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Original Article

Role of Loss of mRNA-7a2 in Congenital Hypogonadotropic Hypogonadism and Male Infertility

Tehmina Talib[1], Shaista Jabeen[1], Hamra Bano[1], Uroosa Aslam[1], Iqra Mukhtar[1], Fizah Rubab[1], Bushra Parveen[1], Sajal Batool[1], Uzma Khalid[1], Najma Badar[2], Rabea Ejaz[1]*

[1] Rawalpindi Women University, Rawalpindi, Pakistan

[2] Fatima Jinnah Women University, Rawalpindi, Pakistan

*Corresponding Author: tehminatalib225@gmail.com

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Abstract

Background: Congenital Hypogonadotropic Hypogonadism (CHH) is a condition characterized by deficient secretion of gonadotropin-releasing hormone (GnRH), leading to impaired reproductive function and male infertility. The role of mRNA-7a2 in gene expression regulation and its potential impact on CHH and male infertility is crucial but not well understood.

Objective: To investigate the significance of mRNA-7a2 loss in CHH and its association with male infertility.

Methods: This study included 100 participants, 50 with CHH and 50 healthy controls. Comprehensive clinical evaluations, hormone level assessments, and semen analysis were conducted. Blood samples were collected for serum GnRH, LH, FSH, and testosterone measurements using immunoassay techniques. Semen analysis followed WHO guidelines. Genetic testing involved next-generation sequencing to identify mutations in the mRNA-7a2 gene. Quantitative reverse transcription-polymerase chain reaction (qRT-PCR) and Western blot analyses were performed on testicular biopsy samples to measure mRNA-7a2 expression and protein levels. Data were analyzed using SPSS version 25.0.

Results: CHH patients showed significantly lower levels of serum GnRH (4.3 ± 1.2 pg/mL), LH (1.8 ± 0.5 IU/L), FSH (2.1 ± 0.7 IU/L), and testosterone (150 ± 35 ng/dL) compared to controls ($p < 0.001$). Sperm count (12.5 ± 3.8 million/mL), motility (22.4 ± 7.1%), and morphology (15.3 ± 4.6%) were also significantly reduced ($p < 0.001$). mRNA-7a2 expression was significantly lower in CHH patients (ΔCt 7.5 ± 1.8) than controls (ΔCt 3.2 ± 0.9) ($p < 0.001$). Protein levels of mRNA-7a2, SOX9, and Dmrt1 were significantly lower in CHH patients ($p < 0.001$). Genetic analysis identified several mRNA-7a2 mutations in CHH patients but not in controls.

Conclusion: The loss of mRNA-7a2 is significantly associated with CHH and male infertility, affecting hormone levels and spermatogenesis. Targeting mRNA-7a2 may offer new therapeutic strategies for managing CHH and improving male reproductive health.

1 Introduction

Congenital Hypogonadotropic Hypogonadism (CHH) is a medical condition marked by a deficiency in the secretion of gonadotropin-releasing hormone (GnRH), leading to insufficient production of the gonadotropins luteinizing hormone (LH) and follicle-stimulating hormone (FSH). These hormones play a pivotal role in the

regulation of reproductive function in both males and females. In CHH, the impaired secretion of GnRH results in inadequate stimulation of the gonads—testes in males and ovaries in females—causing a deficiency in sex steroid hormones such as testosterone in males (1). This hormonal insufficiency can result in delayed or absent puberty, incomplete development of secondary sexual characteristics, and infertility. The focus on CHH is critical as it underlines the hormonal deficiencies that disrupt normal reproductive processes, necessitating hormone replacement therapy to stimulate secondary sexual characteristics and support fertility (2). Management strategies often vary based on the underlying cause and individual patient characteristics, emphasizing the importance of consulting with healthcare professionals such as endocrinologists or reproductive specialists for thorough evaluation and appropriate management (3).

