (83, 84). Localizado principalmente no sistema nervoso central (plexo coriáide, córtex pré-frontal e estruturas límbicas), está relacionado com comportamento alimentar, e sua ausência em camundongos *knockout* leva a um aumento excessivo de peso em comparação com os controles (83, 85).

O gene *5-HTR2C* está localizado no cromossomo Xq24, e apresenta em sua região flanqueadora o polimorfismo -997G>A (rs3813928) (86, 87). Polimorfismos na região promotora (responsável pela regulação da expressão do gene) podem alterá-la, influenciando a densidade desses receptores (88). Alguns estudos buscaram associação entre esse polimorfismo e obesidade (87, 89, 90). Análises do nosso grupo de pesquisa relacionaram esse polimorfismo com consumo alimentar elevado em crianças aos 3-4 anos ($p=0,007$), mas não em um ano e 7-8 anos de idade. Medidas antropométricas observadas aos 7-8 anos foram elevadas: índice de massa corporal ($p=0,007$), soma das dobras cutâneas ($p=0,042$) e circunferência da cintura ($p=0,004$) (77).

2 JUSTIFICATIVA

A prevalência global de sobrepeso e obesidade infantil (<5 anos) aumentou de 4,2% em 1990 para 6,7% em 2010 e estima-se que até 2020 essa prevalência atinja aproximadamente 60 milhões de crianças (9,1%) (1). No Brasil, a prevalência estimada de obesidade infantil é de 21,1% em meninas e 23,1% em meninos, semelhante ao observado em países desenvolvidos (2). O *Obesity Gene Map*, uma revisão detalhada de genética e obesidade, indica que muitos novos estudos sobre a influência de diversas variantes sobre a obesidade em crianças. No entanto, os escassos estudos realizados na população brasileira restringem-se a adultos (91). Os resultados dessa investigação permitirão esclarecer a associação dos componentes genéticos à obesidade e/ou comportamentos alimentares, possibilitando, dessa forma, a fundamentação e elaboração de propostas de intervenção mais eficazes nos primeiros anos de vida. A elucidação das causas genéticas dessa patologia permitirá, no futuro, não somente a identificação a priori dos indivíduos mais susceptíveis ao seu desenvolvimento, mas também o direcionamento de tratamentos específicos para cada indivíduo. Pois, embora a obesidade seja tratada como uma doença única e o manejo dos pacientes seja praticamente o mesmo, os mecanismos fisiológicos que causam ganho de peso nas pessoas podem ser diferentes.

O ponto mais forte do presente estudo foi a possibilidade de comparar os achados de associação de variantes genéticas em uma amostra independente, representativa da mesma população já estudada, oito anos após o início da primeira coorte.

3 OBJETIVOS

3.1 OBJETIVO GERAL

Investigar a associação de variantes em genes relacionados ao sistema dopaminérgico (*COMT* - rs4680; *DRD2/ANKK1* - rs1800497; e *SLC6A3* - rs1048953) e serotoninérgico (*SLC6A4* - *5-HTTLPR*; *SLC6A14* - rs2071877, rs2312054 e rs12391221; e *HTR2C* - rs3813928) com ingestão alimentar e parâmetros de

adiposidade em uma coorte de crianças da cidade de Porto Alegre, acompanhadas do nascimento até os 2-3 anos de idade.

3.2 OBJETIVOS ESPECÍFICOS

- Analisar a frequência dos polimorfismos acima descritos nos genes *COMT*, *DRD2/ANKK1*, *SLC6A3*, *SLC6A4*, *SLC6A14* e *5-HTR2C* em uma coorte de crianças de 2-3 anos de idade acompanhada desde o nascimento.
- Investigar a associação dessas variantes com parâmetros de adiposidade (índice de massa corporal, circunferência da cintura e dobras cutâneas) em crianças.
- Investigar a associação dessas variantes com ingestão alimentar (consumo total de calorias, consumo de lipídeos, açúcares e de alimentos com altamente energéticos).

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MANUSCRITO DO ARTIGO CIENTÍFICO

O artigo intitulado ***Association study of variants in genes related to dopaminergic and serotonergic systems and food intake and nutritional status in children of Southern Brazil.*** está em preparação para submissão para a revista *European Journal of Pediatrics*, ISSN: 0340-6199 (print version), ISSN: 1432-1076 (electronic version), fator de impacto 1.983 (2013).

Association study of variants in genes related to dopaminergic and serotonergic systems with food intake and nutritional status in children of Southern Brazil.

Vanessa Feistauer¹ vfeistauer@gmail.com

Mariana Eick² mari_eick@hotmail.com

Vanessa S Mattevi^{1,3} vmattevi@ufcspa.edu.br

Márcia R Vitolo⁴ vitolo@ufcspa.edu.br

Silvana Almeida^{1,3} salmeida@ufcspa.edu.br

¹Laboratório de Biologia Molecular, Programa de Pós-Graduação em Ciências da Saúde, Universidade Federal de Ciências da Saúde de Porto Alegre, Rua Sarmento Leite, 245/ Anexo 2 – sala 07; CEP 90050-170, Porto Alegre, RS, Brazil, Tel.: +55 51 3303 8763; fax: +55 51 3303 8718

²Curso de Graduação em Biomedicina, Universidade Federal de Ciências da Saúde de Porto Alegre, Rua Sarmento Leite, 245; CEP 90050-170, Porto Alegre, RS, Brazil, Tel.: +55 51 3303 9000; fax: +55 51 3303 9000

³Departamento de Ciências Básicas da Saúde, Universidade Federal de Ciências da Saúde de Porto Alegre, Rua Sarmento Leite, 245 – sala 309; CEP 90050-170, Porto Alegre, RS, Brazil, Tel.: +55 51 3303 8799

⁴Departamento de Nutrição, Universidade Federal de Ciências da Saúde de Porto Alegre, Rua Sarmento Leite, 245/ Anexo 2 – sala 03; CEP 90050-170, Porto Alegre, RS, Brazil, Tel.: +55 51 3303 8798

Corresponding author: S Almeida, Universidade Federal de Ciências da Saúde de Porto Alegre, Rua Sarmento Leite, 245 – sala 309; CEP 90050-170, Porto Alegre, RS, Brazil, Tel.: +55 51 3303 8763; fax: +55 51 3303 8718, E-mail address: salmeida@ufcspa.edu.br

ABSTRACT

Dopaminergic and serotonergic systems have been associated with food intake and associated disorders such as obesity. We aim to investigate associations of variants in genes related to dopaminergic and serotonergic systems with food intake and adiposity parameters in a cohort of 427 children followed from birth until 2-3 years of age. Dietary data and nutritional status were collected at 12 months and 2-3 years old. The polymorphisms rs4680 (*COMT*), rs1800497 (*DRD2/ANKK1*), rs1048953 (*SLC6A3*), rs3813928 (*HTR2C*), rs2071877, rs2312054 and rs12391221 (*SLC6A14*) were analyzed using polymerase chain reaction based techniques. The A1/A1 genotype of *DRD2/ANKK1* gene was associated with higher subscapular skinfold thickness Z-score at 12 months ($p=0.022$) and at 2-3 years ($p=0.020$) and higher BMI Z-score at 2-3 years ($p=0.009$) than A2 allele carriers. At 12 months, girls homozygous G/G of the rs3813928 polymorphism (*5-HTR2C*) had higher average daily energy ($p=0.007$), carbohydrate ($p=0.019$), and lipid ($p=0.005$) intake; and higher tricipital skinfold thickness ($p=0.034$) than A allele carriers. *Conclusion:* The polymorphism rs1800497 (*DRD2/ANKK1*) was associated with adiposity parameters and in girls the polymorphism rs3813928 was associated with food intake and adiposity parameters. The follow-up of this study continues and may elucidate the genetic contribution to obesity throughout the years.

Keywords: obesity, children, dopamine, serotonin, polymorphism.

52 What is Known:

- 53 • Polymorphisms in *COMT*, *DRD2/ANKK1*, *SLC6A14* and *5-HTR2C* genes have
54 been associated with anthropometric measurements and food intake in adults
55 and children.

56

57 What is New:

- 58 • The data of current study suggest that variants in *DRD2* and *5-HTR2C* genes are
59 associated with adiposity parameters and food intake in children at early
60 developmental phase.

61

62• **List of Abbreviations**

63

64 5-HT_{2C} – 5-hydroxytryptamine (serotonin) receptor 2C G protein-coupled

65 5-HTTLPR – Serotonin transporter linked polymorphic region

66 BMI – Body mass index

67 BRATSA I – Ten steps in action randomized trial

68 *COMT* – Catechol-O-methyltransferase gene

69 DNA – Deoxyribonucleic acid

70 *DRD2/ANKK1* – Dopamine receptor D2 and Ankyrin repeat and kinase domain

71 containing 1 genes

72 EDF – Energy dense foods

73 *HTR2C* – 5-hydroxytryptamine (serotonin) receptor 2C G protein-coupled gene

74 LDF – Lipid dense foods

75 MZF – Myeloid zinc finger protein

76 PCR – Polymerase chain reaction

77 SDF – Sugar dense foods

78 *SLC6A14* – Solute carrier family 6 (amino acid transporter) member 14 gene

79 *SLC6A3* – Solute carrier family 6 (neurotransmitter transporter) member 3 gene

80 SP1 – Specificity protein 1

81

INTRODUCTION

The overall prevalence of overweight (including obesity) in preschool children (<5 years) increased from 4.2% in 1990 to 6.7% in 2010 (1). It is estimated that by 2020 the prevalence reaches the mark of 9.1%, or approximately 60 million children (1). Several factors determine childhood obesity, such as inadequate nutrition, few physical activity, socio-cultural factors, and endocrine and/or genetic causes (3, 7, 8). Therefore, the genetic variability may explain the individual predisposition, and could relate them to an early development of obesity.

Animal studies have shown that stimulation of specific brain areas (related to dopamine) may lead to a binge eating, and the feeling of reward (16, 17). In humans, changes in serotonergic neurotransmission have been associated with disorders of food intake (20, 25, 26). These neurotransmitters are related to the feeling of reward after ingestion, however, in some individuals this feeling is greater than in others, which could explain the difference in food intake between individuals exposed to the same environmental factors (27). The identification of candidate genes related to the dopaminergic and serotonergic systems may facilitate the understanding of heterogeneity in obesity development. Few human studies have associated polymorphisms in *COMT* (37, 38), *DRD2/ANKK1* (48, 52, 54, 55), *SLC6A14* (74, 75) and *5-HTR2C* (79, 80) genes with anthropometric measurements, and food intake, indicating a possible relationship of these genotypes with obesity. Nevertheless, to our knowledge, few studies investigate the association of variations in genes of dopaminergic and serotonergic systems with nutritional status and food intake in a cohort of children in early developmental phase. Therefore, we aim to investigate if these association of variants in genes related to dopaminergic (*COMT* - rs4680; *DRD2/ANKK1* - rs1800497, and *SLC6A3* - rs1048953) and serotonergic (*SLC6A4* - 5-HTTLPR; *SLC6A14* - rs2071877, rs2312054, rs12391221; and *HTR2C* - rs3813928)

systems with food intake and nutritional status in a cohort of children in the city of Porto Alegre, followed from birth until 2-3 years of age.

