Gonadotropin-Releasing Hormone Receptor II in Rare Ovarian Neoplasms: Correlation with VEGF and Clinicopathological Features

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Abstract: ***Background*****:** Rare ovarian tumors pose diagnostic and therapeutic challenges due to their heterogeneous biology and limited treatment options. Gonadotropin-releasing hormone receptor type II (GnRHR-II) has been implicated in tumor regulation, including potential roles in angiogenesis, yet its clinicopathological significance in rare ovarian tumors remains unclear. This study aimed to evaluate GnRHR-II expression and its association with vascular endothelial growth factor (VEGF) as an angiogenic marker.

Methods**:** This analytical observational study used a cross-sectional design on tissue microarray samples containing 29 rare ovarian tumors, of which 25 were eligible for analysis. Immunohistochemistry was performed to assess GnRHR-II and VEGF expression, quantified using the H-score method. Associations with clinicopathological parameters were analyzed using t-test, ANOVA, Fisher's exact test, and Pearson's correlation, with $p < 0.05$ considered significant.

Results**:** The mean age of patients was 40.12 ± 19.73 years, with most cases diagnosed at stage I (72%) and classified as malignant (68%). GnRHR-II was consistently expressed across histopathological subtypes, with a mean H-score of 187.50 ± 84.86, while VEGF showed a mean H-score of 103.35 ± 76.94. VEGF expression significantly correlated with tissue type ($p = 0.004$), being lowest in borderline tumors. There was positive expression of GnRHR-II in rare ovarian cancer. However, correlation analysis revealed no significant relationship between GnRHR-II and VEGF ($p = 0.541$), indicating independent signaling pathways.

Conclusion**:** GnRHR-II is widely expressed in rare ovarian tumors but does not correlate with angiogenesis as reflected by VEGF expression. These findings suggest that GnRHR-II may act independently of VEGF-driven pathways, potentially serving a role in tumor biology distinct from angiogenic regulation. Further studies are warranted to explore its functional and therapeutic implications.

Keywords: Gonadotropin-releasing hormone receptor ii; rare ovarian tumor; angiogenesis; vascular endothelial growth factor; immunohistochemistry.

1. INTRODUCTION

Ovarian cancer remains one of the most lethal gynecologic malignancies, largely due to late-stage diagnosis, tumor heterogeneity, and limited targeted treatment options [1]. In 2020, a total of 313,959 ovarian cancer cases were reported, reflecting a 5.9% rise compared to the 295,414 cases recorded in 2018. Additionally, ovarian cancer ranks as the eighth most common cause of cancer-related mortality worldwide, with a five-year survival rate of less than 45% [1, 2].

While the majority of ovarian cancers are of epithelial origin, a significant proportion comprises rare ovarian tumors, including sex cord-stromal tumors, germ cell tumors, and rare epithelial subtypes [3]. These rare entities, although infrequent, present unique biological and clinical challenges due to their diverse molecular profiles, unpredictable clinical courses, and relative resistance to conventional chemotherapy [4, 5]. As such, there is an urgent need to identify novel molecular targets and pathways that could refine diagnostic accuracy, prognostic assessment, and therapeutic strategies specifically tailored for these uncommon ovarian neoplasms.

The gonadotropin-releasing hormone (GnRH) system, traditionally known for its role in regulating the hypothalamic-pituitary-gonadal axis, has emerged as an important extra-hypothalamic signaling pathway in a variety of tumors, including those of the reproductive tract [6-8]. GnRH exerts its effects through binding to GnRH receptors (GnRHR), of which two isoforms have been identified in humans: GnRH receptor type I (GnRHR-I) and the less-characterized GnRH receptor type II (GnRHR-II).. Interestingly, while the therapeutic potential of GnRH-I has been previously documented, GnRH-II demonstrated a greater ability to promote cell pro-

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liferation and exhibited significantly higher receptor binding affinity compared to GnRH-I [9, 10].

Notably, some studies suggest that GnRHR-II may act independently of the classical GnRHR-I pathway, engaging distinct intracellular signaling cascades that influence tumor behavior [11-14]. One hypothesized function of GnRHR-II is its involvement in inhibiting angiogenesis, a critical process in tumor growth and metastasis [15, 16]. Angiogenesis in the tumor microenvironment is driven by a complex interplay of pro-angiogenic factors such as vascular endothelial growth factor (VEGF), inflammatory mediators, and hypoxia-inducible factors [17-19]. Dysregulated angiogenesis is a hallmark of aggressive tumor phenotypes and is often associated with poor clinical outcomes [20]. Given the potential regulatory role of GnRHR-II in vascular remodeling and its presumed anti-angiogenic effects in other cancers, its expression in rare ovarian tumors may provide insight into previously unrecognized mechanisms of tumor progression and vascular support.

