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Pine Pollen Impacts Testosterone-Related Symptoms in Older Men: A Pilot Report

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1. Abstract

1.1. Purpose: The primary goal of this beta study was to determine the effects of consuming a proprietary tincture form of Pine Pollen extract [*Pinus massoniana* pollen] during an eight-week open-label trial on the clinical outcomes in older men with initial symptoms of low testosterone. The secondary goal was to determine the total serum testosterone level changes in the same cohort and analyze the correlation with symptomatology.

1.2. Methods: Men between the ages of 47 and 78 years were randomly recruited. Individuals with confounding factors or specific medical histories that may cause undue risk in participation were prospectively excluded. The remaining subjects were enrolled after appropriate informed consent was obtained. The study consisted of eight weeks of supplement consumption with total testosterone blood measurements and completion of the qualitative Androgen Deficiency in the Aging Male [qADAM] questionnaire at pre-study baseline and at the end of the trial.

1.3. Results: Mean blood testosterone levels increased from 362.50 to 448.40 ng/dL, with a t-test statistical value at $P=0.0584$, demonstrating a positive trend although not reaching statistical significance. Mean total scores on the qADAM survey increased from 23.9 to 31.7 points, highly statistically significant at $P=0.0035$.

1.4. Conclusions: The current beta study demonstrated that a pro-

prietary pine pollen tincture can positively affect total testosterone levels and lifestyle factors in older men within eight weeks. This uniquely constituted pine pollen supplement may be a promising option for older men with low androgenic hormone levels who prefer a natural supplement rather than direct testosterone replacement. Further investigation in larger cohorts is necessary to confirm these results and to assess pine pollen supplementation in other groups with hypogonadal symptoms.

2. Introduction

Testosterone is the primary male androgenic [sex] hormone critical to the properly functioning of a broad spectrum of physiological processes. The primary focus is on sexual functions, such as libido and erectile dysfunction, yet related physical changes, including muscle mass, strength, body fat, and hair and bone loss, warrant attention [1,2]. In addition, other organ systems and emotional or cognitive changes can be affected [3-5]. Average male testosterone levels peak in the mid-20s and decline from that point, with the average decrease approaching one percent per year from age 40 [6]. While the direct linkage between lower testosterone levels in aging males [generally <300 ng/dL] and concomitant symptoms has not been firmly established, it is reasonable to consider that a portion of the issues with low libido, sexual dysfunction, decreased strength, and depressed mood are caused by lower circu-

lating androgen levels [7-9].

Since symptomatic low testosterone, or hypogonadism, negatively affects men's health and quality of life, elevation of this critical hormone has become a readily acceptable potential therapeutic option. The primary rationale for treatment is the alleviation of bothersome symptoms and improvement in overall well-being while elevating serum levels may also be accomplished [10-12]. Validated questionnaire tools are now available with the sensitivity to estimate the success of interventions and may assist in proper clinical substantiation [13, 14]. Testosterone replacement is a powerful strategy for managing deficiency in many clinical settings when it is employed with proper diagnosis, risk assessment, personalized treatment, and shared decision-making [15-17].

However, not every endpoint is positive, especially fracture incidence [18]. Long-term use of exogenous testosterone has been associated with prostate disease and cancer risk, but more recent studies have quieted this concern [19-21]. A similar positive trajectory of thought has occurred in cardiovascular disease risk [22, 23]. Nevertheless, while testosterone replacement therapy is commonly available in several formats, because of persisting safety and fertility preservation concerns, some non-testosterone-based treatments have been employed and proved efficacious [24]. Several vitamins, minerals, and phytonutrients may also elevate testosterone levels and alleviate hypogonadism symptoms.

One of these natural plant-based products is pine pollen from pine tree species, a traditional Chinese medicine used for centuries that has produced recent interest as an androgenic steroid-boosting supplement employed by Western holistic and complementary medical practices. One mechanism presumed responsible for clinical outcomes is the presence of phytoestrogens, plant-derived compounds exhibiting some androgenic effect similar to testosterone [25-27]. The extract also includes polysaccharides, vitamins, phospholipids, amino acids, minerals, enzymes, polyphenols, fiber, and proteins [28-31].

