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ALLIANCE FOR CLINICAL TRIALS IN ONCOLOGY

Randomized Double-Blind Placebo Controlled Study of Testosterone in the Adjuvant Treatment of Postmenopausal Women with Aromatase Inhibitor Induced Arthralgias

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Drug Availability

Supplied Agent: Testosterone IND #114707 (Alliance)

***Investigator having NCI responsibility for this protocol**

✓ Study contributor(s) not responsible for patient care.

ClinicalTrials.gov Identifier: NCT01573442

Participating NCTN Organizations

Alliance/ Alliance for Clinical Trials in Oncology (lead)

ECOG-ACRIN/ ECOG-ACRIN Cancer Research Group

NRG/ NRG Oncology

SWOG/ SWOG

Cancer Trials Support Unit (CTSU)**Contact Information**

For regulatory requirements:	For patient enrollments:	For Study data submission
<p>Regulatory documentation must be submitted to the CTSU via the Regulatory Submission Portal.</p> <p>Regulatory Submission Portal: (Sign in at www.ctsu.org, and select the Regulatory Submission sub-tab under the Regulatory tab.)</p> <p>Institutions with patients waiting that are unable to use the Portal should alert the CTSU Regulatory Office immediately at [REDACTED] to receive further instruction and support.</p> <p>Contact the CTSU Regulatory Help Desk at [REDACTED] for regulatory assistance</p>	<p>Please refer to the patient enrollment section of the protocol for instructions on using the Oncology Patient Enrollment Network (OPEN) which can be accessed at https://www.ctsu.org/OPEN_SYSTEM/ or https://OPEN.ctsu.org.</p> <p>Contact the CTSU Help Desk with any OPEN-related questions at [REDACTED]</p>	<p><i>Legacy NCCTG sites will submit electronic CRFs via: NCCTG Remote Data Entry System.</i></p> <p><i>Sites not previously affiliated with NCCTG will submit paper CRFs to: Alliance Statistics and Data Center Attention: [REDACTED]</i></p> <p>Do <u>not</u> submit study data or forms to CTSU Data Operations. Do <u>not</u> copy the CTSU on data submissions.</p>
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<p><u>For clinical questions (i.e. patient eligibility or treatment-related)</u> Contact the Research Base Quality Assurance Specialist (listed in Protocol Resources table on next page).</p>		
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Protocol Resources

Expedited Adverse Event Reporting
<https://eapps-ctep.nci.nih.gov/ctepaers/>

OPEN (Oncology Patient Enrollment Network)
<https://open.ctsu.org>

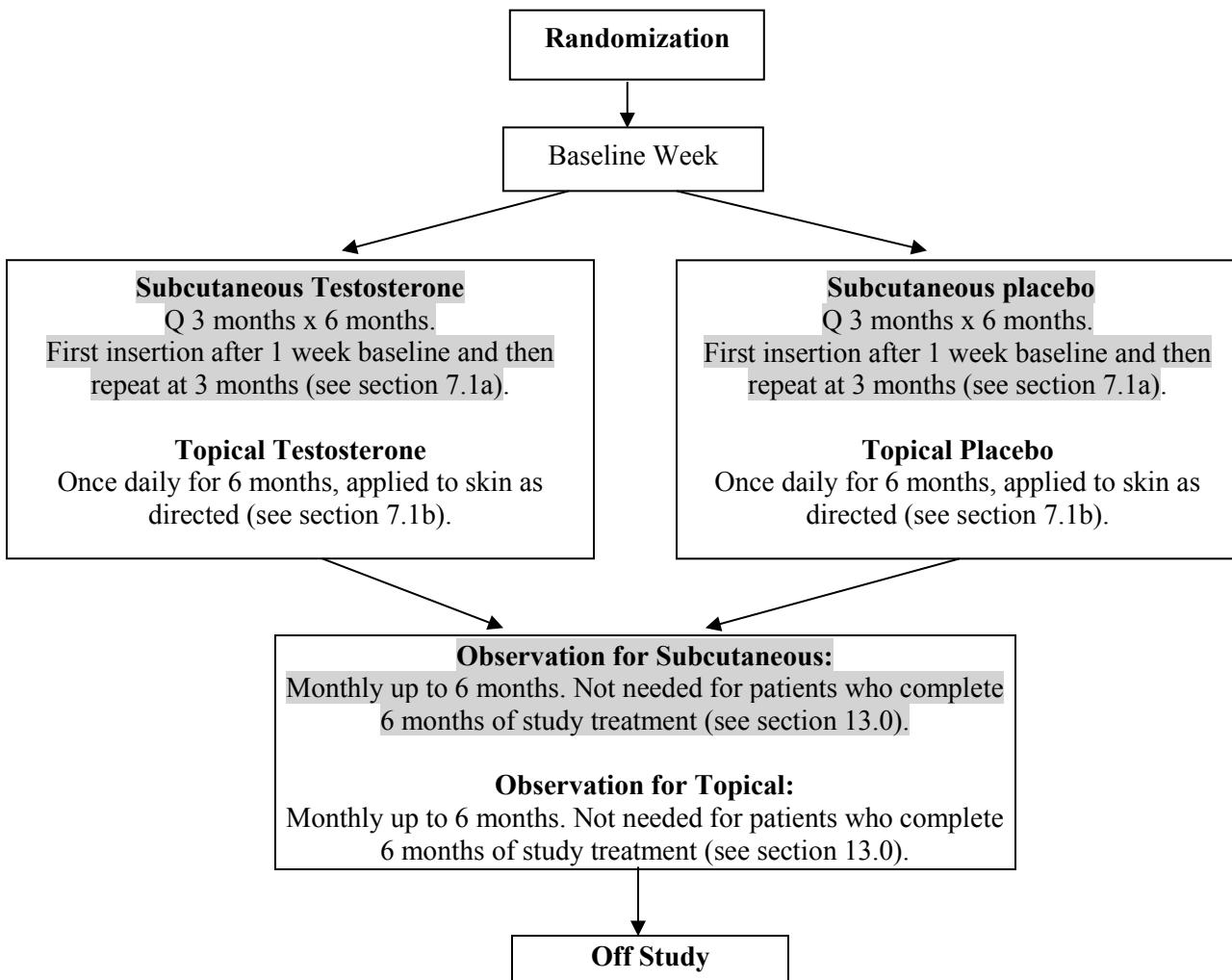
Biospecimen Management System
<http://bioms.allianceforclinicaltrialsinoncology.org>

Questions:	Contact Name:
Patient eligibility*, test schedule, treatment delays/interruptions/adjustments, dose modifications, adverse events, forms completion and submission	[REDACTED]
Drug administration, infusion pumps, nursing guidelines	[REDACTED]
Forms completion and submission	[REDACTED]
Protocol document, consent form, Regulatory issues	[REDACTED]
Adverse Events (CTEP-AERS, MedWatch, Non-AER, AML/MDS)	[REDACTED]
Non-paraffin biospecimens	[REDACTED]

*No waivers of eligibility per NCI

Schema

Prior to Update #3, patients were treated with a pellet form of testosterone, which was inserted via surgical implant. Effective with the issuance and local IRB approval of Update #3 to this protocol, new patients are to be randomized to the topical form of testosterone. Those patients randomized to the pellet form will complete the study according to the original protocol and will receive their second pellet as planned.



Baseline cycle length = 7 days

Cycle 1 length = 90 days

Cycle 2 length = 90 days

NOTE: Cycle is an Alliance data management tool to facilitate consistent remote data entry.

Generic name: Testosterone Brand name(s): Abbreviation: TESTO Availability: Research Base Pharmacy	Generic name: Placebo Brand name(s): Abbreviation: PLACEB Availability: Research Base Pharmacy
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1.0 BACKGROUND

1.1 Executive Summary

Rationale

- Aromatase Inhibitor induced arthralgia (AIA) is a major impediment for the use of this class of drug in breast cancer management.
- There are available data to support that testosterone metabolic anomalies in synovium may be the cause of this syndrome.
- A recent phase II trial (ART2) demonstrated the efficacy of testosterone replacement in treating AIA.
- In a concurrent *in vitro* study, aromatase inhibitor (AI) efficacy was unaffected by testosterone (T) supplementation.

Proposal

This concept application is to validate and refine the application of topical testosterone in women suffering from AIA. This will include the evaluation of T supplementation to overcome other side effects, such as hot flashes, loss of libido, and general sense of well-being (quality of life).

1.2 Review of Related Research

The new generation of aromatase inhibitors (AIs) used in the treatment of breast cancer has resulted in a therapeutic advantage over the gold standard tamoxifen. However, when the original trials were undertaken with AIs, one side effect or adverse event (AE) was not anticipated and, therefore, was not adequately evaluated during the protocol generation. Specifically, this AE is joint pain and stiffness. This AE has been demonstrated to occur in between 30 and 50% of women undergoing AIs therapy, and is resulting in significant compliance problems¹.

This unanticipated syndrome is characterized by joint inflammation similar to rheumatoid arthritis. Similarly, a relatively high incidence of Sjogren's syndrome (well known to be secondary to abnormal androgen metabolism), has also been diagnosed among those undergoing AI therapy². To date, the management of AIA has been with traditional rheumatological therapeutic interventions. Although the etiology of AIA is poorly understood, it may be secondary to alterations in hormonal metabolism; and quite possibly secondary to alteration in androgen metabolism, rather than a direct effect of estrogen deprivation. This may explain a potential relationship of this AIA syndrome to Sjogren's syndrome. The severity of this rheumatoidal adverse effect is not seen in women taking selective estrogen receptor modulators like toremifene or in women receiving LHRH agonist therapy.

1.3 Aromatase Inhibitors and Arthralgia

Studies show that the aromatase enzyme is prevalent in the synovium of joints and is active in metabolizing precursor androgens to estrogen (E). The balance of estrogens and androgens in the joint space seems to be pivotal in regulating levels of pro-inflammatory cytokines such as tumor necrosis factor (TNF)³. TNF has a pivotal role in the pathogenesis of rheumatoid arthritis synovitis. The direct pathogenic effects of TNF are amplified by TNF's ability to induce other pro-inflammatory cytokines, including: interleukin-1 and interleukin-6; and a granulocyte macrophage colony stimulating factor and adhesion molecules such as prostaglandin E2 and also matrix metalloproteinase, which are involved in joint degradation exerted by the synovial tissue. Androgens, especially T and dihydrotestosterone (DHT) have been identified as being especially

important in countering the pro-inflammatory cytokines⁴. Unlike other tissues, the synovium is not effective in producing androgens from precursors such as progesterone or cholesterol; and it is, therefore, very dependent on serum delivery of dihydroepiandrosterone (DHEA) or testosterone³. Hydroxy (OH)-DHEA, a metabolite of DHEA, is pro-inflammatory in the synovium⁵; therefore, it can be asserted that T, and its more active, non-aromatizable metabolite 5α-dihydrotestosterone (5α-DHT) (thought to be anti-inflammatory in the normal synovium) would be a protective androgen against joint inflammation.

Intriguingly, serum T and DHT in women does not rise in the presence of 3rd generation aromatase inhibitors⁶. This seems counter-intuitive as E becomes almost undetectable in the serum. Because of this, one would assume that it would leave more substrate (i.e., T) to become available in the serum. In joints where aromatase is plentiful in cartilage and synovium, there is a complex interplay of hormonal actions. In the synovial fluid of women being treated with aromatase inhibitors, there may be low DHT levels for two reasons: 1) secondary to low levels of bio-available T (as seen in women after chemotherapy, oophorectomy and aging); and 2) ultra low levels of estrogen found in women who are taking aromatase inhibitors that result in a negative feedback on 5-alpha reductase⁷, the enzyme that metabolizes T to 5α-DHT.

Conversion to T from other precursors does not appear to be as efficient in joints when compared with other tissues. Joints seem to require T to ensure adequate DHT levels⁸. Thus, to overcome this negative side-effect it was decided to try testosterone replacement in women experiencing the side-effect of joint pain and stiffness caused by aromatase inhibitors (ART therapy).

1.4 Summary of ART Therapy Data

Two concurrent streams of investigation of AIs and T have been undertaken, an in vitro study and a phase II trial. Both these studies were presented at the San Antonio Breast Cancer Conference 2009.

- 1) **The *In Vitro* Study** is about to be published and the abstract is as follows:

Abstract

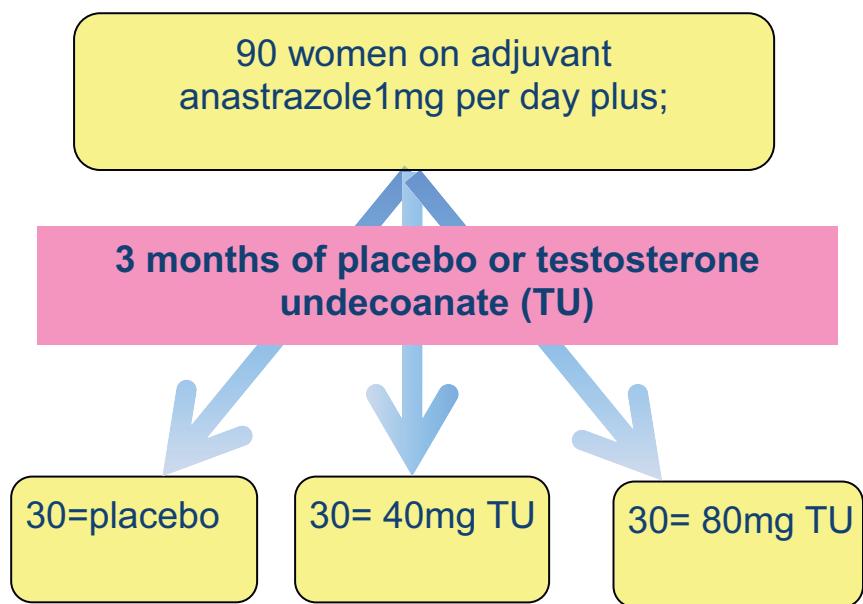
Purpose: Aromatase inhibitors (AI) are the first line adjuvant therapy for postmenopausal women with estrogen receptor (ER) positive breast cancer. Side effects of AI therapy, such as arthralgia, can cause significant patient discomfort leading to compliance issues. Testosterone (T) supplementation has emerged as a potential means to treat AI-associated arthralgia with favourable results in a phase II clinical trial (NCT00497458). Our objective was to determine whether T supplementation has potential to compromise the anti-proliferative effects of AIs in the treatment of breast cancer.

Methods: Fresh breast tumor tissues collected from 26 ER-positive post-menopausal women were cultured for 24h in media with 10% steroid depleted fetal calf serum and treated with vehicle (control), T (5nM) and/or AI-Anastrozole (25ng/ml). Tissues were stained with antibodies for ER, androgen receptor (AR) and Ki67, a marker of cell proliferation.

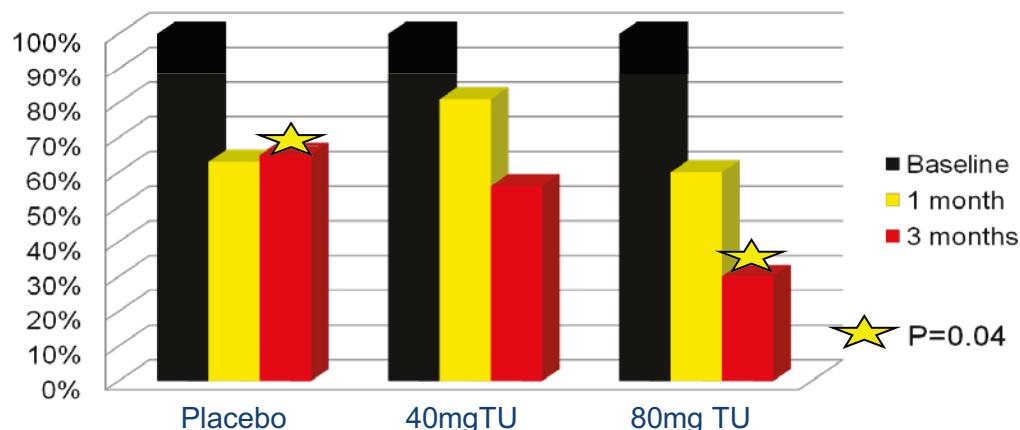
Results: Anastrozole treatment significantly inhibited breast cancer cell proliferation as compared to control ($p=0.003$; Wilcoxon signed rank test) in 11/26 (42%) cases, and this inhibition was unaffected by T supplementation.

Conclusions: Our findings suggest that T supplementation in post-menopausal women on adjuvant AI therapy is not deleterious.

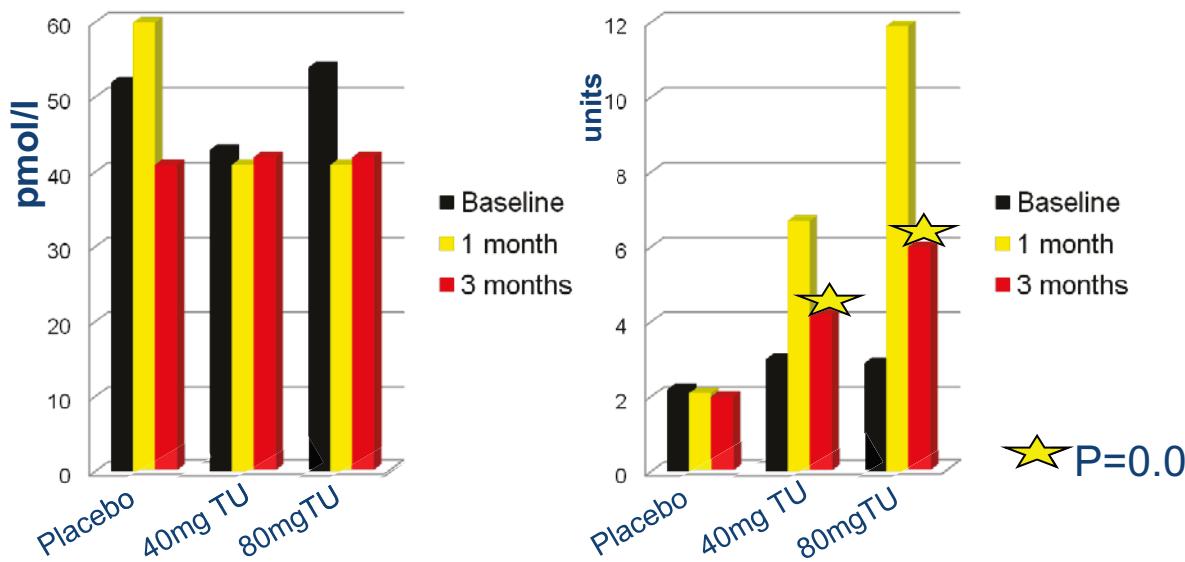
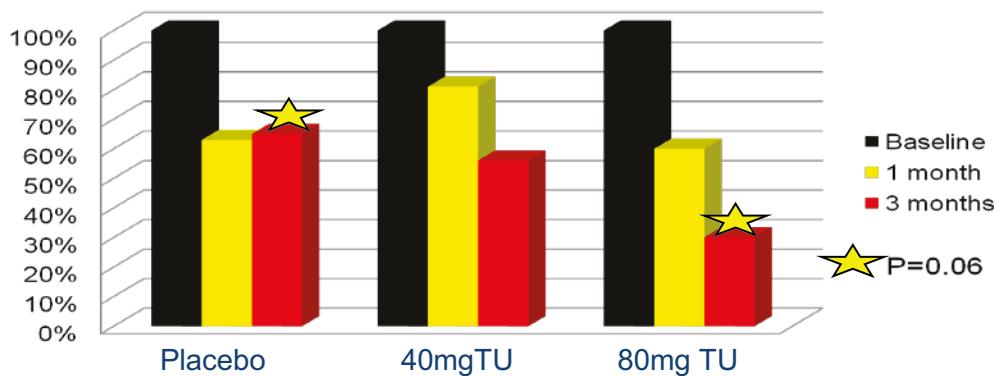
- 2) **The Phase II or ART 2 trial** is about to be published, a summary of the San Antonio posters presentation is as follows:



Graph 1. Percentage of patients with a PAIN VAS >50mm



Graph 2. Percentage of patients with a STIFFNESS VAS >50mm



Graph 3. Serum estradiol levels
(40pmol/l is the lower limit of detection)

Graph 4. Free Testosterone levels
(ratio of SHBG to T)

Serious adverse events (n=1)

- One chest wall recurrence after 4 weeks on 80mg TU in a woman who had received adjuvant chemotherapy, chest wall radiotherapy and 3 weeks of anastrazole for a T3N2M0 ER +ve PR -ve and AR -ve breast carcinoma.

Adverse events

	Placebo	40mg TU	80 mg TU
Total AEs (N=57, all CTC 1 or 2)	21	22	14
Androgenic AEs	2	3	1
Acne (less than 3 pustule count)	2	2	0
Hirsutism (* <8F-G scale)	0	1*	0
Raised BSL (* borderline elevated at baseline)	0	0	1*
Alopecia	0	0	0

- Good tolerability of TU at both doses.
- Despite a large placebo effect, a significant reduction in pain and a trend to a reduction in stiffness was observed with 80mgTU.
- No rise in serum estradiol with either dose of TU.
- Physiological levels of T obtained with once a day dosage with TU.
- No change in Lipid profile with either dose of TU (results not shown).
- No significant reduction in bone absorption (a trend to reduced absorption at 3 months with 80mg TU, results not shown)

The one woman with a recurrence, had very low levels of ER, no PR and, when subsequently tested, no androgen receptors. Thus, it is not likely that the administered testosterone had anything to do with her recurrence.

1.5 The Androgen Receptor, Androgens and Breast Cancer

Androgens and the androgen receptor (AR) have important physiological effects in women, especially in the breast (excellent reviews on this subject can be found in the works of Professor Susan Davis and her colleagues ⁹). However, there is great confusion as to whether the main circulating androgen, T, has a beneficial or detrimental effect on the normal breast and/or on malignant breast tissue. There are two schools of thought:

- That the presence of the AR in normal and malignant breast cells is indicative of the AR's importance in carcinogenesis, or on the survival and progression of that cell when exposed to its cognate ligand, T. (A higher level of serum testosterone is, therefore, not desirable); and
- That the AR in normal and malignant breast cells is an important regulator of estrogen receptor (ER) induced cellular proliferation, and that T that has been 5 α -reduced to 5 α -DHT is an essential ligand for AR induced reduction of cellular proliferation (in normal

breast cells) and increased apoptosis of malignant breast cells. (A higher level of serum testosterone, is therefore, desirable.)

Note: [REDACTED]
recognized international leaders in the field of androgens and breast cancer, concur that the second hypothesis is correct.

1.6 Androgen Receptor Expression in Breast Tissue

It would be unusual in breast malignancies (when AR is co-expressed with ER and PR), for the tumor to behave in a fashion other than the default position found in normal breast tissue (i.e. cellular stimulation by E and inhibition by T and DHT). However, this may not be the case as tumors progress toward hormonal independence where there may be global rearrangement of the cellular basis of hormonal regulation. This has recently been demonstrated by Hanley, et. al.,¹⁰ who examined the co-expression ER, PR and HER2 in relation to AR *in situ* and invasive malignancies and found that loss of AR expression may be important in preventing invasive progression. However, when AR is the sole steroid receptor expressed in a malignancy (as frequently occurs in advanced disease with HER2 over-expression), AR regulated pathways may be stimulated by androgens in an attempt by the cell to maintain hormonal stimulation. This is in keeping with work done on the androgen responsive breast cancer cell line MDA-MB-453 where high levels of AR are associated with amplification of HER2 and no ER and PR expression¹¹.

As a result, it is currently being asked whether malignancies, where the AR is the sole expressed steroid receptor, can be treated using an AI and DHEA¹². This is based on the observation that in some genetically modified breast cancer cell lines, proliferation is inhibited by AR stimulation in cells that express AR as the sole steroid hormone receptor¹³. This is not in keeping with the findings in MDA-MB-453 where AR is the sole steroid hormone receptor and androgens cause a mild cellular proliferation. Thus, the converse is being tested at The Memorial Sloan-Kettering Cancer Center, where bicalutamide, an AR antagonist, is being used to block the AR related pathways in metastatic disease, hoping that this could result in a hormonal regulation of a tumor thought to be hormonally insensitive¹⁴.

The evidence is pointing to the latter as being a sounder hypothesis and the trial results should be available next year; however, these divergent trials add to the uncertainty about androgen action in the breast. Nevertheless, it must be noted that AIs are currently only used in ER and PR positive malignancies and not in the types of cases outlined above.

1.7 Circulating Androgens in Women

In post-menopausal women, up to 25% of T is derived from the adrenal glands, 25% is derived from the ovary, and the remaining 50% is derived from peripheral conversion of the proandrogens, with androstenedione being the main precursor. Circulating T can be converted to DHT by 5α-reductase or to estradiol by the aromatase enzyme. Both these enzymes are present in both normal and malignant breast tissue¹⁵. However, in most malignant breast tissue, these enzymes are present in amplified quantities. In normal breast tissue their presence is less, but there are significant differences among individuals with normal breast tissue in the level of expression of these enzymes.

Although serum T is used as a marker of the androgenic status of a woman, it appears that the tissue levels of T are much more important. This is obviously difficult to measure and we must rely upon serum T measurements, which are fraught with potential difficulties, such as:

- 1) Insensitive techniques for measurement of serum T (especially at the very low serum concentration found in women);
- 2) Variable Sex Hormone Binding Globulin (SHBG) levels that keep T bound and not

- available for tissue activation;
- 3) Cyclical, diurnal and stress-induced variations in both T, E and adrenal precursor; and
 - 4) Variation in tissue enzyme activity (i.e., more aromatase than 5 α -reductase activity).

Many believe that it is the tissue level or intracrine metabolism of T and its precursors that is the essential issue in determining T action on normal and malignant breast tissue ¹⁵. In women where there is an excess of androgen production (such as polycystic ovarian syndrome), it may be that there is a reduction in breast cancer incidence; however, recent epidemiological data suggest that in healthy women with higher T levels the converse applies. The latter may be the result of higher T to E conversion. There are no studies that have addressed this issue in a systematic epidemiological fashion at the tissue level.

1.8 Androgens and Breast Cancer

In the 1940-60s, synthetic androgens were used in the treatment of metastatic breast cancer and were shown to have similar efficacy to anti-estrogens. However, the side-effect profile mitigated against their widespread acceptance and the advent of better hormonal therapies resulted in synthetic androgens being relegated to obscurity. While their mode of action was never fully understood, there is a body of evidence suggesting that the efficacy of androgen therapy resulted from direct interaction with the AR in breast cancer cells. This researcher and others have demonstrated that the synthetic androgen mibolerone ¹⁶ and the native androgen DHT (reviewed in ¹⁶), by binding to the AR ¹⁷, could inhibit breast cancer cell growth. In the ZR-75-1 breast cancer cell line, incubation with physiological concentrations of DHT, similar to those found in normal women ¹⁸, inhibited E induced proliferation ¹⁹. In ovariectomized athymic mice, DHT is a potent inhibitor of the stimulatory effect of E on ZR-75-1 tumor growth, and this effect can be reversed with the androgen antagonist, flutamide ²⁰. Although the DHT effect appears to be AR mediated, DHT suppresses the ER content of ZR-75-1 cells ²¹. However, the 2 most important observations regarding the effect of androgen on the breast come from a primate study which demonstrated that T reversed E-induced epithelial stimulation ²². It has also been shown that women taking T have a reduction in HRT-induced epithelial and stromal proliferative index ²³. Additional data attesting to the safety of testosterone is provided in appendix XI.