Although previous studies have shown that GnRHR-II is expressed extrapituitarily in various ovarian tumor types—including rare subtypes—and that its highest expression occurs significantly in sex cord stromal tumors (SCST), the expression profile and clinicopathological relevance of GnRHR-II in rare ovarian tumors remain poorly understood, especially in relation to angiogenesis and tumor aggressiveness. GnRHR-II expression has been found to correlate significantly with patient age (particularly those over 40 years old) and the primary site of the tumor, although no notable differences have been observed between different tumor types [32, 22]. These findings suggest that higher GnRHR-II expression may be linked to lower proliferative activity, particularly in SCST and tumors confined to the ovary, indicating a potential antiproliferative function in the context of rare ovarian tumors [22]. In light of these observations, the present study aims to comprehensively assess the expression of GnRHR-II receptor type II in rare ovarian tumors and its association with clinicopathological features and angiogenic activity, in order to address a critical gap in current knowledge and explore its potential as a prognostic biomarker or therapeutic target.

2. METHOD

2.1. Study Design

This research is an analytical observational study with a cross-sectional approach, aiming to investigate the association between GnRHR-II expression and the clinicopathological features as well as the proliferative activity of rare ovarian tumors.

2.2. Subjects

The study used commercial Tissue Micro Array (TMA) samples from Novus Bio (US), product code NBP2-30290 – Human Ovary Tissue MicroArray (Cancer), containing 59 ovarian tumor cores and one carbon core as a position marker. The following samples were excluded:

- Ovarian tumor types with a prevalence greater than 6 per 100,000 people per year;
- Ovarian tumor types with a frequency above 10%;
- Incomplete clinicopathological data; and
- Damaged or missing cores.

The research received ethical approval from the Ethics Committee of the Faculty of Medicine, Public Health, and Nursing at Universitas Gadjah Mada No. KE/0806/05/2023.

2.3. Immunohistochemical Staining

After deparaffinization and hydration, antigen retrieval was performed using an EDTA buffer (pH 9.0) at 90°C for 20 minutes. The slides were washed in PBS for 15 minutes and treated with 3% hydrogen peroxide solution for 6 minutes, then washed again in PBS for another 15 minutes. Next, the slides were blocked with serum for 30 minutes, followed by incubation with anti-GnRHR-II (catalog no. bs11403R, Bioss, US) and anti-VEGF polyclonal antibodies for 60 minutes at a dilution of 1:200.. After washing with PBS, the slides were incubated with a secondary antibody (Ultrateck Anti-polyvalent, Biotinylated) for 30 minutes, followed by further washing and incubation with HRP for 30 minutes. The slides were then washed again and incubated with DAB solution for 2 minutes, after which the reaction was stopped with tap water. Counterstaining was performed using Meyer's hematoxylin for 1 minute. The slides were dehydrated through ethanol series (10 dips at each concentration), cleared in xylene (10 dips), and mounted with coverslips.

2.4. Assessment of GnRHR-II and VEGF Expression

The slides were examined under a confocal microscope at 400× magnification, with images taken from four fields of view. The expression of GnRHR-II and VEGF was assessed quantitatively using the H-score method, which combines staining intensity and the percentage of positive cells, with scores ranging from 0 to 300. The H-score is calculated using the formula:

$$\text{H-Score} = (0 \times P0) + (1 \times P1) + (2 \times P2) + (3 \times P3)$$

Where P0 to P3 represent the percentage of cells with staining intensities of 0, 1, 2, and 3, respectively. Intensity and cell percentage measurements were performed semi-automatically using the Immunohistochemical IHC Profiler plugin in ImageJ version 1.53.

2.5. Statistical Analysis

Statistical analysis was performed using SPSS version 27.0. The relationships between GnRHR-II expression and clinicopathological variables were analyzed using the independent t-test for two groups and ANOVA for more than two groups. The correlation between GnRHR-II and VEGF was examined using Pearson's correlation test, while categorical variables were analyzed using Fisher's exact test. A p-value of less than 0.05 was considered statistically significant.

3. RESULT

3.1. Subject Characteristics

Altogether, 29 tissue samples were available, but 4 were excluded owing to sample damage. The mean age of the pa-

tients was 40.12 ± 19.73 years. Most cases were diagnosed at Stage I (72.0%), with the ovary as the predominant site of origin (84.0%). The majority of tissues were malignant (68.0%), and early onset (<45 years) was more frequent (64.0%). Histopathologically, germinal cell tumors (32.0%) and ovarian epithelial tumors (28.0%) were the most common, followed by sex cord stromal cell tumors (24.0%), metastatic tumors (12.0%), and miscellaneous types (4.0%) (Table **1**).