Numerous health-based attributes are ascribed to pine pollen, primarily related to antiaging effects and antioxidant activity [32-35]. Further studies demonstrate potent immune-enhancing properties [36-39]. In addition, potentially beneficial impacts on intestinal disease, arthritis, and hepatic and colon cancer have been reported [40-46]. However, almost all of these observations have been in laboratory-based in vitro systems or in vivo animal models. Human studies are scarce, and because of this limitation, it seems appropriate to demonstrate positive clinical effects and confirming the benefits in older men with symptoms of hypogonadism is a reasonable place to start. Fortunately, pine pollen is quite safe for oral consumption, although it is possible to experience some initial mild stomach upset. While allergic responses may occur, severe reactions are rare and generally limited to individuals with known allergic histories [47].

3. Materials and Methods

Participants were prospectively and randomly recruited from males over age 45 who perceived they had low testosterone levels. They were instructed to review the common symptoms associated with low testosterone to enhance appropriate study subject matching. To qualify, subjects had to note they had at least three of the symptoms, which included low libido, fatigue increasingly pronounced than previously, hair and or muscle loss, low self-esteem/moodiness, poor energy level overall, or depression/lack of ability to focus. These general terms were used rather than the qADAM descriptors for the initial screen since only their presence was deemed necessary, without reference to an actual scale number. In addition, a participant could not be in current or past treatment for prostate cancer. Nine potential subjects were excluded for this condition.

Subjects disclosed all current medications and supplements for clinical and safety review. They also listed their exercise and lifestyle routines for additional review. Participation required that lifestyle, nutrition, supplements and medications remain consistent over the eight-week trial period. If a subject was taking biotin, he was instructed to stop that supplement three days before blood sampling to ensure there was no conflict with the blood test analysis.

The initial study group included 12 males, ages 47 to 78. Two subjects were removed from the study shortly after onset due to non-disclosure of a medical condition in conflict and non-compliance with testing/survey instructions. The remaining subjects [n=10] mean age was 63 years of age.

The supplement used for the study was *Pinus massoniana* pollen [Lost Empire Herbs, Salt Lake City, UT]. The compounds are flash steamed for sterilization, and the pine pollen is hydroethanolic extracted. Precisely this is 50% alcohol by volume, with 50% organic cane alcohol and 50% deionized water. There is the equivalent of approximately 18 grams of pine pollen per bottle extracted. This results in a 300 mg per 1mL dose in a tincture format for oral delivery and absorption under the tongue, Figure 1. This recommended dosing, 1mL, was followed in the study as ½ of a dropper full in the morning and the same volume consumed in the evening. After five days, the subject takes a break from any supplement consumption for two days, then restarts the five- and two-day regimens again the following week.

Figure 1: Lost Empire Herbs Pine Pollen Tincture.

At baseline and after eight weeks, the participants completed the quantitative version of the Androgen Deficiency in the Aging Male [ADAM] questionnaire. The original ADAM survey uses a simple Yes/No system where three or more "Yes" results and two specific "Yes" ratings on libido-related issues correlate with likely low testosterone. The updated version of the questionnaire, the qADAM, expands the responses to include a rating scale for each question

from one to five. The entire 10-question panel was used for each subject. The question about loss of height, while rated, was removed from the tabulation as shown in Figure 2. While that component may be pertinent to a clinician's assessment of a patient over time, in this case, a supplement would not impact height and

was irrelevant to the purposes of this study. The surveys were administered electronically on the same day as the subject blood test samples were collected. For each survey, the subjects were asked to recall their experience, feelings and thus generate a rating for the seven days prior to the survey.



Figure 1: Lost Empire Herbs Pine Pollen Tincture.

Figure 2: The qADAM survey used in the study.

