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REVIEW

Long noncoding RNA variations in cardiometabolic diseases

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Cardiometabolic diseases are characterized as a combination of multiple risk factors for cardiovascular disease (CVD) and metabolic diseases including diabetes mellitus, dyslipidemia, hypertension and abdominal obesity. This cluster of abnormalities individually and interdependently leads to atherosclerosis and CVD morbidity and mortality. In the past decade, genome-wide association studies (GWASs) have identified a series of cardiometabolic disease-associated variants that can collectively explain a small proportion of the variability. Intriguingly, the susceptibility variants imputed from GWASs usually do not reside in the coding regions, suggesting a crucial role of the noncoding elements of the genome. In recent years, emerging evidence suggests that noncoding RNA (ncRNA) is functional for physiology and pathophysiology of human diseases. These include microRNAs and long noncoding RNAs (lncRNAs) that are now implicated in human diseases. The ncRNAs can interact with each other and with proteins, to interfere gene expressions, leading to the development of many human disorders. Although evidence suggests the functional role of lncRNAs in cardiometabolic traits, the molecular mechanisms of gene regulation underlying cardiometabolic diseases remain to be better defined. Here, we summarize the recent discoveries of lncRNA variations in the context of cardiometabolic diseases.
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INTRODUCTION

Cardiometabolic diseases are the leading cause of death in both developed and developing countries.[1] The cardiometabolic diseases are defined as a constellation of several physiological risk factors for cardiovascular disease (CVD) and metabolic disorders including increased blood glucose levels, elevated triglycerides and apolipoprotein B-containing lipoproteins and low high-density lipoproteins, elevations of blood pressure, obesity, prothrombotic and proinflammatory states.[1–3] The cardiometabolic diseases consequently result in increased risk for coronary artery disease, stroke, peripheral vascular disease, renal insufficiency, prothrombotic and inflammatory abnormalities, making it a heavy burden on economies, particularly in low- and middle-income countries.[4] In addition, the prevalence of cardiometabolic diseases is relatively high and is increasing in parallel with rising obesity rates globally.[2] Apart from the traditional and environmental risk factors for cardiometabolic diseases, including diabetes, dyslipidemia, hypertension and obesity, human genome variance is involved in the disease development.[5] To improve understanding of cardiometabolic disease pathogenesis, large efforts employing genome-wide association studies (GWAS) have identified numerous disease-associated single-nucleotide polymorphisms (SNPs). Most of them are enriched within noncoding elements, and very few of them are found in the coding that allows reasoning by the difference in structural change of the protein. Thus, the molecular function of SNPs needs to be interpreted in other ways.[6,7]

In recent years, the Encyclopedia of DNA Elements (ENCODE) project and the Functional ANnoTation Of the Mammalian genome (FANTOM) consortium have advanced our understanding on the principles of genome organization and function, and have identified a large number of novel transcripts, namely noncoding RNAs (ncRNAs).[8] The ncRNAs were previously thought as transcriptional noises from junk DNA, but now they consist of a class of functional RNA molecules that do not translate into proteins.[9,10] Increasing evidence suggests that a large fraction of ncRNAs have an essential function in the regulation of biological processes and play a critical role in disease development.[10,11] Based on the size of the transcript, ncRNAs can be classified into two classes: (1) short ncRNAs (<200 nt) including microRNAs, PIWI-interacting RNAs and transcription initiation RNAs; and (2) long ncRNAs (lncRNAs) (>200 nt) including long intergenic ncRNAs (lincRNAs), natural antisense transcripts, enhancer-like ncRNAs and transcribed ultraconserved regions.[10,12,13] In contrast to the short ncRNAs that are highly conserved and their function primarily involved in post-transcriptional repression, lncRNAs are not well conserved and their functional mechanisms are diverse.[5,13–15] In this review, we focus in particular on the role of recently discovered lncRNAs and their SNPs with respect to cardiometabolic diseases in humans.