1.9 Androgen Replacement Safety

The risks of androgen replacement may be divided into: 1) those which are masculinizing (acne, hirsutism, alopecia, fluid retention, bloating); and 2) those which are potentially more serious (hepatocellular, cardiovascular and cancer risks). There are no large studies quantifying the incidence of the risk of masculinization with injected T. However, this risk does exist and depends on the level of T achieved and the woman's skin sensitivity. When careful monitoring has been carried out, and with lower dose therapy, the overall risk is low. In a safety surveillance study of esterified E and methyltestosterone, the risk of hirsutism was less than 5%; and in one other study, the side effects, with the addition of methyltestosterone, were no different from those in women receiving E alone. Similarly, fluid retention (which tends to be more idiosyncratic) and bloating were rare, and there were no reports of hepatic toxicity, changes in liver function or changes in blood pressure. In terms of cardiovascular disease risk, oral methyltestosterone adversely affects high-density lipoprotein (HDL) cholesterol with values decreasing by up to 20% from baseline; however, low-density lipoprotein (LDL) is lowered to a similar extent as E. Triglycerides, which tend to increase with oral E, decrease by approximately 15% with methyltestosterone ²⁴. In terms of direct effects on the vasculature, there are data suggesting that T may be beneficial for dilating coronary arteries, possibly via conversion by aromatase to E; but it may be a direct androgenic effect. In addition, data in the cynomolgus monkey have shown that methyltestosterone does not negatively influence coronary vasodilatation and decreases

coronary arterial LDL uptake. There is also no difference in the risk of endometrial hyperplasia when comparing E to E/androgen combination therapy.

1.10 Effect of AI and T combination (ART)

Although it was traditionally thought that most of the hormonal effect of testosterone in women resulted from aromatization of T to E, two recent studies have challenged this traditional thought. Studies of women receiving testosterone for sexual dysfunction and cognitive impairment did not have any reversal of the therapeutic benefit of T by the addition of the 3rd generation letrozole ^{25,26}. This indicates that the benefit of the T was not due to aromatization to E.

Utilizing T to overcome the side-effects of an AI in breast cancer therapy is a novel concept. The only current therapeutic utilization of the ART therapy are:

- 1) To reduce the estrogenic effect of T abuse in bodybuilding, in particular gynaecomastia. These data are unpublished as they are related to the illicit use of anabolic steroids, but is widely discussed on body building web sites <http://www.mesomorphosis.com/steroid-profiles/arimidex.htm>;
- 2) To reduce estrogenic side effects in hypogonadal men on T therapy ^{27,28}; and
- 3) Testosterone therapy for female-to-male transsexuals. ²⁹.

It is this last study that fully explores the safety issues of ART therapy. There were no demonstrable effects on the cardiovascular system in a double-blind placebo-controlled study of ART versus T and placebo ²⁹.

In addition, intriguing research by Macedo et al., has demonstrated that aromatase inhibitors may exert their antiproliferative effect in breast cancer cell lines, not only by reducing the intracellular production of estrogens, but also by unmasking the inhibitory effect of androgens acting via the AR ³⁰. Moreover, aromatase suppresses *in situ* production of bioactive androgen, DHT, in breast carcinoma ³¹. Aromatase inhibitors may thus have additional antiproliferative effects through increasing local DHT concentration with recent findings suggesting that intratumoral androgen actions are increased during exemestane treatment ³². Lastly, an abstract presented at the 2010 ASCO Breast Cancer Symposium³³, reported on the results of 43 women treated with anastrazole and testosterone therapy, with the testosterone being provided as subcutaneous pellet inserts. Acknowledging the lack of detail in an abstract, as opposed to a published manuscript, the authors reported that the patients had therapeutic levels of testosterone without elevations of estradiol levels. They also reported that patients did not have evidence of disease recurrence/progression on this therapy and that androgen deficiency symptoms resolved.

1.11 CCOP Feasibility

There are a large number of women on aromatase inhibitors and about 50% of them with get arthralgias, with about 15% getting arthralgias that are severe enough to cause them to stop taking the medication. The legacy NCCTG investigators, when queried, were quite enthusiastic about participating in this study. Thus, this study is quite feasible within the CCOP mechanism.

1.12 Study Duration

The primary endpoint is at 3 months, based on the results of the previous randomized trial. However, if we just stop the study at 3 months, reporting a positive result, the question will surely be: would it have continued to work for a longer period of time? We, thus, opted to study patients for a total period to 6 months.

1.13 Testosterone preparation to use for Pellet Insertion

Prior to Update #3, patients were treated with a pellet form of testosterone, which was inserted via surgical implant. Effective with the issuance and local IRB approval of Update #3 to this protocol, new patients are to be randomized to the topical form of testosterone. Those patients randomized to the pellet form will complete the study according to the original protocol and will receive their second pellet as planned.

We obtained an IND (IND# 114147) for using a subcutaneous preparation of testosterone for alleviating aromatase inhibitor arthralgias and planned to utilize this means to deliver the testosterone for this study. However, we subsequently determined that we had found a better testosterone formulation for conducting this trial, as opposed using to the subcutaneous preparation we were planning to utilize. We now propose to use a subcutaneous preparation formulation of testosterone. This preparation includes testosterone 120mg and anastrozole 8mg (provided to prevent aromatization of testosterone to estrogen). The testosterone /anastrozole pellets will come from a compounding pharmacy in Cincinnati, OH. The pharmacist, [REDACTED] [REDACTED] a colleague of [REDACTED] who manufactures Testopel®, an FDA approved testosterone implant. The testosterone/anastrozole pellets contain 98% active ingredient and about 2% stearic acid.

[REDACTED] provided information at the 2010 ASCO Breast Cancer Symposium regarding her experience with this preparation. A copy of her slides is provided as appendix XIII. To summarize, she treated women with androgen deficiency symptoms with a history of breast cancer. Previous published information supported that continuous testosterone therapy delivered by subcutaneous implant effectively treats hormone/androgen deficiency symptoms in both pre- and post-menopausal patients³⁴.

She presented preliminary information supporting that 12 mg of anastrozole, a third generation aromatase inhibitor, delivered subcutaneously by pellet implant with up to 1200 mg of testosterone effectively prevented the conversion of testosterone to estradiol in men with previously elevated estradiol levels. The amount of anastrazole delivered this way, over 3 months is negligible and should not interfere with the systemic aromatase inhibitor that the patient is receiving.

Subcutaneous testosterone implants provided consistent delivery and consistent absorption. It was a simple procedure to insert the implants. This administration bypasses the liver, thereby avoiding entero-hepatic circulation. There was no problem with compliance and it was well tolerated.

The dose that was provided to women was 120 mg of testosterone and 8 mg of anastrozole.

The ASCO presentation focused on breast cancer survivors who were referred from their oncologist, or self referred with permission from their oncologist, for symptoms of androgen deficiency or bone loss. Data were presented on 75 testosterone/anastrozole inserts performed in 43 breast cancer survivors treated between July 2009 and May 2010.

Serum testosterone and estradiol levels were measured 2 weeks following the implantation.

The results supported that this preparation was effective in treating symptoms with androgen deficiency in breast cancer survivors. Patients achieved relief of symptoms and had therapeutic testosterone levels with a mean of 281 nanograms per deciliter and a range of 120-518 nanograms per deciliter. With 70 out of 75 (93%) insertions in 43 patients, serum estradiol levels measured less than 30 pg/ml. There was one patient who had an estradiol level greater than 40 pg/ml but a subsequent level in this patient measured less than 30 pg/ml.

In a group of patients who received testosterone implants alone (without anastrozole) 42% patients had estradiol levels greater than 30 pg/ml. In contrast, only 7% (5/75) of patients treated with combination of anastrozole and testosterone had estradiol levels greater than 30 pg/ml.

This group reported that they did not have any adverse drug events in over 170 insertions in 67 patients through September 2010. None of the breast cancer survivors, who were without evidence of metastatic disease at presentation, had been diagnosed with recurrent disease with up to 4 years of therapy. There was no evidence of disease progression in two ER+ patients and 1 ER- patient who had metastatic disease treated up to 30 months. Another patient with active disease actually responded to chemotherapy with minimal side effects from the chemotherapy.

In addition, [REDACTED] have had an abstract /poster accepted for presentation at the European Menopause and Andropause Society meeting in Athens, Greece; March of 2012. They report on 1,408 insertions of the testosterone anastrozole combination implant, between July 2009 – July 2011, with 314 insertions in breast cancer survivors. There have been no complication or adverse drug events due to the combination implant (see attached poster as appendix XIV). They are also presenting an abstract/poster on the beneficial effects of subcutaneous testosterone implants on lipid profiles in women (see attached poster as appendix XV).

As of 9 February, 2012 [REDACTED] has performed 430 testosterone anastrozole insertions in breast cancer survivors with no complications or adverse drug events.

The advantages of the currently proposed manner of delivery, compared to subcutaneous delivery, are significant and we feel that this mode of delivery will be utilized in clinical practice and, thus, it is best if it is tested under trial conditions. It does carry the following advantages over subcutaneous application:

- The most reliable application of the therapeutic compound under investigation
- Only 2 applications required for the trial duration rather than daily application
- Minimization of the risk of application of the therapeutic compound under investigation (i.e. testosterone) without any form of aromatase inhibitor delivery (i.e. not taking the oral medication)

This current protocol is not designed for a labeling indication. Rather, it is an effort toward discovery of whether or not this preparation may alleviate aromatase inhibitor arthralgias. Aromatase inhibitor arthralgias are reported in up to 60% of women taking the aromatase inhibitors and are resulting in major compliance issues.

1.14 Summary

This unique and exciting concept has the potential to completely alter the nature of hormonal therapy in breast cancer management. It is recognized that long-term compliance has been an issue with anti-estrogen therapy. The advent of AIA has compounded this problem and made even short-term compliance a problem. By harnessing the unique ability of 3rd generation aromatase inhibitors to completely block the formation of E from T, the benefit of T can be realized without compromising anti-estrogen efficacy and indeed potentially provide the following benefits:

1. Reduce the impact of AIA
2. Enhance anti-estrogen efficacy
3. Reduce the risk of tumor hormone independence
4. Reduce hot flashes
5. Enhance sense of well-being
6. Improve libido

1.15 Reasoning for Topical Application

As of early 2015, a decision was made to switch from the subcutaneous testosterone/anastrozole pellet to a topical preparation applied daily. This change was made due to inadequate accrual with the subcutaneous preparation. This is thought to be because the pellet is inserted via surgical procedure. Thus, to make the therapy more accessible for clinical practice, where surgeons are not seeing patients routinely, the protocol has been modified to utilize a topical preparation of testosterone.

2.0 GOALS

2.1 Primary

To determine whether testosterone will reduce AI-induced arthralgia and associated joint symptoms.

2.2 Secondary

To explore whether testosterone will have an acceptable safety and tolerability profile, with particular reference to androgenic adverse events including acne, hirsutism, and alopecia.

2.3 Ancillary

- 2.3.1 To explore whether testosterone will reduce AI-induced arthralgias and associated joint symptoms and their interference with activity.
- 2.3.2 To explore whether testosterone will reduce the incidence of hot flashes.
- 2.3.3 To explore whether testosterone will modify libido or change quality of life.
- 2.3.4 To explore whether identified SNPs, that were associated with more aromatase inhibitor associated arthralgias in a previous prospective trial⁷⁷, are more commonly seen in the patients entered into the present clinical trial, than are observed in the normal population.

3.0 PATIENT ELIGIBILITY

3.1 Inclusion Criteria

- 3.1.1 Age \geq 18 years.
- 3.1.2 Receiving anastrozole (1mg) or letrozole (2.5 mg) orally once a day, for \geq 21 days prior to registration and plan to continue it throughout the duration of study.
- 3.1.3 Body Mass Index (BMI) between 18 and 35 kg/m².
- 3.1.4 Women who have undergone a total mastectomy or breast conserving surgery for primary breast cancer +/-chemo, +/-radiotherapy.
- 3.1.5 Must have **BOTH** ER and PR receptor-positive tumors and BOTH must be \geq 26% positive. Alternatively, if ER and PR are determined by Allred score, the score needs to be 5 or higher.
- 3.1.6 Women who are postmenopausal by surgery, radiotherapy or presence of natural amenorrhea \geq 12 months.

- 3.1.7** $\geq 5/10$ arthralgia (in hands, wrist, knees, or hips) whilst being treated with anastrozole or letrozole which is felt by the patient to be caused by their aromatase inhibitor, as measured by verbally addressing the following question:

Please rate your pain by picking a number, from 0 to 10 (0 being none and 10 being as bad as you can imagine) that best describes your pain from your aromatase inhibitor breast cancer medication on AVERAGE, over the past week.

Note: Patients may, or may not, be taking non-opioid analgesics:

- 3.1.8** Ability to complete questionnaire(s) by themselves or with assistance.
- 3.1.9** ECOG Performance Status (PS) 0, 1 or 2.
- 3.1.10** Willing to provide informed written consent.
- 3.1.11** Willing to return to an Alliance enrolling institution for follow-up.
- 3.1.12** Willing to provide blood samples for correlative research purposes (see Sections 6.0 and 14.0).
- 3.1.13** Laboratory values obtained $\leq 365^*$ days prior to registration:

Creatinine $\leq 1.5 \times$ ULN

Hemoglobin $> 11 \text{ g/dL}$

WBC > 3.0

Platelet Count $> 100,000$

SGOT (AST) $\leq 1.5 \times$ ULN

*Note: Without medical situations that should change these parameters since they were done.

3.2 Exclusion Criteria

- 3.2.1** Presence of residual or recurrent cancer (locally or metastatic).
- 3.2.2** Diabetes mellitus or glucose intolerance, defined as a fasting glucose $> 125 \text{ mg/dL}$.
- 3.2.3** History of coronary artery disease (angina or myocardial infarction).
- 3.2.4** Patients on hormone replacement therapy (HRT) ≤ 4 weeks prior to registration. This includes the use of vaginal estrogen therapy.
- 3.2.5** Known hypersensitivity to any component of testosterone.
- 3.2.6** Prolonged systemic corticosteroid treatment, except for topical applications (e.g. for rash), inhaled sprays (e.g. for obstructive airway diseases), eye drops or local insertion (e.g. intra-articular). Note: Short duration (< 2 weeks) of systemic corticosteroids is allowed (e.g. for Chronic Obstructive Pulmonary Disease) but not within 30 days prior to registration.
- 3.2.7** Receiving any other investigational agent.
- 3.2.8** History of a deep venous thrombosis or a thromboembolism.
- 3.2.9** Concurrent use of the aromatase inhibitor exemestane, as it is structurally similar to an androgen.
- 3.2.10** Concurrent radiation therapy or chemotherapy.
- 3.2.11** Current or planned use of cyclosporine, anticoagulants, insulin, oral or injectable vitamin D doses over 4,000IU/day, or tamoxifen

4.0 TEST SCHEDULE

Tests and procedures	Active-Monitoring Phase				
	≤ 30 days prior to registration	After registration, Prior to baseline hot flash questionnaire	Baseline Daily x 1 week**	At the end of Months 3 and 6*	Monthly (active-monitoring or observation)
History and exam, weight, performance status, vitals	X			X	
Height	X				
BMI	X				
Chemistry group: SGOT (AST) Creatinine	X ¹				
Hematology Group : HGB WBC Platelet Count	X ¹				
Insertion (testosterone or placebo)				X ⁴	
Adverse event assessment	X			X ⁵	
Mandatory research blood draw (see Section 14.0) ^R		X ²			
Patient Questionnaire ³ : Hot Flash Diary (Appendix IV)			X	A	
Patient Questionnaires ³ : Symptom Experience Questionnaire (App. V) POMS (App. VI) HFRDIS (App. VII) Modified Brief Pain Inventory (App. VIII) MENQOL (App. IX)		X			X ⁶
Nurse/CRA Phone Contact (Appendix X)					B

* +/- 7 days

** Must be completed before initiation of testosterone/placebo gel.

1. ≤365 days prior to registration.
 2. Kits are required for this collection. Blood draw should be done at the same time as the clinical draw, if possible. Will need to be done within 104 days of study entry (ideally ≤ 7 days prior to testosterone), but must be done before the end of the 3 month follow-up clinic visit.
 3. Patient questionnaire booklets **must** be used; copies are not acceptable for this submission.
 4. First implant insertion after 1 week baseline and then repeat after 3 months.
 5. Done whether the patient is in active-monitoring or observation.
 6. If the patient does not receive the second implant insertion, the patient will be encouraged to complete the remaining questionnaire booklets.
 - A. Daily during the first 8 weeks of treatment only.
 - B. First nurse/CRA call is made after the first two weeks on study. Not needed on months that patient is seen at the clinic. Call is to reinforce patient compliance and answer questions. Collect AE's, if applicable.
- R Research funded (see Section 19.0).

5.0 STRATIFICATION FACTORS

- 5.1** Baseline pain score (per eligibility question in section 3.17): 5-6 vs. 7-10.
5.2 Age: < 50 vs. 50-60 vs. > 60.

The stratification factors listed include demographic, prognostic factors that may potentially impact the primary or secondary outcomes, so they need to be distributed evenly among the two arms. The 6 level combinations involved in these stratification factors are within the maximum recommended of one half of the group sample size for the study⁶⁴.

6.0 REGISTRATION/RANDOMIZATION PROCEDURES

This study is supported by the NCI Cancer Trials Support Unit (CTSU).

6.1 CTEP / DCP Registration Procedures

Food and Drug Administration (FDA) regulations and National Cancer Institute (NCI) policy require all individuals contributing to NCI-sponsored trials to register and to renew their registration annually. To register, all individuals must obtain a Cancer Therapy Evaluation Program (CTEP) Identity and Access Management (IAM) account (<https://ctepcore.nci.nih.gov/iam>). In addition, persons with a registration type of Investigator (IVR), Non-Physician Investigator (NPIVR), or Associate Plus (AP) (i.e., clinical site staff requiring write access to OPEN, RAVE, or TRIAD or acting as a primary site contact) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) (<https://ctepcore.nci.nih.gov/rcr>). Documentation requirements per registration type are outlined in the table below.

Documentation Required	IVR	NPIVR	AP	A
FDA Form 1572	✓	✓		
Financial Disclosure Form	✓	✓	✓	
NCI Biosketch (education, training, employment, license, and certification)	✓	✓	✓	
HSP/GCP training	✓	✓	✓	
Agent Shipment Form (if applicable)	✓			
CV (optional)	✓	✓	✓	

An active CTEP-IAM user account and appropriate RCR registration is required to access all CTEP and CTSU (Cancer Trials Support Unit) websites and applications. In addition, IVRs and NPIVRs must list all clinical practice sites and IRBs covering their practice sites on the FDA Form 1572 in RCR to allow the following:

- Added to a site roster

- Assigned the treating, credit, consenting, or drug shipment (IVR only) tasks in OPEN
- Act as the site-protocol PI on the IRB approval

Additional information can be found on the CTEP website at <https://ctep.cancer.gov/investigatorResources/default.htm>. For questions, please contact the RCR Help Desk by email at [REDACTED]

Registration requires the submission of:

- Human Subject Protection (HSP) training certificate

6.2 Site Registration Requirements – IRB Approval

- 6.2.1** This study is supported by the NCI Cancer Trials Support Unit (CTSU).

IRB Approval:

Each investigator or group of investigators at a clinical site must obtain IRB approval for this protocol and submit IRB approval and supporting documentation to the CTSU Regulatory Office before they can be approved to enroll patients.

Assignment of site registration status in the CTSU Regulatory Support System (RSS) uses extensive data to make a determination of whether a site has fulfilled all regulatory criteria including but not limited to:

- an active Federal Wide Assurance (FWA) number,
- an active roster affiliation with the Lead Network or a participating organization,
- a valid IRB approval, and
- compliance with all protocol specific requirements.

In addition, the site-protocol Principal Investigator (PI) must meet the following criteria:

- Active registration status
- The IRB number of the site IRB of record listed on their Form FDA 1572
- An active status on a participating roster at the registering site.

Sites participating on the NCI CIRB initiative and accepting CIRB approval for the study are not required to submit separate IRB approval documentation to the CTSU Regulatory Office. For sites using the CIRB, IRB approval information is received from the CIRB and applied to the RSS in an automated process. Signatory Institutions must submit a Study Specific Worksheet for Local Context (SSW) to the CIRB via IRBManager to indicate their intention to open the study locally. The CIRB's approval of the SSW is then communicated to the CTSU Regulatory Office. In order for the SSW approval to be processed, the Signatory Institution must inform the CTSU which CIRB-approved institutions aligned with the Signatory Institution are participating in a given study.

Requirements for A221102 site registration:

- IRB approval (For sites not participating via the NCI CIRB; local IRB documentation, an IRB-signed CTSU IRB Certification Form, Protocol of Human Subjects Assurance Identification/IRB Certification/Declaration of Exemption Form, or combination is accepted)

- 6.2.2** In addition to submitting initial IRB approval documents, ongoing IRB approval documentation must be on file (no less than annually). If the necessary documentation is not submitted in advance of attempting patient randomization, the randomization will not

be accepted and the patient may not be enrolled in the protocol until the situation is resolved.

6.2.3 When the study has been permanently closed to patient enrollment, submission of annual IRB approvals to the CTSU is no longer necessary.

6.2.4 Downloading Site Registration Documents:

Site registration forms may be downloaded from the A221102 protocol page located on the CTSU members' website. Permission to view and download this protocol and its supporting documents is restricted and is based on person and site roster assignment housed in the CTSU RSS.

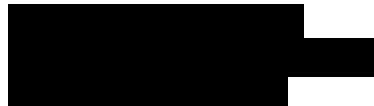
- Go to <https://www.ctsu.org> and log in to the members' area using your CTEP-IAM username and password
- Click on the Protocols tab in the upper left of your screen
- Either enter the protocol # in the search field at the top of the protocol tree, or
- Click on the By Lead Organization folder to expand
- Click on the Alliance link to expand, then select trial protocol #A221102
- Click on LPO Documents, select the Site Registration documents link, and download and complete the forms provided.

6.2.5 Submitting Regulatory Documents:

Submit required forms and documents to the CTSU Regulatory Office via the Regulatory Submission Portal, where they will be entered and tracked in the CTSU RSS.

Regulatory Submission Portal: www.ctsu.org (members' area) → Regulatory Tab → Regulatory Submission

When applicable, original documents should be mailed to:



Institutions with patients waiting that are unable to use the Portal should alert the CTSU Regulatory Office immediately at [REDACTED] in order to receive further instruction and support.

6.2.6 Checking Your Site's Registration Status:

You can verify your site registration status on the members' section of the CTSU website.

- Go to <https://www.ctsu.org> and log in to the members' area using your CTEP-IAM username and password
- Click on the Regulatory tab
- Click on the Site Registration tab
- Enter your 5-character CTEP Institution Code and click on Go

Note: The status given only reflects compliance with IRB documentation and institutional compliance with protocol-specific requirements outlined by the Lead Network. It does not reflect compliance with protocol requirements for individuals participating on the protocol or the enrolling investigator's status with the NCI or their affiliated networks

6.3 Patient Randomization

- 6.3.1** Patient randomization can occur only after pre-treatment evaluation is complete, eligibility criteria have been met, and the study site is listed as 'approved' in the CTSU RSS. Patients must have signed and dated all applicable consents and authorization forms.
- 6.3.2** Patient enrollment will be facilitated using the Oncology Patient Enrollment Network (OPEN). OPEN is a web-based registration system available on a 24/7 basis. To access OPEN, the site user must have an active CTEP-IAM account (check at <<https://eapps-ctep.nci.nih.gov/iam/index.jsp>>) and a 'Registrar' role on either the LPO or participating organization roster.

All site staff will use OPEN to enroll patients to this study. It is integrated with the CTSU Enterprise System for regulatory and roster data. OPEN can be accessed at <https://open.ctsu.org> or from the OPEN tab on the CTSU members' side of the website at <https://www.ctsu.org>.

6.3.3 Prior to accessing OPEN, site staff must verify the following:

- All eligibility criteria must have been met within the protocol stated timeframes. Site staff should use the randomization forms provided on the CTSU web site as a tool to verify eligibility.
- All patients must have signed an appropriate consent form and HIPAA authorization form (if applicable).

6.3.4 Access Requirements for Oncology Patient Enrollment Network (OPEN)

- Site staff will need to be registered with CTEP and have a valid and active CTEP-IAM account. This is the same account (user id and password) used for the CTSU members' web site.
- To perform randomizations, the site user must have been assigned the 'Registrar' role on the relevant Group or CTSU roster.
- To perform randomizations on protocols for which you are a member of the Lead Group, you must have an equivalent 'Registrar' role on the Lead Group roster. Role assignments are handled through the Groups in which you are a member.
- To perform randomizations to trials accessed via the CTSU mechanism you must have the role of Registrar on the CTSU roster. Site and/or Data Administrators can manage CTSU roster roles via the new Site Roles maintenance feature under RSS on the CTSU members' web site. This will allow them to assign staff the "Registrar" role.

NOTE: *The OPEN system will provide the site with a printable confirmation of randomization and treatment information. Please print this confirmation for your records.*

Further instructional information is provided on the OPEN tab of the CTSU members' side of the CTSU website at <https://www.ctsu.org> or at <https://open.ctsu.org>. For any additional questions, contact the CTSU Help Desk at [REDACTED] or [REDACTED]

6.3.5 Correlative Research: A mandatory correlative research component for blood is part of this study, the patient will be automatically registered onto this component (see Sections 3.1.13 and 14.0).

6.3.6 Patient has/had not given permission to store and use his/her sample(s) for use in future research to learn about, prevent, or treat cancer.

6.3.7 Patient has/had not given permission to store and use his/her sample(s) for use in future

research to learn about, prevent, or treat other health problems (for example: diabetes, Alzheimer's disease, or heart disease).

- 6.3.8** Patient has/has not given permission for the Alliance to give his/her stored sample(s) for use in future research to outside researchers.
- 6.3.9** Treatment on this protocol must commence at the accruing membership under the supervision of an Alliance member physician.
- 6.3.10** Study participation cannot begin prior to registration and must begin ≤ 14 days after registration.
- 6.3.11** Pretreatment tests/procedures (see Section 4.0) must be completed within the guidelines specified on the test schedule.
- 6.3.12** All required baseline symptoms (see Section 10.4) must be documented and graded.
- 6.3.13** Blood draw kit is available on site (see Section 14.2.1 for kit ordering instructions).
- 6.3.14** Patient Questionnaire Booklets are available on site; copies are not acceptable for this submission.

6.3.15 Ordering of Patient Questionnaire Booklets

Patient Questionnaire Booklets must be ordered prior to the registration of any patients. Site staff should obtain all necessary patient questionnaire booklets before registering patients. Patient questionnaire booklets should be ordered from CTSU by completing the Supply Request Form on the CTSU website (located under the site registration documents section of the CTSU A221102 Webpage) and faxing the form to the CTSU Data Center. Note: The CTSU will not send questionnaire booklets until the site has submitted a copy of their IRB approval excerpt to the CTSU Regulatory Office.

Copies of questionnaires in the appendices of this protocol are to be submitted to the local IRB for review and approval only and are not acceptable for data submission.

6.3.16 Once the above conditions have been met, access the OPEN website and follow the instructions for enrollment.

The factors defined in Section 5.0, together with the registering membership, will be used as stratification factors. The values of the stratification factors will be recorded. The patient then will be assigned to one of the following treatment groups using the Pocock and Simon⁶⁵ dynamic allocation procedure which balances the marginal distributions of the stratification factors between the treatment groups.

- Testosterone
- Placebo

6.4 Procedures for Double-Blinding the Treatment Assignment

To ensure that both the patient and the medical professionals who care for the patient are blinded to the identity of the treatment assignment, the randomization specialist will follow the double-blinding procedures outlined below.

- 6.4.1** After the treatment assignment has been determined by the registration/randomization application, Alliance registration office personnel will be notified of the treatment assignment, and they will then communicate the treatment assignment to the Alliance Research Base Pharmacy. The registration personnel will also notify the participating institution of the patient-specific kit number to be used on the order form. Institutions will then order testosterone/placebo using the A221102 Clinical Drug Order Form. Upon receipt of the order from, the Alliance Research Base Pharmacy will then ship the

appropriate patient-specific 3-month testosterone/placebo supply directly to the participating institution.

- 6.4.2** Approximately two weeks prior to the patient's second visit (at the end of month 3), institutional staff must place a reorder with the Alliance Research Base Pharmacy for the second 3-month patient specific drug supply, so that it is available when the patient returns. The institutional staff must contact the Registration Office at [REDACTED] for a code number when additional study product is needed for the patient.
- 6.4.3** The kit number assigned to the patient will be recorded on the dosing form.
- 6.4.4** The dose will be prepared and labeled as "testosterone OR placebo" so that the contents are not discernible to the individual administering the treatment.
- 6.4.5** The institutional pharmacist or designated contact person will maintain records that indicate the identity of the patient and their corresponding treatment assignment code.

