RESEARCH ARTICLE

Synergistic Potential of *Ecklonia cava* and Oral Contraceptives in Managing Polycystic Ovary Syndrome: Clinical and Biochemical Evaluation

Asmaa Najm Abd[1], Ahmed R Abu-Raghif [2], Lubna Amer Al-Anbari[3], Hayder Ridha-Salman[4*]

[1]*Diyala University, College of Medicine, Department of Pharmacology, Diyala, Iraq*
[2]*Al-Nahrain University, College of Medicine, Department of Pharmacology, Baghdad, Iraq*
[3]*High Institute for Infertility Diagnosis and Assisted Reproductive Technologies (ART), Al-Nahrain University, Baghdad, Iraq*
[4]*Al-Mustaqbal University, College of Pharmacy, Hillah-51001, Babylon, Iraq*

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ABSTRACT

Background: Polycystic Ovary Syndrome (PCOS) is a complex endocrine condition affecting women of reproductive age, influencing the endocrine, reproductive system, and metabolic processes.

Objectives: To evaluate the anti-inflammatory and hormonal modulation effects of *Ecklonia cava* in PCOS patients.

Methods: In this Randomized controlled study, 50 patients were diagnosed with polycystic ovary syndrome. Their age range (18-40 years) was equally allocated into two groups. The Control group received Diane- 35 (2mg cyproterone acetate/35 Mg ethinyl estradiol) once daily for 3 cycles, and the interventional group received a combination of Diane- 35 plus 300 mg of *Ecklonia cava* once daily for 3 months. Parameters assessed before and after treatments course include: serum LH and FSH, oLH/FSH ratio, free testosterone, leptin HBA1c, IL-8, IL-10.

Results: After 3 months, there was a significant reduction in the weight and waist circumference in the study group in comparison with the control group, with a p-value ≤0.05; significant improvement in LH, FSH, LH/FSH, Free testosterone and leptin hormone, in addition significant reduction in IL-8, and increase IL-10 level.

Conclusions: The addition of Eckonia *cava* to the Diane- 35 in the treatment of PCOS patients gives promising synergistic effects in the regulation of clinical and biochemical parameters in PCOS. Patient

Keywords: Polycystic ovary syndrome, *Ecklonia cava*, Obesity, Hba1c, Free Testosterone, IL-8, IL-10.

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INTRODUCTION

Polycystic ovary syndrome (PCOS) is a prevalent endocrine condition among women, with an incidence ranging from 5% to 21% throughout reproductive years[1]. In females with polycystic ovary syndrome, an elevated level of the luteinizing hormone causes excess production of androgens from the ovarian thecal cell, whereas a decrease in follicle-stimulating hormone causes follicular arrest, polycystic ovarian morphology, and oligo-ovulation[2]. Increased androgen production secretion in PCOS has an adverse effect on folliculogenesis. Increasing androgens in the early stage of gonadotropin-independent stimulation stimulates the production of primordial follicles and increases the number of small antral follicles[3]. Hyperandrogenism, clinically manifested by the presence of hirsutism, acne, and/or androgenic alopecia, the syndrome accounts for 70 – 80 % of hirsutism in females[4,5]. Androgenetic alopecia (AGA) is the most prevalent form of progressive hair loss disorder; in women, Androgenic alopecia is associated with a greater risk of PCOS. This gives a potential link with insulin resistance, obesity, and metabolic syndrome[6,7]. Obesity, menstrual irregularity, and hyperandrogenism have been identified as the primary clinical predictors of increased metabolic burden[8]. Therefore, screening for blood glucose levels is necessary for this syndrome. Glycosylated hemoglobin (HbA1c) levels are an important screening method for IR and DM since day-to-day plasma glucose levels do not influence them and reveal the plasma glucose status 2-3 months before measurement[9]. Obesity is regarded as a cause of low-grade chronic inflammation[10,11]. Visceral fat Accumulation leads to hypoxia and, consequently, necrosis, which in turn leads to the production of inflammatory cytokines[12,13]. Adipose tissue has an endocrine-like function and produces substances called adipokines or adipocytokines that secrete leptin. A higher concentration of leptin inhibits the expression of aromatase mRNA in granulosa cells, thus interrupting androgens' conversion to estrogen. In turn, it is