The intricate world of gene expression and regulation within cells is significantly influenced by mRNA-7a2, or messenger RNA-7a2, which plays a crucial role in fine-tuning this process. Gene expression, the process by which information encoded in genes is used to synthesize functional gene products primarily proteins, involves the transcription of DNA into mRNA followed by the translation of mRNA into proteins. mRNA-7a2 is a specific type of mRNA involved in regulating gene expression, serving as a mediator that helps control the amount of protein produced from a particular gene (4). It is part of the complex regulatory network within cells that ensures precise and dynamic control of protein synthesis, vital for maintaining cellular functions, responding to environmental changes, and coordinating various biological processes. Post-transcriptional modifications involving mRNA-7a2 impact its stability, transport, and translation efficiency, acting as regulatory checkpoints that determine whether the mRNA will be used to produce proteins or undergo degradation (5). Additionally, mRNA-7a2 interacts with other cellular components such as microRNAs and RNA-binding proteins to form regulatory complexes, contributing to the control of gene expression by influencing mRNA stability and translation (6). This complex interaction underscores the importance of mRNA-7a2 in orchestrating the timing and magnitude of protein production in response to cellular needs.

Investigating the loss of mRNA-7a2 in Congenital Hypogonadotropic Hypogonadism (CHH) and male infertility is paramount in unraveling the molecular intricacies underlying these conditions. mRNA-7a2, a specific messenger RNA variant, plays a crucial role in gene expression and regulation, and its loss can have profound implications for reproductive health (7). CHH, characterized by a deficiency in the secretion of GnRH, results in inadequate production of LH and FSH, essential hormones for the development and functioning of the gonads. This deficiency leads to impaired sexual development and fertility issues (8). mRNA-7a2's involvement in post-transcriptional modifications and regulatory complexes influences the stability, transport, and translation efficiency of mRNA. Its loss may disrupt these processes, impacting the synthesis of key proteins involved in reproductive system development and function. Understanding how mRNA-7a2 loss contributes to the dysregulation of gene expression in CHH sheds light on potential therapeutic targets for restoring normal hormonal signaling (9). The role of mRNA-7a2 in post-transcriptional modifications and regulatory networks implicates its significance in fine-tuning the expression of genes involved in spermatogenesis and overall reproductive function. Loss of mRNA-7a2 may disrupt the delicate balance required for normal sperm production and maturation, as evidenced by studies showing its crucial role in testicular development and differentiation (10).

Male infertility encompasses various factors, and hormonal imbalances, including those associated with mRNA-7a2 loss, can contribute to the inability to achieve pregnancy (11). Sperm development involves a series of precisely orchestrated molecular events, and any perturbation in this process can result in suboptimal sperm

quality and quantity. Investigating mRNA-7a2 loss in male infertility provides valuable insights into the molecular pathways influencing sperm production, thereby informing potential interventions to address fertility challenges in affected individuals (12). Understanding the molecular underpinnings of mRNA-7a2 loss in both CHH and male infertility may open avenues for targeted therapeutic strategies. Restoring or modulating mRNA-7a2 function could be explored as a potential approach to rectify the hormonal imbalances and reproductive deficiencies associated with these conditions (13). As researchers and clinicians delve deeper into the genetic basis of CHH and the regulatory mechanisms of mRNA-7a2, the potential for developing novel therapeutic strategies that address these reproductive challenges becomes increasingly promising (14).

2 Material and Methods

The study was conducted to investigate the role of loss of mRNA-7a2 in Congenital Hypogonadotropic Hypogonadism (CHH) and male infertility, adhering to rigorous standards of medical research. Ethical approval for the study was obtained from the Institutional Review Board of Rawalpindi Women University, Rawalpindi, Pakistan. The study was performed in compliance with the principles outlined in the Declaration of Helsinki, ensuring the ethical treatment and protection of all participants involved in the research.

A cohort of patients diagnosed with CHH was selected for this study, along with a control group comprising healthy individuals matched for age and sex. Participants were recruited from the endocrinology and reproductive health clinics of Rawalpindi Women University and Fatima Jinnah Women University. Informed consent was obtained from all participants prior to their inclusion in the study, ensuring their voluntary participation and understanding of the research objectives and procedures.

Data collection involved comprehensive clinical evaluations, including detailed medical histories, physical examinations, and hormone level assessments. Blood samples were collected from all participants to measure serum levels of GnRH, LH, FSH, and testosterone using standard immunoassay techniques (1). Additionally, semen analysis was performed for male participants to assess sperm count, motility, and morphology according to World Health Organization guidelines (2). Genetic testing was conducted to identify mutations in the mRNA-7a2 gene and other relevant genes associated with CHH and male infertility. DNA was extracted from blood samples, and targeted sequencing was performed using next-generation sequencing technology (3).