7.0 PROTOCOL TREATMENT

Prior to Update #3, patients were treated with a pellet form of testosterone, which was inserted via surgical implant. Effective with the issuance and local IRB approval of Update #3 to this protocol, new patients are to be randomized to the topical form of testosterone. Those patients randomized to the pellet form will complete the study according to the original protocol and will receive their second pellet as planned.

7.1a Treatment Schedule for Pellet Insertion (for patients enrolled prior to local IRB approval of Update #3)

Agent	Dose Level	Route	Frequency
Testosterone/anastrozole	TWO pellets (total 120 mg) testosterone/ 8mg anastrozole	Sub Q implant	Once every 3 months for a total of 2 doses
Placebo	TWO pellets	Sub Q implant	Once every 3 months for a total of 2 doses

First insertion 1 week after baseline and then repeat at 3 months.

7.1b Treatment Schedule for Topical Application (for patients enrolled after local IRB approval of Update #3)

Agent	Dose Level	Route	Frequency
Testosterone gel	10.4mg/ 0.264mL	Topical	Daily x 6 months
Placebo	0.264mL	Topical	Daily x 6 months

- 7.11 After the patient has completed the Hot Flash Diary for 7 days, the patient will apply testosterone or placebo to the skin once a day for 6 months. This should start 1 week after study initiation (following the one week of obtaining the baseline hot flash data).
- 7.12 The CRA/Nurse will demonstrate to the patient how to apply the topical study agent.
- 7.13 The detailed instruction sheet (Appendix XVII) will be given to the patient when the

medication is dispensed.

7.2 Unblinding Procedures

Unblinding in this trial may only occur under two circumstances:

1. In case of an emergency for patients on therapy; or
2. After the patient has completed all study-related procedures and at the request of patient/physician.

Follow the directions below to unblind patient treatment. Please note that if a treatment assignment is unblinded, the patient must discontinue protocol therapy.

7.2.1 Emergency Unblinding:

Examples of emergencies include 1) a life-threatening unexpected adverse event that is at least possibly related to the investigational agent and for which unblinding would influence treatment decisions; or 2) medication error, such as accidental overdose. Expected adverse events are listed in the “Toxicities” section below.

Contact the Alliance Executive Officer on call by calling [REDACTED], pressing 1 to speak with an operator, and then asking for pager ID 8625 to return the call.

The institution must provide the following information to the Alliance Executive Officer:

- Alliance study ID (A221102)
- Alliance patient ID number
- Patient initials (e.g., “L,FM”)
- Institution name
- Name and telephone number of treating physician
- Name and contact information of person requesting the unblinding procedure
- Name and contact information of person to inform of treatment assignment
- Reason for emergency unblinding

Please remember that emergency unblinding request may be authorized only by an Alliance Executive Officer, and emergency unblinding applies only if unblinding would influence management of the medical situation.

After the Executive Officer deems unblinding is warranted, the treatment assignment will be provided to the contact person at the treating site.

7.2.2 Protocol-specified unblinding

Trial participants may be unblinded upon study completion. Contact the Alliance Registration Office at [REDACTED] during regular business hours. Upon confirmation by the Primary Statistician (or designee) that all the study completion requirements have been met, the treatment assignment may be unblinded. No Alliance Executive Officer (or designee) approval is required.

7.3 Assessment of patient symptoms, adverse events, and medications

- ### 7.3.1
- Assessment of AI-associated arthralgia and associated joint symptoms will be measured by the change from baseline using data from Modified Brief Pain Inventory for Aromatase Arthralgia (Appendix VIII).

- 7.3.2** Assessment of potential testosterone-associated toxicities will be measured by the Symptom Experience Questionnaire (Appendix V).
- 7.3.3** Assessment of hot flashes will be measured by the Hot Flash Diary (Appendix IV) and the HFRDIS (Appendix VII).
- 7.3.4** Assessment of libido will be measured by the MENQOL, a validated instrument⁷¹ (Appendix IX).
- 7.3.5** Assessment of quality of life will be measured by the POMS (Appendix VI).

8.0 DOSAGE MODIFICATION BASED ON ADVERSE EVENTS

If a patient develops unacceptable toxicity (as determined by the patient and/or attending health care provider) that is determined to potentially be from the study agent (as determined by the attending health care provider), then that toxicity should be recorded on the Adverse Event Form (see section 10) and the medication should be stopped. This includes any grade 2 or worse toxicity from the current version of the CTCAE. These toxicities may be acne, hirsutism, or an unacceptable change in libido. The patient will be asked to continue to complete questionnaires for the entire 6 month period.

→ → Use the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0* unless otherwise specified ← ←			
System Organ Class (SOC)	ADVERSE EVENT	AGENT	ACTION
BASED ON INTERVAL ADVERSE EVENT			
Vascular Disorder	Thromboembolic event (grade ≥ 2)	Testosterone/placebo	Discontinue study agent (by not administering another dose). Patient will continue to complete questionnaires for the 6 month period.
Skin and subcutaneous tissue disorders	Skin reaction to the gel (grade ≥ 2)	Testosterone/placebo	Discontinue study agent (by not administering another dose). Patient will continue to complete questionnaires for the 6 month period.

* Located at http://ctep.cancer.gov/protocolDevelopment/electronic_applications.ctc.htm

9.0 ANCILLARY TREATMENT/SUPPORTIVE CARE

Patients should receive clinically appropriate supportive care while on this study including analgesics. While there is no ban on the use of vitamin D, gabapentin, pregabalin, or antidepressants, these are not recommended treatments for aromatase inhibitor arthralgias. NSAID use is allowed.

10.0 ADVERSE EVENT (AE) REPORTING AND MONITORING

The prompt reporting of adverse events is the responsibility of each investigator engaged in clinical research, as required by Federal Regulations. Adverse events must be described and graded using the terminology and grading categories defined in the NCI's Common Terminology Criteria for Adverse Events (CTCAE), Version 4.0. However, CTCAE v5.0 must be used for serious AE reporting through CTEP-AERS as of April 1, 2018. The CTCAE is available at ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm. The CTCAE is available at <http://ctep.cancer.gov/reporting/ctc.html>. Attribution to protocol treatment for each adverse event must be determined by the investigator and reported on the required forms, using the codes provided.

10.1 Routine Adverse Event Reporting

Adverse event data collection and reporting, which are required as part of every clinical trial are done to ensure the safety of patients enrolled in the studies as well as those who will enroll in future studies using similar agents. Adverse events are reported in a routine manner at scheduled times according to the test schedule in Section 4.0. For this trial, the Adverse Events forms are used for routine AE reporting (see Sections 10.4 and 18.0 for more details).

Adverse events to be graded at each evaluation and pretreatment symptoms/conditions to be evaluated at baseline per CTCAE v4.0 grading unless otherwise stated in the table below:

System Organ Class (SOC)	Adverse event	Baseline	Each evaluation
Vascular Disorder	Thromboembolic event	X	X
Skin and Subcutaneous Tissue	Hirsutism	X	X
	Rash Acneiform Local rash from gel	X	X

Refer to the instructions in the Forms Packet (or electronic data entry screens, as applicable) regarding the submission of late occurring AEs following completion of the Active Monitoring Phase (i.e., compliance with Test Schedule in Section 4.0).

10.2 CTCAE Routine Study Reporting Requirements

- * Combinations of CTCAE Grade & Attribution Required for Routine AE Data Submission on Case Report Forms (CRFs)

Attribution	Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
Unrelated			a	a	a
Unlikely			a	a	a
Possible		a	a, b	a, b	a, b
Probable		a	a, b	a, b	a, b
Definite		a	a, b	a, b	a, b

- a) Adverse Events: Other CRF - Applies to AEs occurring between registration and within 30 days of the patient's last treatment date, or as part of the Clinical Follow-Up Phase.
- b) Adverse Events: Late CRF - Applies to AEs occurring greater than 30 days after the patient's last treatment date.

10.3 Expedited Adverse Event Reporting (CTEP-AERS)

Investigators are required by Federal Regulations to report serious adverse events as defined below. Alliance investigators are required to notify the Alliance Central Protocol Operations Program Office, the Study Chair, and their Institutional Review Board if a patient has an adverse event requiring expedited reporting. All such events must be reported in an expedited manner using the CTEP Adverse Event Reporting System (CTEP-AERS). In the rare occurrence when internet connectivity is lost, a 24-hour notification is to be made to the Alliance Central Protocol Operations Office. Once internet connectivity is restored, the 24-hour notification phoned in must be entered electronically into CTEP-AERS by the original submitter at the site.

CTCAE term (AE description) and grade: The descriptions and grading scales found in the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. The CTCAE is identified and located on the CTEP website at: ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm. All appropriate treatment areas should have access to a copy of the CTCAE. All reactions determined to be “reportable” in an expedited manner must be reported using the Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS).

The Alliance requires investigators to route all expedited adverse event reports through the Alliance Central Protocol Operations Office for Alliance coordinated studies.

Be sure to read this entire protocol section, as requirements are described in both the table and bullet points following the table. Note that the additional instructions or exclusions are protocol specific, and in the case of a conflict, the additional instructions or exclusions supersede the table.

Note: All deaths on study require both routine and expedited reporting regardless of causality. Attribution to treatment or other cause should be provided.

10.3.1 Alliance A221102 Reporting Requirements

Expedited reporting requirements for adverse events that occur on studies under an IND/IDE \leq 30 Days of the last administration of the investigational agent/intervention^{1,2}

FDA REPORTING REQUIREMENTS FOR SERIOUS ADVERSE EVENTS (21 CFR Part 312)

NOTE: Investigators **MUST** immediately report to the sponsor (NCI) **ANY** Serious Adverse Events, whether or not they are considered related to the investigational agent(s)/intervention (21 CFR 312.64)

An adverse event is considered serious if it results in **ANY** of the following outcomes:

- 1) Death
- 2) A life-threatening adverse event
- 3) An adverse event that results in inpatient hospitalization or prolongation of existing hospitalization for \geq 24 hours
- 4) A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- 5) A congenital anomaly/birth defect.
- 6) Important Medical Events (IME) that may not result in death, be life threatening, or require hospitalization may be considered serious when, based upon medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. (FDA, 21 CFR 312.32; ICH E2A and ICH E6).

ALL SERIOUS adverse events that meet the above criteria **MUST** be immediately reported to the NCI via CTEP-AERS within the timeframes detailed in the table below.

Hospitalization	Grade 1 Timeframes	Grade 2 Timeframes	Grade 3 Timeframes	Grade 4 & 5 Timeframes
Resulting in Hospitalization \geq 24 hrs	10 Calendar Days			24-Hour; 5 Calendar Days
Not resulting in Hospitalization \geq 24 hrs	Not required		10 Calendar Days	

Expedited AE reporting timelines are defined as:

- “24-Hour; 5 Calendar Days” - The AE must initially be reported via CTEP-AERS \leq 24 hours of learning of the AE, followed by a complete expedited report \leq 5 calendar days of the initial 24-hour report.
- “10 Calendar Days” - A complete expedited report on the AE must be submitted \leq 10 calendar days of learning of the AE.

¹ Serious adverse events that occur more than 30 days after the last administration of investigational agent/intervention and have an attribution of possible, probable, or definite require reporting as follows:

Expedited 24-hour notification followed by complete report \leq 5 calendar days for:

- All Grade 4, and Grade 5 AEs

Expedited 10 calendar day reports for:

- Grade 2 adverse events resulting in hospitalization or prolongation of hospitalization
- Grade 3 adverse events

Note: All deaths on study require both routine and expedited reporting regardless of causality. Attribution to treatment or other cause should be provided.

- Expedited AE reporting timelines defined:
 - “24 hours; 5 calendar days” – The investigator must initially report the AE via CTEP-AERS \leq 24 hours of learning of the event followed by a complete CTEP-AERS report \leq 5 calendar days of the initial 24-hour report.
 - “10 calendar days” - A complete CTEP-AERS report on the AE must be submitted \leq 10 calendar days of the investigator learning of the event.
- Use the NCI protocol number and the protocol-specific patient ID provided during trial registration on all reports.

Additional Instructions or Exclusions:

- All adverse events reported via CTEP-AERS (i.e., serious adverse events) should also be forwarded to your local IRB.
- Grade 3/4 hematosuppression and hospitalization resulting from such do not require CTEP-AERS, but should be submitted as part of study results.
- Grade 1-3 nausea or vomiting and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 nausea or vomiting does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 diarrhea or constipation and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting
- Grade 3 diarrhea or constipation does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 arthralgias or myalgias and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 arthralgias or myalgias does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 peripheral edema or weight gain and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 peripheral edema or weight gain does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 headache and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 headache does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 peripheral neuropathy and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 peripheral neuropathy does not require AERS reporting, but should be reported via routine AE reporting

- Grade 1-3 vaginal dryness and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 vaginal dryness does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 menopause and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 menopause does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 hot flashes and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 hot flashes does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 fatigue or flu-like symptoms and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 fatigue or flu-like symptoms does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 lymphedema and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 lymphedema does not require AERS reporting, but should be reported via routine AE reporting
- Grade 1-3 radiation dermatitis or radiation recall and hospitalization resulting from such do not require AERS reporting, but should be reported via routine AE reporting.
- Grade 3 radiation dermatitis or radiation recall does not require AERS reporting, but should be reported via routine AE reporting
- Treatment expected adverse events include those listed in Section 15.0.
- All other grade 3, 4, or 5 adverse events that precipitate hospitalization or prolong an existing hospitalization must be reported via CTEP-AERS.
- Death due to progressive disease should be reported as Grade 5 “Disease progression” in the system organ class (SOC) “General disorders and administration site conditions.” Evidence that the death was a manifestation of underlying disease (e.g., radiological changes suggesting tumor growth or progression; clinical deterioration associated with a disease process) should be submitted.
- Any death occurring within 30 days of the last dose, regardless of attribution to the investigational agent/intervention requires expedited reporting within 24 hours.
- Any death occurring greater than 30 days after the last dose of the investigational agent/intervention requires expedited reporting within 24 hours only if it is possibly, probably, or definitely related to the investigational agent/intervention.
- All new malignancies must be reported through CTEP-AERS whether or not they are thought to be related to either previous or current treatment. All new malignancies should be reported, i.e., solid tumors (including non-melanoma skin malignancies), hematologic malignancies, myelodysplastic syndrome /acute myelogenous leukemia, and in situ tumors.

- Secondary Malignancy:

A secondary malignancy is a cancer caused by treatment for a previous malignancy (e.g., treatment with investigational agent/intervention, radiation or chemotherapy). A secondary malignancy is not considered a metastasis of the initial neoplasm.

CTEP requires all secondary malignancies that occur following treatment with an agent under an NCI IND/IDE be reported via CTEP-AERS. Three options are available to describe the event:

- Leukemia secondary to oncology chemotherapy (e.g., acute myelocytic leukemia [AML])
- Myelodysplastic syndrome (MDS)
- Treatment-related secondary malignancy

Any malignancy possibly related to cancer treatment (including AML/MDS) should also be reported via the routine reporting mechanisms outlined in each protocol.

- Second Malignancy:

A second malignancy is one unrelated to the treatment of a prior malignancy (and is NOT a metastasis from the initial malignancy). Second malignancies require ONLY routine reporting unless otherwise specified.

Whenever possible, the CTEP-AERS reports for new malignancies should include tumor pathology, history or prior tumors, prior treatment/current treatment including duration, any associated risk factors or evidence regarding how long the new malignancy may have been present, when and how it was detected, molecular characterization or cytogenetics of the original tumor (if available) and of any new tumor, and new malignancy treatment and outcome, if available.

- Pregnancy loss

- Pregnancy loss is defined in CTCAE as “Death in utero.”
- Any Pregnancy loss should be reported expeditiously, as Grade 4 “Pregnancy loss” under the Pregnancy, puerperium and perinatal conditions SOC.
- A Pregnancy loss should NOT be reported as a Grade 5 event under the Pregnancy, puerperium and perinatal conditions SOC, as currently CTEPAERS recognizes this event as a patient death.

- A neonatal death should be reported expeditiously as Grade 4, “Death neonatal” under the General disorders and administration SOC.
- The reporting of adverse events described above is in addition to, and does not supplant, the reporting of adverse events as part of the reporting of the results of the clinical trial, e.g. routine reporting.

10.4 Contact Information for NCI Safety Reporting

Website for submitting expedited reports	https://eapps-ctep.nci.nih.gov/ctepaers
AEMD Help Desk (for CTEP)*	301-897-7497 Monday through Friday, 7:00 AM to 7:00 PM (US Eastern Time)
Fax for expedited report supporting Medical Documentation for CTEP trials	[REDACTED]

AEMD Help Email:	[REDACTED]
Technical (e.g., IT or computer issues ONLY) Help Phone*	[REDACTED]
CTEP-AERS Technical Help Email	[REDACTED]
CTCAE v4 Help/Questions Email	[REDACTED]
CTEP-AERS FAQs link	https://eapps-ctep.nci.nih.gov/ctepaers/help/webhelp/CTEP-AERS%20FAQ.htm
CTEP-AERS Computer Based Training link	https://betapps-ctep.nci.nih.gov/ctepaers/public/login

*Office phone and fax are accessible 24 hrs per day 7 days a week (The AEMD phone line is staffed from Monday through Friday, 7:00 AM to 7:00 PM ET. Any phone call after these hours will go to voicemail. Please leave contact information and the phone call will be returned the following business day.

10.5 Other Required Expedited Reporting

EVENT TYPE	REPORTING PROCEDURE
Other Grade 4 or 5 Events and/or Any Hospitalizations During Treatment Not Otherwise Warranting an Expedited Report	Complete a Notification Form: Grade 4 or 5 Non-AER Reportable Events/Hospitalization Form electronically via the Remote Data Entry System within 5 working days of the date the clinical research associate (CRA) is aware of the event(s) necessitating the form. If a CTEP-AERS report has been submitted, this form does not need to be submitted.

11.0 TREATMENT EVALUATION

Study endpoints are outlined in section 16.2. Many of the tools are ones that we have commonly used in the symptom intervention program. We will expand, below, information regarding the MENQOL instrument, which we have not previously used.

To measure sexual outcomes and menopause related quality of life, the Menopause Specific Quality of Life Questionnaire, or MENQOL, will be used. The MENQOL was developed in 1996⁷² to fill a void related to menopausal symptoms that other quality of life instruments had. It was initially developed with 29 items, but three additional items were added to the physical subscale in 2005⁷³. There are 4 subscales; sexual, vasomotor, physical and psychosocial. The MENQOL has been used in large US population based studies⁷⁴, has been validated in a breast cancer population⁷⁵ and used in the MA17 trial where it was able to differentiate women receiving letrozole versus placebo on the vasomotor and sexual subscales⁷⁶.

We considered other sex specific outcome measures but decided against these as it was felt these other instruments were too burdensome, asked intimate questions not germane to the purpose of this study, and have not been well validated in breast cancer populations. The MENQOL will provide valid data regarding the secondary endpoints libido, (question 27), sexual function (questions 27-29) and quality of life through the analysis of each subscale.

12.0 DESCRIPTIVE FACTORS

Length of time patients have been on an AI: <6 months vs. 6—12 months vs. >12 months.

13.0 TREATMENT/FOLLOW-UP DECISION AT EVALUATION OF PATIENT

- 13.1 For patients enrolled prior to Update 3 and receiving testosterone pellets: If the patient doesn't get the second dose of the study agent and agrees to complete the remaining questionnaire booklets (up to 6 months), the patient will go to observation.
If the patient discontinues testosterone topical gel prior to 6 months and agree to complete the remaining questionnaire booklets (up to 6 months), the patient will go to observation.
- 13.2 If the patient discontinues the anastrozole or letrozole during first three months, the patient will not receive second dose of testosterone-anastrozole.
If the patient discontinues the anastrozole or letrozole at any time, the patient will discontinue the use of topical testosterone.
- 13.3 For patients enrolled prior to Update 3 and receiving testosterone pellets: If the patient doesn't get the second dose of the study agent and does not agree to complete the remaining questionnaire booklets (up to 6 months), the patient will go off study.
If the patient discontinues testosterone topical gel prior to 6 months and does not agree to complete the remaining questionnaire booklets (up to 6 months), the patient will go off study.
- 13.4 A patient is deemed *ineligible* if, after registration, it is determined that at the time of registration, the patient did not satisfy each and every eligibility criteria for study entry.
The patient may continue protocol treatment, at the discretion of the physician, as long as there are no safety concerns, and the patient was properly registered.
 - If the patient received treatment, all data must be submitted.
 - If the patient never received treatment, on-study material must be submitted.
- 13.5 A patient is deemed a *major violation*, if protocol requirements regarding treatment in cycle 1 of the initial therapy are severely violated that evaluability for primary end point is questionable. All data must be submitted. The patient will go off study. The patient may continue protocol treatment at the discretion of the physician as long as there are no safety concerns, and the patient was properly registered.

13.6 A patient is deemed a *cancel* if he/she is removed from the study for any reason before any study treatment is given. On-study material and the End of Active Treatment/Cancel Notification Form must be submitted. No further data submission is necessary.

14.0 BODY FLUID BIOSPECIMENS

14.1 Specimen Registration and Tracking

USE OF THE ALLIANCE BIOSPECIMEN MANAGEMENT SYSTEM (BioMS) IS MANDATORY AND ALL SPECIMENS MUST BE LOGGED AND SHIPPED VIA THIS SYSTEM.

BioMS is a web-based system for logging and tracking all biospecimens collected on Alliance trials. Authorized individuals may access BioMS at the following URL: <https://bioms.wustl.edu/bioms/login> using most standard web browsers (Safari, Firefox, Internet Explorer). For information on using the BioMS system, please refer to the ‘Help’ links on the BioMS web page to access the on-line user manual, FAQs, and training videos. To report technical problems, such as login issues or application errors, please contact: [REDACTED] For assistance in using the application or questions or problems related to specific specimen logging, please contact: [REDACTED]

After logging collected specimens in BioMS, the system will create a shipping manifest. This shipping manifest must be printed and placed in the shipment container with the specimens.

Summary Table of Body Fluid Biospecimens for This Protocol:

Type of biospecimen to submit	Mandatory or optional	When to collect	Reason for submission (background/methodology section)	Where to find specific details for specimen submission
Blood/blood products	Mandatory	Within 104 days after registration ¹	Genetic Testing (Section 14.4)	Section 14.2

1. Blood draw should be done at the same time as the clinical blood draw, if possible. Will ideally be done ≤ 7 days prior to treatment initiation, but must be done before the end of the 3 month follow-up clinic visit.

14.2 Blood/Blood Products Handling

14.2.1 Kits are required for this study.

- Please log into BioMS, and click on “Kit Requests” to order blood specimen kits.
- Kits will be sent via FedEx® Ground at no additional cost to the participating institutions. **Allow at least two weeks to receive the kits.**
- Kits will not be sent via rush delivery service unless the participating institution provides their own FedEx® account number or alternate billing number for express service. **The Alliance will not cover the cost for rush delivery of kits.**
- Instructions for the collection of samples are included below. Please be sure to use a method of shipping that is secure and traceable. Extreme heat precautions should be taken when necessary.

14.2.2 Collect and process all whole blood according to specific kit instructions and table below.

Summary Table of Research Blood/Blood Products to Be Collected for This Protocol:

Indicate if specimen is mandatory or optional	Collection tube description and/or additive (color of tube top)	Volume to collect per tube (number of tubes to be collected)	Blood product being processed	Within 104 days of study entry (ideally ≤7 days prior to starting testosterone)	Process at site?	Storage/shipping conditions ¹
Mandatory	EDTA (purple)	10 mL (1)	Whole Blood	X	No	Refrigerate/cold pack (DO NOT FREEZE)

- After all samples have been processed according to kit instructions, ship all specimens according to shipping instructions (see Section 14.2.3 for detailed shipping instructions).

14.2.3 Shipping

- All submitted specimens must be labeled with the protocol number (A221102), patient number, patient's initials and date and type of specimen collected (e.g., serum, whole blood).
 - A copy of the shipping manifest produced by BioMS must be printed and placed in the shipment with the specimens.
 - Verify ALL sections of the Blood Specimen Submission Form, Biospecimen Accessioning and Processing (BAP) Blood Specimen Requisition Form (provided in kits), and specimen collection labels are completed and filled in correctly. For Alliance sites, enter information from the Blood Specimen Submission Form into the remote data entry system ≤7 days after specimen collection (see Forms Packet).
 - Specimen must be shipped the same day it is drawn.
 - Ship EDTA tube with a properly prepared cold pack. See kit instructions for specific details for cold pack preparation (i.e., frozen or refrigerated) and proper packing of blood and cold pack to avoid freezing of specimen.
 - Shipment on Monday through Friday by overnight service to assure receipt is encouraged. Do not ship specimens on Saturdays. Samples MUST be sent on the day of collection.
 - The BAP kits will contain a smart shipper label (white barcoded label) affixed to the brown shipping boxes. The smart shipper label is a pre-addressed return label, which replaces the need for an airbill. Shipping costs will be covered by Alliance if this box is used for shipping specimens to BAP.
 - Ship samples to the following address:
- 

[REDACTED]
For questions about blood submission contact:
[REDACTED]
[REDACTED]

- BAP will process specimens according to internal instructions.

14.3 Other Body Fluids Handling

None.

14.4 Study Methodology and Storage Information

Blood/blood product samples will be collected for the following research

A recent manuscript published in the *Journal of Clinical Oncology* looked at genome-wide associations and functional genomic studies in women receiving aromatase inhibitors, in a prospective clinical trial comparing anastrozole with exemestane⁷⁷. Cases were defined as patients with grade 3 or 4 musculoskeletal adverse events or those who discontinued treatment because of a musculoskeletal event within the first 2 years of this prospective MA.27 clinical trial. A nested case control design was utilized, with 2 controls for each case. The study involved a total of 293 cases and 585 controls. Genotyping was performed and a total of 551,358 SNPs were evaluated. Four SNPs were identified on chromosome 14 with the most significant p-values ranging from 10-6 to 10-7. T-cell leukemia 1A was the gene closest to the 4 SNPs. There was further data provided in this manuscript that demonstrated that one of the SNPs created an estrogen response element.

The authors of this manuscript concluded that further research was needed to better understand the genetic associations with musculoskeletal symptoms in patients receiving aromatase inhibitors.

The current trial will study a group of patients who have substantial musculoskeletal problems related to aromatase inhibitors. We will obtain DNA samples on each of the patients on this clinical trial and will plan to evaluate the 4 SNPs which were found to be significantly associated with aromatase inhibitor arthralgias in the above noted, recently-published, clinical trial. We will compare the rates of these 4 SNPs in our protocol population vs. the rates of those in population controls. If we see a much higher incidence of these SNPs in our population, this would further support the data from Ingle⁷⁷.

In addition, the DNA collected in the patients on this clinical trial may be used for looking at other genetic associations with aromatase inhibitor arthralgias that might be discovered by other groups of investigators.

- DNA will be collected from all the consenting patients. SNPs determination will be performed at the Mayo Clinic Genotyping Shared Resource.
- A portion of the DNA will initially be analyzed as described in 14.4.1. According to patient consent information (see Section 6.1.5), remaining DNA will be stored frozen by BAP, until specific analyses are identified. As protocols are developed, they will be presented for Alliance and IRB review and approval. (This collection is part of a general strategy of investigation for the majority of Alliance studies.)

14.5 Return of Genetic Testing Research Results

Because the results generated by the genetic testing included in this section are not currently anticipated to have clinical relevance to the patient or their family members, the genetic results

will not be disclosed to the patients or their physicians.

If, at any time, genetic results are obtained that may have clinical relevance, IRB review and approval will be sought regarding the most appropriate manner of disclosure and whether or not validation in a CLIA-certified setting will be required. Sharing of research data with individual patients should only occur when data have been validated by multiple studies and testing has been done in CLIA-approved laboratories.

15.0 DRUG INFORMATION

Prior to Update #3, patients were treated with a pellet form of testosterone, which was inserted via surgical implant. Effective with the issuance and local IRB approval of Update #3 to this protocol, new patients are to be randomized to the topical form of testosterone. Those patients randomized to the pellet form will complete the study according to the original protocol and will receive their second pellet as planned. Please see sections 15.1-15.3 after the shaded area below for the topical application information.