**Author for Correspondence: hayder80.ridha@gmail.com*

thought that increased leptin levels are related to the absence of folliculogenesis[14]. Physicians tend to use (combined) oral contraceptives, antiandrogen agents, insulin sensitizers, and ovulation inducers[15].

Combined oral contraceptives (COCs) remain the first-line pharmacological treatment, primarily to regulate menstruation and reduce androgen levels[16]. However, the long-term use of hormonal contraceptives may carry risks and side effects. Natural compounds with complementary mechanisms, such as *Ecklonia cava*, offer potential adjunctive benefits. *Ecklonia cava* contains Phlorotannin, a group of polyphenolic compounds unique to brown algae. These compounds exhibit antioxidant, anti-inflammatory, and anti-diabetic effects[15]. They may improve insulin sensitivity, reduce oxidative stress, and modulate estrogen receptor pathways-mechanisms relevant to PCOS pathophysiology[18].

METHODS

Study Design

This prospective randomized controlled study was conducted from March 2023 to December 2024) with 50 Females attending the High Institute for Infertility Diagnosis and Assistant Reproductive Technology. Their age range (18-40 years) suffering from polycystic ovary syndrome, Diagnosed with Polycystic ovary syndrome based on the Rotterdam criteria[19]. Participants were included not pregnant or lactating or had taken hormonal medications within the last six months, and all participants were overweight or obese. Participants with comorbidities such as diabetes mellitus, thyroid disorders, or cardiovascular diseases will be excluded. Ethical approval was obtained from the Institutional Review Board (IRB) of Diyala College of Medicine], and informed consent was obtained from all participants.

Intervention

Eligible participants will be randomly assigned into two groups: (Group1=25): Received Diane-35 from the 2nd day of the menstrual cycle for 21 days for three consecutive cycles, (Group2 = 25): Received a combination of Diane-35 plus *Ecklonia cava* extract 300 mg patients) for 3 months. The dose *of Ecklonia cava* extract is based on prior clinical studies demonstrating efficacy safety[20,21]. Diane-35 was standardized and supplied by [Bayer Schering Pharma, Germany]**.** *Ecklonia cava* extract 300 mg capsule was standardized and supplied by [Bio pure, USA].

Outcome Measures

Primary outcomes included changes in menstrual regularity, body mass index (BMI), and waist circumference assessed via the modified Ferriman-Gallwey score, which evaluates terminal hair growth in nine body areas, with scores ranging from 0 (no hair) to 4 (extensive hair), as originally described by Ferriman and Gallwey[22]. Secondary outcome: hormonal profile: LH, FSH, LH/FSH, free testosterone, leptin hormone, inflammatory markers: IL-8, IL-10, HbA1c, and adverse effects of supplementation.

Statistical Analysis

By using Microsoft Excel 365 and SPSS version 26. The results were explored as mean ±SD of each variable. A comparison between the groups was made using Wilcoxon test, Mann- Whitney U test. Significance was expressed as a value of <0.05 for each parameter.

RESULTS

Effects of Ecklonia cava on Body Mass Index (BMI)

Within group Comparison; Diane- 35 treated group (G1) show a statistically highly significant increase in BMI mean after treatment, P<0.001, While; In (Diane-35 + *Ecklonia cava*) treated group(G2) show Highly significant reduction in BMI P<0.001. Significant difference in BMI after treatment between two groups P <0.01, Table 1.