MATERIALS AND METHODS

Subjects

This is a prospective cohort study of 427 children who participate in a randomized trial, initiated at birth. Twenty district health units in the city of Porto Alegre were randomized into two groups: an intervention group and a control group composed of 9 and 11 health units, respectively. Health professionals in the intervention group received a training based on the guidelines of the program "The ten steps to healthy diet for Brazilian children under two years of age" and printed educational material for reference and distribution. The units in the control group received no information on child's complementary feeding (92).

Recruitment of pregnant women in the last trimester gestation started in 2008 by interviewers blinded to the randomization. In the years 2009-2010, the children's first year of life, dietary and anthropometric assessments were measured. When the children were 2-3 years of old, their nutritional status and DNA samples were collected.

During the visit, mothers were invited to participate in this study to investigate the association of genetic variants with food intake and childhood obesity. After clarifying the objectives of the study, they signed an informed consent for the use of their children's DNA, and samples of the children's oral mucosa squamous cells were collected for DNA extraction. The study was approved by the Research Ethics Committee of the Federal University of Health Sciences, Porto Alegre, Brazil, protocol number 817/11, and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

Nutritional status and Dietary Data

The data collection regarding children's anthropometric and dietary variables was accomplished during home visits in two different development phases: at 1 year and 2-3 years of age. The nutritional status was assessed by means of anthropometric measurements (weight, height, waist circumference, tricipital and sub-scapular skinfold thickness). The BMI variation percent of each child was computed by the formula: $[(BMI_a - BMI_b)/BMI_b] \times 100$ (a = BMI final and b = BMI initial). Dietary data was obtained by two 24 hours dietary recall survey. The dietary recalls were collected for each child on two randomly selected days, and the data obtain was entered into the NutWin, version 1.5 (Nutrition Support Program software from the Escola Paulista de Medicina, Federal University of São Paulo), based on United States Department of Agriculture chemical composition tables. Sugar-dense foods (SDF) were considered those consisting of 50% or more of sugar in food composition, and lipid-dense foods (LDF) if they contained more than 30% fat in food composition. The energy-dense foods (EDF) intake consists of the sum of SDF and LDF intake.

Molecular Analysis

Genomic DNA was extracted of buccal mucosa cells by in-house proteinase K digestion. The *SLC6A4* (5-HTTLPR) was genotyped using polymerase chain reaction (PCR). PCR products were genotyped after electrophoresis on 2.5% agarose gels stained with ethidium bromide by comparison with a 100bp DNA ladder. Single nucleotide polymorphisms (SNPs) were analyzed on Applied Biosystems® StepOnePlus Real-Time PCR System using allelic discrimination (TaqMan®) methodology. SNPs evaluated and the assays used are respectively: *COMT* gene,

rs4680 (assay C_25746809_50); *DRD2/ANKK1* gene, rs1800497 (assay C_7486676_10); *SLC6A3* gene, rs1048953 (assay C_8769965_10); *SLC6A14* gene, rs2071877 (assay C_2474484_10), rs2312054 (assay C_15979351_10) and rs12391221 (assay C_25747165_10); and *HTR2C* gene, rs3813928 (assay C_32343284_10).

Statistical Analysis

Statistical analyzes were performed using the Statistical Package for Social Sciences Version 20.0 (SPSS®, Chicago, IL, USA). The chi-square (χ^2) test was used to verify if the distribution of the genotype frequencies observed agreed with those expected under Hardy-Weinberg equilibrium. General Linear Model, using the type III sums of squares, performed the comparisons of food intake and anthropometric parameters among the genotype groups. Intervention group of the randomized trial Ten Steps in Action (BRATSA I), sex, per capita income and ethnicity were included in each model as covariates for food intake. For anthropometric parameters transformed in Z-score, the same covariates were included in models, except sex already considered on Z-score transformation. The comparisons among SDF, LDF and EDF intake and genotypes were performed by Mann-Whitney U or Kruskal-Wallis. Variants in genes located on the X chromosome were analyzed in girls and boys separately.

RESULTS

Four hundred and twenty-seven children composed this prospective cohort association analysis of food intake and nutritional status with genotype at 12 months and 2-3 years. Some samples could not be analyzed in the laboratory, and so there were losses throughout the study. The sample consisted in 218 (51.1 %) boys and 209

(48.9 %) girls, and the average ages at the two evaluations were 12.03 ± 0.78 and 38.18 ± 2.36 months (mean $\pm$ SD), respectively. The main features of the sample are described in Table 1. Minor allele frequencies of the gene variants were: *COMT* rs4680 0.43 (A/Met); *DRD2/ANKK1* rs1800497 0.25 (T); *SLC6A3* rs1048953 0.25 (A); *SLC6A4* 5-HTTLPR 0.5 (S); *SLC6A14* rs2071877 0.29 (T), rs2312054 0.19 (T) and rs12391221 0.28 (C); and *HTR2C* rs3813928 0.10 (A). The genotype frequencies for all polymorphisms studied in this sample were in agreement with those expected under the Hardy-Weinberg equilibrium, except for the *SLC6A14* rs2071877, discarded for the association analysis.

DRD2/ANKK1 gene

When a recessive model was considered and the sample was grouped for the presence of the A2 allele, or A2 carriers, as follows: A1/A1 (n=30) versus A1/A2 + A2/A2 (n=385). The A1/A1 genotype (rs1800497) was associated with higher subscapular skinfold thickness Z-score at 12 months ($p=0.022$) and at 2-3 years ($p=0.020$) and higher BMI Z-score at 2-3 years ($p=0.009$) than A2 allele carriers (Table 2).

5-HTR2C gene

Girls heterozygous for the polymorphism rs3813928 and homozygous A/A were grouped into carriers of the A allele, thus forming a recessive model (Table 3). At 12 months of age, girls homozygous G/G had higher average daily energy ($p=0.007$), carbohydrate ($p=0.019$) and lipid ($p=0.005$) intake, and higher tricipital skinfold thickness ($p=0.034$) than girls A allele carriers. No association with food intake or anthropometric data was detected in boys (data not shown).

No differences were observed regarding other anthropometric measurements or food intake when compared among genotypes of *COMT* rs4680, *SLC6A3* rs1048953, *SLC6A4 5-HTTLPR* and *SLC6A14* rs2312054, rs12391221 (data not shown).

DISCUSSION

Peripheral signals such as hormones, peptides and nutrients (sugars and lipids) regulate eating behavior mainly through the dopamine reward pathway in the hypothalamus and other autonomic and limbic regions (93). This complex signaling system allows food to be consumed when necessary, preventing the organism to suffer from starvation or over-feed (93). However, excessive and repetitive consumption of palatable foods may, in some individuals (both animal and human), inhibit satiety signals, leading to a binge eating beyond caloric need (10, 18, 19, 93). Despite the importance of the dopaminergic and serotonergic systems in energy consumption, few studies have investigated the association of polymorphisms in genes related to those systems with food intake and the nutritional status, especially in children.

The A1 allele of the rs1800497 (*DRD2/ANKK1*) polymorphism have been related with reduced D2 receptor expression in all areas of the striatum (47). This reduced expression may induce a higher food intake in order to extend the sensation of reward (47). In our study, children homozygous for the A1 allele had higher subscapular skinfold thickness Z-score at 12 months and at 2-3 years and higher BMI Z-score at 2-3 years than A2 allele carriers. The same allele of this polymorphism was associated with lower or no effect of the intervention in weight loss ($p=0.002$) in a one-year intervention study in the lifestyle of German children (12 years) (94). Duran-Gonzalez et al. (95) describe the A1 allele as a risk allele for central obesity in Mexican-American adolescents (16 years). Another study which analyzed the loss and weight maintenance in young adults (21-40 years) found association of the A1 allele

with higher BMI and less weight and BMI loss through the study. These individuals had greater difficulty in maintaining the weight loss and tendency to regain weight (52). However, Van Strien et al. (96) did not observe association of the *DRD2/ANKK1* gene with eating disorders such as emotional eating in adolescents. Davis et al (17) described association of the A2/A2 genotype with binge eating disorder in adults.

Serotonin receptor has been studied as a target for anti-obesity drugs, both in animals and in humans (97-102). 5-HTR_{2C} knockout mice have increased food intake and excessive weight gain (83, 85, 103). The G allele (rs3813928) was associated with reduction in 5-HTR_{2C} expression, indicating that this allele might change the binding of transcription factors of the MZF (myeloid zinc finger) and SP1 (specificity protein 1) in the promoter region of the gene (104). In our sample, girls homozygous G/G had higher energy intake and triceps skinfold thickness. Therefore, is possible that lowered expression of 5-HT_{2C} receptors may induce an increased ingestion of foods in order to increase food reward sensation. In opposite, Miranda et al (2015) observed an association of G/G homozygous of rs3813928 polymorphism with lower food intake than A allele carriers at 3-4 years of age (105). However, the same children homozygous G/G had higher BMI Z-score, sum of triceps and subscapular skinfolds and waist circumference at the 7-8 years of age (105).

The other polymorphisms studied by us in the present research did not indicate any association with anthropometric parameters and food intake. The lack of association in this sample does not necessarily preclude its relation to childhood obesity and food intake. In a similar but smaller sample, evaluated by our research group, there are some interesting findings that could not be replicated in the present sample. Fontana et al (2015) (106) observed that children with the T/T genotype of the polymorphism rs1048953 in the *SLC6A3* gene had a higher average daily energy intake at 3 to 4 years and a higher waist-to-height ratio at 7 to 8 years when compared to children with a different genotype. The *COMT* rs4680 polymorphism was also

associated with food intake, as children with the Val allele had a higher intake of LDF foods (39). Miranda *et al* (2015) (105) observed associations of the A allele carriers of both rs2312054 and rs12391221 polymorphisms (*SLC6A14* gene), with higher food intake at 7 to 8 years. When the 5-HTTLPR (*SLC6A4*) polymorphism was analyzed, children with the S/S genotype presented a higher daily average energy intake at 1 year of age, higher BMI Z-score at 1, 3 to 4 and 7 to 8 years of age and higher sum of skinfolds at 3 to 4 and 7 to 8 years of age (82). Epigenetics could be an explanation for the lack of association of some polymorphisms and adiposity parameters and food intake observed in this sample, and already detected in previous samples of our research group. Obesity has a complex inheritance pattern and so, not only the genetic inheritance, but also the environment these children are inserted should be considered. Some variants have little contribution to the obesity's complex phenotype, and so may be overlapped by environmental factors. Despite the similar minor allele frequencies between both samples studied by us, their environments may have different mechanisms that can alter genetic expression. We hypothesize that the associations detected in previous studies of our group could not be replicated in present investigation due to environmental differences. It is possible that the environment of the present sample is more obesogenic and so may favor obesity despite the children's genotype.