3.2. GnRHR-II Expression in Ovarian Tumor Tissue Samples

The expression of GnRHR-II was assessed through IHC staining, where a positive result was determined by brown staining of the tumor cytoplasm using DAB. Quantitative evaluation of GnRHR-II expression was performed in four microscopic fields based on the H-score. In this analysis, three staining intensities were categorized as positive (Fig. **1A**), weakly positive (Fig. **1B**), and negative (Fig. **1C**). The histogram profile represented the distribution of pixel intensity values and their corresponding counts. The log beneath the histogram illustrated the precise percentage of pixels within each intensity range and their respective scoring zones. Pixel intensity values were classified as follows: 61-120 pixels for the positive zone, 121-180 pixels for the weakly positive zone, and 181–235 pixels for the negative zone.

3.3. Comparison of GnRHR-II and VEGF Expression

The comparison of GnRHR-II and VEGF expression based on clinicopathological parameters showed varied patterns across subgroups (Table **2**). Although VEGF expression appeared to increase with advancing cancer stage, from 196.28 ± 83.95 in stage I to 200.61 ± 0.00 in stage III, the differences were not statistically significant (p = 0.373). Similarly, GnRHR-II expression did not significantly differ among stages (p = 0.567). When analyzed by cancer origin, ovarian tumors and tumors from other organs demonstrated comparable levels of VEGF (99.69 ± 70.45 *vs*. 122.54 ± 116.96) and GnRHR-II (189.18 ± 84.41 *vs*. 178.65 ± 99.90), with no significant associations observed.

In contrast, tissue type showed a significant association with VEGF expression (p = 0.004), where borderline or uncertain malignant potential tumors had the lowest VEGF levels (14.70 ± 14.53) compared to benign tumors (101.92 ± 66.10) and cancers (129.68 ± 71.33). However, GnRHR-II expression did not significantly differ among tissue categories (p = 0.527). While onset (early *vs*. late) and histopathological type displayed numerical differences in both VEGF and GnRHR-II expression, these variations did not reach statistical significance. Overall, these findings suggest that VEGF expression is more strongly influenced by tissue type than by cancer stage, origin, onset, or histopathological subtype, whereas GnRHR-II expression showed no significant associations with any clinicopathological variable (Table **2**).

Table 1. Characteristics of Subjects at Baseline.

Characteristics	Result (n = 25)
Age (Mean ± SD)	40.12 ± 19.73
Cancer Stage (n, %)	-
Stage I	18 (72.0%)
Stage II	4 (16.0%)
Stage III	1 (4.0%)
Stage IV	2 (8.0%)
Origin of Cancer Cell (n, %)	-
Ovarium	21 (84.0%)
Another Organs	4 (16.0%)
Tissue Type (n, %)	-
Benign Tumor	3 (12.0%)
Tumor of borderline malignancy or uncertain malignant potential	5 (20.0%)
Cancer	17 (68.0%)
Onset (n, %)	-
Early Onset (≤45 Years)	16 (64.0%)
Late Onset (>45 Years)	9 (36.0%)

Histopathological Type (n, %)	-
Ovarian Epithelial Tumor	7 (28.0%)
Germinal Cell Tumor	8 (32.0%)
Sex Cord Stromal Cell Tumor	6 (24.0%)
Metastatic Tumor	3 (12.0%)
Miscellaneous	1 (4.0%)

Note: Description: (**a**) Correlation between baseline characteristics and GNRHR-II expression; (**b**) Correlation between baseline characteristics and VEGF expression.

Table 2. Comparison of GnRHR-II and VEGF Expression Based on Clinicopathology.

Parameter	**n (%)**	**VEGF (Mean ± SD)**	***p* value**	**GnRHR-II (Mean ± SD)**	***p* value**
All	25 (100%)	103.35 ± 76.94	–	187.50 ± 84.86	–
Cancer Stage (n, %)	-	-	0.373[a]	-	0.567[a]
Stage I	18 (72.0%)	196.28 ± 83.95	-	98.14 ± 70.97	-
Stage II	4 (16.0%)	164.21 ± 107.75	-	130.56 ± 110.87	-
Stage III	1 (4.0%)	200.61 ± 0.00	-	249.78 ± 0.00	-
Stage IV	2 (8.0%)	123.86 ± 51.26	-	47.17 ± 18.65	-
Origin of Cancer Cell (n, %)	-	-	0.597[c]	-	0.826[c]
Ovarium	21 (84.0%)	99.69 ± 70.45		189.18 ± 84.41	-
Another Organs	4 (16.0%)	122.54 ± 116.96		178.65 ± 99.90	-
Tissue Type (n, %)	-	-	0.004[b*]		0.527[a]
Benign Tumor	3 (12.0%)	101.92 ± 66.10	-	134.00 ± 65.71	-
Tumor of borderline malignancy or uncertain malignant potential	5 (20.0%)	14.70 ± 14.53	-	195.89 ± 98.36	-
Cancer	17 (68.0%)	129.68 ± 71.33	-	194.47 ± 85.04	-
Onset (n, %)	-	-	0.522[c]	-	0.141[d]
Early Onset (≤45 Years)	16 (64.0%)	95.75 ± 71.08	-	169.43 ± 87.42	-
Late Onset (>45 Years)	9 (36.0%)	116.86 ± 89.24	-	219.63 ± 73.89	-
Histopathological Type (n, %)	-	-	0.147[b]	-	0.081[b]
Ovarian Epithelial Tumor	7 (28.0%)	48.49 ± 62.96	-	162.36 ± 89.95	-
Germinal Cell Tumor	8 (32.0%)	127.68 ± 65.27	-	162.66 ± 73.23	-
Sex Cord Stromal Cell Tumor	6 (24.0%)	122.18 ± 59.97	-	255.85 ± 61.79	-
Metastatic Tumor	3 (12.0%)	129.01 ± 142.37	-	131.23 ± 38.43	-
Miscellaneous	1 (4.0%)	181.03 ± 0.00	-	320.92 ± 0.00	-