While not universal in application, many health care professionals use the ADAM scales and similar, alone or in conjunction with total testosterone blood levels in screening for hypogonadism. As the scale evaluates various lifestyle factors, it can chart dysfunction and improvement in measures critical to a specific individual. In this survey, the number “1” equals a rating of “terrible,” while the number “5” equals a rating of “excellent.” Thus, the nine tabulated questions had a potential low score of nine and a potential high score of 45. As the “average” rating in each item was three, 27 points indicated that the subjects considered themselves as average. When the scores were totaled for all questions, a lower aggregate score generally parallels higher dysfunction than average relative to androgenic hormone domains. Conversely, higher

scores above 27 generally correlate with milder dysfunction and more robust total testosterone levels.

For blood sampling, subjects were instructed to use an at-home test kit [LetsGetChecked-LGC, Monrovia, California]. This platform presents a relatively easy means to efficiently obtain clinically reliable diagnostic test results from at-home, self-collected blood samples as depicted in Figure 3. LGC is state-licensed and certified through the federal Clinical Laboratory Improvement Amendments [CLIA] program administered by the Centers for Medicare & Medicaid Services, the agency that regulates all laboratory testing performed on individuals in the U.S. LGC is further accredited by the College of American Pathologists [CAP] program that inspects human testing facilities and ensures their compliance with the highest standards in laboratory medicine and technology.

9 Question qADAM Online Testosterone Supplement Survey

Rating for each question, except question #8:

1- Terrible 2- Poor 3- Average 4- Good 5- Excellent

1. How would you rate your libido or sex drive over the last week?

2. How would you rate your energy level?

3. How would you rate your strength/endurance?

4. How would you rate your enjoyment of life?

5. How would you rate your happiness level?

6. How strong are your erections?

7. How would you rate your work performance over the last week?

8. How often do you fall asleep after dinner?

1- Every Night 2- 5 - 6 X per week 3- 3 or 4 X per week 4- 1- 2 X per week 5- Never

9. How would you rate your sports ability over the last week?

Figure 2: The qADAM survey used in the study.

Figure 3: Blood Test Kit Example, LetsGetChecked.

The methodology utilized in this study with regard to LGC was the Elecsys® Testosterone II assay [Roche Pharmaceuticals]. This test is an immunoassay for the in vitro quantitative analysis of testosterone levels in human serum and plasma [48]. Using patented technology, this electrochemiluminescence technique is employed on the Cobas e 801 immunoassay analyzer, a high throughput module [49]. The method is based on a competitive test principle

using a high-affinity sheep monoclonal antibody directed explicitly against testosterone. This laboratory was selected due to validated accuracy, CLIA/CAP certification and the ease of collecting very early morning samples when testosterone levels are the highest. To keep subjects familiar with the qADAM survey and blood sampling technique, both measures were repeated at the mid-study mark of four weeks, although not tabulated.

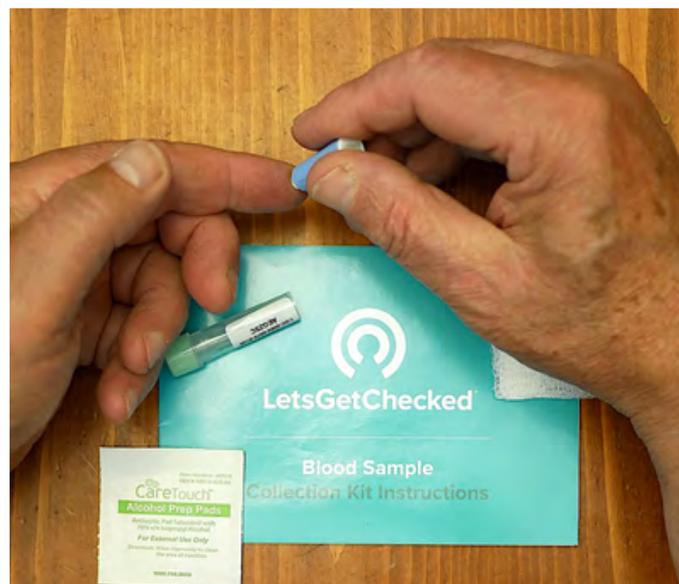


Figure 3: Blood Test Kit Example, LetsGetChecked.