15.1 Testosterone Pellet – Alliance IND #114707

Testosterone-anastrozole pellets for subcutaneous implantation and placebo pellets for subcutaneous implantation. In addition to the information provided below, more detail is provided as appendix XV.

15.1.1 Background:

Testosterone is the primary endogenous androgen in both men and women. Cells in the testis, ovary, and adrenal cortex synthesize endogenous testosterone. Testosterone is used for the palliative treatment of carcinoma of the breast in postmenopausal women. Testosterone levels decline over the lifespan of both men and women. Androgen production in women declines steeply in the early reproductive years, with the testosterone level for a postmenopausal woman about half the normal level for a healthy, nonpregnant woman. Testosterone pellets for subcutaneous implantation have been used in both men and women to effectively treat symptoms of testosterone deficiency. When implanted subcutaneously, the pellets slowly release the testosterone for a long acting androgenic effect.

Anastrozole is a selective, non-steroidal aromatase inhibitor which significantly lowers serum estradiol concentrations. In men and postmenopausal women, estrogens are mainly derived from the action of the aromatase enzyme, which converts androgens (primarily androstenedione and testosterone) to estrone and estradiol. Anastrozole has been used orally as an adjuvant treatment of post-menopausal women with hormone receptor-positive early breast cancer, first-line treatment of postmenopausal women with hormone receptor-positive or hormone receptor unknown locally advanced or metastatic breast cancer, or treatment of advanced breast cancer in postmenopausal women with disease progression following tamoxifen therapy.

The testosterone-anastrozole subcutaneous implant may be useful in any patient who would benefit from testosterone therapy, but in whom elevated estradiol is not desired or is contraindicated. Absorption of testosterone and anastrozole from the subcutaneous implant occurs via uniform erosion of the pellet's surface providing a near linear release of both active ingredients. There is no first pass hepatic inactivation and all released testosterone and anastrozole are absorbed into the circulation.

15.1.2 Formulation

Testosterone-anastrozole pellets for subcutaneous/subdermal implantation will be compounded to contain 60 mg testosterone and 4 mg anastrozole per pellet, with stearic acid as an inactive ingredient.

15.1.3 Preparation and Storage

Pellets are stored at controlled room temperature 20 - 25°C, 68-77°F. Testosterone is a CIII controlled substance. Storage of the pellets must be in compliance with federal and state regulations, and site policies, and procedures. The pellets must be stored in pharmacies or physician offices with current Controlled Substance Registration Certification.

15.1.4 Administration

The testosterone-anastrozole pellets are implanted subcutaneously through a 5 mm incision in the upper gluteal region under local anesthesia, using a disposable trocar kit. The incision is closed with a steri-strip.

15.1.5 Pharmacokinetics

Testosterone in plasma is 98 percent bound to a specific, testosterone-estradiol binding globulin, and about 2 percent is free. Generally, the amount of binding globulin in the plasma will determine the distribution of testosterone between free and bound forms, and the free testosterone will determine the half-life. About 90 percent of a dose is excreted as glucuronic and sulfuric acid conjugates of testosterone and its metabolites. Approximately 6 percent of each dose is excreted in the feces.

15.1.6 Potential Drug Interactions:

There are no known drug interactions with the implanted testosterone-anastrozole pellet.

15.1.7 Known potential toxicities

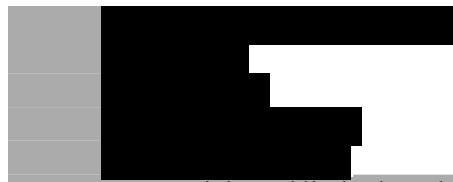
Do not use if the study participant has had previous allergic or adverse reactions to anastrozole or testosterone therapy. Pellet implantation is less flexible for dosage adjustment than oral or intramuscular insertions. If complications arise and the product should be discontinued, the pellets would have to be removed (See Appendix XI).

15.1.8 Study agent procurement

Jungle Jim's Olde Fashioned Pharmacy, Farfield, OH will supply testosterone/anastrozole pellets and placebo pellets to the Alliance research base pharmacy.

Testosterone is a CIII controlled substance. Investigators who prescribe the testosterone-anastrozole pellets must have an up-to-date Controlled Substance Registration Certificate and must be registered to prescribe CIII substances.

Each participating Alliance institution will order a complete course (two doses) of blinded testosterone-anastrozole/placebo pellets from the Alliance Research Base Pharmacy each time a study participant is enrolled. (See Site Ordering Instructions and Order Forms, Appendix XII). Fax the A221102 Clinical Drug Order/Return Form to:



Note: Due to anticipated limited product expiration dating, we will not provide each patient with two doses upon enrollment in the trial. Instead, each participating Alliance institution must order the 2nd dose of blinded testosterone-anastrozole/placebo for each enrolled patient at least one week prior to the next scheduled dose, by faxing a completed A221102 Testosterone-anastrozole/Placebo Re-order Form (see Appendix XII) to the registration office.

Outdated or remaining drug/product should be destroyed on-site per procedures in place at each institution.

15.2 Placebo

A matching placebo will be provided. The placebo pellets will be similar in appearance to the testosterone-anastrozole pellets. The placebo pellets will contain approximately 98% USP cholesterol and 2% stearic acid.

15.3 Nursing Guidelines

- 15.3.1** The pellet implant will be inserted subcutaneously (SC) using local anesthesia in the upper gluteal region. Steri-Strips will be used to close the wound. Instruct the patient to allow the Steri-Strips to wear off or peel off naturally or they can be removed after seven-10 days. To remove them, gently lift the Steri-Strips from the outside edges toward the center of the wound.
- 15.3.2** Instruct the patient to report any skin reaction or signs of infection at insertion site to their physician or study team member.
- 15.3.3** Instruct the patient to report any signs of masculinization or other side effects, although no adverse drug events have been reported in previous related studies as of September 2010.

15.1 Testosterone Topical Gel – Alliance IND #114707

Testosterone gel for topical application and placebo gel for topical application. In addition to the information provided below, more detail is provided as Appendix XVII.

Note: An investigational brochure is not available for the topical testosterone gel formulation as this is a compounded product. Refer to a package insert for a similar product (i.e., AndroGel).

15.1.1 Background

Testosterone has commonly been provided by topical solution.

Testosterone gel was originally developed as a topical delivery for men who have low or no testosterone. Native testosterone is absorbed well from the intestine, but it is metabolized so rapidly by the liver that it is virtually impossible to maintain a normal serum testosterone concentration in a hypogonadal man with oral testosterone. The solutions to this problem that have been developed over many years involve modifying the testosterone

molecule, changing the method of testosterone delivery, or both. Testosterone gels are generally preferred because they typically result in normal and relatively stable serum testosterone concentrations, and most patients prefer them to other preparations.

15.1.2 Formulation

Testosterone gel will contain 10.4mg of testosterone per 0.264ml of gel.

15.1.3 Study agent procurement

When a patient is randomized to the study, the Alliance Research Base Pharmacy will be alerted that a 3 month supply of the study drug is to be sent to the study site upon receipt of an order form. At 2 ½ months, the site will place a reorder of the study drug, using the A221102 Testosterone/Placebo Re-Order Form (found in Appendix XII), and the Alliance Research Base Pharmacy will send the next 3 month supply.

15.1.4 Preparation and Storage

Testosterone gel should be stored at controlled room temperature 20 - 25°C, 68-77°F. Testosterone is a CIII controlled substance. Storage of the gel must be in compliance with federal and state regulations, and site policies, and procedures.

The product will be compounded using the following procedure: For a 100 mL batch, weigh testosterone USP micronized powder (3.939 grams) in an electronic mortar and pestle jar. Tare. Add propylene glycol USP (8 mL) and ethyl alcohol 95% (2 mL). Mix with the electronic mortar and pestle at speed 5 for 1 minute. Tare. Add Versabase®gel (87.57 grams). Mix with the electronic mortar and pestle at speed 5 for 2 minutes. Mill at finest setting. Mix with the electronic mortar and pestle at speed 0 for 6 seconds. Snap an electronic mortar and pestle-to-syringe adapter into the electronic mortar and pestle jar. Remove plunger of an amber oral syringe before filling. Package in Accupen® dispensing device and label. The resulting product delivers approximately 378 doses of testosterone 10.4 mg in 0.264 mL of gel.

Drug and matching placebo will be compounded at Gateway HealthMart Compounding Pharmacy and distributed through the NCCTG Research Pharmacy to NCCTG sites free of charge. The testosterone will be delivered by an Accupen® device which will deliver testosterone 10.4 mg/0.264 mL in two pumps of the device. Each pen will contain 13.5 mL and will dispense 12.5 mL, as approximately 1 mL of product will be left in the pen as residual. This will deliver approximately 47 two pump doses. Two pens will be provided to each patient at baseline and refills will be provided to patients during their visit at the end of month 3 during the 6 month study period.

15.1.5 Administration: Directions for Applying Testosterone Topical Gel

Virilization has been reported in children who were secondarily exposed to testosterone gel. Children should avoid contact with unwashed or unclothed application sites in patients using testosterone gel. Patients should wash hands immediately with soap and water after applying the topical gel and cover the application site(s) with clothing after the gel has dried. Patients should wash the application site thoroughly with soap and water prior to any situation where skin-to-skin contact of the application site with another person is expected. Therefore, patient application instructions have been provided in Appendix XVII.

15.1.6 Pharmacokinetics

Testosterone in plasma is 98 percent bound to a specific, testosterone-estradiol binding globulin, and about 2 percent is free. Generally, the amount of binding globulin in the plasma will determine the distribution of testosterone between free and bound forms, and

the free testosterone will determine the half-life. About 90 percent of a dose is excreted as glucuronic and sulfuric acid conjugates of testosterone and its metabolites. Approximately 6 percent of each dose is excreted in the feces.

15.1.7 Potential Drug Interactions:

Since androgens can decrease glucose concentrations, they may decrease insulin requirements in patients with diabetes.

For patients on vitamin K antagonists, testosterone may alter coagulation, so that this may need to be monitored more.

Concurrent use of testosterone with adrenocorticotropic hormone (ACTH) or corticosteroids may lead to increased fluid retention.

15.1.8 Known potential toxicities

Do not use if the study participant has had previous allergic or adverse reactions to testosterone therapy.

15.2 Placebo

A matching placebo will be provided. The placebo gel will be similar in appearance to the testosterone gel. Since the Versabase® gel base is clear, an agent such as magnesium stearate or lactose will be added to the placebo gel to make it white in appearance to match the testosterone containing gel.

15.3 Nursing Guidelines

- 15.3.1** A specific patient instruction sheet on how to measure out and apply the gel (avoiding the vulvar areas) to maximize surface area and therefore, absorption, is included in the protocol in section 15.1.4 as well as Appendix XVII.
- 15.3.2** Verbally go through the patient instruction sheet.
- 15.3.3** Instruct the patient to report any skin reactions to their physician or study team member.
- 15.3.4** Instruct the patient to report any signs of masculinization or other side effects, although no adverse drug events have been reported in previous related studies as of September 2010.

16.0 STATISTICAL CONSIDERATIONS AND METHODOLOGY

16.1 Study Design

This is a randomized, placebo-controlled, phase III trial evaluating testosterone for the alleviation of aromatase inhibitor induced arthralgia. A parallel group design will be utilized for this two-arm study: topical testosterone vs. placebo.

16.1.1 Study Populations

Intent-to-Treat (ITT) Population: Consists of all randomized patients who had baseline pain measurement. Patients missing the 3 month pain measurement will be imputed by the last value carried forward (LVCF) method to obtain the primary endpoint. The ITT population will be used for the efficacy evaluation at each of the planned analyses. Following the intent-to-treat principle, patients will be analyzed according to the treatment they were assigned at randomization.

Safety Population: Consists of all patients who took any randomized treatment and who have at least one post-baseline safety assessment. The safety population will be used in

the safety data summaries. Note that a patient who had no adverse events (on the Adverse Event CRF) constitutes a safety assessment. Patients who have received at least one dose of study drug but have no post-treatment safety data of any kind would be excluded.

16.2 Study Endpoints

16.2.1 Primary endpoint

The intra-patient change in joint pain at 3 months from baseline as measured by item #3 (average) of the Brief Pain Inventory for aromatase inhibitor arthralgias (BPI-AIA). In addition, we will explore the following:

- The proportion of women with an improvement (reduced pain) of at least 10 points (on a converted 0-100 scale) at 3 months from baseline.
- The intra-patient change in joint pain at 6 months from baseline as measured by item #3 (average) of the Brief Pain Inventory for aromatase inhibitor arthralgias (BPI-AIA).
- The intra-patient changes in joint pain at each month from baseline as measured by the BPI-AIA.

16.2.2 Secondary Endpoints

- 1) The safety and tolerability of testosterone assessed using CTCAE 4.0 and additional questionnaires including:
 - Alopecia as reported by the patient.
 - Acne as reported by the patient.
 - Hirsutism as reported by the patient.
- 2) The intra-patient change in joint pain and its interference of activity for each month from baseline as measured by item #1 (worst), item #2 (least), item #4 (right now) and item #5 (stiffness) item #6A-6G (interference) of the BPI-AIA.
- 3) The change of hot flashes during the first two months from baseline as measured by hot flash diary (appendix IV).
- 4) The change of libido and menopause specific quality of life from baseline as measured by MENQOL and POMS monthly.
- 5) The minor allele and minor allele frequency (MAF) of SNPs in these postmenopausal women with aromatase inhibitor induced arthralgias.

16.2.3 Exploratory Endpoint

- Differential effects of testosterone in subsets defined by race or ethnicity.

16.3 Statistical Analysis

16.3.1 Primary Analysis

The original 0-10 scale in BPI-AIA will be converted to a 0-100 scale with higher value stands for better quality of life. The two-sample *t*-test (or nonparametric Wilcoxon rank-sum test if normality fails) will be applied for comparison of the changes of joint pain at 3 months between testosterone and placebo arms. Multiple regression model will be preferred if more than moderate imbalance between baseline, stratification and other risk factors are found. In addition,

- The Fisher's exact test (or Chi-square test) will be used to compare proportion of women with an improvement (reduced pain) of at least 10 points on a converted 0-100 scale at 3 months from baseline.

- The repeated-measures analysis of variance (RM-ANOVA) model will be used for comparison of the monthly changes of joint pain from baseline.

16.3.2 Secondary Analyses

Because they will be conducted in an exploratory manner, no adjustment of p-value for multiple comparisons from these secondary analyses will be taken. Therefore, interpretation of significant results of secondary analyses shall be interpreted accordingly.

- 1) Descriptive statistics and statistical plot including frequency (percentage) and histogram will be used for summary of safety and tolerability of testosterone and placebo.
- 2) The repeated-measures analysis of variance (RM-ANOVA) model will be used for comparison of the monthly changes from baseline of joint pain, stiffness and its interference with activity between two arms.
- 3) The area-under-the-curve of hot flash score and frequency will be compared by two-sample *t*-test (or nonparametric Wilcoxon rank-sum test if normality fails) between two arms.
- 4) The repeated-measures analysis of variance (RM-ANOVA) model will be used for comparison of the monthly changes from baseline of libido and menopause specific quality of life. The number of months on an aromatase inhibitor may be considered as a covariate in this regression analysis.
- 5) Quality control of the genotypes for the 4 SNPs⁷⁷ will be completed by assessing SNP call rates, Hardy-Weinberg Equilibrium (HWE) and subject call rates prior to exploratory genetic analysis. SNPs/Subjects will be removed if they fall below pre-determined thresholds (SNP and subject call rate of 95%, HWE $p < 0.001$). The minor allele and minor allele frequency (MAF) based on this study will be compared to the MAFs of cases (18.9%, N=293) and controls (10.9%, N=585) found in the MA.27 study⁷⁷, using the Fisher's exact test.

With the planned sample size of 194 patients, we will have approximately 83% power to detect a difference of 8.0% in the estimated MAF (assuming it is 18.9%, the same as cases in MA.27 study) for the postmenopausal women with substantial aromatase inhibitor induced arthralgias in this study and the controls in the MA.27 study with no substantial aromatase inhibitor induced arthralgias at the 5% significance level with a one-sided Fisher's exact test.

16.4 Sample Size and Power Analysis

In the ART 2 trial, the change of joint pain from baseline (at 3 months) measured by the VAS pain instrument were observed as 18.3mm ($SD=29.2mm$) for placebo arm, and 34.8mm ($SD=19.0mm$) for high dose of testosterone arm. The measurement of joint pain by BPI-AIA (item #3) will be converted to a 0-100 scale, a similar but not identical range as that of the VAS pain instrument. We will consider an absolute difference of at least 10 points in the change of joint pain between placebo and testosterone arms as clinically meaningful effect size. Because we do not have extra data on BPI-AIA, we assume the variation of the changes from baseline to 3-month in the BPI-AIA (item #3) will be similar in quantity.

Based on the primary analysis using a two-sided two-sample *t*-test (with unequal variance), we will have 80% power to detect the above effect size with a total sample size of 194 patients (97 patients per arm) at the 5% significance level. This sample size is inflated to a total of 224 patients (112 patients per arm) to account for approximately 15% non-evaluable patients due to ineligibility, cancel or major violations.

16.5 Accrual rate and study duration

We anticipate accruing approximately 20 patients per month. This would mean completing accrual within 12 months from study initiation and completing double-blind data collection 18 months from study initiation.

16.6 Missing data

We will examine the mechanisms of missing data if the proportion of missing assessments is not small ($\geq 5\%$). Graphical presentation, correlation analysis and logistic regression will be performed to examine whether the missing data mechanism depends on the covariates (patient characteristics and other baseline risk factors), observed patient-reported outcome (PRO) scores, and missing PRO scores.

If the missing data mechanism depends on the covariates only (missing completely at random, MCAR) or depends on the covariates and observed PRO scores only (missing at random, MAR)³⁵, we will enhance the primary analysis to incorporate these covariates in the repeated measures model. The repeated measures model is a likelihood-based method that uses all observed data and covariates that explain the missing data mechanism, therefore results in unbiased estimates under MCAR and MAR⁶⁷.

If the missing data mechanism depends on not only the covariates and observed PRO scores, but also the missing PRO (missing not at random, MNAR)⁶⁶, we will explore advanced models that consider both longitudinal PRO assessments and missing data mechanism, such as pattern mixture model⁶⁸ and selection model^{69,70}.

Simple and multiple imputation will be used as part of sensitivity analysis to examine the dependence of the results on specific assumptions about the PRO scores of individuals with missing assessments. Previous experience with imputation in clinical trials have demonstrated that the use of various imputation methods compared to analysis of all available data provides evidence of the degree of robustness of the results relative to the assumptions of the analytical procedure.

16.7 Monitoring

This study will be monitored by the Alliance Data and Safety Monitoring Board (DSMB), an NCI-approved functioning body. Reports containing efficacy, adverse event, and administrative information will be provided to the DSMB every six months as per NCI guidelines.

This study will be monitored by the Clinical Data Update System (CDUS) version 2.0. An abbreviated report containing cumulative CDUS data will be submitted quarterly to CTEP by electronic means. Reports are due January 31, April 30, July 31, and October 31.

16.8 Adverse Event Stopping Rule

The stopping rule specified below is based on the knowledge available at study development. We note that the Adverse Event Stopping Rule may be adjusted in the event of either (1) the study re-opening to accrual or (2) at any time during the conduct of the trial and in consideration of newly acquired information regarding the adverse event profile of the treatment(s) under investigation. The study team may choose to suspend accrual because of unexpected adverse event profiles that have not crossed the specified rule below. CTCAE v4.0 will be used to determine grading for these stopping rules.

Accrual will be temporarily suspended to this study if at any time we observe events considered at least possibly related to study treatment (testosterone)(i.e., an adverse event with attribute specified as “possible”, “probable”, or “definite”) that satisfy the following:

- If 5 or more patients in the first 20 treated patients in the testosterone versus placebo arm (or 25% of all patients after 20 are accrued) experience a grade 2 or higher non-hematologic adverse event, and the toxicity rate is higher in the testosterone arm than the placebo arm.

We note that we will review grade 4 and 5 adverse events deemed “unrelated” or “unlikely to be related”, to verify their attribution and to monitor the emergence of a previously unrecognized treatment-related adverse event.

16.9 Minority Accrual

This study will be available to all eligible patients, regardless of race or ethnic origin. There is no information currently available regarding differential effects of testosterone in subsets defined by race or ethnicity, and there is no reason to expect such differences to exist. Nonetheless, the planned analyses will, as always, look for differences in treatment effect based on racial groupings.

Based on prior studies, we expect about 10% of patients will be classified as minorities by race and 100% of patients will be women in the study accrual. Expected sizes of racial subsets are shown in the following table:

Racial Categories	Ethnic Categories				Total	
	Not Hispanic or Latino		Hispanic or Latino			
	Female	Male	Female	Male		
American Indian/Alaska Native	1	0	0	0	1	
Asian	5	0	0	0	5	
Native Hawaiian or Other Pacific Islander	0	0	0	0	0	
Black or African American	26	0	0	0	26	
White	184	0	8	0	192	
More Than One Race	0	0	0	0	0	
Total	216	0	8	0	224	

Racial Categories: **American Indian or Alaskan Native** – a person having origins in any of the original peoples of North, Central, or South America, and who maintains tribal affiliations or community attachment.

Asian – a person having origins in any of the original peoples of the Far East, Southeast Asia, or the Indian subcontinent including, for example, Cambodia, China, India, Japan, Korea, Malaysia, Pakistan, the Philippine Islands, Thailand, and Vietnam. (Note: Individuals from the Philippine Islands have been recorded as Pacific Islanders in previous data collection strategies.)

Black or African American – a person having origins in any of the black racial groups of Africa. Terms such as “Haitian” or “Negro” can be used in addition to “Black or African American.”

Native Hawaiian or other Pacific Islander – a person having origins in any of the original peoples of Hawaii, Guam, Samoa, or other Pacific Islands.

White – a person having origins in any of the original peoples of Europe, the Middle East, or North Africa.

17.0 PATHOLOGY CONSIDERATIONS/TISSUE BIOSPECIMENS

None

18.0 RECORDS AND DATA COLLECTION PROCEDURES

Copies of forms and a data submission schedule are also available for download from the study page on the Alliance and CTSU Web sites.

18.1 Data collection and submission

Patient questionnaire booklets for A221102 are to be ordered prior to the registration of any patients. Samples of questionnaires/booklets are available in the protocol appendices for reference and IRB submission only. They are not to be used for patient completion. Patient assessment questionnaire/booklets must be given to patients to complete and patients should be instructed to return the booklets to site staff either in person or by mail.

The accompanying tables in Section 18.2 detail the forms completion and submission time points.

18.1.1. Legacy NCCTG

All legacy NCCTG sites will submit electronic CRFs and enter QOL/Patient Assessment Booklet data via the NCCTG Remote Data Entry System.

18.1.2 Sites Not Previously Affiliated with NCCTG

For sites that were not previously affiliated with NCCTG, all paper CRFs and QOL/Patient Assessment Booklets will be forwarded to the following address:



18.2 Submission Timetables

Initial Material(s)

Case Report Form (CRF)	Active-Monitoring Phase (Compliance with Test Schedule Section 4.0)
On-Study Form	≤2 weeks after registration
Baseline Adverse Event Form	Submit ≤2 weeks after registration if withdrawal/refusal occurs prior to beginning protocol therapy
Research Blood Submission Form (for all patients)	Submit ≤104 days after registration. Will be submitted one time only (see section 14.0).
Baseline (Initial) Patient Questionnaire Booklet	≤2 weeks after registration - Patient questionnaire booklet must be used; copies are not acceptable for this submission.
Patient Baseline Hot Flash Diary	≤2 weeks after registration - Patient questionnaire booklet must be used; copies are not acceptable for this submission.
Patient Questionnaire Booklet Compliance Form (Hot Flash Diary)	≤2 weeks after registration - This form must be completed only if the Patient Baseline Hot Flash Diary contains absolutely <u>NO</u> patient provided assessment information.
Patient Questionnaire Booklet Compliance Form	≤2 weeks after registration - This form must be completed only if the questionnaire booklet contains absolutely <u>NO</u> patient provided assessment information

Test Schedule Material(s)

CRF	Active-Monitoring Phase (Compliance with Test Schedule Section 4.0)		
	At each evaluation during treatment	At end of treatment	Observation ⁴ : At each evaluation for patients who discontinue study agent
Nurse/CRA Evaluation/Treatment Form	X ³	X	X ⁴
Adverse Event Form	X	X	X
Nurse/CRA Evaluation/Observation Form			X ⁴
Patient Questionnaire Booklet (Months 1 & 2)	X ¹	X ⁴	X ⁴
Patient Questionnaire – Monthly Booklet (Months 3 – 6)	X		X ^{1,4}
Patient Questionnaire Booklet Compliance Form	X ²		X ^{2,4}
End of Active Treatment/Cancel Notification Form		X	
Notification Form – Grade 4 or 5 Non-AER Reportable Events/Hospitalization Form	At each occurrence (see Section 10.0)		

1. Patient questionnaire booklet **must** be used; copies are not acceptable for this submission.
2. This form must be completed **only** if the Patient Questionnaire Booklet contains absolutely **NO** patient provided assessment information.
3. Complete at each evaluation during Active Treatment (see Section 4.0).
4. Will only be submitted if patient discontinues study agent and is completing booklet questionnaires (see section 13.0).

19.0 BUDGET**19.1** Costs charged to patient: Routine clinical care**19.2** Tests to be research funded: None**19.3** Other budget concerns: Study agent and placebo will be provided to the patient free of charge.

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APPENDIX I: CONSENT FORM

A221102, Randomized Double-Blind Placebo Controlled Study of Testosterone in the Adjuvant Treatment of Postmenopausal Women with Aromatase Inhibitor Induced Arthralgias

This is an important form. Please read it carefully. It tells you what you need to know about this research study. If you agree to take part in this study, you need to sign this form. Your signature means that you have been told about the study and what the risks are. Your signature on this form also means that you want to take part in this study.

This is a clinical trial, a type of research study. Your study doctor will explain the clinical trial to you. Clinical trials include only people who choose to take part. Please take your time to make your decision about taking part. You may discuss your decision with your friends and family. You can also discuss it with your health care team. If you have any questions, you can ask your study doctor for more explanation.

You are being asked to take part in this research study because you were diagnosed with breast cancer, and are receiving a treatment with an aromatase inhibitor (AI), either anastrozole or letrozole (which is standard treatment, not research), and are now experiencing joint symptoms.

Why is this research study being done?

It is well known that treatment with an aromatase inhibitor (AI) can cause side effects that result in symptoms such as joint pain and stiffness, which can interfere with activities of daily living. In some patients, the treatment has to be stopped because of these symptoms.

The purpose of this study is to compare the effects, good and/or bad, of the study agent, testosterone, with a placebo (an inactive agent) on joint pain caused by taking aromatase inhibitors. The testosterone/placebo is a gel that will be applied to the body in non-fat pad areas. In this study, you will receive either the testosterone or placebo. You will not receive both.

The study agent, testosterone, used in this study is considered investigational, which means it has not been approved by the Food and Drug Administration (FDA) for routine clinical use. However, the FDA has allowed the use of this agent for this research study.

How many people will take part in the research study?

About 224 people will take part in this study.

What will happen if I take part in this research study?

Before you begin the study ...

You will be receiving a treatment with an aromatase inhibitor (AI), such as anastrozole or letrozole, for your breast cancer.

You will need to have the following exams, tests or procedures to find out if you can be in the study. These exams, tests or procedures are part of regular cancer care and may be done even if you do not join the study. If you have had some of them recently, they may not need to be repeated. This will be up to your study doctor.

- Medical history and physical exam, including height and weight and rating of how well you perform activities of daily living.
- Body Mass Index (BMI) measurement (height and weight).
- Routine blood tests (liver tests and other tests your doctor thinks should be done). About 2 teaspoons of blood will be drawn from a vein in your arm for the blood tests.

During the study...