Note: Description: **a**) ANOVA test; **b**) Kruskal Wallis test; **c**) Unpaired t test; **d**) Mann-Whitney test.

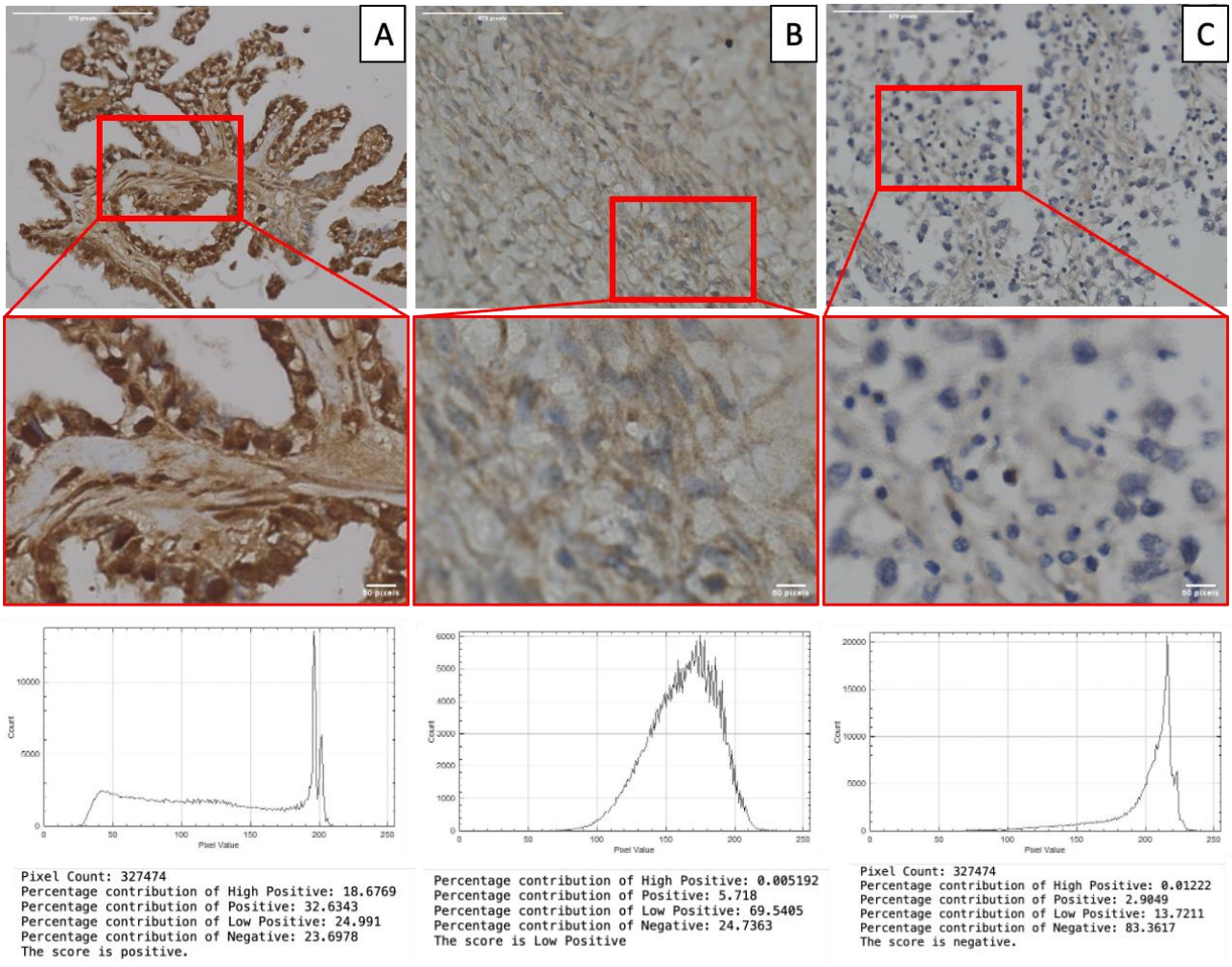

Fig. (1). Immunohistochemical staining of GnRHR-II showing staining intensity, region of interest, histogram distribution, and semiautomated scoring analyzed using the IHC profiler. (**A**) Positive (**B**) Weakly positive (**C**) Negative. Images were captured under 400× magnification.

