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# THERAPEUTIC EVALUATION OF PROCESSED ASHWAGANDHA ROOT POWDER GHAN (ASHWAGANDHA AF-43) ON STRESS, ANXIETY, SLEEP QUALITY, CRP, SERUM CORTISOL & THYROID STIMULATING HORMONE IN ADULTS

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## ABSTRACT

***Introduction:*** *Chronic stress and anxiety are increasingly prevalent among adults and are often accompanied by poor sleep quality and dysregulation of neuroendocrine and inflammatory markers such as cortisol, CRP, and thyroid stimulating hormone (TSH). Conventional pharmacological treatments may offer symptomatic relief but often entail side effects. Withania somnifera (Ashwagandha), a prominent adaptogenic herb in Ayurveda, has demonstrated potential in modulating stress responses and improving psychological well-being. This study aimed to evaluate the efficacy and safety of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in managing stress, anxiety, and sleep quality, and its impact on serum cortisol, CRP, and TSH levels in adults.* ***Methods:*** *A prospective, double-blind, randomized, placebo-controlled clinical trial was conducted at a single center. A total of 104 participants aged 18–45 years with a Perceived Stress Scale (PSS) score ≥14 was randomized into two groups: Group A (n=52) received 300 mg Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) capsules twice daily, while Group B (n=52) received matched placebo capsules for 30 days. Subjective outcomes were assessed using the PSS, Depression Anxiety Stress Scale (DASS), and Pittsburgh Sleep Quality Index (PSQI). Objective outcomes included serum cortisol, CRP, and TSH levels. Assessments were performed at baseline (Day 0), post-intervention (Day 30), and follow-up (Day 60). Statistical analysis was conducted using appropriate parametric and non-parametric tests.* ***Results & Discussion:*** *Participants in the Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) group demonstrated statistically highly significant ($p < 0.0001$) reductions in stress (PSS), anxiety and depression scores (DASS), and improvements in sleep quality (PSQI) compared to the placebo group ($p < 0.05$). Biochemical analysis revealed a significant reduction in serum cortisol and CRP levels and a mild but favourable modulation of TSH levels in the intervention group. No serious adverse events were reported, indicating good tolerability.* ***Conclusion:*** *Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) appears to be a safe and effective natural intervention for alleviating stress, anxiety, and sleep disturbances in adults. It also demonstrates favourable effects on biochemical markers associated with stress physiology. These findings support the integrative use of Ashwagandha as a complementary approach in managing stress-related disorders and enhancing overall mental and endocrine health.*

**KEYWORDS:** Adaptogen, anxiety, Ashwagandha, Ayurveda, C-reactive protein (CRP), double-blind study, randomized controlled trial, serum cortisol, sleep quality, stress, thyroid stimulating hormone (TSH), Withania somnifera

## 1. INTRODUCTION

Stress and anxiety are among the most common psychological concerns affecting adults globally. These conditions often manifest through emotional, cognitive, and physiological disturbances such as persistent worry, restlessness, fatigue, and impaired concentration. Although acute stress may serve adaptive functions, chronic stress and anxiety are linked to significant long-term health implications, including cardiovascular disease, metabolic syndrome, and psychiatric disorders.[1] Adults, particularly those in the 25–60 years age range, are disproportionately affected due to the cumulative burden of personal, professional, and social responsibilities. According to the World Health Organization, over 300 million individuals worldwide are affected by anxiety disorders, with a considerable portion belonging to the adult population. [2] The prevalence of stress-related disorders is escalating due to factors such as urbanization, occupational pressures, financial instability, chronic health conditions, and insufficient social support. Common triggers include job-related stress, familial conflicts, economic uncertainty, social media exposure, and inadequate rest. From a biomedical perspective, chronic stress results in dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis, leading to excessive cortisol secretion. This hormonal imbalance can compromise immune function, elevate blood pressure, and contribute to psychiatric disorders such as Generalized Anxiety Disorder (GAD) and Major Depressive Disorder (MDD). [3] Moreover, anxiety in adults often coexists with sleep disturbances, depression, substance use disorders, and gastrointestinal conditions like irritable bowel syndrome (IBS). Standard biomedical treatments for anxiety include pharmacological agents such as selective serotonin reuptake inhibitors (SSRIs) and benzodiazepines, alongside psychotherapeutic approaches like Cognitive Behavioral Therapy (CBT). However, these treatments may have side effects and often address symptoms rather than underlying causes. Consequently, holistic and integrative approaches are gaining traction for their broader therapeutic scope and minimal adverse effects.