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PHYSIOLOGY AND FUNCTION OF LNCRNAS

Owing to the advent of deeper and more sensitive high-throughput genomic technologies, such as next-generation sequencing and RNA sequencing, the total number of lncRNAs continues to increase.[15,16] According to the latest statistics from GENCODE database version 24 in 2015, there are 19 815 of protein-coding genes and 15 941 lncRNA genes, compared with GENCODE version 3c in 2009, with 22 550 of protein-coding genes and 6496 of lncRNA genes showing a great rise in the number of lncRNAs in less than a decade. This is a reflection of motivated scientists to pay more attention to lncRNAs to understand their roles.[15] However, unlike the well-studied microRNAs, the knowledge on lncRNAs is still at a premature stage.[5,9,14]

The lncRNAs are pervasively abundant and important for cells in every branch of organisms. They can be found in both the nucleus and cytoplasm.[13,17] Similar to mRNAs, lncRNAs are typically transcribed by RNA polymerase II, undergo 5′ capping and are polyadenylated and spliced into different alternative isoforms.[17,18] The categorization of lncRNAs is varied depending on the criteria such as transcript length or association with annotated protein-coding genes or function.[19] The lncRNAs are classified into four groups on the basis of their genomic location relative to nearby encoding regions.[14,17] The statistics retrieved from the GENCODE database version 24 in 2015 shows the following: (1) lincRNAs that are transcribed intergenetically and form the largest subclass of lncRNA molecules in humans, accounting for 7674 lncRNAs; (2) antisense lncRNAs that are transcribed from the opposite strand of a protein-coding gene, accounting for the second most prevalent group with 5564 lncRNAs; (3) sense lncRNAs that are transcribed from the sense strand of a protein-coding gene and can be either sense intronic or sense overlapping, accounting for 1112 lncRNAs; (4) bidirectional or divergent lncRNAs that are transcript pairs that are arranged head to head on the opposite strands and both transcription start sites are spaced <1 kb, and 3 lncRNAs are detected in this category.[9,13,17,20–22] In addition, lncRNAs can be summarized into four main categories based on the mechanism of action, including signals, decoys, guides and scaffolds.[14] Notably, some lncRNAs have more than one mode of action.[14,16]

Signals

The lncRNAs can act as a molecular marker for functional biological conditions in a time- and space-specific manner, responding to intracellular signaling.[14,17] For example, potassium voltage-gated channel, KQT-like subfamily, member 1 opposite strand/antisense transcript 1 (KCNQ1OT1) is essential for parental silencing of multiple imprinted genes in potassium channel, voltage gated kqt-like subfamily q, member 1 (KCNQ1) domain and cyclin-dependent kinase inhibitor 1c (CDKN1C). KCNQ1OT1 is implicated in the development of type 2 diabetes (T2D).[23,24]

Decoys

The lncRNAs can be negative regulators of transcription by interfering or competing with other RNAs or proteins that bind to DNA.[14,17,25] For instance, lncRNA growth arrest-specific transcript 5 (GAS5) is a riborepressor of the glucocorticoid receptor.[26] GAS5 decoys glucocorticoid response element by using an RNA motif that resembles the DNA-binding domain of the glucocorticoid receptor.[26] Thus, GAS5 competes with glucocorticoid response element DNA motifs for binding of the glucocorticoid receptor and prevents transcription of metabolic genes.[26]

Guides

The guide lncRNAs bind to proteins and transport the complex to specific regions of the genome, leading to changes in the gene expression of either neighboring or distantly located genes.[14,25] The guide mechanism may be through direct or indirect interaction to the DNA.[14,17] A prototypical example of guide lncRNA is a functionally well-characterized lncRNA, XIST (X-inactive specific transcript).[13–15,27] XIST is associated with the recruitment of polycomb repressive complex (PRC) that is responsible for the silencing of the X chromosome in female.[14,28]

Scaffolds

The lncRNAs bind to multiple proteins at the same time and bring them in close proximity to form a ribonucleoprotein complex. This may result in transcriptional activation or repression events.[10,14,29] An example for scaffold lncRNA is antisense noncoding RNA in the INK4 locus (ANRIL) that can simultaneously scaffold for the chromatin remodeling complexes PRC1 and PRC2 and initiate the transcriptional silencing of important cell division regulators.[30–32]

EXPRESSION OF LNCRNAS IN CARDIOMETABOLIC PHENOTYPES

Recent evidences demonstrated the presence of lncRNAs in human fluids, including plasma, serum and urine.[33–35] The lncRNAs are expected to be potential diagnostic and prognostic tools for complex multifactorial diseases such as cancer and cardiometabolic diseases.[33–36] Although the utility of lncRNAs as biomarkers in a clinical setting has not yet been established at present, the discovery and detection of circulating lncRNAs holds promise that they may be the new molecule for noninvasive detection and monitoring of diseases in the near future.[35] The following sections describe a representative aberrant expression of lncRNAs in cardiometabolic phenotypes.