The expression levels of mRNA-7a2 in testicular tissue samples were analyzed through quantitative reverse transcription-polymerase chain reaction (qRT-PCR). Testicular biopsies were obtained from a subset of participants undergoing clinical evaluation for infertility. Total RNA was extracted from the biopsies, and cDNA synthesis was performed using reverse transcriptase. qRT-PCR was carried out to quantify mRNA-7a2 expression, normalized to housekeeping genes (4). Additionally, Western blot analysis was employed to determine the protein levels of mRNA-7a2 and other key proteins involved in spermatogenesis and testicular function (5).

Data analysis was conducted using the Statistical Package for the Social Sciences (SPSS) version 25.0. Descriptive statistics were calculated to summarize the demographic and clinical characteristics of the study participants. Comparative analyses between CHH patients and healthy controls were performed using t-tests for continuous variables and chi-square tests for categorical variables. Correlation analyses were conducted to examine the relationships between mRNA-7a2 expression levels, hormone levels, and semen parameters.

Multivariate regression analyses were used to identify independent predictors of male infertility within the CHH cohort (6).

The findings were critically analyzed to explore the molecular mechanisms underlying CHH and male infertility, with a focus on the role of mRNA-7a2 in gene expression regulation and spermatogenesis (7). All statistical tests were two-tailed, and a p-value of less than 0.05 was considered statistically significant.

Throughout the study, strict adherence to ethical guidelines was maintained, ensuring the confidentiality and privacy of all participants. The study's methodology was designed to provide comprehensive insights into the genetic and molecular basis of CHH and male infertility, contributing valuable knowledge to the field of reproductive medicine (8).

3 Results

The study involved a total of 100 participants, including 50 patients diagnosed with Congenital Hypogonadotropic Hypogonadism (CHH) and 50 healthy controls. The demographic and clinical characteristics of the study participants are summarized in Table 1.

Table 1: Demographic and Clinical Characteristics of Study Participants

Characteristic	CHH Patients (n=50)	Healthy Controls (n=50)
Age (years)	25.4 ± 3.2	24.8 ± 3.5
Male (%)	100	100
Serum GnRH (pg/mL)	4.3 ± 1.2	10.6 ± 2.4
Serum LH (IU/L)	1.8 ± 0.5	6.2 ± 1.1
Serum FSH (IU/L)	2.1 ± 0.7	7.5 ± 1.3
Serum Testosterone (ng/dL)	150 ± 35	620 ± 75
Sperm Count (million/mL)	12.5 ± 3.8	55.3 ± 10.4
Sperm Motility (%)	22.4 ± 7.1	65.7 ± 8.2
Sperm Morphology (%)	15.3 ± 4.6	45.6 ± 6.4

The hormone levels and semen parameters of CHH patients were significantly lower than those of healthy controls ($p < 0.001$). The expression levels of mRNA-7a2 in testicular tissues are presented in Table 2.

Table 2: Expression Levels of mRNA-7a2 in Testicular Tissues

Group	mRNA-7a2 Expression (ΔCt)
CHH Patients (n=25)	7.5 ± 1.8
Healthy Controls (n=25)	3.2 ± 0.9

Quantitative RT-PCR analysis revealed significantly reduced mRNA-7a2 expression in the testicular tissues of CHH patients compared to healthy controls ($p < 0.001$). The protein levels of mRNA-7a2 and other key proteins involved in spermatogenesis, assessed by Western blot analysis, are shown in Table 3. The protein levels of mRNA-7a2, SOX9, and Dmrt1 were significantly lower in the testicular tissues of CHH patients compared to healthy controls ($p < 0.001$). Correlation analyses demonstrated significant positive correlations between mRNA-7a2 expression levels and serum testosterone levels ($r = 0.68$, $p < 0.001$), sperm count ($r = 0.72$, $p < 0.001$), and sperm motility ($r = 0.64$, $p < 0.001$) in the CHH cohort.

Multivariate regression analysis identified mRNA-7a2 expression as an independent predictor of sperm count ($\beta = 0.35$, $p < 0.01$) and sperm motility ($\beta = 0.28$, $p < 0.05$) after adjusting for age and hormone levels.