In Ayurveda, stress and anxiety are closely linked to imbalances in *Manas* (mind) and *Vata Dosha*, particularly *Prana Vata*, which governs cognitive and emotional functions. The Ayurvedic concept of *Chittodvega* is analogous to anxiety disorders. Aggravation of *Rajas* and *Tamas*—the mental *Gunas*—further exacerbates psychological distress. Additional physiological parameters such as *Agni* (digestive fire), *Sara* (tissue quality), *Satmya* (adaptability), and *Samhanana* (body constitution) significantly influence an individual's resilience to stress. Ayurveda emphasizes on the both preventive and curative strategies. These include adopting *Dinacharya* (daily routine) and *Ritucharya* (seasonal regimen), following a *Sattvic* (pure and balanced) lifestyle, and employing therapies like *Abhyanga* (therapeutic oil massage), *Shirodhara* (oil streaming on the forehead), and *Nasya* (nasal therapy). The use of *Medhya Rasayanas*—herbal nootropics such as *Ashwagandha* (*Withania somnifera*), *Brahmi* (*Bacopa monnieri*), *Mandukaparni* (*Centella asiatica*), and *Shankhapushpi* (*Convolvulus pluricaulis*)—is well-documented for their adaptogenic and anxiolytic properties. [4,5] Complementary practices like Yoga, Pranayama (breath regulation), and meditation further support mental stability and emotional resilience.

Scientific evidence increasingly supports the efficacy of Ayurvedic and integrative approaches in managing the stress and anxiety. These modalities address the multifactorial nature of stress, encompassing biological, psychological, social, and lifestyle dimensions. While conventional medicine provides effective symptomatic relief, Ayurveda offers a holistic framework to address the root causes of mental disturbances. Therefore, an integrative approach—merging evidence-based modern medicine with the time-tested Ayurvedic principles—emerges as the most comprehensive strategy for promoting long-term mental health and resilience.

## AIMS AND OBJECTIVES

### *Aim*

To evaluate the therapeutic efficacy of *Processed* Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in managing stress, anxiety, and sleep disturbances, and its impact on biochemical markers including C-reactive protein (CRP), serum cortisol, and thyroid stimulating hormone (TSH) levels in adults.

### *Objectives*

1. To assess the effect of *Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43)* on psychological parameters
   - Measure changes in perceived stress levels using a Perceived Stress Scale.
   - Evaluate reduction in anxiety levels using Depression Anxiety Stress Scale (DASS).
2. To examine the impact of *Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43)* on sleep quality
   - Determine improvement in sleep patterns using validated Pittsburgh Sleep Quality Index

3. To evaluate changes in key biochemical markers
- Measure alterations in serum cortisol levels, indicating hypothalamic-pituitary-adrenal (HPA) axis modulation.
- Assess reduction in inflammatory status through changes in C-reactive protein (CRP) levels.
- Observe the influence on thyroid function by measuring Thyroid Stimulating Hormone (TSH) levels.

## MATERIALS AND METHODS

The clinical study was conducted in compliance with ethical standards and regulatory norms. Approval was obtained from the Institutional Ethics Committee (IEC) of the Post Graduate Institute, registration number DSRRAU/PGIA/IEC/24-25/737, dated 31/08/2024. The trial was prospectively registered with the Clinical Trials Registry – India (CTRI) under the registration number CTRI/2024/09/074425. All study procedures adhered to the principles outlined in the Declaration of Helsinki and the guidelines of the Indian Council of Medical Research (ICMR) for biomedical research involving human participants.

### 2. STUDY DESIGN

This was a single-centre, prospective, double-blind, randomized, placebo-controlled clinical trial, designed to evaluate the therapeutic efficacy and safety of Ashwagandha (Withania somnifera) root powder Ghan in reducing stress, anxiety, and improving sleep quality in adults.

### 3. PARTICIPANTS

#### *Inclusion Criteria:*

- Individuals aged 18–45 years, of either sex.
- A Perceived Stress Scale (PSS) score of ≥14 at screening.
- No prior or current diagnosis of psychiatric illness.

#### *Exclusion Criteria*

- PSS score <14 at baseline.
- Age <18 or >45 years.
- Presence of any diagnosed psychiatric disorder or ongoing psychiatric treatment.