If the exams, tests and procedures show that you can be in the study, and you choose to take part, then you will need the following tests and procedures:

- Complete a booklet of questionnaires (about 15 - 30 minutes). This booklet contains several sets of questionnaires that will ask about possible side effects, and other related symptoms such as joint aches, pains, and menopausal symptoms. You will complete this booklet of questionnaires when you are first enrolled in the study and then monthly for 6 months after you start the study treatment.
- You will also complete a hot flash daily diary every day for one week before you start study treatment and every day for 2 months after you start study treatment. You will be asked about the number and intensity or severity of your hot flashes over each 24 hour period.
- Research blood tests. This research blood test will only be obtained one time and is required. Ideally, the blood will be drawn after you go on the study and within 7 days before starting treatment, but must be done before the end of your 3 month follow-up exam. This may be done at the same time as routine blood tests that your doctor has requested. Two (2) teaspoons of blood will be drawn for research purposes. This amount will be in addition to how much blood your doctor needs. You will have the option of allowing a portion of this blood sample to be used in future research. Your options are described later in this form. This research test will look at genetic material. Because the genetic tests in this study are not used for regular medical care, you will not be told the results of the test. The test results will not be put in your medical record either.

The following will be done at your month 3 and month 6 exam visits and is part of your regular cancer care.

- Medical history and physical exam, including height and weight and rating of how well you perform activities of daily living.

You will be "randomized" into one of the study groups described below. Randomization means that you are put into a group by chance, as in the flip of a coin. A computer program will place you in one of the study groups. Neither you nor your doctor can choose the group you will be in. You will have an equal chance of being placed in any group.

If you are in group 1

During the first week of the study, you will not receive any study medication. You will complete a daily diary this week. The daily diary should not take longer than 5 minutes to complete each day. The daily diary will ask you about the number and intensity or severity of your hot flashes over each 24 hour period.

Starting the second week of the study, you will receive the study agent, testosterone, as a topical gel that will be applied using a pen dispenser to non-fat pad areas of the body, including tops of the feet, top of the chest, behind the knees, or inner forearms. The location of the body should be changed with each application and should never be applied to open sores, wounds, or irritated skin. Instructions for using the pen dispenser will be given to you by the study nurse. You will continue to complete the daily diary for 2 months. After the first 2 months, you will no longer complete the daily diary. You will complete a booklet of questionnaires at the end of each month for a total of

6 months. The monthly questionnaire should not take longer than 15 – 30 minutes to complete. This questionnaire will ask about possible side effects, and other related symptoms such as joint aches and pains. You will get a pre-addressed envelope to return your questionnaires to your healthcare provider.

If you are in group 2

During the first week of the study, you will not receive any study placebo. You will complete a daily diary this week. The daily diary should not take longer than 5 minutes to complete each day. The daily diary will ask you about the number and intensity or severity of your hot flashes over each 24 hour period.

Starting the second week of the study, you will receive the placebo as a topical gel that will be applied using a pen dispenser to non-fat pad areas of the body, including tops of the feet, top of the chest, behind the knees, or inner forearms. The location of the body should be changed with each application and should never be applied to open sores, wounds, or irritated skin. Instructions for using the pen dispenser will be given to you by the study nurse. You will complete the daily diary for 2 months. After this first 2 months, you will no longer complete the daily diary. You will complete a booklet of questionnaires at the end of each month for a total of 6 months. The monthly questionnaire should not take longer than 15 – 30 minutes to complete. This questionnaire will ask about possible side effects, and other related symptoms such as joint aches and pains. You will get a pre-addressed envelope to return your questionnaires to your healthcare provider.

Study Schedule

Months	Testosterone/Placebo	Questionnaires
Pre-Treatment Week	No study testosterone/placebo this week	Daily Hot Flash Diary, Return questionnaires at end of the week
1-3	Start applying testosterone/placebo daily	Complete Daily Hot Flash Diary for 2 months and also monthly questionnaire, Return questionnaires at the end of each month
End of Month 3	Continue applying topical testosterone/placebo	Clinic visit and return questionnaires
4-6	Continue applying topical testosterone/placebo	Monthly questionnaire, Return questionnaires at the end of each month

Someone from the study team will call you at the end of week 2 to see how you are doing and to answer questions. You will also be called monthly (except for the months when you are seen at the clinic) for 6 months to see how you are doing and to answer questions.

If during the study you have to stop taking your AI (anastrozole or letrozole), you will discontinue testosterone application, but we ask that you continue to complete the questionnaires for the full 6 months.

How long will I be in the research study?

You will be in the study for 6 months.

Can I stop being in the research study?

Yes. You can decide to stop at any time. Tell the study doctor or nurse if you are thinking about stopping or decide to stop.

It is important to tell the study doctor if you are thinking about stopping to discuss what follow-up care and testing could be most helpful for you.

The study doctor may stop you from taking part in this study at any time if he/she believes it is in your best interest; if you do not follow the study rules; or if the study is stopped.

What side effects or risks can I expect from being in the research study?

You may have side effects while on the study. Everyone taking part in the study will be watched carefully for any side effects. However, doctors don't know all the side effects that may happen. Side effects may be mild or very serious. Your health care team may give you medicines to help lessen side effects. Many side effects go away soon after you stop taking the study agent. In some cases, side effects can be serious, long lasting, or may never go away. There also is a risk of death.

You should talk to your study doctor about any side effects that you have while taking part in the study. Risks and side effects related to the testosterone include those which are:

Likely

- Increase in oily skin
- Acne

Less Likely

- Facial hair
- Deepening of the voice
- Fluid retention
- Increase in blood pressure
- Changes in liver function
- Increase in cholesterol or lipids
- Increase in blood sugar

Rare but serious

- Blood clot

In a study of a similar product, 2% of the patients reported a rash on the area where the gel had been applied.

There is a theoretical risk that administration of testosterone could stimulate tumor growth.

There are potential drug interactions between testosterone and the following drugs: cyclosporine, anticoagulants, or insulin. Therefore you will not be eligible to start this study if you are receiving any of them. If you need to start them after starting the study, which is not very likely if you are not on such drugs now, your physician can advise you.

You should wash hands immediately with soap and water after applying the gel and cover the area with clothing after the gel has dried. You should also wash the area thoroughly with soap and water if you are

going to have skin-to-skin contact with another person. Children should avoid contact with areas of the body that are unwashed or that are not covered with clothes.

As with any medication, allergic reactions are a possibility.

The risks of drawing blood include pain, bruising or rarely infection at the needle site.

Some questions you will be asked to answer in the study questionnaire(s) may make you feel uncomfortable. You may choose not to answer any questions that make you feel uncomfortable.

For more information about risks and side effects, ask your study doctor.

Are there benefits to taking part in the research study?

Taking part in this study may or may not make your health better. While doctors hope the testosterone will be useful in lessening the joint pain caused by aromatase inhibitor therapy, there is no proof of this yet. We do know that the information from this study will help doctors learn more about using testosterone for the joint pain caused by aromatase inhibitors. This information could help future cancer patients or patients with joint pain.

What other choices do I have if I do not take part in this research study?

You do not have to be in this study to receive treatment for your joint pain.

Your other choices may include:

- Getting treatment or care for your joint pain without being in a study
- Taking part in another study
- Getting no treatment

Talk to your doctor about your choices before you decide if you will take part in this study.

Will my medical information be kept private?

We will do our best to make sure that the personal information in your medical record will be kept private. However, we cannot guarantee total privacy. Your personal information may be given out if required by law. If information from this study is published or presented at scientific meetings, your name and other personal information will not be used.

Organizations that may look at and/or copy your medical records for research, quality assurance, and data analysis include:

- Alliance Researchers
- The National Cancer Institute (NCI) and other government agencies, like the Food and Drug Administration (FDA), involved in keeping research safe for people
- The Cancer Trials Support Unit (CTSU), a service sponsored by the National Cancer Institute (NCI) to provide greater access to cancer trials

[Note to Local Investigators: The NCI has recommended that HIPAA regulations be addressed by the local institution. The regulations may or may not be included in the informed consent form depending on local institutional policy.]

What are the costs of taking part in this research study?

The study agents, testosterone and placebo, will be provided free of charge while you are taking part in this study.

You will not need to pay for tests and procedures which are done just for this research study. This test is:

- Research blood test(s)

However, you or your insurance company will need to pay for blood tests that are used for routine clinical care.

You and/or your health plan/ insurance company will need to pay for some or all of the costs of treating your cancer in this study. Some health plans will not pay these costs for people taking part in studies. Check with your health plan or insurance company to find out what they will pay for. Taking part in this study may or may not cost your insurance company more than the cost of getting regular cancer treatment.

You will not be paid for taking part in this study.

For more information on clinical trials and insurance coverage, you can visit the National Cancer Institute's Web site at <http://cancer.gov/clinicaltrials/understanding/insurance-coverage>. You can print a copy of the "Clinical Trials and Insurance Coverage" information from this Web site.

Another way to get the information is to call 1-800-4-CANCER (1-800-422-6237) and ask them to send you a free copy.

What happens if I am injured because I took part in this research study?

It is important that you tell your study doctor, _____ [investigator's name(s)], if you feel that you have been injured because of taking part in this study. You can tell the doctor in person or call him/her at _____ [telephone number].

You will get medical treatment if you are injured as a result of taking part in this study. You and/or your health plan will be charged for this treatment. The study will not pay for medical treatment.

What are my rights if I take part in this research study?

Taking part in this study is your choice. You may choose either to take part or not to take part in the study. If you decide to take part in this study, you may leave the study at any time. No matter what decision you make, there will be no penalty to you and you will not lose any of your regular benefits. Leaving the study will not affect your medical care. You can still get your medical care from our institution.

We will tell you about new information or changes in the study that may affect your health or your willingness to continue in the study.

In the case of injury resulting from this study, you do not lose any of your legal rights to seek payment by signing this form.

Who can answer my questions about the research study?

You can talk to your study doctor about any questions or concerns you have about this study. Contact your study doctor _____ [name(s)] at _____ [telephone number].

For questions about your rights while taking part in this study, call the _____ [name of center] Institutional Review Board (a group of people who review the research to protect your rights) at _____ [telephone number]. [Note to Local Investigator: Contact information for patient

representatives or other individuals in a local institution who are not on the IRB or research team but take calls regarding clinical trial questions can be listed here.]

Please note: This section of the informed consent form is about additional research studies that are being done with people who are taking part in the main study. You may take part in these additional studies if you want to. You can still be a part of the main study even if you say ‘no’ to taking part in any of these additional studies.

You can say “yes” or “no” to each of the following studies. Please mark your choice for each study.

About Using Blood for Research

You have had blood drawn as part of this research study. We would like to keep some of the blood samples that are left over for future research. If you agree, this blood will be kept and may be used in research to learn more about cancer and other diseases. Your blood may be helpful for research whether you do or do not have cancer.

The blood will be sent to laboratories associated with Alliance, where the tests will be done. The results of these tests will not be sent to you or your study doctor and will not be used in planning your care. These tests are for research purposes only and you will not have to pay for them.

Things to Think About

The choice to let us keep the left over blood for future research is up to you. No matter what you decide to do, it will not affect your care.

If you decide now that your blood can be kept for research, you can change your mind at any time. Just contact us and let us know that you do not want us to use your blood. Then any blood that remains will no longer be used for research.

In the future, people who do research may need to know more about your health. While Alliance may give them reports about your health, it will not give them your name, address, phone number, or any other information that will let the researchers know who you are.

Sometimes blood is used for genetic research (about diseases that are passed on in families). Even if your blood is used for this kind of research, the results will not be put in your health records.

Your blood will be used only for research and will not be sold. The research done with your blood may help to develop new products in the future.

Benefits

The benefits of research using blood samples include learning more about what causes cancer and other diseases, how to prevent them, and how to treat them.

Risks

The greatest risk to you from the use of your blood sample is the possible release of information from your health records. We will do our best to make sure that your personal information will be kept private. The chance that this information will be given to someone else is very small.

Making Your Choice

Please read each sentence below and think about your choice. After reading each sentence, mark "Yes" or "No". If you have any questions, please talk to your doctor or nurse, or call our research review board at the IRB's phone number.

No matter what you decide to do, it will not affect your care.

1. My blood sample(s) may be kept for use in research to learn about, prevent, or treat cancer.

Yes No Please initial here: _____ Date: _____

2. My blood sample(s) may be kept for use in research to learn about, prevent or treat other health problems (for example: diabetes, Alzheimer's disease, or heart disease).

Yes No Please initial here: _____ Date: _____

If you want your sample(s) destroyed at any time, contact your study doctor.

The Alliance has the right to end storage of the sample(s) without telling you.

The sample(s) will be stored at the Research Base at Mayo Clinic in Rochester, MN. Outside researchers may one day ask for a part of your sample(s) for studies now or future studies.

How do outside researchers get the sample?

Researchers from universities, hospitals, and other health organizations do research using blood and tissue. They may contact the Alliance Research Base and ask for samples for their studies. The Alliance looks at the way that these studies will be done, and decides if any of the samples can be used. The Alliance sends the samples and some information about you to the researcher. The Alliance will not send your name, address, phone number, social security number, or any other identifying information to the researcher. If you allow your sample(s) to be given to outside researchers, it will be given to them with a code number. If researchers outside the Alliance use the sample(s) for future research, they will decide if you will be contacted and, if so, they would have to contact the researchers at the Alliance. Then the Alliance will contact the clinic where you registered for this study, who will contact you.

Please read the following statements and mark your choice:

3. I permit Alliance to give my sample(s) to outside researchers:

Yes No Please initial here: _____ Date: _____

Where can I get more information?

You may visit the NCI Web site at <http://cancer.gov/> for more information about studies or general information about cancer. You may also call the NCI Cancer Information Service to get the same information at: 1-800-4-CANCER (1-800-422-6237).

A description of this clinical trial will be available on <http://www.ClinicalTrials.gov>, as required by U.S. Law. This Web site will not include information that can identify you. At most, the Web site will include a summary of the results. You can search this Web site at any time.

You will get a copy of this form. If you want more information about this study, ask your study doctor.

I have been given a copy of all _____ [insert total of number of pages] pages of this form. I have read it or it has been read to me. I understand the information and have had my questions answered. I agree to take part in this study.

Printed Participant Name: _____

Participant Signature: _____

Date: _____

Printed name of person obtaining informed consent:

Signature of person obtaining informed consent:

Date _____

Local IRB changes to this document are allowed. Sections "What are the risks of the research study" or "What other choices do I have if I don't take part in this research study?" should always be used in their entirety if possible. Editorial changes to these sections may be made as long as they do not change information or intent. If the institutional IRB insists on making deletions or more substantive modifications to these sections, they may be justified in writing by the investigator and approved by the IRB. Under these circumstances, the revised language and justification must be forwarded to the Alliance Central Protocol Operations Office for approval before a patient may be registered to this study.

Consent forms will have to be modified for each institution as it relates to where information may be obtained on the conduct of the study or research subject. This information should be specific for each institution.

APPENDIX II: PATIENT INFORMATION SHEETS

(Initial Booklet - before baseline week)
PATIENT INFORMATION SHEET
Patient Completed Quality of Life Booklet

You have been given a booklet to complete for this study. The booklet contains some questions to help us better understand how the treatment you are receiving is affecting the way you feel.

1. The booklet contains 5 sets of questions you will complete when you go on this study, before you start the study treatment.
2. Directions on how to complete each set of questions are written on the top of each set.
3. **You will complete the**
 - a. **Symptom Experience Questionnaire**
 - b. **Profile of Mood States (POMS)**
 - c. **Hot Flash Related Daily Interference Scale (HFRDIS)**
 - d. **Modified Brief Pain Inventory for Aromatase Inhibitor Induced Arthralgia**
 - e. **Menopause Specific Quality of Life**
4. Please return this booklet to your study staff when you are done.

We would like to thank you for taking the time to help us.

(Baseline-WEEK 1)
PATIENT INFORMATION SHEET
Patient Completed Quality of Life Booklet

You have been given a booklet to complete for this study. The booklet contains some questions to help us better understand how the treatment you are receiving is affecting the way you feel.

You will not receive any study medication this week.

1. The booklet contains sets of questions you will complete the first week on study, before you start the study treatment.
2. Directions on how to complete each set of questions are written on the top of each set.
3. **Every day during the first week you will complete the Hot Flash Diary**
 - a. The Hot Flash Diary is very important for this study and should be completed daily.
 - b. On page 2 of this booklet, you will find examples of the different hot flash intensities: mild, moderate, severe, and very severe hot flash. This is to help you to decide the intensity of your hot flash, but it is not an absolute rule. Your hot flashes may differ in some ways or may fall just between two descriptions. Try to get as close as you can, but do not worry if your hot flashes do not match exactly what is given.
 - c. It has been helpful for some patients to carry a small notebook and pen with them to record their hot flashes during the day, then to sit down every evening, and transfer their numbers to the diary. Keeping track on a small notebook or paper during the day is the best way to more precisely keep track of the number of hot flashes you are having.
 - d. Write any comments on the bottom of the diary if you wish.
4. It is very important that you return the booklet to us, whether you finish the study or not.
5. **At the end of week 1:** Return the booklet that you completed in the envelope provided.

We would like to thank you for taking the time to help us.

(Treatment Booklet for the FIRST 2 months)

PATIENT INFORMATION SHEET
Patient Completed Quality of Life Booklet

You have been given a booklet to complete for this study. The booklet contains some questions to help us better understand how the treatment you are receiving is affecting the way you feel.

1. The booklet contains a Hot Flash Diary that you will complete every day during the month. It also contains 5 sets of questions you will complete at the end of the month.
2. Directions on how to complete each set of questions are written on the top of each set.
3. **Every day during the month you will complete the Hot Flash Diary**
 - a. The Hot Flash Diary is very important for this study and should be completed daily.
 - b. On page 2 of this booklet, you will find examples of the different hot flash intensities: mild, moderate, severe, and very severe hot flash. This is to help you to decide the intensity of your hot flash, but it is not an absolute rule. Your hot flashes may differ in some ways or may fall just between two descriptions. Try to get as close as you can, but do not worry if your hot flashes do not match exactly what is given.
 - c. It has been helpful for some patients to carry a small notebook and pen with them to record their hot flashes during the day, then to sit down every evening, and transfer their numbers to the diary. Keeping track on a small notebook or paper during the day is the best way to more precisely keep track of the number of hot flashes you are having.
 - d. Write any comments on the bottom of the diary if you wish.
4. **At the end of the month, you will complete the:**
 - a. Symptom Experience Questionnaire
 - b. Profile of Mood States (POMS)
 - c. Hot Flash Related Daily Interference Scale (HFRDIS)
 - d. Modified Brief Pain Inventory for Aromatase Inhibitor Induced Arthralgia
 - e. Menopause Specific Quality of Life
5. It is very important that you return the booklet to us, whether you finish the study or not.
6. **At the end of the month:** Return the booklet that you completed in the envelope provided.

We would like to thank you for taking the time to help us.

(Treatment Booklet Monthly AFTER first 2 months)

PATIENT INFORMATION SHEET
Patient Completed Quality of Life Booklet

You have been given a booklet to complete for this study. The booklet contains some questions to help us better understand how the treatment you are receiving is affecting the way you feel.

1. The booklet contains 5 sets of questions you will complete at the end of the month.
2. Directions on how to complete each set of questions are written on the top of each set.
3. **At the end of the month, you will complete the:**
 - a. Symptom Experience Questionnaire
 - b. Profile of Mood States (POMS)
 - c. Hot Flash Related Daily Interference Scale (HFRDIS)
 - d. Modified Brief Pain Inventory for Aromatase Inhibitor Induced Arthralgia
 - e. Menopause Specific Quality of Life
4. It is very important that you return the booklet to us, whether you finish the study or not.
5. **At the end of the month:** Return the booklet that you completed in the envelope provided.

We would like to thank you for taking the time to help us.

APPENDIX III: PATIENT INFORMATION SHEET FOR HOT FLASH DIARY**Hot Flash Definitions for the Female Patient
Patient Information Sheet**

Please refer to these examples of hot flashes that have been given by cancer survivors in previous studies when describing their hot flash severity. One or more of these descriptions may help to categorize your hot flash as mild, moderate, severe, or very severe.

Mild

- Physical symptoms: Warmth, felt uncomfortable, red face
Emotional symptoms: Not expected
Action needed: Usually no action taken

Moderate

- Physical symptoms: Head, neck, ears, or whole body felt warm; tense, tight muscles; clammy (wet) skin; a change in heart rate or rhythm (heart speeds up or changes beat); some sweating; dry mouth
Emotional symptoms: Felt irritated, felt agitated (restless), felt as though energy was drained out, felt embarrassed when having a hot flash in front of others, felt tired, felt annoyed
Action needed: Needed to use a fan, awakened sometimes at night, needed to uncover, took off layers of clothing, drank water, opened the windows even when cold outside, lighter clothing

Severe

- Physical symptoms: Warmth, sometimes described as a raging furnace or burning up; a change in heart rate or rhythm (heart speeds up or changes beat); felt faint; headache; severe sweating; weakness, a prickling, stinging sensation over skin; chest heaviness
Emotional symptoms: Embarrassment, anxiety, feelings of having a panic attack
Action needed: Needed to stop what was being done at that time, usually awakened at night and removed covers, needed to remove clothes, opened windows, kept the house a cool temperature, frequently used fans

Very Severe

- Physical symptoms: Boiling heat, rolling sweat, difficulty breathing, felt faint, felt dizzy, feet and/or legs cramping, a change in the heart rate or rhythm (heart speeds up or changes beat), felt slightly sick to stomach
Emotional symptoms: Felt distressed, had the urge to escape, had difficulty functioning
Action needed: Awakened frequently at night, needed to change sheets and pajamas, needed to take a cold shower, needed to hold ice on skin

APPENDIX IV: PATIENT QUESTIONNAIRE -HOT FLASH DIARY**Hot Flash Diary****Directions:**

The hot flash diary is divided into 7 sections, one for each day of your week.

1. Write the date your week started in the spaces provided.
2. Write the days of the week in the top row of each section.
3. Every day, fill out the diary for that day.
 - In the # (number) column, write in the number of mild, moderate, severe and very severe hot flashes you had that day in the box next to the hot flash type. Please enter a '0' (zero) if you had no hot flashes of a specific type.

See page 2 of this booklet for the definitions of each type of Hot Flashes.

Day: <i>Tuesday</i>	
#	Type
0	mild
2	moderate
3	severe
0	very severe

Date week started: / /
month **day** **year**

| Day: | |
|------|----------------|------|----------------|------|----------------|------|----------------|------|----------------|------|----------------|------|----------------|
| # | Type |
| | mild |
| | moderate |
| | severe |
| | very
severe |

*A day should be considered to be a 24-hour period (i.e. 7 a.m. to 7 a.m. or midnight to midnight)

APPENDIX V: PATIENT QUESTIONNAIRE -SYMPTOM EXPERIENCE QUESTIONNAIRE**Symptom Experience Questionnaire**

INSTRUCTIONS: Please complete at the end of each study month by circling the one number for each item below that best describes you.

1. Over the past week, have you experienced stomach pain or cramps?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

2. Over the past week, have you experienced diarrhea (loose stools)?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

3. Over the past week, have you experienced nausea?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

4. Over the past week, did you experience dizziness?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

5. Over the past week, did you experience an undesirable decrease in your appetite?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

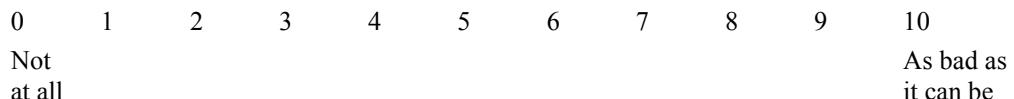
6. Over the past week, did you experience fatigue?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

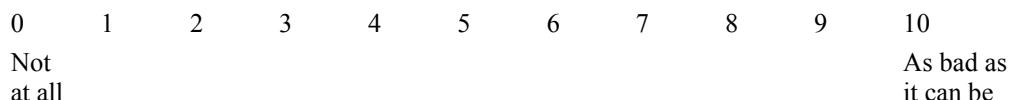
7. Over the past week, did you experience abnormal sweating?

0	1	2	3	4	5	6	7	8	9	10
Not at all										As bad as it can be

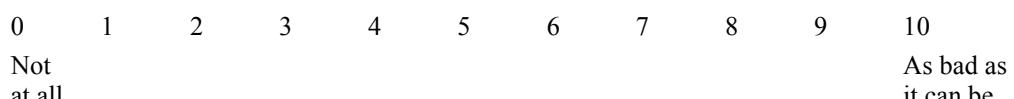
8. Over the past week, did you experience trouble sleeping?



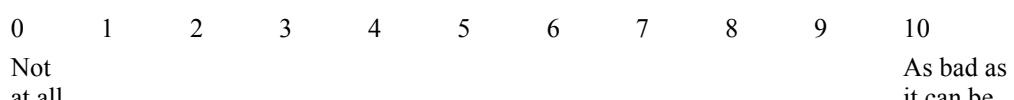
9. Over the past week, did you experience negative mood changes?



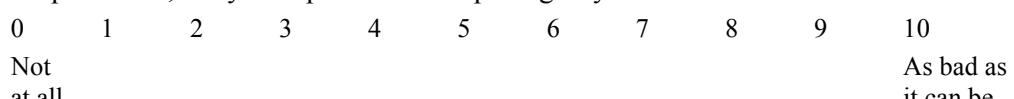
10. Over the past week, have you had trouble concentrating?



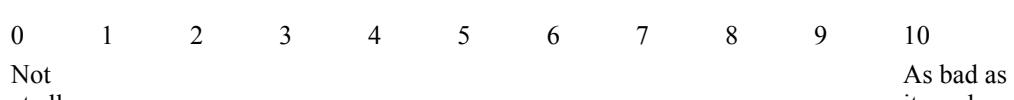
11. Over the past week, how distressing are hot flashes?



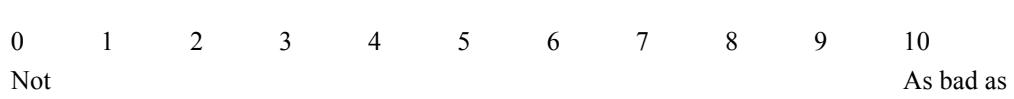
12. Over the past week, did you experience a deepening of your voice?



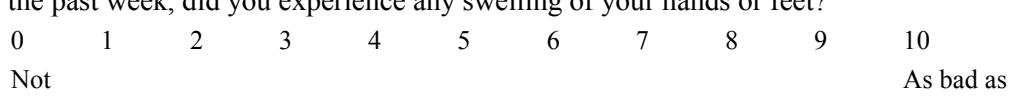
13. Over the past week, have you experienced unwanted weight gain?



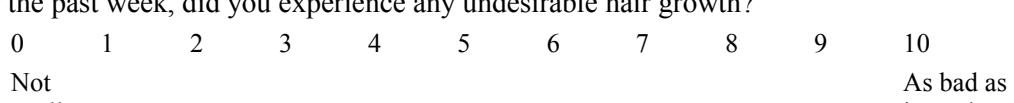
14. Over the past week, have you had any acne-type rash?



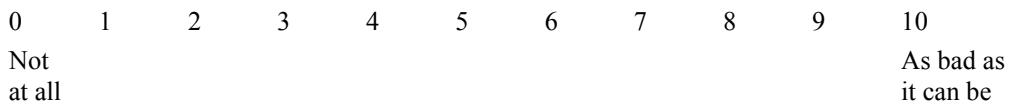
15. Over the past week, did you experience any swelling of your hands or feet?