Effects of Ecklonia cava on Waist Circumference

Within group Comparison; Diane- 35 treated group (G1) show Significant increase in waist Circumference, p <0.01**.** While; In (Diane-35 + *Ecklonia cava*) treated group (G2) show significant decrease in Waist circumference, p <0.01. Significant difference in waist circumference after treatment between two groups. P <0.001, Table 2.

Effects of Ecklonia cava on Hirsutism

Within group Comparison; Diane- 35 treated group (G1) show highly significant reduction in hirsutism score. In (Diane-35 + *Ecklonia cava*) treated group (G2) show highly significant reduction in hirsutism score P<0.001**.** In Comparison between the Two groups; there are a statistically significant difference, P<0.049, Table 3

Effect of Ecklonia cava on Menstrual Regularity

Statistically significant improvement in menstrual cycle regularity demonstrated in both groups, and significant difference demonstrated between groups p- value 0.028.

Effects of Ecklonia cava on LH

Both groups showed significant reductions in LH after treatment (p < 0.0001).

No significant difference between the two groups p- value > 0.05

Effect of Ecklonia cava on Follicular Stimulating Hormone

Within group Comparison; Diane- 35 treated group (G1) show highly significant increase in FSH level after treatment

In (Diane-35 + *Ecklonia cava*) treated group (G2) show highly significant reduction in FSH level P<0.001, In Comparison between the two groups; there are a statistically significant difference, P<0.001, Table 6

Effects of Ecklonia cava on LH/FSH Ratio

Within group Comparison; Diane- 35 treated group (G1) show highly significant reduction in LH/FSH ratio after treatment. In (Diane-35 + *Ecklonia cava*) treated group(G2) show highly significant reduction in LH/FSH ratio P<0.001 In Comparison Between the Two groups; there are NO statistically significant difference, P- value > 0.05, Table: 7

Effects of Ecklonia cava on Free Testosterone

Within group Comparison; Diane- 35 treated group (G1) show highly significant reduction in Free testosterone level after treatment, in (Diane-35 + *Ecklonia cava*) treated group(G2) show highly significant reduction in Free testosterone level P<0.001.

In Comparison between the two groups; there are statistically significant difference, P- value < 0.001, (Table 8)

Effects of Ecklonia cava on Leptin Hormone

Table 1: Assessment of BMI

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
BMI kg/m²	Diane-35 (G1)	27.53 ± 1.17	28.47 ± 1.18	-0.94 kg/m²	0.001	p < 0.01
	Diane-35 plus *Ecklonia cava* (G2)	27.84 ± 1.42	25.88 ± 1.12	1.97 kg/m²	0.001	

Wilcoxon Test, Mann-Whitney U Test p-value: significant < 0.05, highly significant < 0.001, Very highly significant <0.0001

Table 2: Assessment of waist circumference

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
Waist circum-ference	Diane-35 (G1)	85.44 ± 1.61 cm	86.5 ± 1.44 cm	+1.06 *(increase)*	0.01	0.01
	Diane-35 plus *Ecklonia cava* (G2)	87.32 ± 3.41 cm	81.92 ± 2.66 cm	-5.40 *(largest decrease)*	0.01	

Wilcoxon Test, Mann-Whitney U Test p-value: significant < 0.05, highly significant < 0.001.

Table 3: Assessment of hirsutism

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
Hirsutism	Diane-35 (G1)	10.24 ± 1.96	9.36 ± 1.82	0.88	0.001	N.S
	Diane-35 plus *Ecklonia cava* (G2)	9.88 ± 1.59	9.08 ± 1.19	0.8	0.001	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, p-value: significant < 0.05, highly significant < 0.001.

Table 4: Assessment of Menstrual Regularity

Group	Regularity (Before)	Regularity (After)	Change percentages	p-value Within group	P-value Between groups
Diane-35 (G1)	0.25	1.00	+0.75	0.028	N.S.
Diane-35 plus *Ecklonia cava* (G2)	0.28	1.00	+0.72	0.028	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, p-value: significant < 0.05, highly significant < 0.001, N.S (Non-significant).