Table 3. Correlation of GnRHR-II and VEGF Expressions.

Variable	H-Score (Mean ± SD)	R	p value
VEGF	103.35 ± 76.94	0.128	0.541
GnRHR-II	187.50 ± 84.86		

3.4. Correlation of GnRHR-II and VEGF Expressions

Table **3** below demonstrates the correlation between GnRHR-II and VEGF expressions. The mean H-score of VEGF expression was 103.35 ± 76.94, while GnRHR-II showed a higher mean H-score of 187.50 ± 84.86. Statistical analysis revealed a weak positive correlation between GnRHR-II and VEGF expressions (R = 0.128); however, this association was not statistically significant (p = 0.541). These findings suggest that GnRHR-II expression does not have a meaningful correlation with VEGF expression in this study population.

4. DISCUSSION

This study investigated the expression of gonadotropin-releasing hormone receptor type II (GnRHR-II) in rare ovarian tumors and its association with clinicopathological features and angiogenesis, particularly vascular endothelial growth factor (VEGF) expression. Our findings demonstrated that while GnRHR-II was consistently expressed across various histopathological subtypes, its expression did not significantly correlate with clinicopathological characteristics or VEGF levels. Conversely, VEGF expression varied significantly with tissue type, being lowest in borderline tumors compared to benign and malignant counterparts. These

results provide important insights into the complex biology of rare ovarian tumors and the potential role of GnRHR-II as a biological marker.

The presence of GnRHR-II in diverse rare ovarian tumor subtypes aligns with earlier reports indicating extrapituitary GnRH receptor expression in gynecological cancers, including ovarian, endometrial, and cervical tumors. Prior studies have suggested that GnRHR-II can exert antiproliferative effects, potentially through inhibition of growth factor signaling pathways such as Epidermal Growth Factor Receptor (EGFR)-mediated cascades [23, 24]. This mechanism may explain why GnRHR-II expression in our study did not parallel VEGF expression or correlate with more aggressive tumor stages. Instead, its presence across histological categories could indicate a broader regulatory function, possibly acting as a tumor-suppressive modulator rather than a driver of angiogenesis.

Interestingly, VEGF expression was found to be significantly associated with tissue type, with borderline tumors showing remarkably low angiogenic activity. This observation is consistent with the intermediate biological behavior of borderline ovarian tumors, which typically demonstrate limited invasiveness and a more indolent clinical course [25, 26] Elevated VEGF levels in malignant tumors reinforce the central role of angiogenesis in driving tumor aggressiveness and progression [27, 28]. These findings support existing literature highlighting VEGF as a reliable indicator of malignant potential and a therapeutic target in ovarian cancer.

The lack of significant correlation between GnRHR-II and VEGF expression suggests that GnRHR-II may not directly regulate angiogenesis in rare ovarian tumors, particularly not through VEGF-dependent pathways. While in vitro studies have demonstrated that GnRHR-II activation can influence angiogenic processes, including the modulation of VEGF secretion, the weak and non-significant association found in this study indicates that such effects are likely context-dependent [15, 16]. These variations may be influenced by tumor subtype [29], the surrounding microenvironment [30], or differential activity of receptor isoforms [31]. Moreover, GnRHR-II may exert its regulatory effects through alternative angiogenic or anti-angiogenic mediators beyond VEGF, such as fibroblast growth factor (FGF) [32] and platelet-derived growth factor (PDGF) [33].

Mechanistically, GnRHR-II signaling is distinct from the canonical VEGF-driven angiogenic cascade. VEGF expression in tumors is largely controlled by hypoxia-inducible factor-1α (HIF-1α), inflammatory cytokines, and oncogenic signaling pathways such as phosphatidylinositol 3-kinase/protein kinase B (PI3K/AKT) and mitogen-activated protein kinase/extracellular signal-regulated kinase (MAPK / ERK) [34, 35]. In contrast, GnRHR-II activation predominantly engages G protein–coupled signaling, including Gi/Go-mediated inhibition of cyclic adenosine monophosphate (cAMP) and activation of phosphotyrosine phosphatases, which are more closely associated with antiproliferative and pro-apoptotic effects than with VEGF transcriptional control [36, 37]. Although some experimental models have reported that GnRHR-II agonists can suppress VEGF secretion [15, 38]. These effects may appear stronger in hormone-dependent epithelial tumors or endometriosis than in rare ovarian tumor subtypes. Taken together, the absence of a significant correlation in this study suggests that angiogenesis in rare ovarian tumors is driven primarily by VEGF-independent pathways—such as hypoxia and inflammatory mediators—rather than by GnRHR-II signaling, thereby explaining the lack of molecular association between these two markers.