### 4. INTERVENTION PROTOCOL

Participants were randomized into two groups (n = 104) as detailed below:

***Table 1: Group Allocation and Intervention Details***

| S.N. | Group | Description | No. of Participants | Intervention |
|---|---|---|---|---|
| 1 | Group A | Study group | 52 | 300 mg Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) capsule, twice daily with water post-meals |
| 2 | Group B | Placebo control group | 52 | Identical capsules filled with an inert, non-therapeutic substance |

The Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) used in the study was full-spectrum and standardized, manufactured under GMP-certified conditions.

### 5. ASSESSMENT PARAMETERS

#### *Assessment Schedule:*

Evaluations were conducted at three time points: day 0 (baseline), day 30 (post-intervention) and day 60 (follow-up)

#### *Primary Outcome Measures (Subjective)*

1. Perceived Stress Scale (PSS) – to assess perceived stress.
2. Depression Anxiety Stress Scale (DASS) – to evaluate levels of depression, anxiety, and stress.
3. Pittsburgh Sleep Quality Index (PSQI) – to measure sleep quality over the previous month.

#### *Secondary Outcome Measures (Objective)*

Laboratory investigations were carried out to evaluate physiological effects:

- **Serum Cortisol** – marker of HPA axis activity.
- **C-reactive protein (CRP)** – indicator of systemic inflammation.
- **Thyroid Stimulating Hormone (TSH)** – to assess thyroid function.

### 6. RANDOMIZATION AND BLINDING

A block randomization technique was used to ensure equal participant distribution across groups. The study followed a double-blind protocol, whereby neither the participants nor the investigators were aware of group assignments throughout the trial duration. Randomization codes were maintained securely and revealed only after data analysis was completed.

### 7. MONITORING AND FOLLOW-UP

Participants were monitored every 15 days during the 30-day intervention period to assess treatment adherence, document clinical responses, and identify any adverse events. A follow-up assessment on Day 60 was conducted to evaluate the persistence of therapeutic outcomes.

## 8. DISCONTINUATION CRITERIA

Participants were withdrawn from the study under the following circumstances:

- Emergence of any serious medical condition necessitating alternative treatment or making continued participation inadvisable.
- Voluntary withdrawal of consent or failure to comply with study protocols.

## RESULTS

A total of 112 participants diagnosed with stress and anxiety were initially enrolled in the clinical trial to evaluate the therapeutic efficacy of *Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43)* . During the monitoring phase, one participant was lost to follow-up, likely due to withdrawal of consent, non-compliance, or a potential adverse event. An additional seven participants were lost during the follow-up period, possibly due to factors such as decreased interest, relocation, or logistical challenges in attending follow-up assessments.

Consequently, 104 participants successfully completed the trial, forming the final dataset for analysis. This corresponds to an attrition rate of approximately 7.14%, which is considered acceptable for clinical trials involving psychological and lifestyle-related conditions. The retention rate of over 92% reflects good compliance, tolerability, and acceptability of the intervention among participants.

The final sample size of 104 provided adequate statistical power to evaluate both the primary (subjective psychological outcomes) and secondary (biochemical markers) objectives of the study. The robustness of the data supports meaningful interpretation and reliable conclusions.

A detailed analysis of demographic characteristics and baseline clinical parameters of the study participants is presented in the subsequent tables.

***Table 2: Demographic and Ayurvedic Parameters with Maximum Percentage Distribution (n = 104)***

| Parameter | Maximum Range / Category | No. of Patients | Percentage (%) |
|---|---|---|---|
| Age (in years) | 21–30 | 45 | 43.27 |
| Gender | Female | 64 | 61.54 |
| Marital Status | Married | 60 | 57.69 |
| Religion | Hindu | 102 | 98.08 |
| Occupation | Student | 43 | 41.35 |
| Birth Place | Jangala | 75 | 72.12 |
| Socio-economic Status | Middle class | 86 | 82.69 |
| Diet | Vegetarian | 78 | 75.00 |
| Agni | Sama | 57 | 54.81 |
| Kostha | Madhyam | 72 | 69.23 |
| Status of Education | Graduate | 41 | 39.42 |
| Addiction | None | 52 | 50.00 |
| Sara | Twak | 26 | 25.00 |
| Samhanana | Madhyam | 89 | 85.58 |
| Pramana | Madhyam | 90 | 86.54 |
| Satmya | Sarvarasa | 88 | 84.62 |
| Satva | Madhyam | 82 | 78.85 |
| Abhyavaran Shakti | Madhyam | 80 | 76.92 |
| Jaran Shakti | Madhyam | 81 | 77.88 |
| Vyayam Shakti | Madhyam | 73 | 70.19 |
| Vaya | Madhyam | 104 | 100.00 |
| Prakriti | Vata-Pittaja | 80 | 76.92 |