Within group Comparison; Diane- 35 treated group (G1) show highly significant reduction in leptin hormone after treatment.

In (Diane-35 + *Ecklonia cava*) treated group(G2) show highly significant reduction in leptin hormone after treatment P<0.001.

In Comparison Between the Two groups; there are statistically significant difference, P- value < 0.001, (Table 9)

Effects of Ecklonia cava on Glycated Hemoglobin (HbA1c)

Statistically significant increase in HbA1c levels demonstrated in Diane-35(G1) after treatment < 0.001).

Highly Significant reduction in HbA1c in (Diane-35 + *Ecklonia cava*) treated group(G2) (p = 0.001), Table 10.

Effects of Ecklonia cava on Interleukin-8 (IL-8)

No statistically significant change in interleukin 8 level in Diane- 35 treated group (G1) after treatment p- value > 0.05.

In (Diane-35 + *Ecklonia cava*) treated group(G2) show highly significant reduction in interleukin-8

Effect of Ecklonia cava on Interleukin-10 (IL-10)

Significant statistical difference demonstrated in both groups, p value < 0.05

No significant differences demonstrated between groups, (Table 12)

Impact of Ecklonia cava on Side Effects

No statistically significant differences in the incidence of side effects between the Diane-35 treated group (G1) and the Diane-35 plus *Ecklonia cava* group (G2) (p > 0.05). (Table 13)

DISCUSSION

This randomized, controlled clinical trial demonstrated that *Ecklonia cava* supplementation for three months along with oral contraceptives showed significant weight and waist circumference reduction demonstrated in the women who received *Ecklonia cava* extract along with hormonal therapy in comparison to women who received only hormonal therapy. These results are consistent with other clinical studies[23]. These results suggest that *Ecklonia cava* may have beneficial effects in controlling obesity[24], which is regarded as an increasing risk of cardiovascular diseases such as hypertension, diabetes, dyslipidemia, and nonalcoholic fatty liver disease[25,26].

Regarding hirsutism, a significant reduction in hirsutism scores following intervention in both groups, (p = 0.001), but there is no statistically significant difference in hirsutism score changes between groups, suggesting that the addition of *Ecklonia cava* did not produce an additive benefit beyond Diane-35 alone. Diane-35, combining cyproterone acetate and ethinylestradiol, effectively reduces androgen levels and improves clinical hyperandrogenism in PCOS patients[27,28]. *Ecklonia cava*, a

Table 5: Assessment of Luteinizing hormone

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
LH (m. IU/ml)	Diane-35 (G1)	13.28 ± 2.26	11.16 ± 1.82	-2.12	0.0001	N.S
	Diane-35 plus *Ecklonia cava* (G2)	13.84 ± 2.71	11.54 ± 1.89	-2.3	0.0001	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001, N.S (Non-significant)

Table 6: Assessment of Follicular Stimulating Hormone

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
FSH m. IU/ml	Diane-35 (G1)	5.88 ± 1.07	7.92 ± 12.41	+2.04	0.0042	0.001
	Diane-35 plus *Ecklonia cava* (G2)	6.63 ± 1.77	6.25 ± 1.87	-0.38	0.0001	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001

Table 7: Assessment of LH/ FSH ratio in the study groups

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
LH/ FSH ratio	Diane-35 (G1)	2.27 ± 0.17	1.97 ± 1.97	-0.29	0.0001	0.559 N.S.
	Diane-35 plus *Ecklonia cava* (G2)	2.16 ± 0.4	1.96 ± 1.96	-0.19	0.0006	

Wilcoxon Test, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001, N.S. (Non-significant)

Table 8: Assessment of Free Testosterone

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
Free Testo Pg/ml	Diane-35 (G1)	1.75 ± 0.27	1.48 ± 0.27	-0.27	< 0.001	p = 0.0001
	Diane-35 plus *Ecklonia cava* (G2)	1.83 ± 0.13	1.74 ± 0.12	-0.1	<0.001	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001

brown seaweed rich in polyphenolic compounds like phlorotannins, has shown antioxidant, anti-inflammatory, and metabolic regulatory properties in preclinical studies[29,30].