Expression of lncRNAs in coronary artery disease (CAD)

Long intergenic noncoding RNA predicting cardiac remodeling (LIPCAR). Dysregulation of a mitochondrial long noncoding RNA uc022bqs.1 (named as LIPCAR) was found in the plasma of patients with or without left ventricular remodeling after myocardial infarction (MI) using global transcriptome analyses.[33] The circulating level of LIPCAR was reduced during early stage after MI and the level increased in later stages.[33] The increased LIPCAR level was found in chronic heart failure patients and was associated with a higher risk of cardiovascular death.[33]

Expression of lncRNAs in diabetes mellitus (DM)

Growth arrest-specific transcript 5. GAS5 is a riborepressor for glucocorticoid receptor and acts as a putative tumor suppressor and apoptosis-promoting gene in various cancers.[34,37,38] A recent study demonstrated that serum GAS5 level is associated with the prevalence of T2D.[34] A marked decrease in serum GAS5 was found in diabetic groups compared with controls. Individuals with serum GAS5 of $<10\ \mathrm{ng\,\mu l^{-1}}$ represented nearly 12 times higher diabetic risk.[34] A role of GAS5 in T2D development remains to be investigated.

Metastasis-associated lung adenocarcinoma transcript 1 (MALAT1). MALAT1 was initially identified as a transcript associated with metastasis of lung cancer.[39] A recent study suggested that MALAT1 is a potential gene regulator in the human islet.[40] MALAT1 is upregulated by exposure to high glucose in human umbilical vein endothelial cells.[41] The increment of MALAT1 was in parallel with

Table 1 Representatives of lncRNAs associated with cardiometabolic diseases

lncRNA	*SNP ID*	*Nearby gene*	*Effect of variant*	*Reference*
Coronary artery disease				
ANRIL	rs1333049, rs10757278, rs2383206	*CDKN2A, CDKN2B, ARF*	Inhibit expression of CDKN2A, CDKN2B	67,68,76,80,81
MIAT	Intron 1 5338 C>T (rs2331291), exon 3 8813 G>A, exon 3 9186 G>A (rs2301523), exon 5 11 093 G>A, exon 5 11 741 G>A, exon 5 12 311 C>T	*MIATNB*	Alter transcriptional level of MIAT	65
H19	rs217727 rs2067051	*IGF2*	Elevate expression level of IGF2 Interfere binding ability of ICRs to CCCTC-binding factor	54
Linc-VWF	rs1558324	*VWF*	Unknown	49
Diabetes mellitus				
ANRIL	rs10811661	*CDKN2A, CDKN2B, ARF*	Mediate transcriptional silencing of CDKN2A in pancreatic β-cells	70, 71, 100–107, 109
KCNQ1OT1	rs2237895	*KCNQ1*	Increase methylation levels at the CTCF-binding site	24
MEG3	rs941576	*DLK1*	Alter DLK1 expression and interfere with the imprinting mechanism	126
NONHSAG044354	rs3757247, rs597325	*BACH2*	Alter secondary structure of lnc-NONHSAG044354	133,135
Obesity				
H19	rs10732516	*IGF2*	Interfere CpG4 methylation in H19	145
LincRNA XLOC_005690	rs206936	*NUDT3*	Unknown	21

Abbreviations: ANRIL, antisense noncoding RNA in the INK4 locus; ICR, imprinting control region; IGF2, insulin-like growth factor 2; lincRNA, long intergenic noncoding RNA; lncRNA, long noncoding RNA; MEG3, maternal expressed gene 3; MIAT, myocardial infarction associated transcript; SNP, single-nucleotide polymorphism; VWF, Von Willebrand factor.