16. Over the past week, did you experience any undesirable hair growth?



17. Over the past week, did you experience any undesirable scalp hair loss (male pattern baldness)?



18. Did you experience any other symptoms this past week that were not mentioned above?

No _____ Yes _____

If so, please describe _____

APPENDIX VI: PATIENT QUESTIONNAIRE -PROFILE OF MOOD STATES (POMS)**Profile of Mood States (POMS)**

NAME _____	DATE _____																																																												
SEX: Male <input type="radio"/> Female <input type="radio"/>	Identification No. _____																																																												
Below is a list of words that describe feelings people have. Please read each one carefully. Then fill in ONE circle under the answer to the right which best describes HOW YOU HAVE BEEN FEELING DURING THE PAST WEEK INCLUDING TODAY.																																																													
The numbers refer to these phrases.																																																													
① = Not at all ② = A little ③ = Moderately ④ = Quite a bit ⑤ = Extremely																																																													
<table style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: center; width: 33.33%;">Not at all</th> <th style="text-align: center; width: 33.33%;">A little</th> <th style="text-align: center; width: 33.33%;">Moderately</th> <th style="text-align: center; width: 33.33%;">Quite a bit</th> <th style="text-align: center; width: 33.33%;">Extremely</th> </tr> </thead> <tbody> <tr> <td style="text-align: center;">1. Tense</td> <td style="text-align: center;">12. Uneasy</td> <td style="text-align: center;">23. Weary</td> <td style="text-align: center;">Not at all</td> <td style="text-align: center;">Not at all</td> </tr> <tr> <td style="text-align: center;">2. Angry</td> <td style="text-align: center;">13. Fatigued</td> <td style="text-align: center;">24. Bewildered</td> <td style="text-align: center;">A little</td> <td style="text-align: center;">A little</td> </tr> <tr> <td style="text-align: center;">3. Worn out</td> <td style="text-align: center;">14. Annoyed</td> <td style="text-align: center;">25. Furious</td> <td style="text-align: center;">Moderately</td> <td style="text-align: center;">Moderately</td> </tr> <tr> <td style="text-align: center;">4. Lively</td> <td style="text-align: center;">15. Discouraged</td> <td style="text-align: center;">26. Efficient</td> <td style="text-align: center;">Quite a bit</td> <td style="text-align: center;">Quite a bit</td> </tr> <tr> <td style="text-align: center;">5. Confused</td> <td style="text-align: center;">16. Nervous</td> <td style="text-align: center;">27. Full of pep</td> <td style="text-align: center;">Extremely</td> <td style="text-align: center;">Extremely</td> </tr> <tr> <td style="text-align: center;">6. Shaky</td> <td style="text-align: center;">17. Lonely</td> <td style="text-align: center;">28. Bad-tempered</td> <td colspan="2"></td> </tr> <tr> <td style="text-align: center;">7. Sad</td> <td style="text-align: center;">18. Muddled</td> <td style="text-align: center;">29. Forgetful</td> <td colspan="2"></td> </tr> <tr> <td style="text-align: center;">8. Active</td> <td style="text-align: center;">19. Exhausted</td> <td style="text-align: center;">30. Vigorous</td> <td colspan="2"></td> </tr> <tr> <td style="text-align: center;">9. Grouchy</td> <td style="text-align: center;">20. Anxious</td> <td colspan="3"></td> </tr> <tr> <td style="text-align: center;">10. Energetic</td> <td style="text-align: center;">21. Gloomy</td> <td colspan="3"></td> </tr> <tr> <td style="text-align: center;">11. Unworthy</td> <td style="text-align: center;">22. Sluggish</td> <td colspan="3"></td> </tr> </tbody> </table>		Not at all	A little	Moderately	Quite a bit	Extremely	1. Tense	12. Uneasy	23. Weary	Not at all	Not at all	2. Angry	13. Fatigued	24. Bewildered	A little	A little	3. Worn out	14. Annoyed	25. Furious	Moderately	Moderately	4. Lively	15. Discouraged	26. Efficient	Quite a bit	Quite a bit	5. Confused	16. Nervous	27. Full of pep	Extremely	Extremely	6. Shaky	17. Lonely	28. Bad-tempered			7. Sad	18. Muddled	29. Forgetful			8. Active	19. Exhausted	30. Vigorous			9. Grouchy	20. Anxious				10. Energetic	21. Gloomy				11. Unworthy	22. Sluggish			
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POMS-B, by Douglas M. McNair, Ph.D., Joan Lorr Ph.D., Leo F. Droppleman, Ph.D. Copyright © 2003, 1989, Douglas M. McNair, Ph.D., Joan Lorr, Ph.D., Leo F. Droppleman, Ph.D., under exclusive license to Multi-Health Systems Inc. All rights reserved. In the USA, P.O. Box 950, North Tonawanda, NY 1420-0950, 1-800-456-3003. In Canada, 3770 Victoria Park Ave., Toronto, ON M2H 3M6, 1-800-268-6011. Internationally, +1-416-492-2627. Fax, +1-416-492-3343.																																																													
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APPENDIX VII: PATIENT QUESTIONNAIRE -HOT FLASH RELATED DAILY INTERFERENCE SCALE**HOT FLASH RELATED DAILY INTERFERENCE SCALE (HFRDIS)**

Please circle one number to the right of each phrase to describe how much **DURING THE PAST WEEK**, hot flashes have **INTERFERED** with each aspect of your life.

	Do not interfere										Completely interfere
1. Work (work outside the home and housework)	0	1	2	3	4	5	6	7	8	9	10
2. Social activities (time spent with family, friends, etc.)	0	1	2	3	4	5	6	7	8	9	10
3. Leisure activities (time spent relaxing, doing hobbies, etc.)	0	1	2	3	4	5	6	7	8	9	10
4. Sleep	0	1	2	3	4	5	6	7	8	9	10
5. Mood	0	1	2	3	4	5	6	7	8	9	10
6. Concentration	0	1	2	3	4	5	6	7	8	9	10
7. Relations with others	0	1	2	3	4	5	6	7	8	9	10
8. Sexuality	0	1	2	3	4	5	6	7	8	9	10
9. Enjoyment of life	0	1	2	3	4	5	6	7	8	9	10
10. Overall quality of life	0	1	2	3	4	5	6	7	8	9	10

Carpenter, J.S. (2001). The Hot Flash related Daily Interference Scale: A tool for assessing the impact of hot flashes on quality of life following breast cancer. Journal of Pain and Symptom Management, 22(6), 979-989

APPENDIX VIII: PATIENT QUESTIONNAIRE - MODIFIED BRIEF PAIN INVENTORY FOR AROMATASE INHIBITOR INDUCED ARTHRALGIA

Modified Brief Pain Inventory for Aromatase Inhibitor Induced Arthralgia

Please answer these questions as they relate to symptoms regarding your joints that you think is from your aromatase inhibitor breast cancer medication.

1. Please rate your pain by circling the one number that best describes your pain from your aromatase inhibitor breast cancer medication at its WORST in the last 24 hours.

0	1	2	3	4	5	6	7	8	9	10
No pain										Pain as bad as you can imagine

2. Please rate your pain from your aromatase inhibitor breast cancer medication by circling the one number that best describes your pain at its LEAST in the last 24 hours.

0	1	2	3	4	5	6	7	8	9	10
No pain										Pain as bad as you can imagine

3. Please rate your pain by circling the one number that best describes your pain from your aromatase inhibitor breast cancer medication on the AVERAGE.

0	1	2	3	4	5	6	7	8	9	10
No pain										Pain as bad as you can imagine

4. Please rate your pain by circling the one number that best tells how much pain from your aromatase inhibitor breast cancer medication that you have RIGHT NOW.

0	1	2	3	4	5	6	7	8	9	10
No pain										Pain as bad as you can imagine

5. Please rate your joint stiffness by circling the one number that best describes any joint stiffness from your aromatase inhibitor breast cancer medication on the AVERAGE.

0	1	2	3	4	5	6	7	8	9	10
No joint stiffness										Joint stiffness as bad as you can imagine

6. Circle the one number that describes during the past 24 hours how pain from your aromatase inhibitor breast cancer medication has interfered with you:

A. General Activity

0	1	2	3	4	5	6	7	8	9	10
Does not interfere										Completely interferes

B. Mood

0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes
C. Walking Ability											
0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes
D. Normal work includes both work outside the home and housework											
0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes
E. Relations with other people											
0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes
F. Sleep											
0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes
G. Enjoyment of life											
0	1	2	3	4	5	6	7	8	9	10	
										Does not interfere	Completely interferes

7. Think about the pain in your joint(s) that you did not have prior to starting anastrozole or letrozole, how much of this pain have you had over this past week? (**circle one number**)?

0	1	2	3	4	5	6	7	8	9	10	
										No pain	Pain, as bad as it can be

Answer the following question only if you have started the study treatment:

8. In the last 24 hours how much RELIEF has the study agent provided (compared to what you had prior to starting study medication)?
Please circle the one percentage that most shows how much relief you have received.

0%	10%	20%	30%	40%	50%	60%	70%	80%	90%	100%
										Complete relief
No relief										

APPENDIX IX: PATIENT QUESTIONNAIRE - MENOPAUSE SPECIFIC QUALITY OF LIFE (MENQOL)
Menopause Specific Quality of Life (MENQOL)

For each of the following items, please indicate whether you have experienced the problem in the **PAST WEEK**. If you have experienced the problem, please indicate how much you have been bothered by it.

1. Over the past week, have you been bothered by hot flashes or flushes? (circle no/yes)

No Yes

If yes, how bothered are you by hot flashes or flushes?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

2. Over the past week, have you been bothered by night sweats? (circle no/yes)

No Yes

If yes, how bothered are you by night sweats?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

3. Over the past week, have you been bothered by sweating? (circle no/yes)

No Yes

If yes, how bothered are you by sweating?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

4. Over the past week, have you been bothered by dissatisfaction with your personal life? (circle no/yes)

No Yes

If yes, how bothered are you by dissatisfaction with your personal life?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

5. Over the past week, have you been bothered by feeling anxious or nervous? (circle no/yes)

No Yes

If yes, how bothered are you by feeling anxious or nervous?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

6. Over the past week, have you been bothered by poor memory? (circle no/yes)

No Yes

If yes, how bothered are you by poor memory?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

7. Over the past week, have you been bothered by accomplishing less than you used to? (circle no/yes)

No Yes

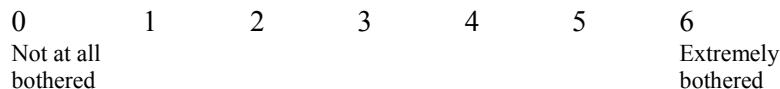
If yes, how bothered are you by accomplishing less than you used to?

0	1	2	3	4	5	6
Not at all bothered						Extremely bothered

8. Over the past week, have you been bothered by feeling depressed, down or blue? (circle no/yes)

No Yes

If yes, how bothered are you by feeling depressed, down or blue?



9. Over the past week, have you been bothered by being impatient with other people? (circle no/yes)

No Yes

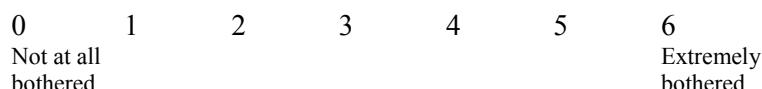
If yes, how bothered are you by being impatient with other people?



10. Over the past week, have you been bothered by feelings of wanting to be alone? (circle no/yes)

No Yes

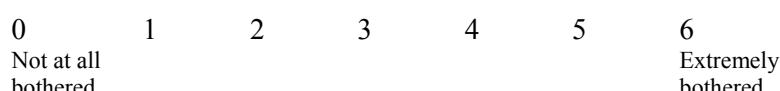
If yes, how bothered are you by feelings of wanting to be alone?



11. Over the past week, have you been bothered by flatulence (wind) or gas pains? (circle no/yes)

No Yes

If yes, how bothered are you by flatulence (wind) or gas pains?



12. Over the past week, have you been bothered by aching in muscles and joints? (circle no/yes)

No Yes

If yes, how bothered are you by aching in muscles and joints?

1 2 3 4 5 6

0

Not at all
bothered

Extremely
bothered

13. Over the past week, have you been bothered by feeling tired or worn out? (circle no/yes)

No Yes

If yes, how bothered are you by feeling tired or worn out?

0 1 2 3 4 5 6

Not at all
bothered

Extremely
bothered

14. Over the past week, have you been bothered by difficulty sleeping? (circle no/yes)

No Yes

If yes, how bothered are you by difficulty sleeping?

0 1 2 3 4 5 6

Not at all
bothered

Extremely
bothered

15. Over the past week, have you been bothered by aches in back of head or neck? (circle no/yes)

No Yes

If yes, how bothered are you by aches in back of head or neck?

0 1 2 3 4 5 6

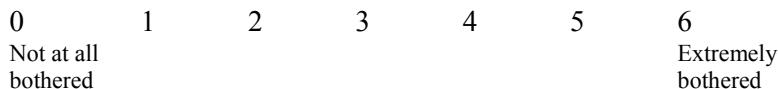
Not at all
bothered

Extremely
bothered

16. Over the past week, have you been bothered by decrease in physical strength? (circle no/yes)

No Yes

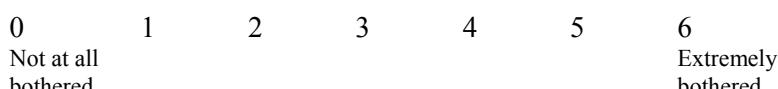
If yes, how bothered are you by decrease in physical strength?



17. Over the past week, have you been bothered by decrease in stamina? (circle no/yes)

No Yes

If yes, how bothered are you by decrease in stamina?



18. Over the past week, have you been bothered by lack of energy? (circle no/yes)

No Yes

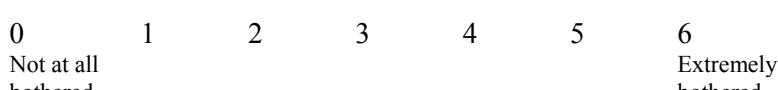
If yes, how bothered are you by lack of energy?



19. Over the past week, have you been bothered by dry skin? (circle no/yes)

No Yes

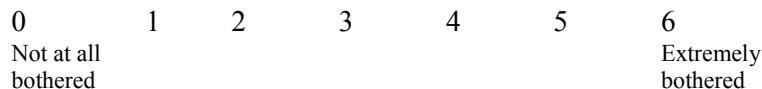
If yes, how bothered are you by dry skin?



20. Over the past week, have you been bothered by weight gain? (circle no/yes)

No Yes

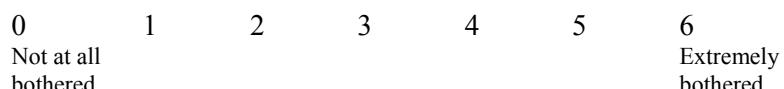
If yes, how bothered are you by weight gain?



21. Over the past week, have you been bothered by increased facial hair? (circle no/yes)

No Yes

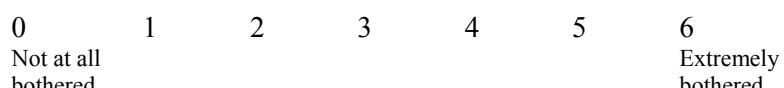
If yes, how bothered are you by increased facial hair?



22. Over the past week, have you been bothered by changes in appearance, texture or tone of your skin?
(circle no/yes)

No Yes

If yes, how bothered are you by changes in appearance, texture or tone of your skin?



23. Over the past week, have you been bothered by feeling bloated? (circle no/yes)

No Yes

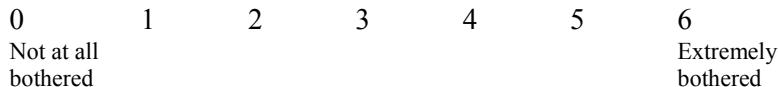
If yes, how bothered are you by feeling bloated?



24. Over the past week, have you been bothered by low backache? (circle no/yes)

No Yes

If yes, how bothered are you by low backache?



25. Over the past week, have you been bothered by frequent urination? (circle no/yes)

No Yes

If yes, how bothered are you by frequent urination?



26. Over the past week, have you been bothered by involuntary urination when laughing or coughing? (circle no/yes)

No Yes

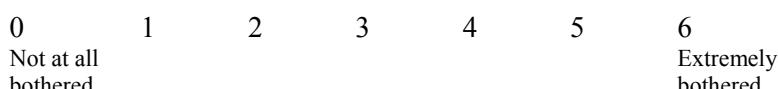
If yes, how bothered are you by involuntary urination when laughing or coughing?



27. Over the past week, have you been bothered by a decrease in your sexual drive? (circle no/yes)

No Yes

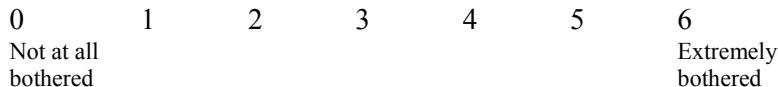
If yes, how bothered are you by a decrease in your sexual drive?



28. Over the past week, have you been bothered by vaginal dryness? (circle no/yes)

No Yes

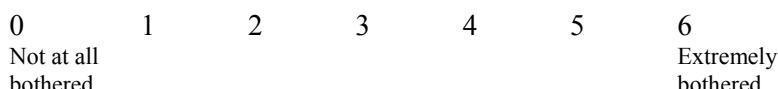
If yes, how bothered are you by vaginal dryness?



29. Over the past week, have you been bothered by avoiding intimacy? (circle no/yes)

No Yes

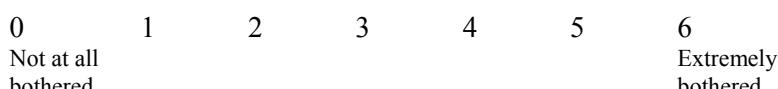
If yes, how bothered are you by avoiding intimacy?



30. Over the past week, have you been bothered by breast pain or tenderness? (circle no/yes)

No Yes

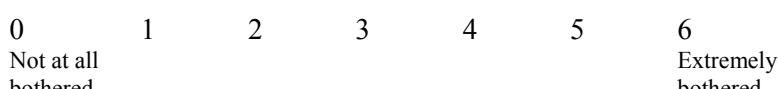
If yes, how bothered are you by breast pain or tenderness?



31. Over the past week, have you been bothered by vaginal bleeding or spotting? (circle no/yes)

No Yes

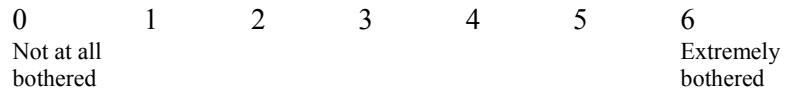
If yes, how bothered are you by vaginal bleeding or spotting?



32. Over the past week, have you been bothered by leg pains or cramps? (circle no/yes)

No Yes

If yes, how bothered are you by leg pains or cramps?



APPENDIX X: NURSE/CRA PHONE CONTACT GUIDE

Nurse/CRA Phone Contact Guide

Patient Phone No. _____
Best Dates/Times to call _____

FOLLOW UP:

1. Phone call schedule: Call patient at home monthly to document compliance, encourage completion of the booklet, and address problems. If the patient is seen in clinic, this phone call can be omitted.

- It is important to reinforce “real time” capture of hot flashes and daily completion of diary as well as weekly side effects on symptom experience questionnaire.

2. Items to document:

- Date of phone call
- Study week
- Questions/Comments
- AE assessment every 3 months

If the patient reports any side effects, please document:

- Side effects:
 - Thromboembolic event - Severity and attribution, if applicable
 - Hirsutism - Severity and attribution, if applicable
 - Acne - Severity and attribution, if applicable
 - Any others - Severity and attribution, if applicable.

3. Reinforce compliance with study medication.

4. Reinforce completion of questionnaires and request return of them at end of each month.

APPENDIX XI: TESTOSTERONE SAFETY ISSUES

Testosterone Safety Issues

1) Breast cancer-specific safety.

a) Animal studies

Zhou et al.²² examined the long-term effects of estrogen (E) exposure in the presence and absence of progesterone (P) or testosterone (T) on mammary epithelial cell proliferation and steroid receptor gene expression in oophorectomized rhesus monkeys. Mammary epithelial proliferation (MEP) was measured using the Ki-67 index. In the presence of E alone, there was a 6-fold increase in MEP and a 50% increase in ER mRNA expression. Similar results were found in E plus P treated animals. In contrast, the combination of E plus T decreased MEP by 40% when compared with E alone²². The combination of E plus T also completely inhibited the E-induced increase in ER mRNA expression. This study demonstrates the anti-proliferative and anti-estrogenic effects of exogenously administered T, and supports the concept that T can have a protective effect against breast cancer development by down-regulating ER mRNA expression and reducing epithelial cell proliferation.

Dimitrakakis et al.³⁷ also investigated the role of long-term hormonal replacement on oophorectomized monkeys treated with E, E + P, E + T or placebo. In the E alone and E + P groups, there was a 3.5-fold increase in MEP, while in the E + T group there was no increase in the MEP above placebo. Estrogen binds to both ER-alpha and ER-beta; ER-alpha receptor stimulates cell proliferation whereas ER-beta may inhibit proliferation. In the E group, the ratio of ER-alpha/ER-beta was 2.5. In the presence of T, this ratio decreased to 0.7, suggesting the anti-prolifertive effects of T may be via interactions with ER-beta. In addition, this study evaluated the role of flutamide, an AR antagonist, and found that it resulted in a two-fold increase in MEP. These findings support the hypothesis that endogenous androgens suppress the growth of mammary epithelium and, therefore, result in reduced susceptibility to cancer-promoting agents in these cells. Of note, is the recently recognized importance of ER-beta in reducing the risk of hormonal failure with long-term endocrine therapies, thus the importance of continued exposure of hormonal responsive tumors to T³⁸.

b) Endogenous androgens and breast cancer risk

i) De-novo cancer risk

Many studies have shown that endogenous levels of estrogen and T exhibit a positive correlation with the development of breast cancer³⁹⁻⁴². However, despite the fact that estrogens are derived from androgens, few studies have controlled for estrogen as a possible confounder when studying the effect of T on the breast. In most of the studies that have controlled for estrogen, the relative risk (RR) of T exposure decreases, and may become non-significant. Zeleniuch-Jacquotte et al^{43,44} performed two studies over a period of seven years on postmenopausal women. The RR of T and breast cancer was found to have an odds ratio (OR) of 2.7 for the highest quartile of serum T. However, after controlling for both E and percent of E bound to SHBG, the risk dropped to 1.2 and was no longer significant. In a continued analysis of this cohort with additional matched cases, the OR for breast cancer associated with the doubling of T was found to be 1.23 (P=0.001). After adjusting for E, this OR decreased to 1.17 (P=0.04), and after adjusting for estrone, the OR decreased to 1.08 and was non-significant (P=0.31).

Additionally, the Endogenous Hormones and Breast Cancer Collaborative group performed a meta-analysis of the results of nine case-control studies performed prior to 2002⁴⁵ examining sex hormones and breast cancer risk. The analysis found an overall increased risk of breast cancer with each of the hormones studied and that the risk increased in a dose-dependent manner. When comparing the hormone levels from the highest quintile to the lowest quintile, the RR of breast cancer for T was 2.22 (CI 1.59–3.10), DHEA RR 2.04 (CI 1.21–3.45) and E 2.00 (CI 1.47–2.71), (P<0.02), while SHBG had a RR of 0.66 (confidence

interval [CI] 0.42–1.00). E positively correlated with T ($r=0.37$), DHEA ($r=0.20$) and DHEA-S ($r=0.29$). After adjusting for E, the RR of breast cancer with doubling of T decreased from 1.42 to 1.32 (CI 1.15–1.51). For DHEA and DHEA-S, the RR also decreased from 1.24 to 1.19 (CI 0.98–1.45) and 1.20 to 1.15 (CI 1.04–1.27), respectively. The reductions in RR after adjusting for E provides further evidence that, when assessing hormonal trends, T needs to be adjusted for the independent effects of E. Of note, T was measured in seven of the nine studies and only two of these seven studies had a sample size of >100 . The largest study included in this meta-analysis was a nested case-control study from the Nurse's Health Study, in which a significant association between DHEA-S and breast cancer (RR 2.15, CI 1.11–4.17) was found, but no relationship between T or DHEA and breast cancer risk³⁹. This was also confirmed in a cross-sectional analysis of women in the Nurse's Health Study that found that breast cancer risk was not associated with any of the androgens and, interestingly, that T and free-T levels were approximately 30% lower among women with a hysterectomy when compared to those without ($p < 0.01$)⁴⁶. This low T level in women that have had their ovaries removed is in keeping with a recent study that showed a low bioavailable T levels in post-menopausal women with breast cancer, many who have had total hysterectomies or who have had chemotherapy induced ovarian ablation⁴⁷. In this study, T levels were determined from saliva, thus representing tissue T rather than serum T. Obviously this could be the result of therapeutic intervention, but validates our concerns about the low levels of bio-available T in women with breast cancer.

ii) Recurrence Risk

In women that have had breast cancer, the recurrence risk is not associated with serum T levels. In a nested case-control cohort study from a randomized diet trial (Women's Healthy Eating and Living Study), recurrence free survival in women followed for more than 7 years after diagnosis was determined according to baseline serum concentrations of E, T, and sex hormone binding globulin (SHBG). In 153 case-control pairs of perimenopausal and postmenopausal women, there was a significant increased risk for reduced survival time to recurrence with increased baseline total E [hazard ratio (HR), 1.41 95% CI, 1.01-1.97], bioavailable E (HR, 1.26; 95% CI, 1.03-1.53), and free estradiol (HR, 1.31; 95% CI, 1.03-1.65). These concentrations were significantly associated with risk recurrence. In women who relapsed in the follow-up period, the average total E concentration was double that in women who did not have recurrence during the period (22.7 versus 10.8 pg/mL; $P = 0.05$). T and SHBG concentrations did not differ between cases and controls and were not associated with risk for recurrence⁴⁸.

c) Polycystic ovaries and breast cancer risk

The natural physiological example of hyperandrogenism in patients with polycystic ovarian syndrome (PCOS) can be used to elucidate the role of endogenous T in breast cancer. In a case-control study by Gammon et al.⁴⁹ with over 4700 matched subjects, the age-adjusted OR for breast cancer in self-reported physician-diagnosed PCOS was 0.52 (CI 0.32 –0.87). Using data available from this patient population, evidence suggests that an elevation of endogenous T does not increase breast cancer risk and, further, that the significantly reduced OR with increasing levels of T suggests a protective effect. When adjusted for age at first birth, history of infertility, number of spontaneous abortions before first birth and menopausal status, the OR for breast cancer with increasing levels of T decreased to 0.47 (CI 0.25–0.85). In addition, in a prospective cohort study with six years follow-up, there was no increased risk of breast cancer in patients with PCOS after adjusting for cofounders, such as age at menarche, parity and age at menopause, with the RR being 1.0 (CI 0.6–1.9)³⁶.

d) Exogenous androgen and breast cancer risk

i) Retrospective studies

In a retrospective observational study, of women receiving HRT and implantable T pellets and followed for an average of 5.8 years⁵⁰, the incidence of breast cancer (per 100,000 woman-years) was less than that reported in the Women's Health Initiative and the Million Women's studies, which followed women treated with HT without T. Although this study sample size was small and the study design lacked a concurrent

control group, the breast cancer rate was similar to that in women that had never used HT after adjusting for age.

Additionally, Ewertz et al.⁵¹ performed a case-control study in 1486 women with breast cancer and 1336 controls. Women completed a self-reported questionnaire on their use of sex hormones. With the sequential therapy of E+P there was not a significant increase in risk of breast cancer RR 1.36 (CI 0.98–1.87). The RR of breast cancer with estrogens plus androgen use was 2.31 (CI 1.37–3.88) and with estrogen, progesterone and androgen, there was no increased risk RR 1.26 (CI 0.58–2.74). This study did not control for estrogen to assess for the risk of androgens alone. The authors noted that 23% of the women did not state the brand name of the hormone they were receiving, therefore, leading to more cautious interpretations of the analyses.

In a case-control study by Brinton et al.⁵² there was no increased risk of breast cancer with estrogen use at varying doses or in combination with T, with the RR for E and methyltestosterone being 1.05 (CI 0.6–1.8). In addition, when compared with non-users of hormones, the risk of breast cancer with 10 or more years of use of conjugated equine estrogens with methyl-testosterone was decreased, with a RR of 0.66, although the finding was not statistically significant.

ii) Prospective studies

In the first randomized study to directly measure the effects of T on breast cell proliferation in postmenopausal women, Hofling et al.⁵³ randomized women starting HRT with E+P to either subcutaneous T delivered via a patch or versus a placebo treatment. Fine needle aspiration breast tissue biopsies were taken at baseline and six months and the ki-67 proliferative index of both the epithelial and stromal cells were determined. There was a five-fold increase in breast cell proliferation after six months in women on placebo patch when compared to baseline values. There was no significant difference between the six month and baseline proliferation index in breast tissues from women who were randomized to the T treatment. These results are consistent with those reported from animal studies, which support the hypothesis that androgens play a role in suppressing the proliferation effects of E and P on mammary cell lines (reviewed in⁵⁴).