Our study has some limitations. First, the sample was derived from another study, which aimed to evaluate the impact of an intervention in primary health care in the nutritional status of preschool children, but association statistical analysis were controlled by this intervention. Second, many losses occurred during the study, despite the care of researchers to maintain contact with the subjects of the study. Third, the children who participated in the study may have not chosen their food on their own. Parents have a great influence on their children's choices, and if the options are not healthy, it is more likely that they become obese, despite their genotype. Finally, the

lack of association of the analyzed polymorphisms with obesity parameters may be due to an overlap of the environmental influence in this population, which was not quantified. Nevertheless, despite these limitations, the relatively large sample and the monitoring of children allows the results obtained to be reliable. In addition, the follow-up of this sample may give further information on the influence of these and other polymorphisms in parameters related to obesity and overweight in children. More studies in different populations are needed to identify the genetic factors that influence in weight gain.

Acknowledgements We are thankful to all the funding agencies for supporting this work. We are also thankful to Clarissa Löhder Salgado for the assistance in laboratory techniques.

Funding This study was funded by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, Brazil; Universal grant number 472353/2011-0), Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS, Brazil; PRONEM grant number 11/2062-0), Programa de Bolsas CAPES, PIBIC/CNPq and Produtividade em Pesquisa CNPq.

Conflicts of Interest The authors declare that they have no conflicts of interest.

Author's contributions MRV, VSM and SA: conceived the project and planned the design of the study. MRV: coordinated the recruitment of the participants and data collection. VF and ME: carried out the research. VF and SA: analyzed data and wrote the manuscript; SA had the primary responsibility for final content. All authors discussed the results and commented on and approved the final manuscript.

324 **Compliance with ethical standards**

325 **Ethical approval** All procedures performed in studies involving human
326 participants were in accordance with the ethical standards of the institutional and/or
327 national research committee and with the 1964 Helsinki declaration and its later
328 amendments or comparable ethical standards.

329

330 **Informed consent** Informed consent was obtained from all individual
331 participants included in the study.

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661 **Table 1** Main characteristics of the studied sample.

Characteristics	Boys	Girls	P
Controls ^a	48.6 (106)	47.4 (99)	0.795 ^c
Ethnicity ^a			
Whites	59 (128)	60.3 (126)	
Blacks	19.4 (42)	15.8 (33)	0.595 ^c
Asian, Pardos and Amerindians	21.6 (47)	23.9 (50)	
Per capita income (R\$) ^b	313.06 ± 255.55	306.66 ± 249.28	0.796 ^d
Height at birth (cm) ^b	49.31 ± 2.43	48.61 ± 2.53	0.004 ^d
Weight at birth (g) ^b	3301.12 ± 500.69	3270.07 ± 535.52	0.540 ^d
Age at 12 months (months) ^b	12.14 ± 0.83	11.91 ± 0.71	0.003 ^d
Child's BMI at 12 months (Z score) ^b	0.985 ± 1.024	1.023 ± 1.066	0.717 ^d
Average energy intake daily at 12 months (kcal) ^b	1077.95 ± 474.22	961.87 ± 440.15	0.013 ^d
Tricipital skinfold thickness at 12 months (Z-score) ^b	0.765 ± 1.260	0.887 ± 1.239	0.353 ^d
Subscapular skinfold thickness at 12 months (Z-score) ^b	0.680 ± 1.359	0.832 ± 1.439	0.284 ^d
Energy dense food intake at 12 months (kcal) ^b	29.06 ± 55.32	32.79 ± 84.49	0.607 ^d

Age at 2-3 years (months) ^b	38.19 ± 2.37	38.16 ± 2.35	0.912 ^d
Child's BMI at 2-3 years (Z-score) ^b	0.910 ± 1.540	0.929 ± 1.258	0.890 ^d
Waist circumference at 2-3 years (cm) ^b	52.04 ± 4.08	52.07 ± 4.64	0.935 ^d
Average energy intake daily at 2-3 years (kcal) ^b	1553.64 ± 449.20	1463.46 ± 426.64	0.037 ^d
Tricipital skinfold thickness at 2-3 years (Z-score) ^b	0.440 ± 1.327	0.663 ± 1.152	0.069 ^d
Subscapular skinfold thickness at 2-3 years (Z-score) ^b	-0.050 ± 1.567	0.452 ± 1.387	0.001 ^d
Average daily energy intake per kilogram (kcal/kg) ^b	99.61 ± 33.80	96.19 ± 30.20	0.281 ^d
Energy dense food intake at 2-3 years (kcal) ^b	170.84 ± 200.20	159.31 ± 170.74	0.531 ^d

662 ^aData presented as % (n). ^bData presented as mean ± standard deviation. ^cPearson's
663 chi-square test with yates continuity correction. ^dStudent's T test

Table 2 Anthropometric data according to rs1800497 polymorphism of *DRD2/ANKK1* gene in children at twelve months and two to three years old.

rs1800497 Genotypes (<i>DRD2/ANKK1</i>)			
Twelve months			
	A1/A1 (n=30)	A1/A2 and A2/A2 (n=385)	p
BMI Z-score ^a	1.200 [0.400 – 2.150]	0.800 [0.000 – 1.600]	0.305 ^c
Tricipital skinfold thickness Z-score ^b	1.084 ± 1.292	0.636 ± 1.270	0.343 ^d
Subscapular skinfold thickness Z-score ^b	1.329 ± 1.216	0.470 ± 1.442	0.022 ^d
Two to three years			
	A1/A1 (n=30)	A1/A2 and A2/A2 (n=385)	p
BMI Z-score ^b	1.565 ± 1.366	0.878 ± 1.226	0.009 ^d
Waist circumference (cm) ^b	53.20 ± 3.96	52.11 ± 4.12	0.302 ^d
Tricipital skinfold thickness Z-score ^b	0.819 ± 1.495	0.482 ± 1.297	0.251 ^d
Subscapular skinfold thickness Z-score ^b	0.630 ± 1.644	0.066 ± 1.509	0.020 ^d

BMI indicates body mass index. ^aMedian [interquartile range]; ^bMean ± standard deviation; ^cMann-Whitney U; ^dGeneral Linear Model (GLM)

Table 3 Food intake and anthropometric data according to rs3813928 polymorphism of 5-*HTR2C* gene in boys and girls at twelve months old.

rs3813928 Genotypes (5- <i>HTR2C</i>)						
Twelve months	Boys			Girls		
	A (n=138)	C (n=56)	p	AA e AG (n=100)	GG (n=88)	p
Average daily energy intake (kcal) ^b	1047.39 ± 391.38	1088.21 ± 471.41	0.871 ^d	829.43 ± 428.80	987.73 ± 422.19	0.007 ^d
Average carbohydrate intake (kcal) ^b	589.78 ± 213.45	631.99 ± 296.01	0.677 ^d	480.90 ± 250.07	574.78 ± 253.77	0.019 ^d
Average protein intake (kcal) ^b	165.49 ± 68.96	164.33 ± 73.15	0.991 ^d	130.84 ± 64.00	149.36 ± 66.15	0.059 ^d
Average lipid intake (kcal) ^b	272.61 ± 114.79	292.38 ± 138.78	0.806 ^d	220.22 ± 140.51	261.29 ± 264.56	0.005 ^d
SDF (kcal) ^a	33.60 [0.00 – 95.08]	39.89 [7.28 – 94.68]	0.979 ^c	35.66 [0.00 – 62.34]	33.40 [1.38 – 71.82]	0.576 ^c
LDF (kcal) ^a	0.00 [0.00 – 0.00]	0.00 [0.00 – 36.05]	0.076 ^c	0.00 [0.00 – 29.26]	0.00 [0.00 – 31.92]	0.779 ^c
EDF (kcal) ^a	0.00 [0.00 – 33.60]	0.00 [0.00 – 35.13]	0.567 ^c	0.00 [0.00 – 40.82]	0.00 [0.00 – 34.58]	0.990 ^c
BMI Z-score ^a	1.000 [0.625 – 1.650]	0.800 [0.100 – 1.700]	0.299 ^c	0.700 [0.000 – 1.500]	1.000 [0.000 – 1.725]	0.353 ^c
Tricipital skinfold	0.628 ± 1.133	0.748 ± 1.241	0.654 ^d	0.773 ± 1.387	0.865 ± 1.196	0.034 ^d

thickness Z-score ^a						
Subscapular skinfold thickness Z-score ^a	0.703 ± 1.433	0.636 ± 1.326	0.746 ^d	1.083 ± 1.521	0.725 ± 1.435	0.697 ^d

BMI indicates body mass index. SDF indicates sugar dense foods. LDF indicates lipid dense foods. EDF indicates energy dense foods. ^aMedian [interquartile range]; ^bMean ± standard deviation; ^cMann-Whitney U; ^dGeneral Linear Model (GLM)

CONCLUSÃO

A obesidade é usualmente tratada como uma patologia única e o manejo dos pacientes acaba sendo muito semelhante. No entanto, fica cada vez mais evidenciado que esta é uma doença complexa e heterogênea, com particularidades inclusive em pessoas dentro da mesma família. Estudos de associação permitem identificar melhor essas individualidades de modo que se possa selecionar, a partir do genótipo do paciente, medidas mais eficientes de tratamento. Além disso, identificar quais fatores genéticos influenciam na obesidade infantil pode ser determinante para o diagnóstico, possibilitando intervenções ainda nos primeiros anos de vida. Variantes em genes diretamente relacionados aos sistemas dopaminérgico e serotoninérgico, de um ponto de vista fisiológico, poderiam explicar as variações interindividuais nos comportamentos alimentares e obesidade. O polimorfismo rs1800497 do gene *DRD2* foi associado com parâmetros de adiposidade e, em meninas, o rs3813928 do gene *HTR2C* foi associado com ingestão alimentar e parâmetros de adiposidade. Os demais polimorfismos não tiveram associações estatisticamente significativas, como pode ser observado nas tabelas do anexo.

No entanto, a falta de associação observada nesse estudo, não elimina a possibilidade dos polimorfismos analisados estarem relacionados com a obesidade quando controlados para alguns fatores ambientais. Os resultados obtidos das análises dos genes *DRD2* e *HTR2C* são promissores, e permitem que novos estudos sejam iniciados em diferentes amostras.

ANEXO

TERMO DE CONSENTIMENTO LIVRE E ESCLARECIDO

Título da Pesquisa: Investigação da associação de variantes genéticas com a ingestão alimentar e obesidade infantil.

I. Por que realizar essa pesquisa e qual o objetivo dela?

O número de crianças que estão acima do peso ou são obesas vem aumentando ao longo dos últimos anos, como consequência da predisposição genética associada ao aumento da ingestão de alimentos altamente calóricos e à falta de atividade física diária. O objetivo deste estudo é verificar se determinadas diferenças genéticas estão associadas com a obesidade infantil.

II. Qual o procedimento que será utilizado?

Uma escovinha descartável (swab) será introduzida na boca de seu filho e com movimentos giratórios delicados será esfregada na parte interna da bochecha para coleta de células. Esta coleta será feita por um indivíduo treinado. O swab será colocado em um tubo, identificado apenas por números e encaminhado ao laboratório para a análise do DNA. Ao final desse trabalho as amostras de DNA serão guardadas para, eventualmente, serem utilizadas em futuras pesquisas sobre esse assunto. A utilização em estudos futuros somente será realizada mediante nova aprovação do Comitê de Ética em Pesquisa. Todos os dados que vinculem a identidade de seu filho (a) com os dados obtidos serão separados em diferentes bancos de dados

III. Quais os riscos em participar?

Os riscos em participar da pesquisa são mínimos. A coleta será feita cuidadosamente e não causará dor, lesão ou machucado na boca de seu filho, podendo apenas causar um pequeno desconforto durante a coleta.

