MicroRNA-122: A New Player in the Negative Regulation of LH Receptor Expression by the LH Receptor mRNA Binding Protein (LRBP)

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The LH receptor (LHR) plays an essential role in mediating LH actions in the steroidogenic cells of the ovary (theca, granulosa, and luteal cells) and testis (Leydig cells). It belongs to the G protein-coupled superfamily of receptors with seven transmembrane domains (1) and it is one of the most studied peptide hormone receptors. After extensive characterization of its hormone binding properties in the early to mid 1970s (2–10), in the ensuing 40 years or so, tremendous progress has been made in understanding various functions of the LHR, including the regulation of cellular signaling (11–14), expression of steroidogenic acute regulatory protein (StAR), the protein that transports cholesterol to the inner mitochondrial membrane for side-chain cleavage (P450scc, CYP11A) (14–17), endocytic and selective delivery of exogenous cholesterol to the steroidogenic cells mediated by low-density lipoprotein receptor, and scavenger receptor class B, type, respectively (18–21), and the events connected with the mobilization of cholesterol from the intracellular stores (22, 23) and the cellular cholesterol trafficking (17, 24). However, relatively less is known about the molecular and cellular regulation of LHR expression (11, 12). Until recently, based on the available data, it was generally believed that LHR expression is primarily regulated at the level of gene transcription (1, 11). In recent years, however, the seminal studies carried out by Dr Jerry Menon’s laboratory at the University of Michigan have led to the realization that LHR is also subject to posttranscription (25, 26). In fact, the overwhelming evidence now suggests that these mechanisms are major contributors to the LH-induced down-regulation of LHR. Initial studies reported by Menon and colleagues demonstrated that the expression of mevalonate kinase (MVK), a key enzyme involved in cholesterol biosynthesis, that catalyzes the conversion of mevalonic acid to 5-phosphomevalonic acid, is up-regulated under the conditions that promote the repression of the LHR expression (27–29). The follow-up studies demonstrated that MVK can directly bind to the coding region of LHR mRNA and inhibits its translational capacity (27–31); because of this specialized function, MVK was renamed by the Menon laboratory as “LHR mRNA binding protein (LRBP).” Both rat and human granulosa cells also express high levels of LRBP and the functional studies demonstrated that it interferes with the LHR expression in a manner similar to that seen in the luteinized ovary (32–34). The presence of MVK (LRBP) activity has been demonstrated in the interstitial cells of rat testis (35) but so far no functional studies have been carried out in LH responsive Leydig cells. Recent evidence suggests that in addition to gonadotropin, MVK/LRBP, a member of the oxysterol responsive gene family, is also regulated transcriptionally by the cholesterol responsive transcription factors, sterol regulatory element-binding protein-1a (SREBP-1a) and SREBP-2 (36). These various findings can be interpreted to suggest that ovarian steroid hormone biosynthesis, which is under LH regulation via LHR and is dependent on the cholesterol metabolism for the availability of cholesterol substrate, are integrated through a common regulatory mechanism acting at the level of LHR mRNA expression.

A previous study has shown that hepatic MVK is a target of microRNA-122 (miR-122); it inhibits the expression of MVK along with several other genes involved in

Abbreviations: hCG, human chorionic gonadotropin; LHR, LH receptor; LRBP, LH receptor mRNA binding protein; miRNAs, microRNAs; miR-122, microRNA-122; MVK, mevalonate kinase; SREBP, sterol regulatory element-binding protein; StAR, steroidogenic acute regulatory protein.
cholesterol biosynthesis (37), although another study suggests that it is the phosphomevalonate kinase and not the MVK, whose expression is negatively impacted by miR-122 (38). MicroRNAs (miRNAs) comprise a novel class of endogenous non-protein-coding single-stranded small RNAs of 20 to 25 nucleotides that regulate gene expression posttranscriptionally. miRNAs cause posttranscriptional repression of protein synthesis by pairing perfectly or imperfectly with the complementary sequences in the 3‘-untranslated regions of target mRNAs, leading to mRNA cleavage and degradation, translation repression, or compartmentalization and sequestration of target mRNAs (39–41). The liver-specific miR-122 is the most abundant microRNA in the liver (accounting for 70% of all the miRNAs expressed in the liver) (42). Considering this, it is not surprising that miR-122 is involved in multiple hepatic metabolic processes as well as in liver disease (42–44). MiR-122 affects hepatic cholesterol and lipid metabolism and other metabolic processes and exerts anti-inflammatory actions (37, 38, 45). It promotes stability and propagation of hepatitis C virus RNA, whereas it inhibits replication of hepatitis B virus and suppresses the hepatocarcinogenesis. The serum levels of miR-122 can serve as a potential biomarker of liver injury (44). Increased levels of miR-122 have been reported in hepatitis C virus infection, hepatitis B virus infection, hepatocarcinogenesis, drug-induced liver injury, nonalcoholic fatty liver diseases/nonalcoholic steatohepatitis, necroinflammation, and hyperlipidemia (44, 46).