However, its direct effects on androgen-related symptoms such as hirsutism remain unclear, which might explain the lack of additional clinical improvement observed.

Moreover, the pathogenesis of hirsutism is multifactorial, involving androgen excess, receptor sensitivity, and peripheral metabolism, factors that may not be fully modulated by antioxidant supplementation alone[31,32].

Previous clinical trials combining conventional treatments with natural supplements have reported mixed results, reflecting t the need for larger, longer-duration studies to clarify potential synergistic effects[33,34].

Both treatment groups showed significant improvement in menstrual regularity after intervention p-value of 0.028. Although both groups reached complete menstrual regularity at the end of the study, the difference in improvement between groups was not statistically significant. These findings indicate that Diane-35 effectively restores menstrual cyclicity in women with PCOS, consistent with its established role in suppressing androgen excess and normalizing hypothalamic-pituitary-ovarian axis function[28,35]. The addition of *Ecklonia cava* showed a similar degree of improvement, suggesting it may contribute to supporting menstrual regularity, potentially through its antioxidant and metabolic effects; however, the current data do not demonstrate a clear additive benefit beyond Diane-35 alone. Given the complexity of menstrual regulation in PCOS and the promising but limited evidence on *Ecklonia cava*'s clinical effects, further large-scale and longer-term studies are suggested to explore its potential synergistic role in combination therapies [36,37].

In the present comparison LH, both treatment groups, Diane-35 alone and Diane-35 combined with *Ecklonia cava* led to a statistically significant reduction in LH levels (p = 0.0001 within groups). The mean LH reduction was slightly greater in the combination group compared to Diane-35 alone, although this difference was not statistically significant, which contradicts previous results obtained from animal studies[38] but still there is a trend toward reduction.

Regarding FSH hormone, significant increase in FSH in the Diane-35 group which may reflect a compensatory pituitary response to suppressed LH and androgen levels, possibly restoring aspects of normal feedback signaling from the hypothalamic-pituitary-ovarian (HPO) axis[39]. Oral contraceptives like Diane-35 have been shown to modulate gonadotropin secretion, although their primary effect is on LH suppression[40]. Some evidence suggests that low

Table 9: Assessment of Leptin hormone in the study groups

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
Leptin hormone ng/ml	Diane-35 (G1)	3.75 ± 0.57	3.51 ± 0.61	-0.23	0.001	N.S.
	Diane-35 plus *Ecklonia cava* (G2)	3.38 ± 0.65	3.22 ± 0.65	-0.16	0.0015	

Table 10: Assessment of HbA1c

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
HbA1c%	Diane-35 (G1)	5.92 ± 0.72	6.25 ± 0.72	0.33	$p < 0.001$	$p = 0.001$
	Diane-35 plus *Ecklonia cava* (G2)	5.52 ± 0.71	4.98 ± 0.63	-0.54	$p < 0.001$	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant

Table 11: Assessment of IL-8

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
IL-8 ng/ ml	Diane-35 (G1)	86.71 ± 10.75	87.76 ± 13.45	1.05	0.871	0.0001
	Diane-35 plus *Ecklonia cava* (G2)	89.3 ± 9.69	88.87 ± 22.86	-0.43	0.0004	

Wilcoxon Test, Mann-Whitney U Test with Bonferroni correction, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001.

Table 12: Assessment of IL-10 in the study groups

Parameter	Group	Mean ± SD Before	Mean ± SD After	Mean Change (Δ)	p-value Within group	p-value Between groups
IL-10 ng/ ml	Diane-35 (G1)	46.68 ± 7.12	49.7 ± 5.9	3.02	0.001	0.258
	Diane-35 plus *Ecklonia cava* (G2)	45.17 ± 4.25	46.85 ± 4.62	1.68	0.001	

Wilcoxon Test, Kruskal-Wallis Test, p-value: significant < 0.05, highly significant < 0.001.

estrogen doses in combined pills can mildly elevate FSH, supporting follicular growth indirectly[41].