IV. Quais os benefícios para o paciente que participar deste estudo?

Embora esta pesquisa não possa gerar nenhum benefício imediato aos participantes, ela poderá trazer vários benefícios a longo prazo, quando será possível conhecer quais pessoas possuem características genéticas associadas com a obesidade. Por fim, a sua participação ajudará no desenvolvimento de novos conhecimentos, que poderão eventualmente beneficiar você e outras pessoas no futuro.

V. Com quem você pode esclarecer suas dúvidas?

Em caso de qualquer outra dúvida quanto à pesquisa ou sobre os seus direitos, você poderá contatar com as professoras Silvana de Almeida ou Vanessa Suñé Mattevi, Doutoradas em Genética e Biologia Molecular e professoras da Universidade Federal de Ciências da Saúde de Porto Alegre, responsáveis pelo estudo, pelo telefone (51) 3303-8763. Assim como, com o Comitê de Ética em Pesquisa da UFCSPA (51) 3303-8804, Rua Sarmento Leite, 245.

VI. Quais são os seus direitos?

A participação de seu filho no estudo é voluntária. Caso você decida não participar, isto não afetará no tratamento normal que vocês tem direito. Além disso, você tem liberdade para abandonar a pesquisa a qualquer momento sem nenhum prejuízo, devendo apenas contatar a responsável pelo estudo (citada logo acima). Os seus registros médicos serão sempre tratados confidencialmente. Os resultados deste estudo poderão ser usados para fins científicos, mas você não será identificado pelo nome.

Assinatura do Paciente

Assinatura do Pesquisador

Nome do Paciente

Nome do Pesquisador

**FORMULÁRIO DE CONSENTIMENTO PARA PACIENTES DE ACORDO EM
PARTICIPAR DE UM ESTUDO EM GENÉTICA**

Eu, fui informado dos objetivos da pesquisa acima de maneira clara e detalhada. Recebi informação a respeito do procedimento e esclareci minhas dúvidas. Sei que em qualquer momento poderei solicitar novas informações e terei liberdade de retirar meu consentimento de participação na pesquisa. O Dr. certificou-me de que todos os dados desta pesquisa serão confidenciais, bem como o meu tratamento não será modificado em razão desta pesquisa.

Caso tiver novas perguntas sobre este estudo, posso chamar Silvana de Almeida ou Vanessa Suñé Mattevi no telefone (51) 3303-8763. Para qualquer pergunta sobre os meus direitos como participante deste estudo ou se penso que fui prejudicado pela minha participação, posso chamar Silvana de Almeida ou Vanessa Suñé Mattevi no telefone (51) 3303-8763 ou o Comitê de Ética em Pesquisa da UFCSPA (Rua Sarmiento Leite, 245, telefone (51) 3303-8804).

Declaro que recebi cópia do presente Termo de Consentimento.

Assinatura do Paciente

Assinatura do Pesquisador

Assinatura da
testemunha

Nome do Paciente

Nome do Pesquisador

Nome da testemunha

Porto Alegre, ____/____/____

QUESTIONÁRIO APLICADO NAS MÃES AOS 2-3 ANOS DE IDADE DA CRIANÇA

FICHA DA CRIANÇA

Entrevistador _____

1. Data ____/____/____	Data3: ____/____/____
------------------------	-----------------------

Identificação (criança):

2.Telefones para contato _____

3.Numero de identificação _____	Ident3: ____
4.Nome da criança _____	
5.Nome da mãe _____	
6.Endereço: _____ _____ _____ _____	
7.Data de Nascimento: ____/____/____	

Dados Maternos e Socioeconômicos:

8.Qual a sua idade? _____anos	IdMae3:_____
09.Data de nascimento da mãe ____/____/____	DNm3_____
10.Qual o seu estado civil? Casada/ou mora junto (1) Viúva (2) Solteira (3) Separada (4)	EstCivil3 _____
11.Você teve outros filhos? (1) Sim (2) Não (pule para a questão 14)	Filhos3 _____ Quant3:_____
12. Se sim:	DNf1:____/____/____
Quantos:_____	DNf2____/____/____
DN____/____/____	DNf3____/____/____
DN____/____/____-	
DN____/____/____	
13.Quantas pessoas moram na sua casa? _____	Famí3:_____
14.Qual o grau de parentesco? (1) Família nuclear (2) Família não nuclear	Adul3:_____ Parente3 _____

15.Qual a sua ocupação? (1) Desempregada (2) Empregada c/ carteira assinada (3) Empregada s/ carteira assinada (4) Do lar (5) Estudante 16.Qual a ocupação do pai do seu (sua) filho (a)? (1) Desempregado (2) Empregado c/ carteira assinada (3) Empregado s/ carteira assinada (4) Aposentado (5) Estudante 17.Qual a renda total da família? R\$ _____ 18.Qual o gasto familiar mensal com alimentação? R\$ _____ 19.Qual o gasto familiar mensal com transporte? R\$ _____	OcupaMae3: _____ OcupaPai3: _____ RendaT3: _____ GFA: _____ GFT: _____
---	---

20.Você é fumante? (1) Sim (2) Não (pule para a 22) (3) Parou de fumar (pule para a 22)	Vcfum3: _____
---	----------------------

21.Quantos cigarros você fuma por dia? _____	Ncd3: _____
---	--------------------

22.Alguém que mora na sua casa é fumante?	Ncd3: _____
--	--------------------

Sim (1) Não (2) (Pule para a pergunta 24)	
---	--

Se sim:

23. Quem é fumante na sua casa? Pai (1) Outros moradores da casa (2) Anotar quantos (3) Pai e outros	QuemFuma: _____
---	-----------------

24. Você fumou durante a gestação do seu filho que participou do projeto?

(1) Sim (2) Não (pule para a 26)

Fgest3: _____

Se sim:

25. Quantos cigarros você fumava por dia? _____	Ncfum3 _____
---	--------------

26. Alguém na família tem ou teve? (**referente a criança**)

Para a pergunta quem: coloque 1 quando sim e 2 quando não

26.a Obesidade: (1) Sim (2) Não ou (3) Não Sabe (9) IGN

Se sim: Quem? () Pai () Mãe () Avós () Tios

() Irmãos (88) NSA (99) IGN

Obesi: _____

Obpai: _____

Obmãe: _____

Obavós: _____

Obatio: _____

Obairm: _____

26.b Colesterol Alto: (1) Sim (2) Não ou (3) Não Sabe (9) IGN Se sim: Quem? () Pai () Mãe () Avós () Tios () Irmãos (88) NSA (99) IGN	ColAlto: ____ Colpai: ____ Colmãe: ____ Colavós: ____ Colatio: ____ Colairm: ____
26.c Doença cardiovascular: (1) Sim (2) Não (3) Não Sabe (9) IGN Se sim: Quem? () Pai () Mãe () Avós () Tios () Irmãos (88) NSA (99) IGN	DCV: ____ DCVpai: ____ DCVmãe: ____ DCVavós: ____ DCVtio: ____ DCVirm: ____
26.d Diabetes Melitus: (1) Sim (2) Não ou (3) Não Sabe (9) IGN Se sim: Quem? () Pai () Mãe () Avós () Tios () Irmãos (88) NSA (99) IGN	DM: ____ DMpai: ____ DMmãe: ____ DMavós: ____ DMtio: ____ DMirm: ____

26.e Hipertensão (Pressão Alta):(1) Sim (2) Não (3) Não Sabe (9) IGN Se sim: Quem?() Pai () Mãe () Avós () Tios () Irmãos (88) NSA (99) IGN	PA: _____ PApai: _____ PAmãe: _____ PAavós: _____ PAtio: _____ PAirm: _____
---	---

27. A criança realizou algum exame de sangue após o realizado quando o seu filho estava com 1 ano de idade, através do nosso projeto? Sim (1) Não (2)	Exam3 _____
---	--------------------

Se sim anotar:

Data: __/__/__

Data: __/__/__

Hb: _____g/dl

Hb: _____g/dl

Ht: _____g/dl

Ht: _____g/dl

VCM: _____fl

VCM: _____fl

HCM: _____pg

HCM: _____pg

Sim (1) Não (2) Não sabe (3) 38. Seu (sua) filho (a) teve infecção urinária nos últimos 6 meses? Sim (1) Não (2) Não sabe (3) 39.O seu (sua) filho (a) apresentou algum problema respiratório? Sim (1) Não (2) (pule para a 67)	InfUri3_____ Resp3_____
--	---------------------------------------

Leia as alternativas para o entrevistado

40. Qual ou quais problema (s) que seu (sua) filho (a) apresenta? Tosse () Coriza () Obstrução Nasal () Respiração rápida ou difícil () Para o quadro ao lado preencher 1 para sim e 2 para não	Tosse3_____ Coriza3_____ Obstru3_____ Respd3_____
---	---

Preencher se a criança recebe leite de vaca: (se não recebe pule para a 72)

41. Qual o volume da preparação? _____ml 42. Qual a freqüência que seu filho toma leite no dia? _____ vezes 43. Volume total de leite ingerido no dia: _____ml (descontar se sobra)	Vol3_____ freqleit_____ Volleite_____
---	--

44. A criança vai a creche? Sim (1) Não (2) 45. Período: meio turno (1) dia inteiro (2) 46 Desde que idade (em meses): _____ 47. Se não, no lugar onde ela fica, tem outras crianças junto? Sim (1) Não (2)	Creche3 _____ Períod3 _____ temp3 _____ idcre3 _____ ondfica3 _____
--	--

48. O (a) seu (sua) filho (a) bebe água? Sim (1) Não (2) (Pule para a pergunta 51)	Água3 _____
--	--------------------

Se sim:

49. Quanto bebe? _____ 50. Qual o “tipo” da água? Filtrada / Fervida / Torneira tratada e fervida / Mineral (próprias para o consumo) (1) Torneira não tratada (impróprias para o consumo) (2)	Quant3 _____ Tagua3 _____
---	---

51. Seu (sua) filho (a) comeu/come terra ou objetos não alimentares? Sim (1) Não (2) (pule para a 80) Não sabe (3) (pule para a 53)	Objet3 _____
---	---------------------

Se sim:

52. Quais os objetos não alimentares que seu (sua) filho (a) comeu? (1) Terra (2) Sabão/Sabonete (3) Terra + sabão (5) casca da mandioca (4) Outras substancias _____(qual?)	Quais3_____
--	-------------

Estado Nutricional:	
53. Peso _____ gramas	Peso3_____
54. Comprimento _____ cm	Compri3_____
Dobra cutânea subescapular: _____ mm	Subscap3 _____
Dobra cutânea tricipital: _____ mm	Tricip3 _____

Hemoglobina	
Valor 1:	Hb13_____
Valor 2:	Hb23_____
Avaliação da saúde bucal	
Número de cáries:	Carie3_____
Número de superfícies atingidas:	SAi3_____

Atividades diárias(ontem) :	
55. Que horas foi dormir ontem ____Que horas acordou hoje ____/ horas de sono:_____	Hsonoit_____
56. O que fez ontem pela manha:	

() Creche / Tempo_____	
() Assistiu TV / Tempo:_____	MCrecheT____
() Brincou fora de casa / Tempo:_____	MTvT_____
() Brincou dentro de casa / Tempo:_____	MBrifT_____
() Dormiu / Tempo:_____	MBridT_____
	MDormT_____
57. O que fez ontem de tarde:	
() Creche / Tempo_____	
() Assistiu TV / Tempo:_____	TCrecheT____
() Brincou fora de casa / Tempo:_____	TTvT_____
() Brincou dentro de casa / Tempo:_____	TBrifT_____
() Dormiu / Tempo:_____	TBridT_____
	TDormT_____
58. O que fez ontem de noite:	
() Assistiu TV / Tempo:_____	
() Brincou dentro de casa / Tempo:_____	NTvT_____
() Dormiu / Tempo:_____	NbrinT_____
OBS:Colocar sempre o tempo de HORAS	NdormT_____
59. Tem alguma atividade física regular na semana:	
() Sim ()Não	
Se sim qual:_____	Ativ_____
Frequência na semana:_____	Qual_____

60. Você considera seu filho: 1. muito calmo 2. calmo 3. ativo 4. muito ativo 5. agitado	FreqS_____ ConFi_____
--	--

INQUÉRITO DE FREQUÊNCIA DE ALIMENTOS

Com que frequência o seu filho consome os seguintes alimentos?