4. Results

Of the 12 initial subjects, ten completed the entire eight weeks and all testing/assessments. The subjects’ total testosterone levels are listed in Table 1. The mean aggregate levels increased from 362.50 ng/dL to 448.40 ng/dL, an increase of 85.9 ng/dL, graphically depicted in Figure 4. Statistical significance was trending positive at $p= 0.0584$. All participants except one showed at least some increase over the study period.

Table 1: Blood Test Results, Pre and Post, ng/dL

Figure 4: Graphic Depiction of Blood Testosterone Mean Changes

The mean aggregate score for the nine measures on the qADAM test increased from 23.90 [below average] to 31.7 [above average]. Individual changes are listed in Table 2, with graphic illustration of the mean change for the subject group in Figure 5. Statistical significance was high at $p= 0.0035$. All subjects reported at least some improvement in each survey question. No adverse pine pollen side effects were noted in any participant throughout the study duration.

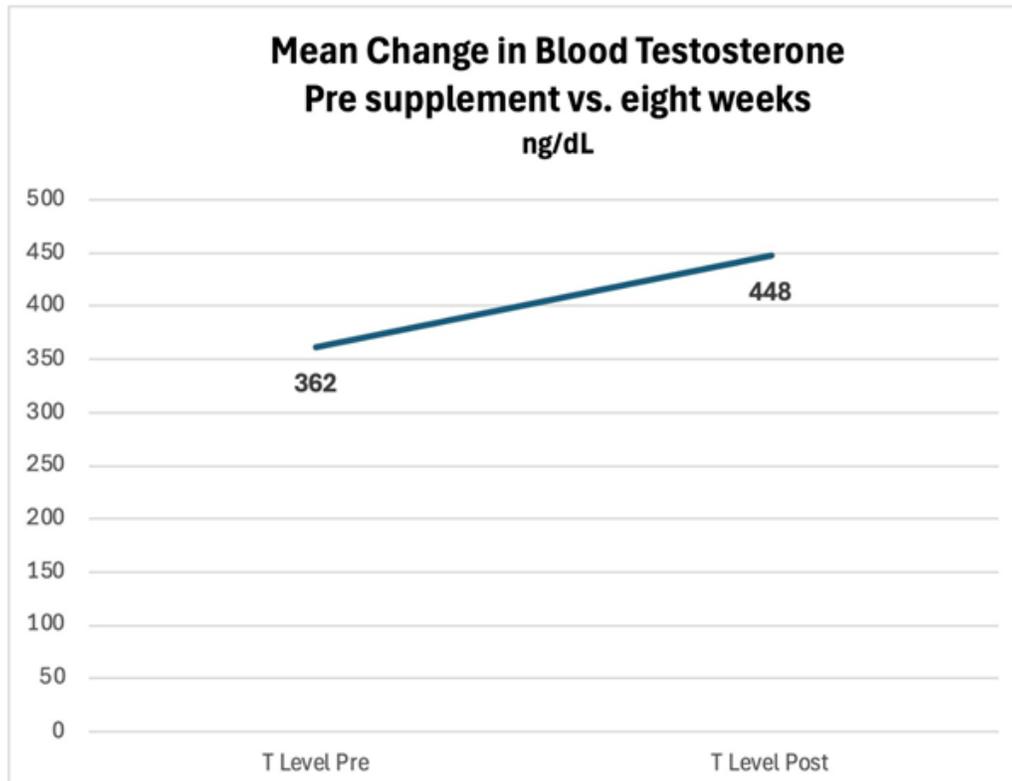


Figure 4: Graphic Depiction of Blood Testosterone Mean Changes

Table 1: Blood Test Results, Pre and Post, ng/dL

Subject	Pre T Level	Post T Level	Change
1	351	429	78
2	282	703	421
3	209	263	54
4	316	316	0
5	563	575	12
6	418	430	12
7	469	616	147
8	318	382	64
9	224	275	51
10	475	495	20
Mean	362.5	448.4	85.9

Table 2: qADAM Scores and Changes by Subject**Figure 5: Graphic Depiction of qADAM Mean Changes****5. Discussion**

This prospective beta observational trial is one of the first studies to describe the impact of a unique pine pollen extract on serum testosterone levels and related clinical symptoms in older men. The importance of these observations is related to the fact that while extensive historical information exists in traditional Eastern medicine, empiric human data in Western allopathic medicine is scarce. While in a limited pilot study, the current findings demonstrated clinical improvement without testosterone replacement within the relatively short period of time of eight weeks.