those of serum amyloid antigen, interleukin-6 and tumor necrosis factor-α.[41] Even after the level of MALAT1 was normalized, interleukin-6 and tumor necrosis factor-α mRNA levels were sustained at high level in the late stage.[41] Hyperglycemia may initiate the inflammatory response through MALAT1 and mediated the upregulation of serum amyloid antigen, interleukin-6 and tumor necrosis factor-α.[41] MALAT1 is also implicated in diabetes-induced microvascular dysfunction in rodents.[42]

HI-LNC25. HI-LNC25 is a multiexonic transcript and located nearby the *MAFB* (*V-Maf Avian Musculoaponeurotic Fibrosarcoma Oncogene Homolog B*) gene, an important regulator of human β-cell function.[43–45] A transcriptome analysis of human pancreatic islets and β-cells identified 1128 islets lncRNAs genes, most of which are located near islet-enriched protein-coding genes.[43] Among them, knockdown of a β-cell-specific lncRNA named HI-LNC25 subsequently down-regulated Gli-similar 3 (GLIS3) mRNA that encodes an important islet transcription factor.[43,46,47] The GLIS3 gene region resides in a DM susceptibility risk locus and reduced expression of GLIS3 promotes apoptosis in cultured human β-cell line.[43] Notably, HI-LNC25 and GLIS3 are located on separate chromosomes of chromosome 20 and chromosome 9, respectively, suggesting a *trans*-action of this lncRNA in diabetes pathophysiology.[48]

Expression of lncRNAs in obesity

A deep RNA sequencing of adipose tissue from lean and obese human identified lincRNA named linc doublesex and Mab-3 related transcription factor 2 (linc-DMRT2) and linc tumor protein p53 inducible protein 13 (linc-TP53I13).[49] They are adipose tissue-specific lincRNAs.[49] Lipopolysaccharide administration downregulated linc-DMRT2 and linc-TP53I13 in obese but not lean subjects.[49] The linc-DMRT2 is located on chromosome 9p24.3 that is in close proximity to a well-known multiple disease-associated lncRNA, ANRIL.[49] Aberrant expression of lncRNAs associated with cardiometabolic traits warrants more study.

VARIATIONS OF LNCRNAS IN CARDIOMETABOLIC PHENOTYPE

At present, there are more than 60 million human genetic variations in the database.[50] The SNPs, which are the result of point mutations that produce single base-pair differences, represent the most widespread variants.[51] SNPs can be found throughout the genome at a frequency of ~1 in 1000 bp.[51] They may influence promoter activity, resulting in aberrant gene expression, or alter the affinity of the regulatory protein such as transcription factor, resulting into different mRNA levels, or affect conformation and subcellular localization of mRNAs and/or proteins and hence may lead to diseases.[51] The GWAS catalog identified over 10 000 SNPs associated with various human traits and diseases, and 88% of these SNPs are located in the noncoding regions.[52] As many lncRNAs have been reported that they are dysregulated in a number of complex human diseases, SNPs may affect human diseases through expression or functions of lncRNAs.[17,39,53–62]

Clarifying the correlation between genetic variations in lncRNAs and the disease pathophysiology is still a challenging task. SNPs could play regulatory roles by altering the lncRNA expression levels, causing alternative splicing or influencing the secondary structure.[17,63]

SNPs located in the promoter region can directly decrease the expression levels of lncRNAs. SNPs may interfere with the binding site of inhibitory complexes, resulting in the increase of lncRNA expression levels.[17,64,65] On the other hand, SNPs in lncRNA transcripts can cause alternative splicing or influence the secondary structure of the lncRNA, leading to aberrant function.[17,62] Recently, the lncRNA-SNP database has been constructed in order to provide comprehensive information on lncRNA- related SNPs and their potential functions with a hope to elucidate the biological functions of lncRNAs.[62] We will present examples in which the effects of lncRNA SNPs in cardiometabolic diseases are well studied and function interpreted (Table 1).