In this issue of Endocrinology, Menon et al (47), using several different novel approaches, for the first time provide evidence that liver-specific miR-122 can regulate the expression of MVK/LRBP in a nonhepatic tissue, the rat ovary. Using a quantitative RT-PCR technique, the authors first demonstrated that human chorionic gonadotropin (hCG) treatment of animals up-regulates the expression of ovarian miR-122 expression in a time-dependent manner along with the reciprocal changes (down-regulation) in the mRNA levels of LHR. This in itself is a unique observation, given that until now it was thought that miR-122 is expressed exclusively in the liver (42). It is also demonstrated that hCG induction of miR-122 levels requires the participation of both cAMP-PKA and ERK signaling pathways. Quantitative RT-PCR measurements show that hCG induction of LRBP mRNA follows a pattern similar to that of miR-122 except that LRBP mRNA expression is preceded by miR-122. The authors also show that suppression of miR-122 by specific miR-122 antagonomir leads to complete inhibition of MVK/LRBP expression. Furthermore, miR-122 expression shows a temporal relationship to the expression of SREBP-1a and SREBP-2, consistent with its potential role in the regulation of MVK/LRBP.

Many important questions remain to be answered. Although Menon et al (47) report that co-treatment of rats with hCG and miR-122 leads to almost complete inhibition of MVK/LRBP protein expression, more direct evidence is needed to demonstrate that miR-122 alters the expression of MVK/LRBP. One potential approach could be to assess the effects of pre-miR-122 directly on the reporter (eg, luciferase) activity of the 3’-untranslated regions of MVK/LRBP. Also, because SREBP-1a and SREBP-2 contribute to the miRNA-122 regulation of MVK/LRBP, it would be interesting to examine the effects of miR-122 on the expression of StAR and scavenger receptor class B proteins, both of which play a crucial role in the regulation of steriodogenesis (15–18) and are targets of SREBP transcription factors (48, 49). Finally, a just published report suggests that miR-136–3p mediates hCG-induced repression of LHR expression in granulosa cells by targeting the LHR mRNA directly (50). Again, it will be interesting to know the relative contributions of miRNA-122 and miR-136–3p in hormone-induced down-regulation of LHR expression and to delineate the underlying mechanisms involved. Although much still needs to be done, the studies by Menon et al (47) laid a strong foundation, which can form the basis for future studies aimed at delineating the underlying mechanisms involved in gonadotropin-mediated posttranscriptional/posttranslational down-regulation of LHR in the steroidogenic cells of the ovary and possibly in the testicular Leydig cells and other related systems (eg, ACTH-mediated regulation of the adrenal ACTH receptor), and the roles played by the specific miRNAs in these processes. More importantly, the current studies have introduced a new concept that a noncoding region of the mRNAs, coupled with the relevant miRNAs, plays a crucial role in endocrine physiology by regulating the expression of RNA binding proteins such as MVK/LRBP shown here. In broader terms, the current study has raised the possibility that the acute hormonal stimulation of steriodogenesis is not only dependent on StAR-mediated transport of appropriate amounts of the cholesterol substrate from the outer mitochondrial membrane to the inner mitochondrial membrane, where CYP11A is located, and efficient steroidogenic machinery, but also involves the active participation of relevant mRNA binding proteins, noncoding regions of mRNAs as well as specific miRNAs.

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References