Alimentos	Diária (vezes)	Quantidade (total)	Semanal (vezes)	Quantidade (total)	Esporádica	Sabor	Marca
Pão							
Bolo							
Biscoito recheado							
Biscoito sem recheio							
Mucilon/Neston/Farinha Láctea							

Petit Suisse							
Chocolate							
Sorvete							
Açúcar							
Chiclete							
Balas							
Doces							
Gelatina () Diet () Normal							
Salgadinho							
Carne de vaca							
Carne de frango							
Carne de peixe							
Fígado							
Refrigerante () Diet () Normal							
Suco artificial () Diet () Normal							
Suco natural							
Folhas verdes escuras							
Alimentos amarelos							

Frutas							
--------	--	--	--	--	--	--	--

Inquérito recordatório de 24 horas

Horário	Alimentos/e ou Preparação	Medidas caseiras

LEMBRE-SE: ANOTE NO VERSO DESTA FOLHA A COMPOSIÇÃO DOS

PRODUTOS INDUSTRIALIZADOS “ MENOS CONHECIDOS”

Critério ABIPEME (Nova Proposta para o Critério ABA/ABIPEME)

			Nº de ítems possuídos						
Ítem			0	1	2	3	4	5	6 e +
Televisor			0	4	7	11	14	18	22
Rádio			0	2	3	5	6	8	9
Banheiro			0	2	5	7	10	12	15
Automóvel			0	4	9	13	18	22	26

Empregada Mensalista			0	5	11	16	21	26	32
Posse de:								Pontos	
Aspirador de Pó								6	
Máquina de Lavar Roupa								28	
VCR - vídeo cassete								10	
Geladeira								7	
Grau de instrução do chefe da família								Pontos	
Analfabeto ou primário incompleto								0	
Primário completo								5	
Ginásio Completo								10	
Colégial Completo								15	
Superior Completo								21	
Classe A (rica)	89 ou +								
Classe B (média alta)	59 a 88								
Classe C (média média)	35 a 58								
Classe D (média pobre)	20 a 34								

Classe E (pobre)	0 a 19								
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TABELAS RESULTADOS

TABELA 1: Ingestão alimentar e dados antropométricos para o polimorfismo 5-HTTLPR (gene SLC6A4) em crianças aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A4 5-HTTLPR			p
	C/C (n=62)	C/L (n=110)	L/L (n=72)	
Ingestão energética diária média (kcal) ¹	1056,05 ± 403,73	1032,56 ± 472,43	975,45 ± 478,28	0,459 ³
Média de carboidratos (kcal) ¹	608,62 ± 252,24	599,91 ± 279,30	559,16 ± 284,43	0,384 ³
Média de proteínas (kcal) ¹	157,32 ± 58,80	157,28 ± 75,04	153,27 ± 76,18	0,850 ³
Média de lipídios (kcal) ¹	286,91 ± 129,06	275,15 ± 150,47	264,10 ± 149,42	0,620 ³
ADA (kcal) ²	36,84 [2,52 – 80,40]	35,88 [5,96 – 79,29]	29,36 [0,00 – 66,73]	0,512 ⁴
ADG (kcal) ²	0,00 [0,00 – 56,83]	0,00 [0,00 – 49,27]	0,00 [0,00 – 0,00]	0,042 ⁴
ADE (kcal) ²	0,00 [0,00 – 39,20]	0,00 [0,00 – 30,77]	0,00 [0,00 – 34,22]	0,997 ⁴
IMC Z-score ¹	1,044 ± 0,876	1,028 ± 0,973	0,965 ± 1,169	0,766 ³
Dobra cutânea tricipital Z-score ¹	0,735 ± 1,098	0,947 ± 1,179	0,807 ± 1,368	0,493 ³
Dobra cutânea subescapular Z-score ¹	0,775 ± 1,310	0,888 ± 1,326	0,637 ± 1,384	0,315 ³

Dois a três anos		C/C (n=62)	C/S (n=110)	L/L (n=72)	
Ingestão energética diária média (kcal) ¹		1460,24 ± 422,74 ^c	1515,16 ± 445,45	1484,79 ± 422,10	0,893 ³
Média de carboidratos (kcal) ¹		799,88 ± 263,14	838,42 ± 272,32	821,67 ± 234,52	0,677 ³
Média de proteínas (kcal) ¹		222,78 ± 84,53	224,24 ± 73,80	224,54 ± 65,59	0,966 ³
Média de lipídios (kcal) ¹		432,26 ± 145,19	452,79 ± 170,98	431,78 ± 154,70	0,792 ³
ADA (kcal) ²		108,02 [46,28 – 167,79]	92,36 [43,32 – 157,18]	82,14 [43,20 – 160,84]	0,879 ⁴
ADG (kcal) ²		50,82 [0,00 – 173,31]	0,00 [0,00 – 185,32]	17,00 [0,00 – 163,51]	0,923 ⁴
ADE (kcal) ²		99,10 [28,01 – 256,51]	98,02 [0,00 – 271,30]	135,42 [48,42 – 230,26]	0,482 ⁴
Ingestão energética por peso (Kcal/Kg) ¹		93,67 ± 29,14	97,99 ± 31,09	96,15 ± 33,11	0,887 ³
IMC Z-score ¹		0,894 ± 1,131 ^e	0,936 ± 1,296	0,925 ± 1,733	0,821 ³
Circunferência da cintura (cm) ¹		52,08 ± 4,30	51,94 ± 4,11	52,43 ± 5,42	0,619 ³
Dobra cutânea tricipital Z-score ¹		0,423 ± 1,251	0,565 ± 1,181	0,542 ± 1,435	0,622 ³
Dobra cutânea subescapular Z-score ¹		0,216 ± 1,383	0,217 ± 1,436	0,118 ± 1,693	0,773 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. C indica o alelo curto. L indica o alelo longo.

¹Média ± desvio padrão; ²Mediana [intervalo interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 2: Ingestão alimentar e dados antropométricos para o polimorfismo *Va/158Met* (gene *COMT*, rs4680) em crianças aos doze meses e dois a três anos de idade.

Doze meses	Genótipos <i>COMT</i> rs4680			p
	A/A (n=66)	A/G (n=172)	G/G (n=100)	
Ingestão energética diária média (kcal) ¹	1071,77 ± 505,12	1005,92 ± 443,59	1022,58 ± 446,71	0,407 ³
Média de carboidratos (kcal) ¹	627,40 ± 298,13	597,95 ± 270,88	594,97 ± 274,85	0,380 ³
Média de proteínas (kcal) ¹	159,20 ± 75,20	156,98 ± 71,88	151,69 ± 65,92	0,541 ³
Média de lipídios (kcal) ¹	285,02 ± 155,96	264,53 ± 133,34	277,70 ± 142,99	0,291 ³
ADA (kcal) ²	39,88 [9,97 – 74,68]	33,22 [0,68 – 76,31]	38,44 [1,35 – 92,40]	0,832 ⁴
ADG (kcal) ²	0,00 [0,00 – 26,14]	0,00 [0,00 – 28,86]	0,00 [0,00 – 41,99]	0,873 ⁴
ADE (kcal) ²	0,00 [0,00 – 36,02]	0,00 [0,00 – 44,13]	0,00 [0,00 – 34,15]	0,968 ⁴
IMC Z-score ¹	1,051 ± 1,069	1,061 ± 1,118	0,864 ± 0,893	0,272 ³
Dobra cutânea tricipital Z-score ¹	0,859 ± 1,252	0,801 ± 1,353	0,799 ± 1,074	0,825 ³
Dobra cutânea subescapular Z-score ¹	0,833 ± 1,534	0,733 ± 1,467	0,676 ± 1,175	0,732 ³
Dois a três anos				
	A/A (n=66)	A/G (n=172)	G/G (n=100)	
Ingestão energética diária média (kcal) ¹	1481,59 ± 424,34	1546,27 ± 425,59	1490,24 ± 481,03	0,280 ³