VARIATIONS OF LNCRNAS IN CARDIAC DISEASES

Variations of lncRNAs in atherosclerosis and CAD

ANRIL. A prime example of disease-associated lncRNA is ANRIL that contains 19 exons and transcribed from a 126.3-kb region on chromosome 9p21. This region harbors the strongest genetic risk for CAD.[66] ANRIL is transcribed in an antisense orientation of the CDKN2B/CDKN2A/ARF cluster that is involved in cell-cycle regulation.[64,66] Several SNPs within or nearby ANRIL have been reported to associate with CAD, diabetes, aneurysm and various cancers.[67–75] ANRIL expression has been demonstrated in multiple atherosclerosis-related tissues, including the heart, coronary smooth muscle cells, vascular endothelial cells and monocyte-derived macrophage cells.[76–78] Silencing of ANRIL results in increased expression of CDKN2A and CDKN2B and decreased cell growth.[31,79] Overexpression of ANRIL enhances cell adhesion and cell proliferation, the key mechanisms in atherosclerosis lesions.[78] Multiple research groups have reported the association of several polymorphisms in the 58 kb interval of noncoding CAD region on chromosome 9p21, including rs1333049, rs10757278 and rs2383206.[67,68,80,81] We have also replicated the association of rs1333049 with pathologically defined atherosclerosis level in Japanese patients.[69] Importantly, the polymorphisms are not located in the exon regions of ANRIL, but are located in intron and downstream regions.[82] Thus, these SNPs may influence the expression of ANRIL and affect the downstream cell proliferation pathway.[53,82] The presence of the CAD risk allele was associated with decreased expression of ANRIL long transcript and increased expression of ANRIL short transcript. These lead to decreased expression of CDKN2B and CDKN2A.[76] However, the association between SNPs and expression levels of ANRIL, CDKN2B and CDKN2A are still controversial.[64,76,81,83–85]

A mouse model study has demonstrated the critical role of ANRIL in the regulation of CDKN2A and CDKN2B expression. ANRIL acts through a *cis*-acting mechanism to regulate proliferation and senescence.[53,79] ANRIL is involved in PRC-mediated epigenetic regulation of CDKN2B/CDKN2A/ARF locus.[30,31] Specifically, ANRIL directly binds to chromobox 7 (CBX7), a subunit of the PRC1 complex, allowing the recognition of methylation of histone 3 in the lysine 27 (H3K27me). This methylation event leads to monoubiquitination at histone 2A at lysine 119 (H2AK119), and subsequent silencing of the CDKN2A locus.[30] ANRIL also initiates repression of CDKN2B by binding to Suppressor of Zeste 12 (SUZ12) in PRC2 to induce methylation of H3K27.[31] Polymorphisms in ANRIL are thought to affect the repressing ability of the PRC that induces or inhibits the expression of CDKN2B/CDKN2A/ARF.[5] ANRIL is also implicated in atherosclerosis development by regulation of target genes in *trans*-action, resulting in activating atherogenic cell functions, including increased cell proliferation/adhesion and decreased apoptosis. Notably, Alu motifs mark the promoter regions of ANRIL *trans*-regulated target genes, suggesting that Alu motifs may have an essential role in the *trans*-regulation and the pro-atherogenic functions.[78] ANRIL also regulates cell cycle and apoptosis in DNA damage response.[86] ANRIL is upregulated by transcription factor E2F1, and subsequently represses the expression of CDKN2B/CDKN2A/ARF at the late stage of DNA damage response, allowing cells to return to their normal state.[86] Further *in vitro* and *in vivo* studies are needed for the comprehensive understanding of the role of ANRIL in various biological processes.

Myocardial infarction associated transcript (MIAT). MIAT was identified through a large-scale case–control association study of the samples from 3435 MI patients and 3774 controls using 52 608 haplotype-based SNP markers.[65] MIAT is located on chromosome 22q12.1 and contains 5 exons with no translational product.[65] Six SNPs in MIAT conferred risk for MI, including intron 1 5338 C>T (rs2331291), exon 3 8813 G>A, exon 3 9186 G>A (rs2301523), exon 5 11 093 G>A, exon 5 11 741 G>A and exon 5 12 311 C>T.[65] The six SNPs were in strong linkage disequilibrium. Among the six SNPs, the minor variant of exon 5 (11 741 G>A SNP) increased the transcriptional level of MIAT, suggesting altered expression of MIAT by the SNP might have a role in the pathogenesis of MI.[65]