Concerns about exogenous testosterone consumption has led to the search for alternative strategies that more indirectly elevate androgen levels. Since many men of all age groups seek benefits without drug intervention, the natural supplement route may be appealing. Although pine pollen has enjoyed a centuries-long history of success in Chinese medicine, it is derived from many pine tree species and its composition, stability and bioavailability are not easily determined [50]. Therefore, this pilot trial utilized a proprietary pine pollen extract in tincture form [Lost Empire Herbs] that was formulated by a unique extraction technique to provide quality, consistency, efficacy and safety.

Total testosterone determination is the usual initial screening test in males who present with symptoms typical of low androgenic hormone levels. However, baseline serum testosterone levels may vary due to individual genetics, lifestyle differences and other factors [51, 52]. In this study, the initial value range was 209-563 ng/dL. Therefore, health care professionals strongly consider symptoms in guiding possible treatment and measuring therapeutic success [8, 9, 11, 12]. Nevertheless, blood levels in the current investigation did increase on average for the entire cohort from 362.50 ng/dL to 448.40 ng/dL, representing a positive t-test trend at the P=0.0584 level. While the participants did increase their serum levels as the study progressed, the wide baseline level ranges

and limited subject numbers may have precluded reaching standard statistical significance. All blood samples were analyzed by the same CLIA/CAP certified laboratory and supervised by an academic pathologist, thereby producing acceptable ranges for the results and reducing confounding variances. This was particularly important since actual total testosterone levels have never been measured in response to pine pollen supplementation.

In addition, the beneficial outcomes related to symptoms is a definitive strength of this study. A sensitivity-validated instrument, the quantitative Androgen Deficiency in the Aging Male [qADAM] survey scale, was employed to effectively assess the changes in clinical symptoms and quality of life factors over time [13, 14, 53, 54]. After completing the eight-week trial, brief interviews with the subjects revealed that the improvements were readily noticeable by themselves and available spouses. Every individual reported positive benefit across the scales of the nine combined items queried. The mean score for the group was 23.9 points initially and increased to 31.7 points by the end of eight weeks, a highly statistically significant t-test change at the P=0.0035 level. It was further observed that no participants experienced any allergic reactions or other adverse side effects to the pine pollen.

This beta pilot investigation supports consideration of consuming uniquely selected and produced natural products to elevate androgen levels and improve related hypogonadal symptoms in older men. The study is limited by the open label design, the limited number of subjects and the variability of baseline testosterone levels. However, the participants were personally monitored to minimize recall bias, and the proper tincture consumption was carefully and frequently confirmed. Subsequent trials of longer duration and with larger subject numbers may be warranted to further confirm the present findings. In addition, evaluation of a younger male cohort [e.g. ages 25-45 years] could facilitate more widely generalizing these results. While not the primary study purpose, the study demonstrated that an at home, blood sampling and testing as performed by LGC has promise in both research and individual patient utilization and monitoring.

Table 2: qADAM Scores and Changes by Subject

Subject	Pre Supplement	Post Supplement	Score Change
1	27	41	14
2	23	26	3
3	31	36	5
4	23	39	13
5	22	30	8
6	28	34	6
7	27	32	5
8	17	23	6
9	21	35	14
10	20	21	1
Mean Score	23.9	31.7	7.8

6. Conclusion

An orally consumed proprietary pine pollen extract increased total testosterone levels in older men over an eight-week prospective clinical trial. The unique supplement also significantly improved a broad spectrum of personally relevant hypogonadal symptoms during the same time-period. The extract in tincture form was easily tolerated and did not cause any adverse side effects.

7. Acknowledgements

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8. Conflicts of Interest

None.

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