Our findings also showed that GnRHR-II expression tended to be higher in sex cord stromal tumors compared to epithelial and germ cell tumors, although this trend did not reach statistical significance. This pattern aligns with prior studies suggesting that sex cord stromal tumors may be particularly enriched in GnRHR-II expression, possibly reflecting their unique hormonal microenvironment [22, 39]. Given that many SCSTs retain endocrine activity, the presence of GnRHR-II may represent a physiological remnant of gonadotropin responsiveness or a compensatory mechanism regulating proliferation. Another important aspect is the lack of association between GnRHR-II expression and tumor stage. Unlike VEGF, which is strongly linked to tumor progression and metastatic potential, GnRHR-II expression appeared relatively stable across early and advanced stages. This finding may support the hypothesis that GnRHR-II expression is more reflective of intrinsic tumor biology rather than tumor aggressiveness [22, 40]. Consequently, GnRHR-II might serve as a diagnostic or classification marker rather than a prognostic one in rare ovarian tumors.

The clinical implications of these results are twofold. First, VEGF remains a critical biomarker for malignancy and a therapeutic target, consistent with the established role of anti-angiogenic therapies such as bevacizumab in ovarian cancer treatment. Second, GnRHR-II could represent an adjunct biomarker for refining tumor classification, particularly in rare ovarian tumor subtypes where diagnostic challenges often arise due to overlapping histological features. Furthermore, if future studies confirm its antiproliferative role, GnRHR-II agonists or modulators could represent a novel therapeutic avenue, complementing existing treatment strategies.

This study also raises several questions. The weak correlation between GnRHR-II and VEGF suggests that larger cohorts and mechanistic studies are needed to clarify whether GnRHR-II influences angiogenesis indirectly or through non-VEGF pathways. Additionally, functional assays examining the effect of GnRHR-II activation or inhibition on cell proliferation, apoptosis, and angiogenic factor secretion in rare ovarian tumor models would provide critical mechanistic insights. From a methodological standpoint, the use of tissue microarray (TMA) provided efficiency in analyzing multiple rare tumor subtypes but also introduced certain limitations, such as heterogeneity within tumor sections and the small sample size of each histological group [21, 22]. These factors may partially explain why some associations did not reach statistical significance. Expanding the cohort size, particularly for less common tumor subtypes, would enhance the robustness of future analyses.

Another limitation is the cross-sectional design, which restricts causal inference. Longitudinal studies correlating GnRHR-II expression with clinical outcomes such as recurrence, survival, and treatment response would be essential to

establish its true prognostic or predictive value. Additionally, exploring co-expression patterns of GnRHR-II with other signaling molecules or receptors (e.g., GnRHR-I, EGFR, or hormone receptors) may reveal synergistic or antagonistic roles in tumor biology. Despite these limitations, this study contributes novel evidence by demonstrating that GnRHR-II is expressed in a spectrum of rare ovarian tumors but is not significantly associated with angiogenesis as measured by VEGF. These findings suggest that GnRHR-II may play a role distinct from conventional angiogenic regulation and warrant further exploration as a biomarker and potential therapeutic target.

In summary, while VEGF expression strongly reflects malignant potential and angiogenic activity, GnRHR-II appears to act independently of VEGF-related pathways in rare ovarian tumors. Its consistent expression across subtypes highlights its biological relevance, but its precise functional role remains to be elucidated. Future investigations combining molecular, functional, and clinical outcome analyses are needed to unlock the potential of GnRHR-II as a diagnostic and therapeutic target in the management of rare ovarian tumors.

CONCLUSION

This study demonstrates that GnRHR-II is consistently expressed in rare ovarian tumors but shows no significant association with clinicopathological features or VEGF expression. While VEGF levels varied according to tissue type and reflected angiogenic activity, GnRHR-II expression appeared independent of VEGF-driven pathways. These findings suggest that GnRHR-II may play a role in tumor biology distinct from conventional angiogenic regulation. Further studies with larger cohorts and functional analyses are warranted to clarify its potential as a diagnostic marker and therapeutic target in rare ovarian tumors.

REFERENCES

[1] Webb PM, Jordan SJ. Epidemiology of epithelial ovarian cancer. Best Pract Res Clin Obstet Gynaecol. 2017;41:3–14. Available from: http://dx.doi.org/10.1016/j.bpobgyn.2016.08.006

[2] Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA Cancer J Clin. 2021;71(3):209–49.