Média de carboidratos (kcal) ¹	831,46 ± 257,88	857,22 ± 261,10	814,21 ± 283,11	0,360 ³
Média de proteínas (kcal) ¹	222,84 ± 75,36	231,64 ± 75,35	223,33 ± 89,69	0,203 ³
Média de lipídios (kcal) ¹	430,09 ± 156,62	448,55 ± 161,99	465,42 ± 175,57	0,521 ³
ADA (kcal) ²	98,27 [37,80 – 144,75]	92,12 [40,88 – 171,13]	97,68 [49,88 – 181,47]	0,778 ⁴
ADG (kcal) ²	44,66 [0,00 – 147,29]	21,49 [0,00 – 182,40]	48,68 [0,00 – 187,98]	0,640 ⁴
ADE (kcal) ²	116,87 [15,35 – 238,45]	119,07 [27,46 – 221,34]	118,94 [14,05 – 275,92]	0,836 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	94,66 ± 29,81	100,67 ± 33,02	97,59 ± 32,65	0,280 ³
IMC Z-score ¹	1,010 ± 1,478	0,938 ± 1,511	0,763 ± 1,207	0,343 ³
Circunferência da cintura (cm) ¹	52,32 ± 4,41	52,36 ± 4,71	51,41 ± 3,84	0,107 ³
Dobra cutânea tricipital Z-score ¹	0,594 ± 1,219	0,541 ± 1,290	0,468 ± 1,223	0,472 ³
Dobra cutânea subescapular Z-score ¹	0,152 ± 1,529	0,263 ± 1,517	0,092 ± 1,479	0,268 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo

interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 3: Ingestão alimentar e dados antropométricos para o polimorfismo rs1048953 (gene SLC6A3) em crianças aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A3 rs1048953			p
	A/A (n=22)	A/G (n=134)	G/G (n=195)	
Ingestão energética diária média (kcal) ¹	1080,61 ± 480,71	1016,71 ± 488,43	1017,71 ± 440,22	0,957 ³
Média de carboidratos (kcal) ¹	622,53 ± 289,36	592,61 ± 298,62	591,04 ± 268,30	0,986 ³
Média de proteínas (kcal) ¹	167,28 ± 78,65	154,14 ± 74,35	154,35 ± 67,45	0,853 ³
Média de lipídios (kcal) ¹	277,62 ± 125,63	267,77 ± 144,36	273,53 ± 140,15	0,911 ³
ADA (kcal) ²	33,60 [5,98 – 69,52]	39,00 [3,42 – 76,23]	35,13 [0,00 – 79,76]	0,985 ⁴
ADG (kcal) ²	0,00 [0,00 – 38,39]	0,00 [0,00 – 0,00]	0,00 [0,00 – 36,05]	0,307 ⁴
ADE (kcal) ²	0,00 [0,00 – 52,00]	0,00 [0,00 – 37,70]	0,00 [0,00 – 32,96]	0,533 ⁴
IMC Z-score ¹	0,912 ± 1,007	0,914 ± 1,072	1,077 ± 1,026	0,369 ³
Dobra cutânea tricipital Z-score ¹	0,773 ± 1,519	0,957 ± 1,251	0,738 ± 1,212	0,373 ³
Dobra cutânea subescapular Z-score ¹	0,521 ± 1,587	0,792 ± 1,331	0,755 ± 1,427	0,742 ³
Dois a três anos				
	A/A (n=22)	A/G (n=134)	G/G (n=195)	
Ingestão energética diária média (kcal) ¹	1604,22 ± 370,07	1515,54 ± 430,16	1497,56 ± 455,21	0,795 ³

Média de carboidratos (kcal) ¹	890,12 ± 221,44	835,85 ± 251,66	829,77 ± 279,31	0,930 ³
Média de proteínas (kcal) ¹	245,61 ± 58,74	229,61 ± 88,04	222,22 ± 74,75	0,519 ³
Média de lipídios (kcal) ¹	466,80 ± 153,25	443,79 ± 159,33	446,03 ± 169,00	0,753 ³
ADA (kcal) ²	101,40 [43,13 – 161,81]	85,15 [37,56 – 186,40]	93,35 [47,20 – 152,82]	0,930 ⁴
ADG (kcal) ²	106,50 [0,00 – 181,00]	29,46 [0,00 – 177,98]	35,06 [0,00 – 180,91]	0,283 ⁴
ADE (kcal) ²	118,81 [14,35 – 289,66]	112,12 [22,22 – 236,81]	115,68 [20,72 – 243,19]	0,889 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	97,92 ± 26,89	99,74 ± 33,26	96,90 ± 32,00	0,634 ³
IMC Z-score ¹	1,227 ± 1,892	0,959 ± 1,476	0,848 ± 1,293	0,309 ³
Circunferência da cintura (cm) ¹	52,73 ± 5,52	52,15 ± 4,67	51,90 ± 4,00	0,490 ³
Dobra cutânea tricipital Z-score ¹	0,673 ± 1,426	0,547 ± 1,302	0,529 ± 1,197	0,732 ³
Dobra cutânea subescapular Z-score ¹	-0,005 ± 1,859	0,243 ± 1,478	0,198 ± 1,476	0,375 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo

interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 4: Ingestão alimentar e dados antropométricos para o polimorfismo *TaqIA* (gene *DRD2*, rs1800497) em crianças aos doze meses e dois a três anos de idade.

Doze meses	Genótipos <i>DRD2/ANKK1</i> rs1800497			p
	A1/A1 (n=23)	A1/A2 (n=129)	A2/A2 (n=188)	
Ingestão energética diária média (kcal) ¹	930,52 ± 367,47	1020,27 ± 497,08	1034,36 ± 432,56	0,713 ³
Média de carboidratos (kcal) ¹	521,66 ± 187,94	598,61 ± 312,66	599,79 ± 258,63	0,603 ³
Média de proteínas (kcal) ¹	147,87 ± 69,39	152,62 ± 72,01	158,25 ± 69,56	0,632 ³
Média de lipídios (kcal) ¹	259,63 ± 133,74	264,27 ± 145,28	278,35 ± 137,89	0,708 ³
ADA (kcal) ²	33,40 [9,60 – 63,76]	39,75 [0,00 – 79,71]	36,84 [6,05 – 78,41]	0,797 ⁴
ADG (kcal) ²	0,00 [0,00 – 55,89]	0,00 [0,00 – 37,32]	0,00 [0,00 – 10,30]	0,262 ⁴
ADE (kcal) ²	0,00 [0,00 – 63,12]	0,00 [0,00 – 34,28]	0,00 [0,00 – 36,55]	0,598 ⁴
IMC Z-score ¹	1,160 ± 1,035	0,955 ± 0,978	0,992 ± 1,084	0,539 ³
Dobra cutânea tricipital Z-score ¹	1,084 ± 1,292	0,636 ± 0,978	0,871 ± 1,233	0,139 ³
Dobra cutânea subescapular Z-score ¹	1,329 ± 1,216	0,636 ± 1,270	0,748 ± 1,395	0,065 ³
Dois a três anos				
	A1/A1 (n=23)	A1/A2 (n=129)	A2/A2 (n=188)	
Ingestão energética diária média (kcal) ¹	1457,85 ± 442,33	1539,09 ± 445,25	1502,70 ± 442,30	0,721 ³

Média de carboidratos (kcal) ¹	825,10 ± 286,36	859,11 ± 277,02	823,81 ± 257,77	0,728 ³
Média de proteínas (kcal) ¹	219,69 ± 60,86	226,52 ± 77,15	228,16 ± 84,16	0,878 ³
Média de lipídios (kcal) ¹	417,66 ± 143,75	449,13 ± 158,15	448,31 ± 172,80	0,790 ³
ADA (kcal) ²	70,78 [41,36 – 145,60]	83,15 [38,80 – 159,15]	98,88 [49,33 – 181,88]	0,335 ⁴
ADG (kcal) ²	0,00 [0,00 – 164,43]	22,94 [0,00 – 181,31]	50,00 [0,00 – 178,50]	0,454 ⁴
ADE (kcal) ²	81,77 [36,73 – 263,43]	115,78 [27,39 – 248,25]	118,26 [11,11 – 225,97]	0,764 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	89,86 ± 32,15	99,12 ± 32,27	98,92 ± 32,32	0,518 ³
IMC Z-score ¹	1,565 ± 1,366	0,878 ± 1,226	0,820 ± 1,520	0,029 ³
Circunferência da cintura (cm) ¹	53,20 ± 3,96	52,11 ± 4,12	51,85 ± 4,56	0,504 ³
Dobra cutânea tricipital Z-score ¹	0,819 ± 1,495	0,482 ± 1,297	0,517 ± 1,190	0,505 ³
Dobra cutânea subescapular Z-score ¹	0,630 ± 1,644	0,066 ± 1,509	0,193 ± 1,477	0,039 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo

interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 5: Ingestão alimentar e dados antropométricos para o polimorfismo -995G>A (gene 5-*HTR2C*, rs3813928) em meninas aos doze meses e dois a três anos de idade.

Doze meses	Genótipos -995G>A 5- <i>HTR2C</i> (rs3813928)		
	Portadoras alelo A (n=34)	G/G (n=133)	p
Ingestão energética diária média (kcal) ¹	829,43 ± 428,80	987,73 ± 422,19	0,007 ³
Média de carboidratos (kcal) ¹	480,90 ± 250,07	574,78 ± 253,77	0,019 ³
Média de proteínas (kcal) ¹	130,84 ± 64,00	149,36 ± 66,15	0,059 ³
Média de lipídios (kcal) ¹	220,22 ± 140,51	261,29 ± 136,56	0,005 ³
ADA (kcal) ²	35,66 [0,00 – 62,34]	33,40 [1,38 – 71,82]	0,242 ⁴
ADG (kcal) ²	0,00 [0,00 – 29,26]	0,00 [0,00 – 31,92]	0,654 ⁴
ADE (kcal) ²	0,00 [0,00 – 40,82]	0,00 [0,00 – 34,58]	0,865 ⁴
IMC Z-score ¹	0,880 ± 1,071	1,042 ± 1,076	0,489 ³
Dobra cutânea tricipital Z-score ¹	0,773 ± 1,387	0,865 ± 1,196	0,034 ³
Dobra cutânea subescapular Z-score ¹	1,083 ± 1,521	0,725 ± 1,435	0,697 ³
Dois a três anos	Portadoras alelo A (n=34)	G/G (n=133)	

Ingestão energética diária média (kcal) ¹	1480,79 ± 399,36	1459,25 ± 433,87	0,738 ³
Média de carboidratos (kcal) ¹	799,13 ± 250,25	804,20 ± 263,47	0,544 ³
Média de proteínas (kcal) ¹	215,03 ± 69,17	219,50 ± 81,94	0,798 ³
Média de lipídios (kcal) ¹	453,04 ± 157,79	431,34 ± 156,30	0,121 ³
ADA (kcal) ²	74,46 [30,43 – 157,01]	99,90 [44,46 – 169,78]	0,476 ⁴
ADG (kcal) ²	61,64 [0,00 – 200,08]	28,24 [0,00 – 180,24]	0,928 ⁴
ADE (kcal) ²	70,46 [8,18 – 209,05]	125,10 [20,38 – 248,04]	0,703 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	100,36 ± 29,96	95,20 ± 29,89	0,383 ³
IMC Z-score ¹	0,731 ± 1,063	0,966 ± 1,303	0,415 ³
Circunferência da cintura (cm) ¹	51,94 ± 4,44	52,16 ± 4,77	0,781 ³
Dobra cutânea tricipital Z-score ¹	0,364 ± 1,247	0,733 ± 1,138	0,146 ³
Dobra cutânea subescapular Z-score ¹	0,305 ± 1,439	0,472 ± 1,380	0,839 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 6: Ingestão alimentar e dados antropométricos para o polimorfismo -995G>A (gene 5-*HTR2C*, rs3813928) em meninos aos doze meses e dois a três anos de idade.