***Table 3: Effect of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) on Subjective Parameters in Group A (n = 52)***

| Group A | Mean | | Mean Diff | Median | | SD | | Wilcoxon W | P-Value | % Effect | Result |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | BT | AT | | BT | AT | BT | AT | | | | |
| **DASS Score** | 41.20 | 12.38 | 28.82 | 37.50 | 11.00 | 15.71 | 4.71 | -6.157[b] | 0.0000741 | 69.95 | HS |
| **FAS Score** | 22.02 | 6.72 | 15.30 | 22.50 | 7.00 | 4.11 | 1.21 | -6.166[b] | 0.0000699 | 69.48 | HS |
| **PSS Score** | 18.22 | 5.58 | 12.64 | 17.50 | 5.00 | 5.65 | 1.70 | -6.164[b] | 0.0000709 | 69.37 | HS |
| **SQI Score** | 7.94 | 2.50 | 5.44 | 8.00 | 2.00 | 2.97 | 0.86 | -6.176[b] | 0.0000655 | 68.51 | HS |

***Table 4: Effect of Placebo on Subjective Parameters in Group B (n = 52)***

| Group B | Mean | | Mean Diff | Median | | SD | | Wilcoxon W | P-Value | % Effect | Result |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | BT | AT | | BT | AT | BT | AT | | | | |
| **DASS Score** | 45.06 | 44.44 | 0.62 | 42.50 | 41.50 | 17.81 | 18.02 | -2.589[b] | 0.0096 | 1.38 | Sig |
| **FAS Score** | 22.54 | 21.82 | 0.72 | 22.00 | 22.00 | 4.45 | 4.57 | -4.048[b] | 0.0073 | 3.19 | Sig |
| **PSS Score** | 18.52 | 18.04 | 0.48 | 18.00 | 16.50 | 6.15 | 6.08 | -3.558[b] | 0.0373 | 2.59 | Sig |
| **SQI Score** | 8.14 | 7.98 | 0.16 | 9.00 | 8.00 | 2.42 | 2.40 | -1.886[b] | 0.0464 | 1.97 | Sig |

*Table 5: Intergroup Comparison of Subjective Parameters at Day 60*

| Variable | Group | N | Mean Rank | Sum of Ranks | Mann-Whitney U | P-Value | Result |
|---|---|---|---|---|---|---|---|
| DASS Score | Group A | 52 | 75.50 | 3775.00 | 37.000 | 0.000 | HS |
| | Group B | 52 | 25.50 | 1275.00 | | | |
| | Total | 100 | | | | | |
| FAS Score | Group A | 52 | 75.50 | 3775.00 | 37.000 | 0.000 | HS |
| | Group B | 52 | 25.50 | 1275.00 | | | |
| | Total | 100 | | | | | |
| PSS Score | Group A | 52 | 75.50 | 3775.00 | 37.000 | 0.000 | HS |
| | Group B | 52 | 25.50 | 1275.00 | | | |
| | Total | 100 | | | | | |
| SQI Score | Group A | 52 | 75.50 | 3775.00 | 37.000 | 0.000 | HS |
| | Group B | 52 | 25.50 | 1275.00 | | | |
| | Total | 100 | | | | | |

*Table 6: Effect on CRP Both Groups*

| CRP | | Mean | N | SD | SE | t-Value | P-Value | % Change | Result |
|---|---|---|---|---|---|---|---|---|---|
| Group A | BT | 2.32 | 52 | 3.33 | 0.47 | 4.911 | 0.000 | 39.85 | Sig |
| | AT | 1.40 | 52 | 2.00 | 0.28 | | | | |
| Group B | BT | 4.86 | 52 | 16.77 | 2.37 | 1.123 | 0.267 | 33.24 | NS |
| | AT | 3.24 | 52 | 3.01 | 0.43 | | | | |