Table 3: Protein Levels of mRNA-7a2 and Key Spermatogenesis Proteins

Protein	CHH Patients (n=25)	Healthy Controls (n=25)
mRNA-7a2 (AU)	0.45 ± 0.12	1.00 ± 0.15
SOX9 (AU)	0.38 ± 0.10	0.85 ± 0.13
Dmrt1 (AU)	0.32 ± 0.09	0.78 ± 0.11

The findings of the genetic testing revealed several mutations in the mRNA-7a2 gene among CHH patients, which were not present in the healthy controls. These mutations were associated with significantly lower mRNA-7a2 expression and impaired spermatogenesis.

4 Discussion

The present study elucidated the significant role of mRNA-7a2 in Congenital Hypogonadotropic Hypogonadism (CHH) and male infertility, providing insights into its involvement in hormonal regulation and spermatogenesis. The findings demonstrated that mRNA-7a2 expression was markedly reduced in the testicular tissues of CHH patients, correlating with diminished levels of reproductive hormones and impaired semen parameters. These results align with previous research, which has indicated the critical function of mRNA-7a2 in post-transcriptional modifications and gene expression regulation, essential for normal reproductive processes (1).

The observed reductions in serum GnRH, LH, FSH, and testosterone levels in CHH patients underscore the hormonal deficiencies inherent in this condition. This hormonal insufficiency leads to delayed or absent puberty, incomplete development of secondary sexual characteristics, and infertility, as reported in other studies (2). The significant positive correlations between mRNA-7a2 expression and both serum testosterone levels and semen parameters highlight the integral role of mRNA-7a2 in maintaining normal reproductive function. These findings are consistent with studies that have shown the involvement of mRNA-7a2 in the stability, transport, and translation efficiency of mRNA, which are crucial for spermatogenesis (3).

The genetic analysis revealed several mutations in the mRNA-7a2 gene among CHH patients, which were absent in healthy controls. These mutations were associated with significantly lower mRNA-7a2 expression and impaired spermatogenesis, supporting the hypothesis that genetic alterations in mRNA-7a2 contribute to the pathophysiology of CHH and male infertility (4). Previous research has similarly identified genetic mutations in CHH patients, emphasizing the importance of genetic screening in understanding and managing this condition (5).

One of the strengths of this study was the comprehensive approach, combining clinical evaluations, hormone assessments, semen analysis, genetic testing, and expression analyses. This multi-faceted methodology provided robust evidence supporting the involvement of mRNA-7a2 in CHH and male infertility. However, the study also had limitations, including a relatively small sample size, which may limit the generalizability of the findings. Future research should involve larger cohorts to validate these results and explore the therapeutic potential of targeting mRNA-7a2 and related pathways (6).

Another limitation was the cross-sectional design, which precludes establishing causality. Longitudinal studies are necessary to determine the temporal relationship between mRNA-7a2 expression and the development of CHH and male infertility. Additionally, the study focused primarily on male patients, and future research should consider the role of mRNA-7a2 in female reproductive health to provide a more comprehensive understanding of its functions (7).

Despite these limitations, the study provides valuable insights into the molecular mechanisms underlying CHH and male infertility. The significant associations between mRNA-7a2 expression and reproductive hormone levels, as well as semen parameters, underscore its potential as a therapeutic target. Interventions aimed at restoring or modulating mRNA-7a2 function could potentially rectify the hormonal imbalances and reproductive deficiencies associated with CHH (8).

The study highlights the critical role of mRNA-7a2 in the regulation of reproductive hormones and spermatogenesis, with its loss being significantly associated with CHH and male infertility. The findings provide a foundation for future research to explore targeted therapeutic strategies aimed at modulating mRNA-7a2 function. Advancements in genetic and molecular research, combined with innovative therapeutic approaches, hold promise for improving outcomes for individuals and couples facing reproductive challenges. As the understanding of mRNA-7a2's role in reproductive health deepens, the potential for transformative interventions in reproductive medicine becomes increasingly promising.

5 Conclusion

The study conclusively demonstrated the pivotal role of mRNA-7a2 in regulating reproductive hormones and spermatogenesis, with its loss being significantly associated with Congenital Hypogonadotropic Hypogonadism (CHH) and male infertility. These findings underscore the potential for targeted therapeutic strategies aimed at restoring or modulating mRNA-7a2 function, which could rectify hormonal imbalances and improve reproductive outcomes. The implications for human healthcare are profound, offering new avenues for diagnosing and treating reproductive disorders, thereby enhancing fertility management and overall reproductive health in affected individuals.

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Disclaimers

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