[3] Debuquoy C, Romeo C, Vanacker H, Ray-Coquard I. Rare ovarian tumors: an update on diagnosis and treatment. Int J Gynecol Cancer. 2020;30(6):879–87. Available from: https://www.sciencedirect.com/science/article/pii/S1048891X24001622

[4] Ray-Coquard I, Trama A, Seckl MJ, Fotopoulou C, Pautier P, Pignata S, et al. Rare ovarian tumours: Epidemiology, treatment challenges in and outside a network setting. Eur J Surg Oncol. 2019;45(1):67–74. Available from: https://www.sciencedirect.com/science/article/pii/S0748798317309472

[5] Varghese A, Lele S. Rare Ovarian Tumors. In: Lele S, editor. Ovarian Cancer. Brisbane (AU): Ovarian Cancer; 2022.

[6] Filicori M. Pulsatile gonadotropin-releasing hormone: clinical applications of a physiologic paradigm. F&S reports. 2023;4(2 Suppl):20–6.

[7] Wickramasuriya N, Hawkins R, Atwood C, Butler T. The roles of GnRH in the human central nervous system. Horm Behav. 2022;145:105230. Available from: https://www.sciencedirect.com/science/article/pii/S0018506X22001246

[8] Wu H-M, Chang H-M, Leung PCK. Gonadotropin-releasing hormone analogs: Mechanisms of action and clinical applications in female reproduction. Front Neuroendocrinol. 2021;60:100876. Available from: https://www.sciencedirect.com/science/article/pii/S0091302220300674

[9] Gründker C, Günthert AR, Millar RP, Emons G. Expression of gonadotropin-releasing hormone II (GnRH-II) receptor in human endometrial and ovarian cancer cells and effects of GnRH-II on tumor cell proliferation. J Clin Endocrinol Metab. 2002;87(3):1427–30.

[10] Huang H, Li YJ, Lan CY, Huang QD, Feng YL, Huang YW, et al. Clinical significance of ascites in epithelial ovarian cancer. Neoplasma. 2013;60(5):546–52.

[11] Eicke N, Günthert AR, Emons G, Gründker C. GnRH-II agonist [D-Lys6]GnRH-II inhibits the EGF-induced mitogenic signal transduction in human endometrial and ovarian cancer cells. Int J Oncol. 2006;29(5):1223–9.

[12] Gründker; Emons G. Role of Gonadotropin-Releasing Hormone (GnRH) in Ovarian Cancer. 2021;

[13] Gründker C, Völker P, Schulz KD, Emons G. Luteinizing hormone-releasing hormone agonist triptorelin and antagonist Cetrorelix inhibit EGF-induced c-fos expression in human gynecological cancers. Gynecol Oncol. 2000;78(2):194–202.

[14] Gründker C, Völker P, Emons G. Antiproliferative signaling of luteinizing hormone-releasing hormone in human endometrial and ovarian cancer cells through G protein αI-mediated activation of phosphotyrosine phosphatase. Endocrinology. 2001;142(6):2369–80.

[15] Huang F, Wang H, Zou Y, Liu Q, Cao J, Yin T. Effect of GnRH-II on the ESC proliferation, apoptosis and VEGF secretion in patients with endometriosis in vitro. Int J Clin Exp Pathol. 2013;6(11):2487–96.

[16] Filindris T, Papakonstantinou E, Keramida M, Panteris E, Kalogeropoulos S, Georgopoulos N, et al. The effect of GnRH-a on the angiogenesis of endometriosis. Hormones. 2024;23(3):509–15. Available from: https://doi.org/10.1007/s42000-024-00559-6

[17] Verheul HMW, Pinedo HM. The Role of Vascular Endothelial Growth Factor (VEGF) in Tumor Angiogenesis and Early Clinical Development of VEGFReceptor Kinase Inhibitors. Clin Breast Cancer. 2000;1:S80–4. Available from: https://www.sciencedirect.com/science/article/pii/S1526820911701549

[18] Srinivasan S, Chitalia V, Meyer RD, Hartsough E, Mehta M, Harrold I, et al. Hypoxia-induced expression of phosducin-like 3 regulates expression of VEGFR-2 and promotes angiogenesis. Angiogenesis. 2015;18(4):449–62.

[19] Medzhitov R. Origin and physiological roles of inflammation. Nature. 2008;454(7203):428–35.

[20] Li X-Y, Ma W-N, Su L-X, Shen Y, Zhang L, Shao Y, et al. Association of Angiogenesis Gene Expression With Cancer Prognosis and Immunotherapy Efficacy. Front cell Dev Biol. 2022;10:805507.