The Nurses' Health Study⁵⁵ was a large prospective cohort study that analyzed the association between combined estrogen and androgen therapies and development of breast cancer. Over the 24-year follow-up, representing 1,359,323 person-years, 4610 cases of breast cancer were found among postmenopausal women. Analysis gave an adjusted RR of breast cancer of 1.77 (CI 1.22–2.56) for women taking estrogen and T when compared to those never using hormones, and a RR of 1.58 (CI 1.44–1.73) for women taking E+P when compared to those who never took hormones. There was no statistically significant difference found between these two RRs. For women taking E only there was a 1.15 RR (CI 1.05–1.27) of breast cancer when compared to those who never took hormones, while for women taking T alone there was no significant increased RR 2.69, (CI 0.86–8.43). There were only 3 cases of breast cancer reported in this group with 360 person-years, compared with the estrogen only arm with 246,830 person years. This study was never designed to compare the relationship between androgens and breast cancer. In addition, there was a reversal of the anticipated positive association between the duration of hormone use and the risk of breast cancer; in those women who used T for less than five years had an 81% increase in risk (RR 1.81, CI 1.21–2.70), while women who used T for greater than five years had a non-significant increased risk (RR 1.96, CI 0.93–4.14). Some of the design flaws in this study relate to the duration and the type of hormones used. Since follow-up took place with questionnaires every two years, if a woman answered the survey as 'yes' to starting on T in any of the time periods, despite being on E+P or estrogen only for a decade or more, she was then placed into the T group. In addition, if that same woman used only T for four weeks, it was counted as a two-year period since the follow-up surveys occurred every two years. The latest evaluation of E + T HRT in the Women's Health Initiative failed to reveal any increase risk of breast cancer⁵⁶ and they could not substantiate the findings of the Nurses Health Study⁵⁵. Long-term E+T usage in the

WHI study revealed a non-significant RR of 1.02 (CI, 0.58-1.78) (reviewed in⁵⁷.

The latest evaluation of E + T HRT in the Women's Health Initiative failed to reveal any increased risk of breast cancer⁵⁶ and they could not substantiate the findings of the Nurses Health Study⁵⁵. Long-term E+T usage in the WHI study revealed a non-significant RR of 1.02 (CI, 0.58-1.78) (reviewed in⁵⁷.

The largest study evaluating the safety of methyl-T in HRT was a post-marketing analysis done by Solvay Pharmaceuticals, Inc., on the Estratest brand used between January 1989 and August 2002⁵⁸. Exposure to the Estratest brand during the 13-year assessment period is estimated at >3.0 million patient-years. A total of 1372 unique case reports containing 2556 AEs were found and there was no evidence of increased breast cancer incidence prevalence.

iii) Testosterone only therapy and breast cancer risk

There are limited studies on this subject but the most important include subcutaneous application for low libido, transgender studies and treatment of existing breast cancer with androgens

Subcutaneous application

One of the emerging surrogate end-points for breast cancer risk is mammographic density and many exogenous hormones have been associated with an increase in density. A recent randomised placebo-controlled study of 52 weeks of subcutaneous T alone or with E demonstrated no increase in mammographic density⁵⁹. In another prospective analysis of combined HRT, an increase in mammographic density was recorded in approximately 50% of the women. Increased density showed a positive correlation with estradiol, estrone, and SHBG and showed a negative association to free T⁶⁰. As well, a retrospective cohort study of 631 women ever treated with subcutaneous T alone for low libido between January 1989 and December 2007 demonstrated no associated increase in breast cancer⁶¹.

Transgender studies

Another illustration of the effect of T on the breast is female-to-male (FTM) transgenderism, in which genotypic women are treated with doses of T that result in serum levels in the mid-normal male range⁵⁷. Analysis of mastectomy tissue from 29 women who had received prolonged androgen exposure prior to transgender surgery revealed no histological features suggestive of breast cancer or precancerous cellular changes⁶². There were no differences in the morphology or number of acini and ducts, or the amount of fibrosis, or numbers of cysts, estrogen or progesterone receptors, or apocrine metaplasia, when compared to mammoplasty tissue from women on no therapy. The only difference observed was an increase in non-specific microcalcifications in the tissues from the women on the prolonged androgen exposure. Another prospective study conducted between 1975 and 2006, studied 876 FTM transsexuals who were on long-term treatment with T. There were no cases of breast cancer reported⁶³.

Androgen as a treatment for breast cancer

Testosterone propionate,⁶⁴ fluoxymesterone,⁶⁵ and calusterone, a 17-methylated active androgenic, have all been evaluated for the treatment of advanced breast cancer, used since the 1950s. The prevalence of remission (if use rate indicate time – preferable in this sentence) ranged from 20–50% in cohorts, which is equivalent to other endocrine-modulating therapies⁶⁶. In a phase III trial, fluoxymesterone and tamoxifen was evaluated for the treatment of advanced breast cancer in postmenopausal women. In this trial, the remission rates was 15% for tamoxifen alone, and 38% in patients on fluoxymesterone and tamoxifen (P=0.016). This result suggests that T is an independent antiproliferative agent with regards to breast cancer⁶⁷. In preclinical animal models, it has been shown that the predominant mechanism of androgen action is due to a direct interaction of the androgen with the AR within the breast cancer cell, rather than a perturbation in the hypothalamic–pituitary–gonadal axis⁶⁶. Previous studies have shown that AR is commonly expressed at high levels in breast cancer tissue (reviewed in⁶⁸. In a recent Nurses Health Study, a positive association was found between AR status (or levels?) of breast cancer tissues and duration of

survival of breast cancer patients, and that this association was dependent on the ER status of the tissues. In particular, AR expression was associated with a more favorable prognosis among women with ER-positive tumors⁶⁹. In addition, a recent study of 3093 tissue microarrays prepared from breast cancer tissue from women in the Nurses' Health Study found that 64% of tissues were luminal A, 15% luminal B, 6% HER2 positive and 11% basal-like with the frequency of AR expression varying significantly across these molecular phenotypes ($P<0.0001$). In particular, high AR expression was commonly found in luminal A (91%) and B (68%) cancers, but was less frequent in HER2 cancers (59%). Among 246 cases of ductal carcinoma *in situ*, 86% were AR positive, but the frequency of AR expression differed significantly across the molecular phenotypes ($P=0.001$), and high nuclear grade lesions were less likely to be AR positive compared with lower-grade lesions⁷⁰.

The important consideration for this current proposed study is that AR, as a ligand target, is only present in these AR+/ER+ tumors that would be receiving the AI/T combination, as the inclusion criteria includes patients with tumors that are positive for ER and PR and thus have a good prognosis. Therefore, maintenance of AR levels in the tumor appears to be beneficial and there is good data to support the hypothesis that prolonged exposure to T increases the levels of AR within breast tumors⁷¹.

e) *in vitro* studies of androgen treatment in breast cancer cell lines

i) Breast cancer cell lines

Studies on androgens and breast cell line proliferation have multiple variables that affect the outcome of those studies. The predominant data, however, show that androgens have apoptotic and antiproliferative effects (reviewed in ⁹). It should be noted that cell line studies examine only the effect of hormones directly added to the culture medium. It is well recognized the importance all the stromal and supporting tissue in the hormonal regulation of breast tumors. Especially This is especially important when considering aromatase action, as it is well known that aromatase is present in all cellular components making up a breast tumor. To this end we tested the AI/T combination in a whole tumor explant system. This is explained in more detail below in section 2b.

2) Aromatase inhibitor efficacy

a) Human trials of AI and T

By far the most common usage of this combination has been for men using anabolic steroids and using AI to block the induction of gynaecomastia as a consequence of estrogen. As this is an illicit activity, little has been published on the pharmacokinetics of this combination in men. By far the most extensive use of this drug combination has been in studies by Dr Susan Davis at the Monash University, Australia aimed at reducing postmenopausal symptoms in women. In these studies, AI has been used to help determine the effect of aromatization of T to estrogen plays in T replacement therapy. In one double-blind randomized placebo-controlled trial of T replacement in postmenopausal women, increases in total and free T in the physiologic range in postmenopausal women were associated with improved sexual satisfaction, well-being, and mood. In this study, AI therapy did not influence any of these outcomes, implying that these affects of T were mediated by the AR. Short-term subcutaneous T therapy did not modify fasting lipids, lipoprotein, or C-reactive protein ²⁶. Recently, Glaser evaluated the efficacy of subcutaneous anastrozole and T, in two 3.1 x 6.1 mm implants containing a total of 120 mg testosterone and 8 mg of anastrozole, implanted in the upper gluteal area of 55 post-menopausal women with breast cancer ⁷². Serum levels of T and E were measured 2 weeks following implantation. They concluded that T/anastrozole therapy was effective in treating symptoms of androgen (hormone) deficiency. In all but 5 of 75 T/anastrozole patients, serum estradiol levels measured < 30 pg/ml with therapeutic T levels (mean: 281, range: 120-518 ng/dl). A single patient had an estradiol level > 40 pg/ml, but a subsequent level in the same patient measured < 30 pg/ml. No adverse drug events were recorded from subcutaneous T/anastrozole therapy in over 110 implantations. No relapses in symptoms were diagnosed at up to 3 years of therapy and there was no progression of disease in 2 patients with metastasis ⁷². In another trial investigating T with an AI, 21

postmenopausal women with breast cancer who were on AIs and with had symptoms of vaginal atrophy were treated with T cream applied to the vaginal epithelium daily for 28 days. Ten women received a dose of 300 µg, 10 received 150 µg, and one was not evaluable. E and T levels, symptoms of vaginal atrophy, and results of gynecologic examinations and pH and vaginal cytology before and after therapy were compared. E levels remained suppressed to <8 pg/mL after treatment. A 4-week course of vaginal T was associated with a reduction in symptoms of vaginal atrophy related to AI therapy, without increasing estradiol levels⁷³.

b) Human explant study

In this study using a novel breast tumor explant model, breast tumor tissue excised at surgery qA was incubated in the presence of T and T+anastrozole. Of 26 tumor explants, epithelial cell proliferation, as measured by the ki67 Ki-67 index, decreased in 24 of them in the presence of T when compared to vehicle control treated tissue, but the decrease was only significant in 7 tissues. This decrease in proliferation also occurred in the same tissues treated by T + anastrozole. When the same tissues were incubated with T and an androgen antagonist, there was no significant decrease in Ki-67, suggesting that the reduction in cell proliferation was secondary to a direct interaction of the androgen with the androgen receptor. In two of the tumor tissues, there was an increase in the proliferative index with T alone. When these tissues were incubated with T + anastrozole, there was no increase in proliferation, and there was no effect of an androgen antagonist on this response. This finding suggests that this increase in proliferation was secondary to conversion of T to E⁷⁴

c) Human breast cancer cell-lines

Two recent studies in ER+ cell lines have demonstrated that any increase in the proliferation of human breast cancer cells, that may occur in the presence of T, is the result of aromatase conversion of T to E. The increase in proliferation was reversed by the addition of ER antagonists and AIs, and was unaffected by the addition of AR antagonists. At physiological concentrations, DHT had no effect on proliferation^{75,76}. Of equal importance is the finding that T reduces aromatase levels in human breast cancer cell lines⁷⁵. This physiological feedback mechanism has been noted in several other tissues and is of importance when examining the efficacy of an AI in combination with T. Indeed, Dr Angela Brodie's group have reported findings that suggest that part of the mode of action of AIs in inhibiting proliferation and inducing apoptosis of breast cancer cells is via the un-masking of the androgen/AR interaction within the cell³⁰.

3) General safety issues with testosterone replacement

The general safety issues of T replacement are best addressed in the application of Proctor and Gamble to the FDA for the Intrinsa patch in the women with low libido. This slide set is an accurate analysis of the data available in 2004 www.fda.gov/ohrms/dockets/ac/04/slides/2004-4082S1_03_P&G-Intrinsa.ppt

One of the major concerns about T replacement is cardiovascular disease (CVD). Despite the entrenched belief that higher blood levels of T increase the risk of CVD in women, data from recent observational studies mostly show an inverse relationship between testosterone and CVD risk (reviewed in⁷⁷). The largest study to date, examining the safety of T replacement comes from the Solvay Pharmaceuticals Inc. safety study, which failed to demonstrate any clinically important side-effects⁵⁸.

4) General comment regarding safety of testosterone/aromatase inhibitor combination.

It is of increasing concern that the gold standard endocrine treatment for breast cancer has switched from tamoxifen to aromatase inhibitors when such a significant side-effect is resulting in severe compliance problems. The 40% dropout in the active treatment arms on AIs in the recent MA27 trial is almost unprecedented in an adjuvant setting⁷⁸. Compliance in the IBIS-I trial of tamoxifen in the chemoprevention of breast cancer was 85% at five years in healthy women. Therefore, although it is well known that compliance is an issue with both tamoxifen and AIs, if 40% of women drop out of a clinical trial, it causes pause to consider what the “real-world” compliance rate must be. Therefore, when evaluating the relative dangers of the addition of T to women with AI-induced arthropathy, it must be weighed up against the

potential increase in mortality and morbidity from cessation of proven hormonal therapy. It is not known how many of the women dropping out of AI therapy will actually take up alternative endocrine treatment. As well, although vitamin D and calcium supplementation is being investigated as a modality for treatment of AI induced arthropathy, there are recent concerns about an apparent excess of cardiovascular events in women on long-term use of these agents. This is of particular importance when there are signs of adverse lipid events occurring with AIs⁷⁹.

Much of the concern regarding testosterone and breast cancer is because of the potential for T to be converted to E₂ by aromatase within serum and in tissues. The elegance of the T/AI combination therapy is that the possibility for this conversion to occur is eliminated, thus ensuring available T and allowing the evaluation of the therapeutic effect of T. We believe that there is ample evidence that T does not initiate or promote breast cancer or reduce the efficacy of AIs and indeed T may add the potential for additional therapeutic effect due to the inhibitory effect of T, or its more active 5-alpha reductase metabolite DHT, in the breast.

1. Agent selection:

COMMENT: Justification for the dosage of the subcutaneous testosterone is not clear, as well as how it relates (dosage, concentration) to the two dosages used in the ART 2 phase II trial. More discussion is needed on whether the subcutaneous route of administration produces serum concentrations similar to the injected and oral forms of testosterone. In describing the formulation, the investigators refer to 8% octisalate, but never discuss this component.

RESPONSE:

In the ART2 trial⁸⁰, oral TU was used due to the ease of access to the compound. There was a clear benefit of using TU at a dose of 80mg versus 40mg in terms of both the clinical response and serum T levels achieved; these being more commensurate with that of premenopausal woman. However, the need to refrigerate the compound, the variability in serum levels achieved and the possible need to administer twice daily doses meant that other routes may be preferred for both pharmacokinetic and convenience reasons. The subcutaneous spray formulation provides advantages over oral administration and circumvents some of the drawbacks of subcutaneous patches.

In addition to orally (through the buccal mucosa), T may also be administered by insertion, by or subcutaneous implant, and subcutaneously. Subcutaneous administration circumvents first pass liver effects, provides a more sustained physiologic serum concentration of T, and a favorable safety profile with regards to lipid profile and liver effects⁸¹⁻⁸³. For example, the subcutaneous T aerosol appears to be less likely to affect the lipid profile of patients⁸⁴. The subcutaneous spray has added ease of use advantages over the subcutaneous patch for administration of T. Skin reactions may occur in response to the substrate material used for the patch. Patch allergy occurs at a prevalence of more than 30% and is the most commonly reported adverse event in women undergoing subcutaneous therapy⁸⁵. One population study of low dose T use in women with androgen insufficiency syndrome concluded that no absolute cut-off level of T was appropriate⁸⁵. Further, in studies of postmenopausal women with low serum T, the TTS can provide sustained increases in serum T concentrations within the normal range for premenopausal women, as shown below

Title	Indication	Dose, duration & study population	Outcome	Side Effects
Testosterone patch for low sexual desire in surgically menopausal women: a randomized trial	Hypoactive sexual desire disorder (HSDD)	300 ug/day testosterone or placebo for 24 weeks 533 women with HSDD, hysterectomy, and bilateral oophorectomy INTIMATE SM2	Total satisfying sexual activity significantly improved, as well as sexual desire and decreased personal distress	The rate of androgenic adverse events was higher in the testosterone group, mostly mild. Overall, no difference in the AE incidence rate compared with placebo
Testosterone patch increases sexual activity and desire in surgically menopausal women with HSDD ⁸⁶	HSDD	300 ug/day testosterone or placebo for 24 weeks 562 women with HSDD, hysterectomy, and bilateral oophorectomy INTIMATE SM1	Significant improvement in total satisfying sexual activity significantly improved, as well as sexual desire and decreased personal distress	No difference in the incidence or type of adverse events
Subcutaneous testosterone treatment in women with impaired sexual function after bilateral oophorectomy ⁸⁷	Impaired sexual function	Placebo, 150 ug or 300 ug per day for 12 weeks 75 women aged 31 – 56 who received all three treatments in a cross over study without a washout	Mean serum testosterone increased final dosages The 300ugmcg dose group reported improvements in several measures of sexual function, as well as improvements in psychological measures	Treatment related adverse events led four women to withdraw. These events included two women become agitated or anxious, one woman who had a pink discharge from the nipple and one subject had a site reaction
PK of a novel testosterone matrix subcutaneous system in healthy premenopausal women and women infected with the Human Immuno-deficiency virus ⁸⁸	Testosterone deficiency in HIV infected women	300 ug daily over an application period of 3-4 days 9 healthy women and 8 HIV infected women	A regimen of two testosterone patches applied twice weekly can maintain serum total and free testosterone levels in the mid to upper normal range, respectively in HIV infected women with low testosterone	One healthy woman experienced exacerbation of acne, one HIV infected woman experienced irregular vaginal bleeding. Later evaluation found this patient to have uterine fibroids
Randomized controlled	Decreased libido	2% topical cream, 10mg oral,	Increased serum	No statistically significant

trial to evaluate subcutaneous testosterone in female cancer survivors with decreased libido: North Central Cancer Treatment group Protocol N02C3 ⁸⁹		or placebo, daily for 4 weeks, crossover 150 postmenopausal women who had survived cancer and reported reduced libido	testosterone, but no statistical difference libido	difference in testosterone related AEs
Testosterone treatment for HSDD in postmenopausal women (Intimate 1 & 2) ⁹⁰	Reduced sexual function	300 ug/day in twice a week patches Surgically menopausal women receiving concomitant estrogen therapy	Significant increase in total satisfying sexual activity	Application site reactions due to transdermal patch
Testosterone aromatization and cognition in women: a randomized, placebo- controlled tria ²⁵	Cognition	400 uL, 0.5% testosterone or placebo, daily for 16 weeks 61 postmenopausal women using subcutaneous estrogen	No effects on cognition were observed	No adverse effects were observed
Effects of subcutaneous testosterone administration on insulin sensitivity, fat mass and distribution and markers of inflammation and thrombolysis in human immune-deficiency virus infected women ⁹¹	Insulin sensitivity, fat or markers of inflammation and thrombolysis	300 ug testosterone or placebo daily, for 24 weeks 52 HIV infected menstruating women with >5% weight loss over the prior 6 months	Testosterone levels increased into the high normal female range. Insulin sensitivity, whole body-fat mass or distribution, or markers of inflammation and thrombolysis were not adversely affected	
Effects of aromatase inhibition on sexual function and well-being in	Reduced sexual function and mood	400 uL, 0.5% testosterone gel, and letrozole or placebo daily for 16 weeks 60 postmenopausal women	Increase of testosterone to the physiologic range in postmenopausal women was associated with	No adverse treatment effects were reported

postmenopausal women treated with testosterone: a randomized placebo controlled trial ²⁶		using subcutaneous estrogen and reporting low sexual satisfaction completed the trial	improved sexual satisfaction, well-being and mood	
Testosterone for low libido in postmenopausal women not taking estrogen ⁹²	Reduced sexual function	double-blind, placebo-controlled, 52-week trial in which 814 postmenopausal women 150/300 uL patch	modest but meaningful improvement in sexual function with just testosterone and no estrogen replacement	Good tolerance and slight increase in hirsutism in women on the higher dose of testosterone

FemPharm Pty Ltd (West Melbourne, Victoria, Australia) developed TTS for use in women with androgen deficiency. The TTS is intended to provide aerosol delivery of a clinically relevant systemic dose of T (approximately 100 to 300 µg per day) after topical application of a rapidly drying solution from a metered-dose spray to intact skin. The formulation is a single-phase liquid containing 5% T and excipients, one of which enhances delivery. The application area is defined by means of a complete plastic housing around the application nozzle.

This product has been approved in the US for treatment of hypogonadism and T deficiency (in male patients). The transdermal enhancer component used in the formulation is octisalate (see more below), which is widely used as a sunscreen agent, and as is an excipient in Evamist.

An initial clinical study using T MDTs was completed in 6 postmenopausal women, who received daily treatment with approximately 10 mg T ($4 \times 70 \mu\text{L}$ sprays) for 5 days. The mean steady-state serum levels of free-T were elevated, from 1.9 pg/mL at baseline to 7.1 pg/mL on Day 5. (The normal laboratory range for premenopausal women is 1.1 to 6.3 pg/mL). In a subsequent study, conducted in 14 postmenopausal women with low serum T, who received daily treatments of $2 \times 91 \mu\text{L}$ sprays to the abdomen for 5 days, the mean serum concentrations of free-T increased to 5.3 pg/mL (Data on file, VIVUS, Inc.).

A further pharmacokinetic study, in naturally postmenopausal women with low serum T, confirmed that application of the chosen dose ($2 \times 91 \mu\text{L}$ sprays) elevated average steady-state serum concentrations to 5.3 pg/mL.

Crux Pty Ltd undertook a phase 2 study of TTS Formulation in premenopausal women (Protocol FHRT11). In this double-blind, placebo controlled study, a total of 261 women with low serum T were treated for 16 weeks with one of 3 doses or placebo. A statistically significant increase in the frequency of satisfactory sexual events was observed for subjects in the $1 \times 90 \mu\text{L}$ dose group. No major safety issues were observed.

Octisalate is a compound that is currently used as a sunscreen ingredient. Over many years of use as topical sunscreens, these agents have shown a very low incidence of local skin reactions. The FDA has issued a final monograph for sunscreen drug products including octisalate. The monograph establishes conditions under which the sunscreen drug products are generally recognized as safe. The monograph approves the use of concentrations of octisalate of up to 5%⁹³.

Transdermal Testosterone Spray formulation A contains concentrations of octisalate above the 5% normally used in sunscreen agents. However, it will be applied in much smaller amounts and over a smaller skin area than most sunscreens. Therefore an exposure comparison between a typical sunscreen and the excipient in the TTS demonstrated that the exposure to octisalate from the TTS is less than 1.0% of the exposure when applied as a sunscreen ingredient.

APPENDIX XII: SITE ORDERING INSTRUCTIONS AND FORMS (TO ORDER STUDY AGENT)

Site Ordering Instructions and Order Forms: Testosterone and Placebo

Background:

For this trial, the research base pharmacy will send the first 3 month study drug supply directly to a participating Alliance main member institution or directly to an affiliate site upon receipt of an order from the participating site. This order must be completed each time a study participant is enrolled. The research base pharmacy will send a 3 month supply directly to each participating Alliance main member institution or affiliate site after the registration office receives a request for a patient-specific reorder.

First 3 month treatment period: Each treating location, must request one patient-specific dose (2 pens) each time a patient is registered in the trial by completing the Alliance Clinical Drug Order/Return Form located on page 2 of this appendix. Note: A sample form is included (page 3 of this appendix).

Please be certain to include a ***complete shipping address*** for the treatment site.

Please include the DEA Registration Number of your pharmacy or investigator on the order form, along with the patient's study ID number, patient initials, and date needed.

Note: *Allow three business days for the delivery of the 3 month drug supply.*

Second 3 month treatment period: When it has been determined the study participant will be returning for the 2nd half of the 6 month treatment period, each treating location must request a second 3 month drug supply, by faxing a completed copy of the A221102 Testosterone/Placebo gel Re-Order Form (page 4 of this appendix) to the registration office at [REDACTED] The site must also complete the Alliance clinical drug order/return form located on page 2 of this appendix, and fax it to the number listed on the form.

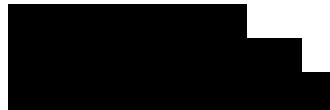
Note: *Allow one week for delivery of the 2nd 3 month drug supply.*

A221102 CLINICAL DRUG ORDER FORM			The drugs listed below are requested for the use of (please type or print):					Gonda 10 USE ONLY	
FAX orders to: Cancer Treatment Pharmacy Svc.			Dr. _____						
MAIL orders/returns to: [REDACTED] [REDACTED] [REDACTED]			Designee/Requester (if other than investigator) (please type or print): Name: _____ Title: _____					filled by _____ checked by _____ date _____	
			Telephone Number: _____ Fax Number: _____						
			Investigator/Designee Signature _____ Date _____					SITE DEA REGISTRATION NUMBER: _____ (Example: FM1234567)	
Protocol Number	PATIENT STUDY ID NUMBER	Patient Initials	Drug Name	Strength & Dosage Form (vials, tablets, etc.)	Quantity Ordered (vials, bottles, etc.)	Date Needed	-----	KIT NUMBER	
A221102			TESTOSTERONE 10.4 MG/0.264 mL OR PLACEBO GEL	10.4 MG/0.2 64 mL Syringe	2 X 13.5 mL Accupen for ~ 94 doses				
SHIPPING ADDRESS:			NOTE: Urgent Shipments must be accompanied by an express courier account number. Express Courier Name: Express Courier Acct No.: _____ - _____ - _____ - _____ - _____				INSTRUCTIONS: 1. One item or protocol per line 2. When requesting drugs: Fill in all sections completely, except shaded areas. 3. When returning drugs: Fill in all sections completely, including shaded areas. 4. Must include official shipping address. 5. Sign and date the order/return.		

A221102 CLINICAL DRUG ORDER FORM			The drugs listed below are requested for the use of (please type or print):					Gonda 10 USE ONLY	
FAX orders to: Cancer Treatment Pharmacy Svc.			Dr. <u>Investigator Name</u>						
MAIL orders/returns to: [REDACTED] [REDACTED] [REDACTED] [REDACTED]			Designee/Requester (if other than investigator) (please type or print): Name: <u>Ordering Designee</u> Title: <u>As needed</u> Telephone Number: <u>Complete number</u> Fax Number: <u>Complete number</u>					filled by _____ checked by _____ date _____	
			Investigator/Designee Signature Date (dd/Mon/yyyy): _____					SITE DEA REGISTRATION NUMBER: _____ (Example: FM1234567)	
Protocol Number	PATIENT STUDY NUMBER	Patient Initials	Drug Name	Strength & Dosage Form (vials, tablets, etc.)	Quantity Ordered (vials, bottles, etc.)	Date Needed	-----	KIT NUMBER	
A221102	2123456	ABC	TESTOSTERONE 10.4 MG/0.264 mL OR PLACEBO GEL	10.4 MG/0.264 mL	2 X 13.5 mL Accupen for ~94 doses	12/25/2012			
SHIPPING ADDRESS: Site Name Facility Department Street Address City, State Zip Code			NOTE: Urgent Shipments must be accompanied by an express courier account number. Express Courier Name: Express Courier Acct No.: _____ - _____ - _____ - _____					INSTRUCTIONS: 1. One item or protocol per line 2. When requesting drugs: Fill in all sections completely, <u>except</u> shaded areas. 3. When returning drugs: Fill in all sections completely, <u>including</u> shaded areas. 4. Must include official shipping address. 5. Sign and date the order/return.	