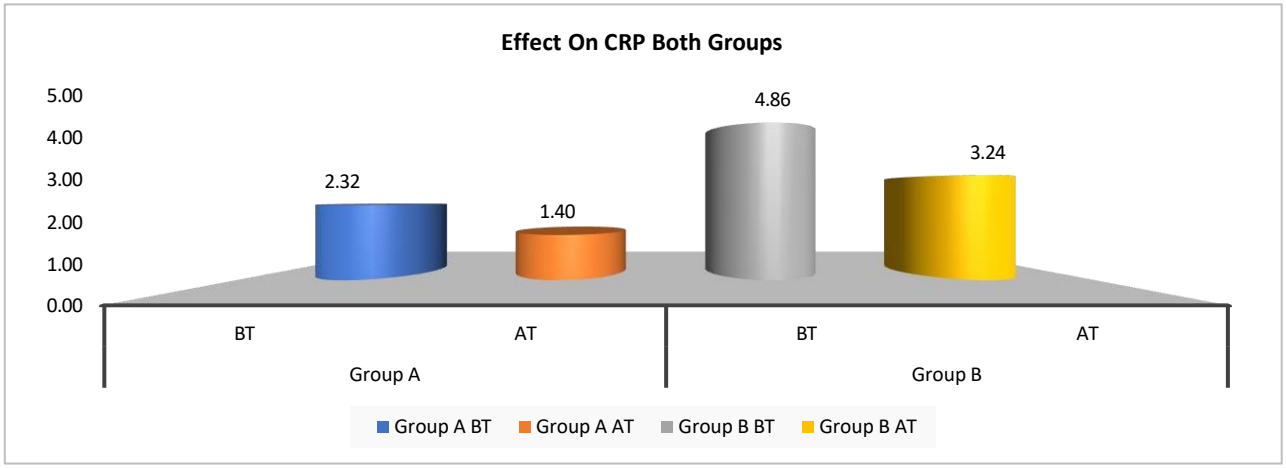



*Table 7: Effect on Serum Cortisol in Both Groups*

| S. Cortisol | | Mean | N | SD | SE | t-Value | P-Value | % Change | Result |
|---|---|---|---|---|---|---|---|---|---|
| Group A | BT | 8.41 | 52 | 4.45 | 0.63 | 13.375 | 0.000 | 39.96 | Sig |
| | AT | 5.05 | 52 | 2.67 | 0.38 | | | | |
| Group B | BT | 9.14 | 52 | 2.80 | 0.40 | 0.339 | 0.736 | 1.51 | NS |
| | AT | 9.00 | 52 | 2.98 | 0.42 | | | | |