Genótipos 5- <i>HTR2C</i> -995G>A (rs3813928)			
Doze meses			p
	A (n=17)	G (n=152)	
Ingestão energética diária média (kcal) ¹	1047,39 ± 391,38	1088,21 ± 471,41	0,871 ³
Média de carboidratos (kcal) ¹	589,78 ± 213,45	631,99 ± 296,01	0,677 ³
Média de proteínas (kcal) ¹	165,48 ± 68,96	164,33 ± 73,15	0,991 ³
Média de lipídios (kcal) ¹	272,61 ± 114,79	292,38 ± 138,78	0,806 ³
ADA (kcal) ²	33,60 [0,00 – 95,08]	39,89 [7,28 – 94,68]	0,979 ⁴
ADG (kcal) ²	0,00 [0,00 – 0,00]	0,00 [0,00 – 36,05]	0,076 ⁴
ADE (kcal) ²	0,00 [0,00 – 33,60]	0,00 [0,00 – 35,13]	0,567 ⁴
IMC Z-score ¹	1,104 ± 0,884	0,960 ± 1,045	0,604 ³
Dobra cutânea tricipital Z-score ¹	0,628 ± 1,133	0,748 ± 1,241	0,654 ³
Dobra cutânea subescapular Z-score ¹	0,703 ± 1,433	0,636 ± 1,326	0,746 ³
Dois a três anos			
	A (n=17)	G (n=152)	
Ingestão energética diária média (kcal) ¹	1574,98 ± 487,08	1561,28 ± 447,75	0,408 ³

Média de carboidratos (kcal) ¹	867,31 ± 298,22	874,53 ± 270,04	0,526 ³
Média de proteínas (kcal) ¹	230,12 ± 67,14	235,08 ± 81,06	0,684 ³
Média de lipídios (kcal) ¹	473,58 ± 174,16	452,65 ± 167,30	0,298 ³
ADA (kcal) ²	135,00 [59,28 – 226,52]	93,35 [47,76 – 149,67]	0,562 ⁴
ADG (kcal) ²	70,62 [0,00 – 177,48]	47,33 [0,00 – 181,19]	0,551 ⁴
ADE (kcal) ²	127,00 [12,34 – 337,51]	117,36 [31,72 – 222,42]	0,574 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	97,66 ± 30,40	100,70 ± 34,30	0,799 ³
IMC Z-score ¹	1,080 ± 1,512	0,848 ± 1,551	0,410 ³
Circunferência da cintura (cm) ¹	51,68 ± 3,68	52,03 ± 4,10	0,877 ³
Dobra cutânea tricipital Z-score ¹	0,403 ± 1,150	0,421 ± 1,334	0,952 ³
Dobra cutânea subescapular Z- score ¹	0,190 ± 1,735	-0,109 ± 1,562	0,435 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 5: Ingestão alimentar e dados antropométricos para o polimorfismo 501C/A (gene SLC6A14, rs12391221) em meninas aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A14 rs12391221			p
	A/A (n=83)	A/C (n=59)	C/C (n=15)	
Ingestão energética diária média (kcal) ¹	958,94 ± 405,38	993,68 ± 528,18	847,36 ± 263,92	0,648 ³
Média de carboidratos (kcal) ¹	559,79 ± 248,41	575,91 ± 313,38	502,11 ± 155,87	0,669 ³
Média de proteínas (kcal) ¹	152,66 ± 70,27	146,28 ± 67,75	120,76 ± 45,15	0,303 ³
Média de lipídios (kcal) ¹	247,85 ± 120,77	271,85 ± 179,36	220,52 ± 92,19	0,678 ³
ADA (kcal) ²	19,53 [0,00 – 59,50]	45,56 [1,86 – 79,28]	43,29 [12,39 – 96,34]	0,060 ⁴
ADG (kcal) ²	0,00 [0,00 – 24,02]	0,00 [0,00 – 26,62]	0,00 [0,00 – 53,52]	0,846 ⁴
ADE (kcal) ²	0,00 [0,00 – 20,70]	0,00 [0,00 – 58,81]	0,00 [0,00 – 22,24]	0,050 ⁴
IMC Z-score ¹	1,050 ± 1,035	0,989 ± 1,100	1,031 ± 1,274	0,980 ³
Dobra cutânea tricipital Z-score ¹	0,812 ± 1,145	0,975 ± 1248	1,097 ± 1,468	0,365 ³
Dobra cutânea subescapular Z-score ¹	0,739 ± 1,412	0,934 ± 1,460	1,104 ± 1,649	0,413 ³
Dois a três anos				
Ingestão energética diária média (kcal) ¹	1447,94 ± 435,53	1503,52 ± 402,83	1420,10 ± 430,39	0,535 ³

Média de carboidratos (kcal) ¹	795,58 ± 258,18	824,85 ± 256,72	792,70 ± 246,41	0,568 ³
Média de proteínas (kcal) ¹	218,09 ± 74,94	220,86 ± 81,70	203,06 ± 96,82	0,898 ³
Média de lipídios (kcal) ¹	419,18 ± 147,56	462,43 ± 169,64	429,26 ± 135,63	0,132 ³
ADA (kcal) ²	77,22 [36,25 – 163,97]	95,80 [38,40 – 170,52]	113,54 [51,72 – 150,24]	0,705 ⁴
ADG (kcal) ²	15,42 [0,00 – 201,54]	104,72 [0,00 – 180,71]	72,70 [0,00 – 135,38]	0,387 ⁴
ADE (kcal) ²	132,50 [25,80 – 266,73]	122,50 [10,45 – 225,38]	86,35 [17,83 – 239,92]	0,682 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	94,43 ± 31,11	100,01 ± 28,24	96,97 ± 35,97	0,214 ³
IMC Z-score ¹	0,985 ± 1,298	0,820 ± 0,946	1,010 ± 2,038	0,683 ³
Circunferência da cintura (cm) ¹	52,67 ± 4,76	51,23 ± 3,91	52,14 ± 7,09	0,121 ³
Dobra cutânea tricipital Z-score ¹	0,873 ± 1,122	0,373 ± 1,167	0,652 ± 1,306	0,027 ³
Dobra cutânea subescapular Z-score ¹	0,632 ± 1,376	0,085 ± 1,350	0,689 ± 1,563	0,050 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo

interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 8: Ingestão alimentar e dados antropométricos para o polimorfismo 501C/A (gene SLC6A14, rs12391221) em meninos aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A14 rs12391221		
	A (n=116)	C (n=43)	p
Ingestão energética diária média (kcal) ¹	1086,50 ± 468,34	1094,87 ± 442,92	0,832 ³
Média de carboidratos (kcal) ¹	633,27 ± 296,92	623,19 ± 258,36	0,714 ³
Média de proteínas (kcal) ¹	166,44 ± 70,46	166,62 ± 79,65	0,648 ³
Média de lipídios (kcal) ¹	282,17 ± 132,74	310,27 ± 146,50	0,099 ³
ADA (kcal) ²	39,88 [7,48 – 94,40]	29,36 [5,98 – 95,52]	0,702 ⁴
ADG (kcal) ²	0,00 [0,00 – 16,39]	0,00 [0,00 – 55,89]	0,027 ⁴
ADE (kcal) ²	0,00 [0,00 – 36,96]	0,00 [0,00 – 25,64]	0,803 ⁴
IMC Z-score ¹	1,050 ± 1,063	0,883 ± 0,978	0,444 ³
Dobra cutânea tricipital Z-score ¹	0,804 ± 1,385	0,742 ± 0,954	0,807 ³
Dobra cutânea subescapular Z-score ¹	0,660 ± 1,441	0,712 ± 1,093	0,853 ³
Dois a três anos			
Ingestão energética diária média (kcal) ¹	A (n=116)	C (n=43)	
	1598,44 ± 443,52	1436,68 ± 422,46	0,107 ³

Média de carboidratos (kcal) ¹	886,66 ± 258,63	795,23 ± 279,00	0,179 ³
Média de proteínas (kcal) ¹	240,61 ± 77,82	229,10 ± 86,15	0,773 ³
Média de lipídios (kcal) ¹	466,54 ± 166,29	433,92 ± 160,77	0,689 ³
ADA (kcal) ²	94,77 [47,77 – 162,51]	84,58 [46,36 – 136,21]	0,382 ⁴
ADG (kcal) ²	31,87 [0,00 – 165,05]	54,24 [0,00 – 201,54]	0,385 ⁴
ADE (kcal) ²	115,78 [36,10 – 251,26]	126,96 [0,00 – 215,57]	0,376 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	102,74 ± 33,68	90,89 ± 30,91	0,045 ³
IMC Z-score ¹	0,824 ± 1,685	1,025 ± 1,305	0,413 ³
Circunferência da cintura (cm) ¹	51,98 ± 4,15	52,32 ± 4,37	0,301 ³
Dobra cutânea tricipital Z-score ¹	0,299 ± 1,365	0,778 ± 1,213	0,039 ³
Dobra cutânea subescapular Z- score ¹	-0,091 ± 1,611	0,048 ± 1,367	0,510 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 9: Ingestão alimentar e dados antropométricos para o polimorfismo 303A/T (gene SLC6A14, rs2312054) em meninas aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A14 rs2312054			p
	A/A (n=105)	A/T (n=46)	T/T (n=7)	
Ingestão energética diária média (kcal) ¹	963,90 ± 410,92	988,22 ± 528,69	797,67 ± 284,19	0,560 ³
Média de carboidratos (kcal) ¹	559,71 ± 244,10	581,65 ± 323,33	445,34 ± 155,98	0,450 ³
Média de proteínas (kcal) ¹	151,54 ± 68,04	143,26 ± 66,85	119,66 ± 56,78	0,342 ³
Média de lipídios (kcal) ¹	253,67 ± 134,43	263,31 ± 168,99	227,22 ± 89,06	0,832 ³
ADA (kcal) ²	23,92 [0,00 – 64,00]	45,37 [5,40 – 75,53]	75,82 [7,38 – 115,28]	0,090 ⁴
ADG (kcal) ²	0,00 [0,00 – 10,60]	0,00 [0,00 – 43,63]	0,00 [0,00 – 82,88]	0,416 ⁴
ADE (kcal) ²	0,00 [0,00 – 22,46]	0,00 [0,00 – 52,97]	0,00 [0,00 – 89,80]	0,400 ⁴
IMC Z-score ¹	1,020 ± 1,065	1,004 ± 1,123	1,025 ± 0,805	0,982 ³
Dobra cutânea tricipital Z-score ¹	0,787 ± 1,176	1,085 ± 1,260	1,172 ± 1,239	0,320 ³
Dobra cutânea subescapular Z-score ¹	0,722 ± 1,460	1,053 ± 1,427	1,376 ± 1,063	0,316 ³
Dois a três anos				
Ingestão energética diária média (kcal) ¹	A/A (n=105)	A/T (n=46)	T/T (n=7)	
	1457,22 ± 418,39	1511,45 ± 442,64	1243,66 ± 211,04	0,159 ³

Média de carboidratos (kcal) ¹	801,40 ± 251,86	828,27 ± 269,50	678,91 ± 197,68	0,237 ³
Média de proteínas (kcal) ¹	219,59 ± 79,99	218,02 ± 80,73	196,20 ± 45,14	0,785 ³
Média de lipídios (kcal) ¹	424,46 ± 145,00	472,09 ± 180,97	369,20 ± 49,78	0,019 ³
ADA (kcal) ²	90,48 [36,89 – 171,33]	96,40 [32,03 – 156,98]	80,09 [47,20 – 152,28]	0,945 ⁴
ADG (kcal) ²	19,84 [0,00 – 203,00]	94,67 [0,00 – 175,12]	19,84 [0,00 – 98,84]	0,485 ⁴
ADE (kcal) ²	126,94 [20,20 – 252,25]	120,82 [11,20 – 268,82]	68,58 [11,79 – 173,62]	0,493 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	96,28 ± 29,88	100,80 ± 32,25	73,04 ± 14,86	0,061 ³
IMC Z-score ¹	0,942 ± 1,236	0,837 ± 1,365	1,204 ± 0,844	0,821 ³
Circunferência da cintura (cm) ¹	52,28 ± 4,67	51,40 ± 4,88	53,36 ± 4,50	0,582 ³
Dobra cutânea tricipital Z-score ¹	0,754 ± 1,164	0,404 ± 1,201	0,974 ± 0,694	0,319 ³
Dobra cutânea subescapular Z-score ¹	0,485 ± 1,362	0,182 ± 1,471	1,240 ± 1,182	0,175 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica

alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo

interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

TABELA 10: Ingestão alimentar e dados antropométricos para o polimorfismo 303A/T (gene SLC6A14, rs2312054) em meninos aos doze meses e dois a três anos de idade.