H19. *H19* is a maternally expressed gene, located on chromosome 11p15, transcribing a 2.3 kb lncRNA.[87] It is abundantly expressed during embryogenesis but downregulated after birth.[88,89] Re-expression of H19 has been reported in atherosclerotic plaques of CAD patients.[90] Elevated expression levels of H19 was found in human vascular smooth muscle cells induced by homocysteine, an independent risk factor for atherosclerosis.[91] Expression of H19 in vascular smooth muscle cells may affect the development of atherosclerosis.[90,92] Polymorphisms in lncRNA H19, rs217727 and rs2067051 have been reported to confer CAD susceptibility in Han Chinese.[54] H19 resides proximal to *insulin-like growth factor 2 (IGF2)*, a maternally imprinted gene.[93] *H19* and *IGF2* genes are expressed unevenly from the two parental alleles.[94] H19 and IGF2 share a common set of downstream enhancer sequences, and H19 can downregulate the expression of IGF2 both in *cis* and *trans*-acting modes.[89,93] As H19 and IGF2 are crucial for the regulation of cell proliferation, the alteration of H19/IGF2 locus is possibly related to the formation of atherosclerosis lesions.[89] The rs217727 in exon 5 of H19 was associated with elevated plasma IGF2 levels that was implicated in the development of atherosclerosis.[89,95,96] The rs217727 may disrupt the inhibitory effect of H19 on IGF2 that may lead to the elevation of IGF2 levels and the risk for CAD.[54] On the other hand, rs2067051 in exon 1 of H19 is located near the imprinting control regions, crucial for the epigenetic regulation of H19/IGF2 locus.[94] The polymorphism might affect the binding ability of imprinting control regions to CCCTC-binding factor, resulting in the alteration of H19 expression levels.[54,89,94]

The linc-Von Willebrand factor (linc-VWF). The linc-VWF is located on chromosome 12 in the proximity of VWF, an endothelial and platelet-derived plasma glycoprotein and a key regulator of hemostasis/thrombosis.[97,98] The linc-VWF contains two exons and rs1558324 SNP located in intron 1 influences the mean platelet volume, a biomarker for CVD.[49,97,99] The polymorphisms on linc-VWF may alter the function of VWF and lead to CAD lesions. However, the mode of action of linc-VWF SNP needs to be elucidated.

VARIATIONS OF LNCRNAS IN METABOLIC DISEASES

Variations of lncRNAs in DM

ANRIL. As previously described, ANRIL is associated not only with CAD trait but also metabolic traits, suggesting a common mechanism for this locus in cardiometabolic diseases. Indeed, rs10811661 on ANRIL locus was found associated with T2D risk by three independent GWASs of Caucasians and replicated later across different ethnicities.[70,71,100–107] The rs10811661 was located 13 kb downstream from ANRIL.[108] Interestingly, rs1333049 and rs10811661, associated with CVD and T2D respectively, are in a different linkage disequilibrium block that is separated by only 1 kb.[109] The two SNPs are not in linkage disequilibrium ($r^2 < 0.01$) according to the HapMap data of CEU and JPT.[109] ANRIL may be involved in T2D development via transcriptional silencing of CDKN2B/CDKN2A/ARF locus in pancreatic β-cells and liver glucose metabolism.[82,110] The β-cell dysfunction and insulin resistance are the hallmarks of DM.[111] The rs10811661 polymorphism is associated with impaired insulin secretion, suggesting that SNP is involved in pancreatic β-cell function.[112,113] Both CDKN2B and CDKN2A are regulators of cell replication in pancreatic β-cells and several tissues.[114] Studies in aging mice demonstrated that overexpression of CDKN2A was associated with the reduced binding of B lymphoma Mo-MLV insertion region 1 homolog (Bmi1), and subsequent loss of H2A ubiquitylation at CDKN2A locus.[115,116] Decreased Bmi1 binding leads to decreased levels of H3K27 trimethylation (H3K27me3) and expression of Enhancer of zeste homolog 2 (Ezh2) that leads to reduced β-cell proliferation in aged mice.[114–117] Bmi-1 is a member of PRC1 that can repress CDKN2A and is essential for stem cell renewal.[116,117] Importantly, the repression ability of Bmi1 on CDKN2A is dependent on Ezh2 and other components of PRC2.[115,116] CDKN2A expression contributes to aging through limiting the regenerative capacity of β-cells in mice, but still lacks evidence in humans.[114] Although the mechanisms that account for ANRIL and T2D risk remain unclear, ANRIL may repress the CDKN2A expression, and subsequently promote β-cell proliferation, resulting in glucose homeostasis. On the contrary, a polymorphism nearby ANRIL might alternate the repressive state of CDKN2A and limit the replicative capacity of β-cells. Moreover, CDKN2A is also involved in the control of hepatic glucose metabolism by regulating the hepatic gluconeogenesis genes through the cAMP-response element-binding protein (CREB), an peroxisome proliferator-activated receptor gamma coactivator 1-α (PGC1α).[118] At present, there is a lack of information related to T2D-associated SNPs and CDKN2B/CDKN2A expression in the liver.[82] Additional investigation is required to clarify the mechanisms of ANRIL in DM pathogenesis.