In the group receiving Diane-35 with *Ecklonia cava* demonstrated a significant reduction in FSH. While *Ecklonia cava* is known for its antioxidant, anti-inflammatory, and insulin-sensitizing properties[15,42], the mechanism through which it influences FSH levels is not fully established. It is hypothesized that its impact on systemic metabolic and endocrine modulation may lead to decreased stimulation of the pituitary, thereby reducing FSH secretion[43,44].

Given the statistically significant difference between groups, it is suggested that *Ecklonia cava* may exert a modulatory effect on gonadotropin dynamics, possibly through enhancing insulin sensitivity, reducing inflammation. A statistically significant reduction in LH/FSH ratio after treatment demonstrated in both groups, confirming the effect of Diane-35 on downregulating LH secretion via hypothalamic negative feedback mechanisms However, the difference between-groups was not statistically significant, but still there is a trend toward LH/FSH ratio reduction.

Regarding free testosterone, a statistically significant reduction in free testosterone levels demonstrated in Both groups demonstrated after treatment. However, the magnitude of reduction was much greater in the Diane-35-only group compared to the combination group with a significant difference between-group (*p* = 0.0001), Diane-35 contains cyproterone acetate, a potent anti-androgen that blocks androgen receptors and inhibits ovarian androgen production by suppressing LH secretion, Therefore, the significant decrease in free testosterone in Group G1 is consistent with its known mechanism of action of Diane-35.

In contrast, the attenuated reduction in free testosterone in combination group suggests that *Ecklonia cava* might partially counteract the anti-androgenic effects of Diane-35. This interpretation is supported by preclinical studies showing that *Ecklonia cava* may increase testosterone in male animal models through stimulation of Leydig cell function and androgen biosynthesis[45,46].

Although *Ecklonia cava* has antioxidant and metabolic benefits, some of its components (phlorotannin) have shown testosterone-boosting activity in male animals, which may not be beneficial in PCOS patients where androgen excess is pathogenic[42].

These conflicting results are due to the higher dose of *Ecklonia cava* used in animal studies to elucidate such normalization regarding LH, FSH, and free testosterone. Endocrine changes, particularly in LH and free testosterone, often require prolonged treatment to become measurable. A treatment duration of 8–12 weeks may be too short for *Ecklonia cava* to demonstrate significant hormonal modulation, especially compared to medications like combined oral contraceptives or anti-androgenic drugs that show more immediate effects.

Diane-35 is not primarily known for leptin modulation, but the small yet significant reduction in leptin levels may be an indirect consequence of improved insulin sensitivity, reduced inflammation, or mild effects on adipocyte function[47,48]. Additionally, Diane-35's anti-androgenic activity may enhance leptin sensitivity and ovarian feedback signaling[49].

Ecklonia cava is rich in phlorotannin, which possess strong anti-inflammatory, antioxidant, and insulin-sensitizing properties[50,51]. These mechanisms likely contribute to the further reduction in leptin seen in Group 2. Although the mean change (–0.16 ng/mL) is slightly smaller than in Group 1, both are statistically significant.

Although there was no significant difference in -group comparison, it's not possible to conclude superiority of either intervention in lowering leptin., the combination may offer complementary metabolic benefits.

HbA1c reflects average blood glucose levels over the past 2–3 months and is a reliable marker of insulin sensitivity and glycemic control[52]. In PCOS, insulin resistance and compensatory hyperinsulinemia are common, even in non-diabetic women[53].

The significant rise in HbA1c in Group 1 supports these concerns and highlights a potential metabolic drawback of using Diane-35 alone in PCOS patients, particularly those with prediabetic tendencies.