Doze meses	Genótipos SLC6A14 rs2312054	
	A (n=116)	T (n=43)
Ingestão energética diária média (kcal) ¹	1078,85 ± 420,33	1182,01 ± 598,48
Média de carboidratos (kcal) ¹	631,19 ± 260,49	666,63 ± 371,93
Média de proteínas (kcal) ¹	163,28 ± 64,90	180,78 ± 97,39
Média de lipídios (kcal) ¹	281,78 ± 123,50	343,33 ± 175,19
ADA (kcal) ²	40,36 [9,97 – 102,26]	23,68 [5,98 – 74,68]
ADG (kcal) ²	0,00 [0,00 – 18,03]	0,00 [0,00 – 78,89]
ADE (kcal) ²	0,00 [0,00 – 39,20]	0,00 [0,00 – 22,78]
IMC Z-score ¹	0,924 ± 1,049	1,074 ± 0,945
Dobra cutânea tricipital Z-score ¹	0,742 ± 1,297	0,889 ± 1,003
Dobra cutânea subescapular Z-score ¹	0,583 ± 1,413	0,903 ± 1,061
Dois a três anos		T (n=43)
Ingestão energética diária média (kcal) ¹	1562,54 ± 430,93	1521,78 ± 483,25

0,126³

0,325³

0,119³

0,006³

0,233⁴

0,080⁴

0,275⁴

0,311³

0,443³

0,248³

Média de carboidratos (kcal) ¹	864,87 ± 251,08	856,24 ± 325,52	0,809 ³
Média de proteínas (kcal) ¹	232,97 ± 69,46	234,52 ± 93,55	0,499 ³
Média de lipídios (kcal) ¹	454,78 ± 159,05	449,35 ± 177,37	0,694 ³
ADA (kcal) ²	95,84 [47,74 – 165,35]	86,74 [48,25 – 134,61]	0,650 ⁴
ADG (kcal) ²	50,02 [0,00 – 177,48]	37,19 [0,00 – 200,09]	0,812 ⁴
ADE (kcal) ²	118,26 [32,82 – 268,02]	147,02 [0,00 – 217,62]	0,727 ⁴
Ingestão energética por peso (Kcal/Kg) ¹	100,52 ± 32,42	95,33 ± 36,73	0,513 ³
IMC Z-score ¹	0,833 ± 1,630	1,144 ± 1,364	0,336 ³
Circunferência da cintura (cm) ¹	51,96 ± 4,09	52,62 ± 4,65	0,220 ³
Dobra cutânea tricipital Z-score ¹	0,359 ± 1,381	0,748 ± 1,180	0,098 ³
Dobra cutânea subescapular Z- score ¹	-0,137 ± 1,609	0,164 ± 1,303	0,393 ³

ADA indica alimentos com alta densidade de açúcar. ADG indica alimentos com alta densidade de gordura. ADE indica alimentos com alta densidade energética. IMC indica índice de massa corporal. ¹Média ± desvio padrão; ²Mediana [intervalo interquartil]; ³Modelo Linear Generalizado; ⁴ Kruskal-Wallis

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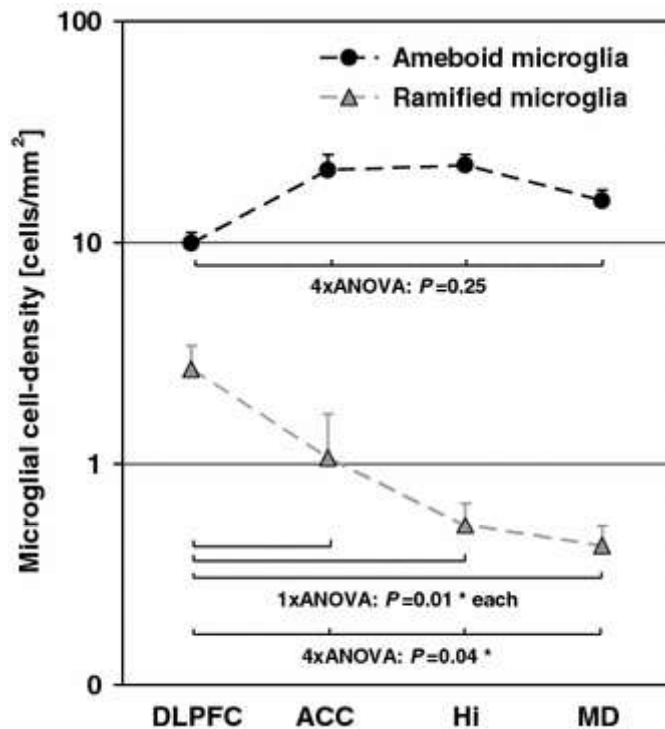
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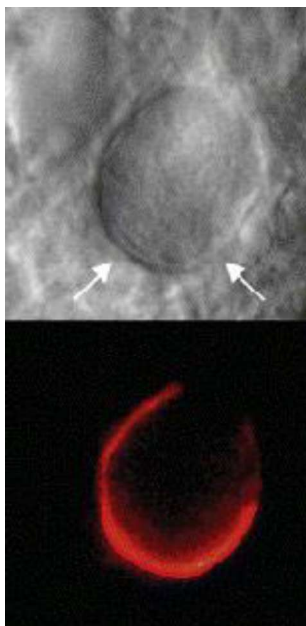
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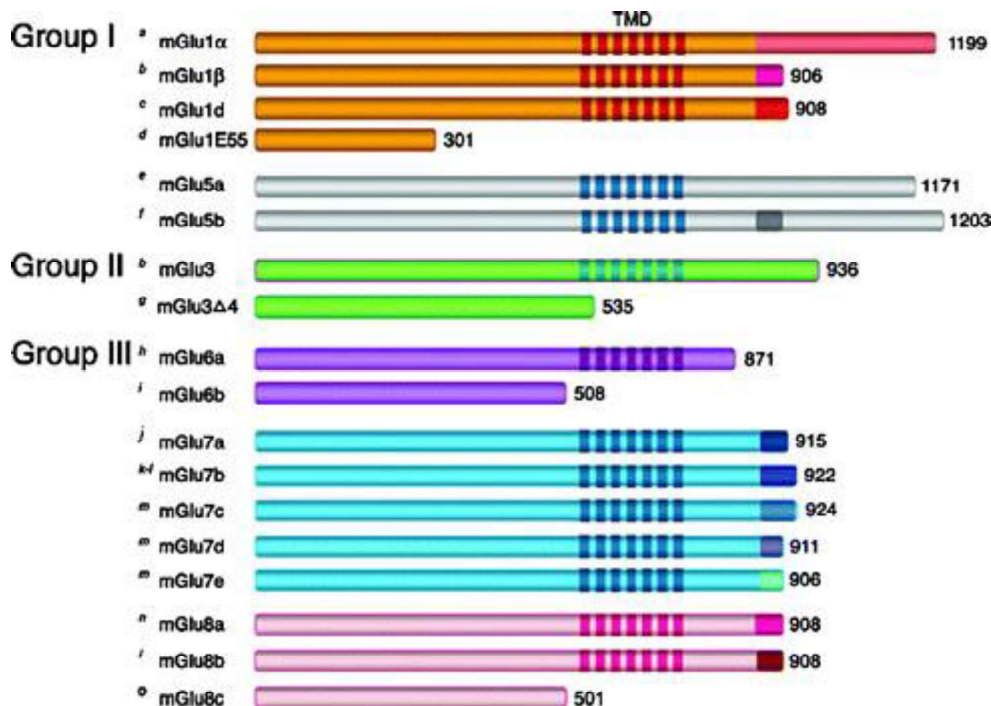
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Parecer Consubstanciado de Projeto de Pesquisa

Título do Projeto: Investigação da associação de variantes genéticas com a ingestão alimentar e obesidade infantil

Pesquisador Responsável Silvana de Almeida

Parc. 1414-11

Data da Versão 22/06/2011

Cadastro 817/11

Data do Parecer 21/07/2011

Grupo e Área Temática III - Projeto fora das áreas temáticas especiais

Objetivos do Projeto

Analisar os efeitos de variantes nos genes LEP, LEPR etc em relação aos parâmetros antropométricos e de ingestão alimentar em 600 pré-escolares do município de Porto Alegre.

Sumário do Projeto

As crianças participarão do projeto Impacto de intervenção na atenção primária à saúde nas condições nutricionais de crianças em idade pré-escolar: segunda fase de avaliação de ensaio de campo randomizado por conglomerados, o qual já recebeu aprovação do CEP. Serão coletadas amostras de células descamativas da mucosa bucal para extração de DNA pelos entrevistadores. As médias dos parâmetros antropométricos e dietéticos serão analisados em relação as genótipos.

Itens Metodológicos e Éticos	Situação
Título	Adequado
Autores	Adequados
Local de Origem na Instituição	Adequado
Projeto elaborado por patrocinador	Não
Aprovação no país de origem	Não necessita
Local de Realização	Própria instituição
Outras instituições envolvidas	Não
Condições para realização	Adequadas

Comentários sobre os itens de Identificação

Introdução	Adequada
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Comentários sobre a Introdução

Objetivos	Adequados
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Comentários sobre os Objetivos

Claros, objetivos e factíveis de serem respondidos.

Pacientes e Métodos	
Delineamento	Adequado
Tamanho de amostra	Total 600 Local escol
Cálculo do tamanho da amostra	Adequado
Participantes pertencentes a grupos especiais	Não
Seleção equitativa dos indivíduos participantes	Adequada
Critérios de inclusão e exclusão	Adequados
Relação risco- benefício	Adequada
Uso de placebo	Não utiliza
Período de suspensão de uso de drogas (wash out)	Não utiliza
Monitoramento da segurança e dados	Adequado
Avaliação dos dados	Adequada - quantitativa
Privacidade e confidencialidade	Adequada
Termo de Consentimento	Adequado
Adequação às Normas e Diretrizes	Sim

Comentários sobre os itens de Pacientes e Métodos

Cronograma	Adequado
Data de início prevista	

Data de término prevista	
Orçamento	Adequado
Fonte de financiamento externa	Não

Comentários sobre o Cronograma e o Orçamento

Referências Bibliográficas	Adequadas
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Comentários sobre as Referências Bibliográficas
atual e pertinente para a pesquisa

Recomendação

Aprovar

Comentários Gerais sobre o Projeto