KCNQ1OT1. KCNQ1OT1 is a 91-kb long regulatory lncRNA paternally expressed and localized exclusively in the nucleus.[23] The KCNQ1OT1 is located in the imprinted region that consists of several maternally expressed protein-coding genes, including *KCNQ1, CDKN1C, solute carrier family 22, member 18 (SLC22A18), solute carrier family 22, member 18 antisense (SLC22A18AS)* and *pleckstrin homology-like domain, family a, member 2 (PHLDA2)*.[24,119,120] KCNQ1 is a strong candidate for conferring susceptibility to T2D by impaired islet function when maternally inherited.[24] The imprinted gene expression of this region is regulated by a maternally methylated imprinting control region named potassium voltage-gated channel differentially methylated region 1 (KvDMR1) that is located at the promoter of KCNQ1 and overlaps with KCNQ1OT1.[121] Paternally hypomethylated KvDMR1 leads to expression of KCNQ1OT1 and silences downstream maternally expressed genes, including *CDKN1C*, a negative regulator of β-cell development.[48,119,120] More importantly, PRCs are thought to be involved in the paternal repression of the imprinted genes.[122] Specifically, KCNQ1OT1 interacts with Ezh2, SUZ12 subunit of PRC2, and G9a histone methyltransferase, resulting in the enrichment of H3K27me3 and H3K9me3 in a lineage-specific manner.[23] It is speculated that the disruption of methylation and imprinted gene expression in this imprinted region may lead to T2D risk.[24] A large-scale association analysis demonstrated that rs231362, which overlaps both intron of KCNQ1 and exon of antisense lncRNA KCNQ1OT1 on 11p15, reached genome-wide significant association with T2D.[123] However, no association was found between the rs231362 risk allele and the expression or methylation of KCNQ1OT1, KCNQ1, CDKN1C, PHLDA2 and SLC22A18.[24] Another variant rs2237895 located in intron 15 of KCNQ1 is a T2D susceptibility locus found by GWASs in the Japanese population and replicated in several ethnicities.[124–126] The rs2237895 risk allele is associated with increased methylation levels at the KvDMR, and subsequently reduced KCNQ1OT1 expression levels in fetal but not adult islet.[24] Similar to rs231362, rs2237895 has no relationship with expression of KCNQ1OT1, KCNQ1, CDKN1C, PHLDA2 and SLC22A18.[24] Increased expression of KCNQ1OT1 level in human islets was reported in T2D donors compared with the nondiabetic group.[43] Thus, the relationship of genetic variants on KCNQ1OT1 and T2D development warrants further investigation.