In contrast, the group receiving Diane-35 with *Ecklonia cava* showed a significant reduction in HbA1c, suggesting a glycemic-protective effect of the algae extract.

Ecklonia cava contains phlorotannin, which have been demonstrated to improve glucose uptake, enhance insulin sensitivity and inhibit α-glucosidase and DPP-4 enzymes, helping reduce postprandial glucose spikes[51,54].

Interleukin-8 (IL-8) is a pro-inflammatory cytokine involved in neutrophil activation and chemotaxis[53,54]. In PCOS, chronic low-grade inflammation contributes to insulin resistance, anovulation, and hyperandrogenism, and IL-8 levels are often found to be elevated compared to healthy controls[57,58].

Diane-35 did not significantly alter IL-8 levels ($p = 0.871$). This aligns with literature indicating that oral contraceptives may not consistently improve inflammatory markers, and may sometimes even worsen inflammation due to hepatic synthesis of acute-phase proteins[59,60].

In contrast, the addition of *Ecklonia cava* led to a statistically significant reduction in IL-8 ($p = 0.0004$). While the absolute change (–0.43 ng/mL) appears small, the large increase in standard deviation post-treatment might indicate individual variability in response to the anti-inflammatory component.

Ecklonia cava is rich in phlorotannin, which inhibits NF-κB activation, reducing the transcription of pro-inflammatory cytokines including IL-8 (5); scavenges ROS, which are upstream inducers of IL-8; and suppresses macrophage and endothelial cell IL-8 secretion, as shown in in vitro and animal models[61].

These mechanisms support the observed anti-inflammatory effect of *Ecklonia cava* in reducing IL-8, and suggest it may help alleviate the chronic low-grade inflammation seen in PCOS.

Table 13: Side Effects

Side effects	Diane-35 treated group G1	Diane-35 plus *Ecklonia cava* Treated group G2	p-value Between groups
Nausea	16%	16%	N. S.
Headache	20%	24%	
Mood Swings	24%	24%	
Abdominal bloating	4%	8%	
Dizziness	12%	8%	
Menstrual irregularities (spotting)	12%	8%	
Breast Tenderness	12%	12%	

L-10 is an anti-inflammatory cytokine that plays a crucial role in suppressing the production of pro-inflammatory cytokines (e.g., IL-6, TNF-α, IL-8) and regulating immune response and protecting tissue from inflammatory damage[62-64]. Women with PCOS typically exhibit lower or dysregulated IL-10 levels, reflecting a state of chronic low-grade inflammation and immune imbalance[65,66].

Both groups showed statistically significant increases in IL-10 levels post-treatment ($p = 0.001$), but the increase was greater in the Diane-35 treated group than in the combination treated group).

No significant difference was found between groups ($p = 0.258$).

Regarding side effects profile, no significant difference between the two groups, the absence of significant differences in side effect profiles between the Diane-35 and Diane-35 plus *Ecklonia cava* groups suggests that the addition of *Ecklonia cava* does not notably alter the tolerability or adverse effect burden of Diane-35 therapy. While some variations were observed in individual symptoms such as dizziness and abdominal bloating, these differences were not statistically meaningful and may be attributed to interindividual variability rather than a pharmacological interaction. These findings indicate that *Ecklonia cava* may be safely co-administered with Diane-35 without increasing the risk of common side effects.

CONCLUSION

Ecklonia cava supplementation appears to improve multiple metabolic and endocrine parameters in PCOS, supporting its potential as a natural therapeutic agent. Incorporation of *Ecklonia cava* in PCOS management protocols may enhance patient outcomes by managing weight, insulin resistance, leptin hormone, and perhaps leptin resistance.

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Ethical approval

The study was conducted following the ethical principles originating in the Declaration of Helsinki. It was applied

with patients' verbal and analytical approval before engagement in the study. The study protocol was reviewed and approved by a local ethics committee, Diyala College of Medicine Code No. 2025 ANA 917.

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