[21] Wicaksono AW, Anggorowati N, Kusumanto A, Kurniadi A, Zucha MA. Clinicopathological Profiles and Proliferative Activity of GnRH Receptor Type II Expressing Ovarian Cancer. Indian J Gynecol Oncol. 2025;23(1):48. Available from: https://doi.org/10.1007/s40944-025-00978-2

[22] Ariesaka K, Kusumanto A, Zucha M, Anggorowati N, Wicaksono A, Yunus M, et al. Expression of gonadotropin-releasing hormone receptor type-II correlates with proliferation activity in tissue microarray of rare ovarian tumor. Indones J Med Lab Sci Technol. 2024;6:95–106.

[23] Cho-Clark MJ, Watkins A, Wu TJ. The role of GnRH metabolite, GnRH-(1-5), in endometrial cancer. Front Endocrinol (Lausanne). 2023;14:1183278.

[24] Cho-Clark M, Larco DO, Semsarzadeh NN, Vasta F, Mani SK, Wu TJ. GnRH-(1-5) transactivates EGFR in Ishikawa human endometrial cells via an orphan G protein-coupled receptor. Mol Endocrinol. 2014;28(1):80–98.

[25] Waluyo ST, Tjokroprawiro BA. The clinicopathological characteristics of borderline ovarian tumors at Dr. Soetomo Hospital, Surabaya [Internet]. Vol. 12, a single center study . 2023. p. 615–20. Available from: https://www.scopus.com/pages/publications/85153783422

[26] Lalwani N, Shanbhogue AKP, Vikram R, Nagar A, Jagirdar J, Prasad SR. Current Update on Borderline Ovarian Neoplasms. Am J Roentgenol. 2012;194(2):330–6. Available from: https://doi.org/10.2214/AJR.09.3936

[27] Masoumi Moghaddam S, Amini A, Morris DL, Pourgholami MH. Significance of vascular endothelial growth factor in growth and peritoneal dissemination of ovarian cancer. Cancer Metastasis Rev. 2012;31(1–2):143–62.

[28] Ramakrishnan S, Subramanian I V, Yokoyama Y, Geller M. Angiogenesis in normal and neoplastic ovaries. Angiogenesis. 2005;8(2):169–82.

[29] Cheung LWT, Wong AST. Gonadotropin-releasing hormone: GnRH receptor signaling in extrapituitary tissues. FEBS J. 2008;275(22):5479–95. Available from: https://doi.org/10.1111/j.1742-4658.2008.06677.x

[30] Poon SL, Lau MT, Hammond GL, Leung PCK. Gonadotropin-releasing hormone-II increases membrane type I metalloproteinase production via β-catenin signaling in ovarian cancer cells. Endocrinology. 2011;152(3):764–72.

[31] Chien CH, Chen CH, Lee CYG, Chang TC, Chen RJ, Chow SN. Detection of gonadotropin-releasing hormone receptor and its mRNA in primary human epithelial ovarian cancers. Int J Gynecol Cancer. 2004;14(3):451–8.

[32] Di Lieto A, De Falco M, Pollio F, Mansueto G, Salvatore G, Somma P, et al. Clinical response, vascular change, and angiogenesis in gonadotropin-releasing hormone analogue-treated women with uterine myomas. J Soc Gynecol Investig. 2005;12(2):123–8.

[33] Di Lieto A, De Rosa G, De Falco M, Iannotti F, Staibano S, Pollio F, et al. Relationship between platelet-derived growth factor expression in leiomyomas and uterine volume changes after gonadotropin-releasing hormone agonist treatment. Hum Pathol. 2002;33(2):220–4.

[34] Magar AG, Morya VK, Kwak MK, Oh JU, Noh KC. A Molecular Perspective on HIF-1α and Angiogenic Stimulator Networks and Their Role in Solid Tumors: An Update. Int J Mol Sci. 2024;25(6).

[35] Karar J, Maity A. PI3K/AKT/mTOR Pathway in Angiogenesis. Front Mol Neurosci. 2011;4:51.

[36] Naor Z. Signaling by G-protein-coupled receptor (GPCR): studies on the GnRH receptor. Front Neuroendocrinol. 2009;30(1):10–29.

[37] Bliss SP, Navratil AM, Xie J, Roberson MS. GnRH signaling, the gonadotrope and endocrine control of fertility. Front Neuroendocrinol. 2010;31(3):322–40.

[38] Huang F, Liu Q, Wang H, Zou Y. Effect of GnRH II and GnRH I on secretion of VEGF by eutopic and ectopic endometrial stromal cells of endometriosis patients. Zhong nan da xue xue bao Yi xue ban = J Cent South Univ Med Sci. 2010;35(5):409–18.

[39] Schultz KAP, Harris AK, Schneider DT, Young RH, Brown J, Gershenson DM, et al. Ovarian Sex Cord-Stromal Tumors. J Oncol Pract. 2016;12(10):940–6.

[40] Chen CP, Lu X. Gonadotropin-releasing hormone receptor inhibits triple-negative breast cancer proliferation and metastasis. J Int Med Res. 2022;50(3).

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