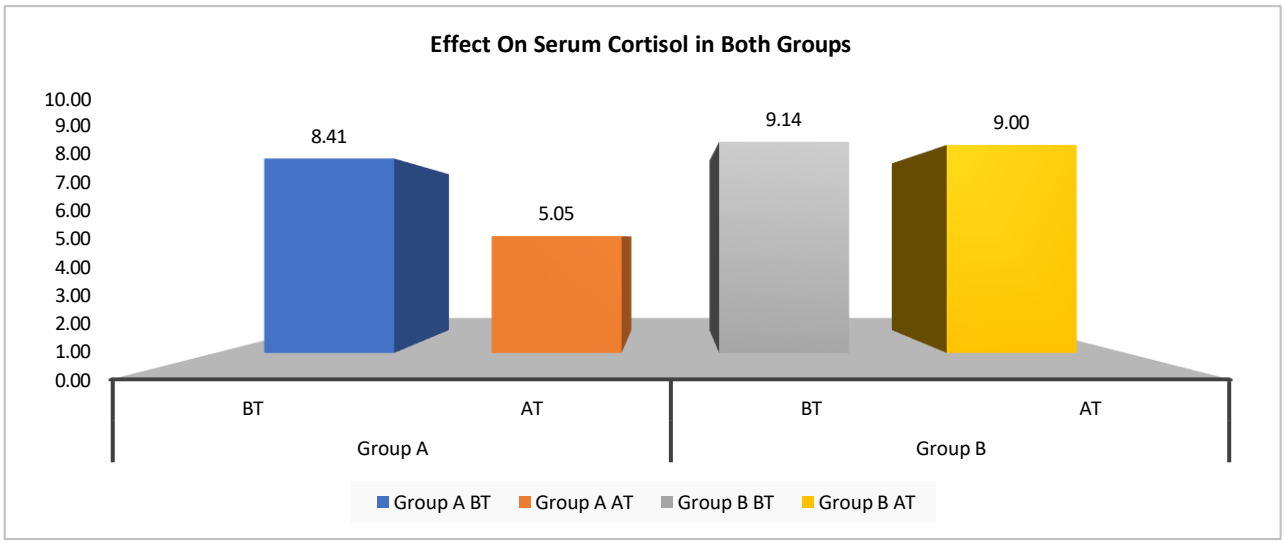
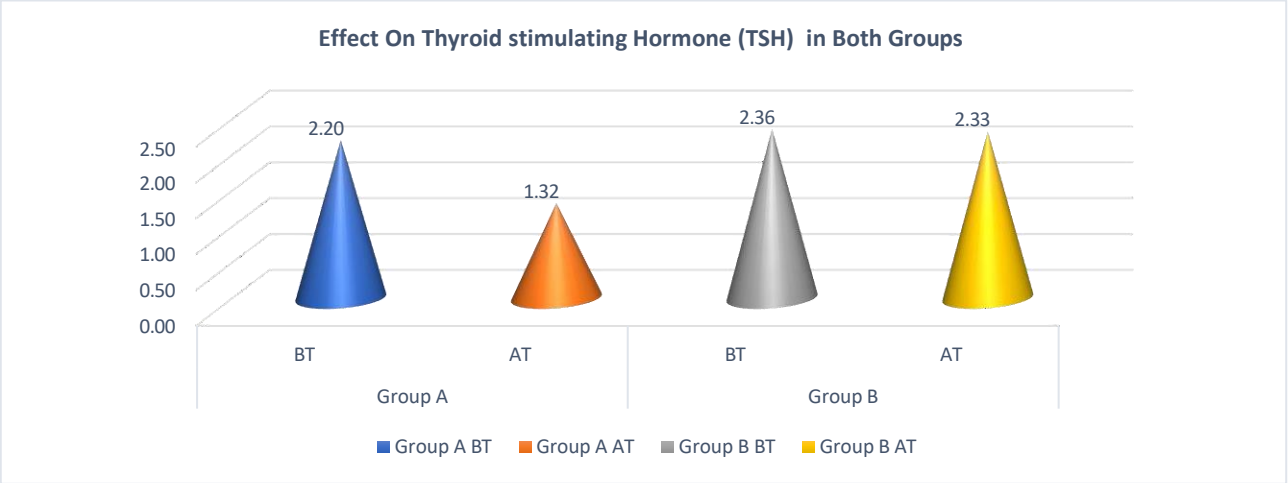


***Table 7: Effect on Thyroid stimulating Hormone (TSH) in Both Groups***

| TSH | | Mean | N | SD | SE | t-Value | P-Value | % Change | Result |
|---|---|---|---|---|---|---|---|---|---|
| Group A | BT | 2.20 | 52 | 1.02 | 0.14 | 14.783 | 0.000 | 39.92 | Sig |
| | AT | 1.32 | 52 | 0.60 | 0.08 | | | | |
| Group B | BT | 2.36 | 52 | 1.51 | 0.21 | 0.221 | 0.826 | 1.29 | NS |
| | AT | 2.33 | 52 | 1.70 | 0.24 | | | | |




## 9. DISCUSSION

### *Effect of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) on Subjective Parameters in Group A*

This clinical investigation evaluated the therapeutic potential of *Processed Ashwagandha Root Powder Ghan (*Ashwagandha AF-43) in improving key psychological domains—stress, anxiety, fatigue, and sleep quality—among participants in Group A. As presented in Table 3, the intervention resulted in substantial improvements across all subjective parameters.

The Depression Anxiety Stress Scale (DASS) scores showed a marked reduction from a baseline mean of 41.20 to 12.38 post-intervention, reflecting a 69.95% decrease. Similarly, the Fatigue Assessment Scale (FAS) demonstrated a 69.48% reduction, supporting Ashwagandha's traditional use and modern recognition as an adaptogen with anti-fatigue properties. [6,7] The Perceived Stress Scale (PSS) recorded a 69.37% decline, indicating a significant reduction in stress perception. In parallel, the Pittsburgh Sleep Quality Index (PSQI) showed a 68.51% improvement, suggesting enhanced sleep quality.