DLK1/maternal expressed gene 3 (MEG3). Apart from T2D, lnRNAs are also involved in type 1 diabetes (T1D) development. MEG3 is a maternally expressed ncRNA.[127,128] An imputation study using the data set from three large-scale GWASs identified rs941576 for paternally inherited T1D.[127] The rs941576 is located in intron 6 of the MEG3 on chromosome 14q32.2.[127] Several of the imprinted genes are located including paternally derived gene such as *Delta-Like 1 homolog (DLK1), RINGLET1 (RLT1)* and *type III iodothyronine deiodinase gene (DIO3)*, and maternally derived genes such as *MEG3* and *MEG8*. The rs941576 altered T1D risk only through paternal transmission and suggested that DLK1 is a functional candidate gene.[127] DLK1 is also known as fetal antigen 1 (FA1)[129] and is involved in cell differentiation of pancreatic β-cells and B lymphocytes.[130–133] The rs941576 may directly alter the expression of DLK1 or may interfere the imprinting mechanism to change the expression of paternally or maternally inherited copies of target genes.[127]

NONHSAG044354. A recent study has identified two SNPs rs3757247 and rs597325 that disrupt the secondary structure of NONHSAG044354, a sense exonic lncRNA in T1D locus.[134] These lncRNA structure-disruptive SNPs are located in both NONHSAG044354 and basic leucine zipper transcription factor 2 (BACH2), of which the latter is a T1D candidate gene.[134–136] Notably, the rs3757247 and rs597325 SNPs are in high ($r^2 = 0.96$) and middle ($r^2 = 0.48$) linkage disequilibrium, respectively, with a T1D susceptibility SNP rs11755527 that was identified by GWAS ($p = 4.7 \times 10^{-12}$).[109,135,137] BACH2 is thought to be involved in T1D by controlling the balance between tolerance and immunity through regulation of $CD4^+$ T-cell differentiation.[138] BACH2 is expressed in human islet β-cells and plays an antiapoptotic role.[139] The expression of the NONHSAG044354 is highly correlated with BACH2 in 14 human tissues.[134] These findings suggest that genetic variants within the lncRNAs can alter the secondary structure of lncRNAs, and subsequently interfere with their molecular functions, resulting in disease phenotype.

Variations of lncRNAs in obesity

H19. Besides the involvement in CAD development, H19 has also been implicated in obesity in a different manner. Several lines of evidence suggest that imprinting and methylation are involved in obesity, particularly in childhood.[140–145] A pilot study in 90 obese Mexican–American children showed a significant difference in the frequency of rs10732516 in CpG4 of H19 DMR between obese boys and girls.[146] A higher frequency of CpG4 methylation and a higher frequency of obesity were found among boys than girls.[146] Similarly, higher levels of methylation of H19 DMR was demonstrated in obese infants.[147] The CpG4 methylation status may be associated with childhood obesity in a gender-specific manner.[146] Among five CpG sites in H19, only CpG4 is polymorphic and the CpG4 methylation status was absent in homozygous variant of rs10732516, suggesting a destructive role of the polymorphism in the CpG site.[146] Larger-scale studies are needed to confirm this association.

The lincRNA XLOC_005690. A number of disease-associated SNPs were found to associate with expression levels of lincRNAs, using genome-wide gene expression analysis.[21] An expression quantitative trait loci study for body mass index has identified that lincRNA XLOC_005690 expression is associated with rs206936.[21] The lincRNA XLOC_005690 is located in the nucleoside diphosphate linked moiety X (nudix)-type motif 3 (NUDT3) in chromosome 6.[21] The effect of rs206936 in NUDT3 on obesity is replicated in cross-ethnicity including Europeans, Japanese and Koreans.[148–150] Specifically, the G-allele of NUDT3 rs206936 was significantly associated with increased body mass index in the obese Japanese women.[149] The mechanism of SNP and disease association remains to be clarified.

SUMMARY

GWASs have identified a number of cardiometabolic trait-associated variants. The majority of imputed SNPs lie in the noncoding region, suggesting that the noncoding genes may play an essential role. Under this context, lncRNAs are gaining attention for their biological roles and accumulating evidence indicates that lncRNAs have the potential to explain the pathogenesis of cardiometabolic diseases. The present knowledge of lncRNAs in cardiometabolic diseases is still in its infancy and exploration of them may provide deeper understanding of the regulatory mechanism of the noncoding regions. This understanding will be fundamental for exploring new diagnostic and therapeutic opportunities.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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