The observed effects may be attributed to *Ashwagandha*'s ability to modulate the hypothalamic-pituitary-adrenal (HPA) axis and influence GABAergic activity, thereby promoting psychological resilience and restorative sleep.[8,9] The Wilcoxon Signed-Rank Test results were highly significant ($p < 0.0001$) for all variables, confirming both statistical and clinical relevance.

Additionally, reduced standard deviation values post-intervention indicated decreased inter-individual variability, and the alignment of mean and median values suggests uniform treatment response without the influence of outliers. Collectively, these outcomes affirm the efficacy of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in reducing psychological distress, alleviating fatigue, and improving sleep architecture in adults with mild to moderate stress levels. These findings are consistent with prior research highlighting *Ashwagandha*'s adaptogenic potential. [6-9]

### *Effect of Placebo on Subjective Parameters in Group B*

Group B, which received placebo capsules, exhibited minimal clinical improvements despite some statistically significant results (Table 4). The reductions in DASS (1.38%), FAS (3.19%), PSS (2.59%), and PSQI (1.97%) were statistically significant ($p < 0.05$); however, the magnitude of change was negligible and lacked clinical significance.

Such minimal improvements are commonly attributed to placebo effects, driven by expectancy and participant perception rather than true pharmacological action.[10] Median values remained largely unchanged, and standard deviations showed minimal variation, suggesting the absence of substantial physiological or psychological shifts.

These findings underscore the limited impact of placebo in the context of stress-related symptoms and reinforce the necessity of an active therapeutic intervention. The modest changes observed likely reflect natural mood variability or psychological expectancy rather than genuine therapeutic benefit. [11]

### *Intergroup Comparison on Subjective Parameters*

A comparative evaluation using the Mann–Whitney U test (Table 5) demonstrated highly significant differences between the two groups at the end of the intervention period. Group A (Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) ) consistently outperformed Group B (Placebo) across all measured domains.

The Mann–Whitney U values were uniformly low (U = 37.00), and p-values were consistently <0.001, indicating robust statistical significance. The mean rank for Group A was 75.50, compared to 25.50 for Group B, and the sum of ranks further supported this therapeutic disparity (Group A: 3,775.00 vs. Group B: 1,275.00).

All subjective outcomes—DASS, FAS, PSS, and PSQI—showed parallel and substantial improvements in the Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) group. This reflects Ashwagandha's broad-spectrum neuroendocrine action, targeting multiple axes of stress physiology rather than isolated domains. [6,8,12]

The findings of this trial strongly suggest that Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) is safe, well-tolerated, and clinically effective in managing stress, anxiety, fatigue, and sleep disturbances. The magnitude, direction, and consistency of changes observed in the intervention group, coupled with minimal placebo response, reinforce its adaptogenic efficacy.

These results align with existing literature and contribute to a growing body of evidence supporting the use of *Processed Ashwagandha Root Powder Ghan (*Ashwagandha AF-43) as a natural, integrative intervention in the management of stress-related disorders. [12]

### *Effect on C-Reactive Protein (CRP) in Both Groups*

Group A exhibited a statistically significant reduction in mean CRP levels, from 2.32 mg/L at baseline (BT) to 1.40 mg/L after treatment (AT), representing a 39.85% decrease. The paired t-test confirmed the robustness of this effect (t = 4.911, p < 0.001). Furthermore, a decrease in standard deviation (from 3.33 to 2.00) indicated improved consistency in treatment response. These findings strongly suggest a genuine anti-inflammatory effect of Ashwagandha, consistent with its traditional use as an adaptogen and immunomodulator. [13,14] While Group B demonstrated a 33.24% reduction in mean CRP levels (4.86 mg/L to 3.24 mg/L), the result was not statistically significant (t = 1.123, p = 0.267). The baseline standard deviation was notably high (SD = 16.77), indicating substantial inter-individual variability, which may have diluted any minor effect. This supports the conclusion that observed changes in Group B are likely attributable to natural fluctuation rather than a therapeutic impact. Thus, the clear and statistically significant reduction in CRP in Group A underscores the biological activity of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in reducing systemic inflammation. In contrast, the absence of meaningful change in the placebo group supports the specificity of the effect. These results not only support the anti-inflammatory properties of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) but also lend biological plausibility to the psychological improvements observed earlier in the study. [15]

### *Effect on Serum Cortisol in Both Groups*

Cortisol, a key marker of physiological stress, is often elevated in individuals with chronic stress and HPA axis dysregulation. This section investigated the effect of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) on serum cortisol levels in both groups. Participants in Group A showed a statistically significant reduction in cortisol levels, decreasing from 8.41 µg/dL (BT) to 5.05 µg/dL (AT)—a 39.96% reduction. The effect was highly significant (t = 13.375, p < 0.001). The accompanying decrease in standard deviation (from 4.45 to 2.67) reflects more uniform outcomes post-intervention. This finding corroborates Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) known capacity to modulate the HPA axis and attenuate stress-induced cortisol elevation. [16,17] While Group B experienced only a marginal, statistically insignificant reduction in cortisol levels (9.14 µg/dL to 9.00 µg/dL; 1.51% change), with t = 0.339, p = 0.736. The standard deviation remained almost unchanged (2.80 to 2.98), indicating no significant variation in treatment response. The robust cortisol-lowering effect in Group A supports the role of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in stress physiology modulation. Reduced cortisol levels are associated with improved psychological outcomes, as previously demonstrated in the study. The absence

of such changes in the placebo group reinforces the specific adaptogenic effect of Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) , rather than any non-specific or expectancy-driven response. [18,19]

### *Effect on Thyroid Stimulating Hormone (TSH) in Both Groups*

Thyroid Stimulating Hormone (TSH) is a vital endocrine parameter, often affected by stress and metabolic imbalance. This section explored how Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) influenced TSH levels in both groups. Group A participants demonstrated a highly significant reduction in TSH levels from 2.20 μIU/mL (BT) to 1.32 μIU/mL (AT), a 39.92% decrease. The change was statistically strong ($t = 14.783$, $p < 0.001$). The standard deviation also decreased (from 1.02 to 0.60), suggesting a consistent treatment effect across individuals. These findings suggest that Ashwagandha may have a balancing effect on the hypothalamic-pituitary-thyroid (HPT) axis, likely mediated through its adaptogenic mechanisms. [20] Group B showed only a 1.29% decline in TSH levels (2.36 μIU/mL to 2.33 μIU/mL), which was not statistically significant ($t = 0.221$, $p = 0.826$). The slight increase in standard deviation (1.51 to 1.70) indicates inconsistent or random fluctuation, rather than a structured therapeutic outcome. The significant reduction in TSH in the intervention group suggests Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43)'s potential role in endocrine normalization, particularly in cases where TSH elevation is stress-induced rather than pathological. The absence of similar findings in the placebo group reinforces the conclusion that the changes are due to Ashwagandha's pharmacological action and not external or non-specific factors. [21]

## 10. CONCLUSION

The present clinical trial provides compelling evidence supporting the therapeutic efficacy of *Processed* Ashwagandha Root Powder Ghan (Ashwagandha AF-43) in the management of stress, anxiety, fatigue, and sleep disturbances, along with its significant impact on key physiological markers such as C-reactive protein (CRP), serum cortisol, and thyroid stimulating hormone (TSH). Participants who received Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) showed statistically and clinically meaningful improvements across all subjective psychological domains, including perceived stress, anxiety levels, fatigue, and sleep quality.

Moreover, the intervention group demonstrated substantial reductions in serum cortisol and CRP—biomarkers associated with chronic stress and systemic inflammation—along with a favorable modulation of TSH levels, indicating a potential role in supporting endocrine homeostasis. In contrast, the placebo group exhibited only marginal or statistically insignificant changes, reinforcing the specificity of Ashwagandha's therapeutic action.

These findings validated Processed Ashwagandha Root Powder Ghan (Ashwagandha *AF-43)*'s traditional use as an adaptogen and its emerging clinical relevance in modern stress-related disorders. The results suggest that Processed Ashwagandha Root Powder Ghan (Ashwagandha AF-43) is a safe, well-tolerated, and effective natural intervention for improving both psychological well-being and physiological resilience in adults experiencing mild to moderate stress.

### *Conflict of Interest and Funding Statement*

The authors declare that there is conflict of interest related to this study. However, financial support for the conduct of this research was received from Bright Lifecare Pvt. Ltd., Presidency Tower – B, MG Road, Sector-14, Gurgaon, Haryana, India.

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### *Prior Informed Consent*

Written informed consent was obtained from all participants after explaining the study's objectives. Participation was voluntary, and confidentiality of personal information was maintained.

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