A221102 Testosterone/Placebo Re-Order Form

FAX to:



Date (MM/DD/YYYY): _____
Alliance Patient ID number (7 digits, begins with a '2') _____

Patient Initials (Last, First, Middle) _____
Kit # _____

Site Contact/ Shipping Information:

Contact Person: _____

Institution: _____

Street Address: _____

City/State/Postal Code: _____

Telephone (with area code): _____

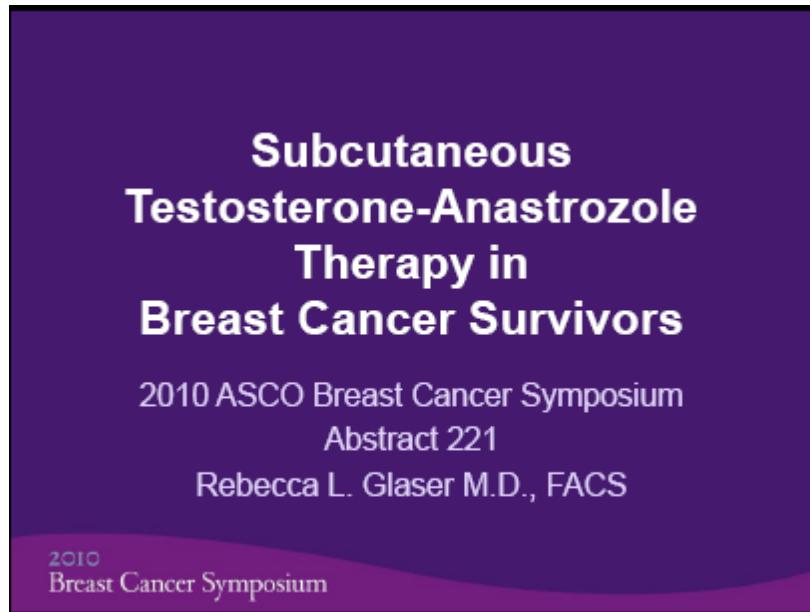
Telefax (with area code): _____

Email: _____

Complete this form and fax to Registration Office at 

APPENDIX XIII: ASCO SLIDES REGARDING TESTOSTERONE/ANASTRAZOLE PRESENTATION

ASCO Slides regarding testosterone/anastrazole presentation



The slide has a dark purple background with white text. The title "Learning Objectives" is at the top. Below it, a statement reads: "After reading and reviewing this material, the participant should be better able to:". A bulleted list follows:

- Identify symptoms of androgen deficiency in pre and post menopausal breast cancer survivors
- Recognize the potential role of subcutaneous testosterone-anastrozole implant therapy in safely treating those symptoms

At the bottom left, there is a small logo for the "2010 Breast Cancer Symposium".

Outline

- Background
- Methods
- Results
- Conclusion
- Future

2010
Breast Cancer Symposium

Background

- Both pre and post menopausal breast cancer survivors commonly experience symptoms of hormone deficiency that can adversely affect their health and quality of life

2010
Breast Cancer Symposium

Efficacy of Testosterone Therapy

- Continuous testosterone therapy, delivered by subcutaneous (SC) implant, effectively treats hormone/androgen deficiency symptoms as measured by the HRQOL, Menopause Rating Scale (MRS) in both pre and post menopausal patients¹

2010
Breast Cancer Symposium

Symptoms improved with SC continuous testosterone therapy

- Hot flashes, sweating
- Heart discomfort
- Insomnia , sleep problems
- Depressive mood, Irritability, Anxiety
- Physical fatigue, Memory loss
- Sexual dysfunction
- Incontinence, bladder problems
- Vaginal dryness
- Joint and muscular pain

2010
Breast Cancer Symposium

Additional potential benefits in breast cancer survivors

- Testosterone protects against bone loss
- Testosterone stimulates bone marrow and enhances immune function

2010
Breast Cancer Symposium

Background

- Evidence supports that testosterone is breast protective^{2,3}
- Testosterone can be aromatized to estradiol which may have adverse effects on breast cancer proliferation
- Third generation aromatase inhibitors effectively inhibit the aromatization of testosterone to estradiol

2010
Breast Cancer Symposium

Preliminary data: 35 male patients

- 12 mg of anastrozole, a third generation aromatase inhibitor (AI), delivered *subcutaneously* by pellet implant, with up to 1200 mg of testosterone, effectively prevented the conversion of testosterone to estradiol in male patients with previously elevated estradiol levels

2010
Breast Cancer Symposium

Subcutaneous delivery (implants)

- Consistent delivery and consistent absorption
- Effective therapy
- Avoids entero-hepatic circulation
 - Bypasses liver
 - Does not affect clotting factors
 - Absence of GI side effects
- Circadian release
- No compliance issues
- Well tolerated
- Simple procedure to insert



2010
Breast Cancer Symposium

Testosterone-Anastrozole Implant

- 3.1 x 6.1 mm implant
 - 60 mg testosterone
 - 4 mg anastrozole

Powdered is compressed and sterilized

- Dose females: 2 implants
 - 120 mg testosterone
 - 8 mg of anastrozole



2010
Breast Cancer Symposium

Simple 2 minute Procedure



2010
Breast Cancer Symposium

Methods

- Breast cancer survivors were referred from their oncologists or self-referred (with permission from oncologist) for symptoms of androgen deficiency including bone loss
- Prior to July 2009, oral AI therapy was prescribed in conjunctions with SC testosterone in ER positive patients

2010
Breast Cancer Symposium

Methods

- Data was available on 75 testosterone-anastrozole inserts performed in 43 of 55 breast cancer survivors treated between July 2009 and May 2010

2010
Breast Cancer Symposium

Patient Demographics

- 38/43 patients were > 5 years from diagnosis
- 40/43 tumors were ER pos / non-invasive Ca
- Tumor Stage
 - 8 DCIS, 1 LCIS
 - 19 Stage I
 - 10 Stage II
 - 1 Stage III
 - 4 Stage IV

2010
Breast Cancer Symposium

Methods: procedure, testing

- Two anastrozole-testosterone (A-T) implants (120 mg testosterone, 8 mg anastrozole) were inserted subcutaneously (SC) using local anesthesia in the upper gluteal area
- Serum testosterone and estradiol levels were measured two weeks following implantation

2010
Breast Cancer Symposium

Results (Clinical)

- Subcutaneous testosterone-anastrozole therapy was effective in treating symptoms of hormone/androgen deficiency in breast cancer survivors
- All patients achieved relief of symptoms with therapeutic testosterone levels
 - Mean: 281 ng/dl, range: 120-518 ng/dl

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Breast Cancer Symposium

Results

- In 70 of 75 (93.3%) testosterone-anastrozole pellet insertions (43 patients), serum estradiol measured ≤ 30 pg/ml
- A single post-menopausal patient on A-T had an estradiol level > 40 pg/ml
 - Subsequent level measured < 30 pg/ml

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Breast Cancer Symposium

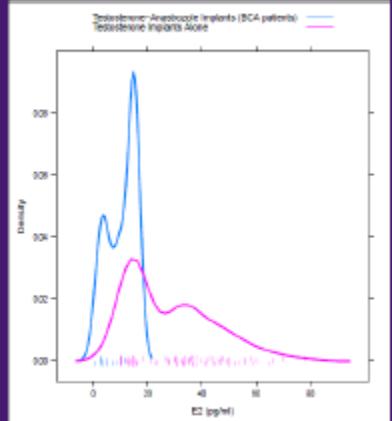
Results: E2 levels T alone vs. A-T

- Control group (n=119)
 - Post menopausal females treated with Testosterone implants alone (T)
- Estradiol levels: T vs. A-T
 - 42% (50/119) of patients treated with Testosterone alone had an E2>30 pg/ml
 - 6.7% (5/75) of patients treated with Anastrozole in combination with Testosterone (A-T) had an E2>30 pg/ml

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Breast Cancer Symposium

Estradiol Density Plot

The levels of Estradiol (E2) in the group with the aromatase inhibitor is significantly less than in the group without it (2-sample Wilcoxon rank sum test, P<0.0001). The separation of E2 in both groups is almost disjoint as illustrated by the kernel density plot.



2010
Breast Cancer Symposium

Clinical follow up

- There have been no adverse drug events in over 170 insertions in 67 breast cancer survivors (Through September 2010)
- No breast cancer survivor treated with subcutaneous testosterone therapy has been diagnosed with recurrent disease in up to 4 years of therapy

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Breast Cancer Symposium

Results

- There has been no progression of disease in 2 ER pos patients and 1 ER neg patient with metastatic disease treated for up to 30 months
 - The 4th patient presented with active disease and has responded to chemotherapy with minimal side effects from the chemotherapy. She continues on therapy and disease is stable.

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Breast Cancer Symposium

Conclusion

- The combination of testosterone with anastrozole, delivered subcutaneously as a pellet implant, provides therapeutic levels of testosterone without elevating estradiol levels

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Breast Cancer Symposium

Current & Future Studies

- Testosterone Implant-Breast Cancer Incidence Trial (Current) Glaser, Dimitrakakis
 - IRB approved, 10 year prospective study looking at the incidence of breast cancer in pre and post menopausal women treated with subcutaneous testosterone therapy
- ATTICA Breast* Trial (Future) Glaser, Dimitrakakis
 - Randomized, placebo controlled trial treating BrCa survivors on no current therapy, with SC A-T implants

*Anastrozole-Testosterone Therapy in CA Breast
Pending IRB approval and Funding

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Breast Cancer Symposium

References

1. Glaser R, Wurtzbacher, D, Dimitrakakis C. Efficacy of Testosterone Therapy Delivered by Pellet Implant. *Maturitas* 2009; 63(Suppl 1):283.
2. Dimitrakakis C, Bondy C. Androgens and the breast. *Breast Cancer Research* 2009;11(5):212.
3. Traish AM, Fetten K, Minor M, Hansen ML, Guay A. Testosterone and risk of breast cancer: appraisal of existing evidence. *Hormone Molecular Biology and Clinical Investigation*. 2010; 2 (1): 177

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Breast Cancer Symposium

APPENDIX XV: SUBCUTANEOUS TESTOSTERONE IMPLANTS ON LIPID PROFILES**Subcutaneous testosterone implants on lipid profiles**

Beneficial effects of subcutaneous testosterone therapy on lipid profiles in women

EMAS
Athens
2012

R. Glaser, C. Dimitrakakis

Introduction

This study was designed to evaluate the effect of subcutaneous testosterone implant therapy on lipid profiles in female patients.

Methods

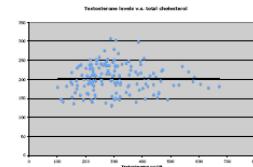
As part of a 10-year IRB approved trial on the effect of testosterone implant therapy on the incidence of breast cancer (Glaser, Dimitrakakis), testosterone levels and lipid profiles were examined in 154 pre- and post-menopausal patients treated at the clinic February-April 2010. All participants had been on testosterone therapy for a minimum of one year (mean 28.1 ± 10.4 months, range 12-56 months).

Spearman's rank correlation coefficient (ρ) was used to determine the relationship between total testosterone levels and total cholesterol, HDL, LDL, VLDL and TG. Significance was determined by a 't' transformation of ' ρ ' to a Student's t-statistic, n-2 degrees of freedom.

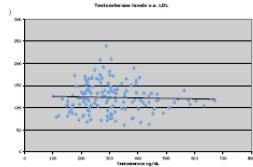
Results

Four weeks following testosterone pellet implantation:

- The mean total testosterone level was 299.36 ± 107.34 ng/dL.
- There was no correlation between testosterone levels and total cholesterol ($\rho = 0.014$, $P = 0.863$)

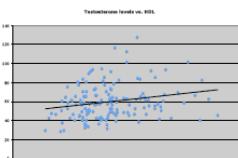


- There was no correlation between testosterone levels and LDL ($\rho = -0.033$, $P = 0.692$).

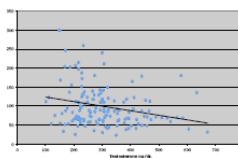
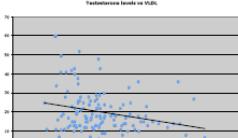


Results

• There was a significant positive correlation between testosterone levels and HDL ($\rho = 0.223$, $P = 0.006$).



• There was a significant inverse correlation between testosterone levels and VLDL ($\rho = -0.283$, $P = 0.027$) and TG ($\rho = -0.334$, $P = 0.006$).



Results

Only one cardiac event was reported in over 3,000 women years of therapy.

- A 103 kg. smoker developed a DVT on testosterone implant therapy and was subsequently diagnosed with 'Lupus antibody prothrombotic disorder'. Therapy with testosterone implants was continued.

Conclusion

Long-term, subcutaneous testosterone therapy has a beneficial effect on lipid profiles in female patients.

Higher levels of testosterone were associated with higher levels of HDL, lower levels of VLDL and lower levels of TG.

There has been no evidence that subcutaneous testosterone therapy has an adverse effect on the cardiovascular system in our patient population.

Keywords: Androgens, testosterone implant, women, lipid profiles

Abbreviations: HDL, high density lipoproteins; LDL, low density lipoproteins; VLDL, very low density lipoproteins; TG, triglycerides

APPENDIX XVI: DRUG INFORMATION PROVIDED BY [REDACTED] FEBRUARY 15, 2012

Drug Information for [REDACTED]
15 February 2012
[REDACTED]

Testosterone-Anastrozole (TE-A) Subcutaneous Implant

SUMMARY PRODUCT INFORMATION

Route of Administration	Dosage Form/Strength ^{a,b}	Non medical ingredients
Subcutaneous/subdermal	60 mg Testosterone 4 mg Anastrozole per implant	Stearic acid

^a Males: 4 TE-A implants (240 mg TE, 16 mg A)*

^b Females: 2 TE-A implants (120 mg TE, 8 mg A)**

INDICATIONS AND CLINICAL USE

Orphan Drug Indication

Hormone resistant ER positive, metastatic breast cancers

Additional Indications

The Testosterone-Anastrozole (TE-A) subcutaneous implant may be useful in any patient who would benefit from testosterone therapy in whom elevated estradiol is not desired or is contraindicated.

Male patients

Male patients with symptoms of androgen deficiency who would benefit from testosterone replacement therapy (TRT) who have elevated estradiol levels (baseline), elevated estradiol levels on testosterone therapy, prostate issues (BPH, elevated PSA, prostate cancer), breast cancer and/or symptoms of elevated estrogen that include:

- Weight gain, increased abdominal fat
- Lack of effect from TE
- Anxiety, irritability, aggression
- Breast discomfort or enlargement, gynecomastia
- Prostate problems (BPH, prostate cancer, elevated PSA)
- Elevated HbA1c or diabetes, usually in association with weight
- Fluid retention

Breast cancer survivors or women at increased risk for breast cancer

Pre and post-menopausal breast cancer survivors or women at an increased risk for breast cancer with symptoms of androgen (hormone) deficiency in who elevated estrogen levels are contraindicated.

- *Pending randomized controlled trials:* First-line therapy for breast cancer, second-line therapy in women who become resistant to aromatase inhibitor (AI) therapy or in patients who cannot tolerate oral AI therapy.

Pre and post-menopausal women with gynecologic pathology

Pre and post-menopausal women with endometriosis, uterine fibroids or dysfunctional uterine bleeding (DUB).

Obese patients

Male or female patients with elevated estradiol to testosterone ratio and are unable to lose weight.

BACKGROUND

Testosterone, delivered by subcutaneous pellet implant has been used in the United States, Europe and Australia in both men and women to effectively treat symptoms of testosterone deficiency. The sustained release Testosterone-Anastrozole (TE-A) implant provides continuous, controlled release and consistent absorption of both testosterone and anastrozole (an aromatase inhibitor).

Potential benefits of testosterone delivered by subcutaneous implantation include increased bone density, increased energy, relief of lethargy, relief of depression and anxiety, improved memory and concentration, improved sleep, increased muscle mass, decreased fat mass, relief of aches and pains, relief of breast pain, relief of migraine headaches, restoration of sex drive and libido, menopausal syndrome relief (hot flashes, night sweats), relief of nocturia, urinary incontinence and vaginal symptoms (Glaser). These results are not documented with other methods of delivery. In pre and post-menopausal women, testosterone has been successfully used in dysmenorrheic patients with endometriosis or small fibroids and to prevent uterine bleeding caused by estrogens. In addition, testosterone, delivered by pellet implant and has been used as a palliative measure in carcinoma of the breast. Testosterone has been shown to lower the risk of breast cancer in women on estrogen/progestin therapy, decrease proliferation of breast tissue, reduce levels of ER (estrogen receptor) alpha, enhance the immune system, and increased red blood cell production.

However, higher levels of estradiol, from the aromatization of testosterone, may counteract the benefits of testosterone therapy. In addition, local over-expression of aromatase and subsequent elevated estradiol may stimulate breast tissue, breast cancer cell growth and adversely affect the prostate gland.

Anastrozole is an aromatase inhibitor and has been used orally as an adjuvant therapy in breast cancer in post-menopausal women. By blocking the enzyme 'aromatase', anastrozole inhibits the conversion of testosterone to estradiol thereby preventing the stimulation of breast tissue and breast cancer cells by estradiol. Oral anastrozole has been used in male patients to prevent the conversion of testosterone to estradiol, raising testosterone levels and lowering estradiol levels.

The combination TE-A implant provides continuous, effective, yet significantly lowered dosing of anastrozole with fewer side effects than oral anastrozole therapy. In addition, by avoiding the GI tract, there is no nausea or adverse effects to the GI tract. Subcutaneous delivery also avoids adverse effects from 'first pass' metabolism in the liver including hepatic-toxicity or increase in clotting factors. Subcutaneous delivery of the combination of testosterone and anastrozole also eliminates patient non-compliance.

CLINICAL PHARMACOLOGY

Testosterone is the primary endogenous androgen in both men and women. Testosterone exerts its effect through the androgen receptor. Androgen receptors are located in the brain, bones, breast, muscle, nerves, heart, vascular tissue, skin, hair follicles, ovaries, testicles, and fatty tissue.

Cells in the testis, ovary, and adrenal cortex synthesize endogenous testosterone. Therapeutically, testosterone is used in the management of hypogonadism (androgen deficiency), either congenital or acquired. Testosterone is also the most effective exogenous androgen for the palliative treatment of carcinoma of the breast in postmenopausal women. Testosterone levels decline over the lifespan in both men and women. Androgen production in women declines steeply in the early reproductive years (Davison). A woman of 40 has half the mean plasma total testosterone of a 21 year old (Zumoff).

In males, (high levels of) endogenous androgens are responsible for the normal growth and development of the male sex organs and for maintenance of secondary sex characteristics. These effects include the growth and maturation of prostate, seminal vesicles, penis, and scrotum; the development of male hair distribution, such as beard, pubic, chest and axillary hair; laryngeal enlargement, vocal chord thickening, alterations in body musculature, and fat distribution. Drugs in this class also cause retention of nitrogen, sodium, potassium, phosphorous, and decreased urinary excretion of calcium. Androgens have been reported to increase protein anabolism and decrease protein catabolism. Testosterone also affects the formation of erythropoietin, and blood glucose. Androgens have high lipid solubility, enabling them to rapidly enter cells of target tissues. Within the cells, testosterone undergoes enzymatic conversion to 5-alpha-dihydrotestosterone and forms a loosely bound complex with cytosolic receptors. Androgen action arises from the initiation of transcription and cellular changes in the nucleus brought about by this steroid-receptor complex. Effect from testosterone has been shown to be dose dependent in both male and female patients.

Anastrozole: Anastrozole is a potent and selective non-steroidal aromatase inhibitor. It significantly lowers serum estradiol concentrations and has no detectable effect on formation of adrenal corticosteroids or aldosterone. In men and postmenopausal women, estrogens are mainly derived from the action of the aromatase enzyme, which convert androgens (primarily androstenedione and testosterone) to estrone and estradiol. The suppression of estrogen biosynthesis in peripheral tissues and in the cancer tissue itself can therefore be achieved by specifically inhibiting the aromatase enzyme.

Subcutaneous implants: Subdermal pellet implantation has been proven an effective treatment modality and method of delivery **since 1940**. Absorption of testosterone and anastrozole from the **subcutaneous combination implant** occurs via uniform erosion of the pellets surface providing a near linear release of both active ingredients. The bioavailability of testosterone and anastrozole from the combination implant is virtually complete (i.e. 100% bioavailability). There is no first pass hepatic inactivation and all released testosterone and anastrozole are absorbed into the circulation. There is no increase in clotting factors or increased risk of blood clots with subcutaneous testosterone-anastrozole.

The duration of action of the **testosterone-anastrozole implant** is 4-5 months in men and 3-4 months in women. Implants provide steady state, sustained release of both TE and A. In women, approximately 0.08 mg of anastrozole is released daily compared to a daily oral dose of 1 mg. In men, approximately 0.12 mg of anastrozole is released daily. Release rates from subcutaneous testosterone implants have been well documented. Because the anastrozole is combined/mixed with the testosterone (ratio of 1:15), the release rates would be identical to those established in the literature for testosterone alone.

Preparation and Storage

Testosterone-anastrozole pellets for subcutaneous/subdermal implantation will be compounded to contain 60 mg testosterone and 4 mg anastrozole per pellet, with stearic acid as an inactive ingredient. The placebo pellets will be similar in appearance to the testosterone-anastrozole pellets. The placebo pellets will contain approximately 98% USP cholesterol and 2% stearic acid. The finished pellets are stored at controlled room temperature (20-25°C).

Administration

The Testosterone-Anastrozole pellets are implanted subcutaneously through a 5 mm incision in the upper gluteal region under local anesthesia using a disposable trocar kit. The incision is closed with a steri-strip.

Male patients are treated with four TE-A implants. Each TE-A implant is placed ‘end on end’ in one of four tracks along with 2-3 additional 75 or 100 mg TE implants. Each row of implants is advanced into the subcutaneous tissue through a cannula.

The TE-A implants release adequate levels of testosterone and anastrozole for 3 months, on average, in women and 4-5 months in men. The procedure takes 2-3 minutes in females and 3-4 minutes in males.

No change in dose in patients with renal or hepatic impairment based on studies with significantly higher doses of oral anastrozole (1 mg po daily vs. 0.08 mg SC released daily).

DRUG INTERACTIONS

No known drug interactions.

CLINICAL EFFICACY

Male patients

Preliminary data (7/2009-3/2010)

- 35 TE-A implants in male patients with previously elevated estradiol.
- In male patients, the mean estradiol level was **63.9 pg/ml** prior to SC anastrozole therapy vs. **17.4 pg/ml** following subcutaneous anastrozole therapy. The **ratio of T/E2** (testosterone/estradiol) prior to subcutaneous anastrozole therapy (baseline and testosterone treated patients) was 10.26 (ng/dl: pg/ml) vs. 59.37 following SC anastrozole/testosterone therapy, approximately a 6-fold reduction.

Male patients treated Oct 2010-Jan 2011.

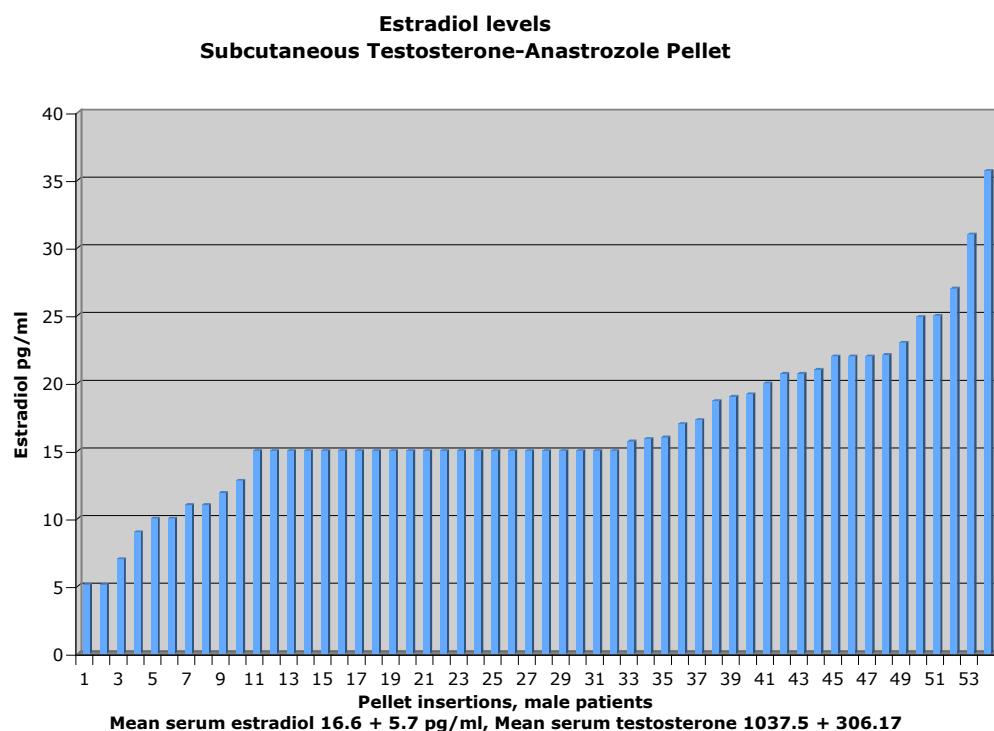


Fig. 1. Estradiol levels in 54 male patients, with previously elevated (> 30 pg/ml) estradiol levels, treated with 12 - 16 mg anastrozole in combination with testosterone as a subcutaneous implant.

Results: Mean serum estradiol^a 16.6 ± 5.7 pg/ml, Mean serum testosterone 1037.5 ± 306.17 ng/dl

^aEstradiol levels of <30 pg/ml were given a value of 15 pg/ml.

Beginning Jan. 2011, a dose of 16 mg of anastrozole (4, implants of 60 mg TE, 4 mg A with additional TE) is being prescribed. There have been no side effects or adverse drug events from the 16 mg dose of anastrozole.

Pharmacokinetics of TE-A implant in an obese, diabetic male patient:

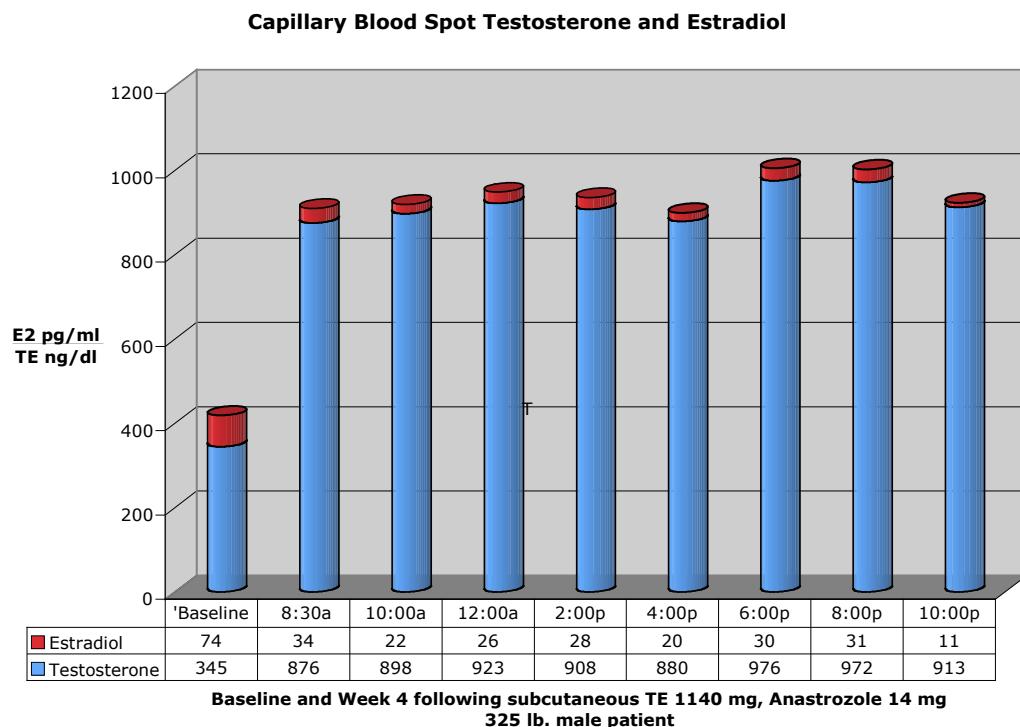


Fig.2 Capillary blood spot was measured at baseline and every two hours 4 weeks following insertion of TE 1140 mg with Anastrozole 14 mg. (total dose)

Results: Baseline E2 was 74 pg/ml. Baseline TE was 345 ng/dl. Mean E2 level on therapy was 17.4 pg/ml. Mean testosterone on therapy was 918 ng/dl.

Female Patients

Data in breast cancer patients presented at the American Society of Clinical Oncology (ASCO) Breast Cancer Symposium October 2010.



Subcutaneous testosterone-anastrozole implant therapy in breast cancer survivors.

Sub-category:

Hormones and Hormone Receptor Biology

Category:

Prevention, Survivorship & Health Policy

Meeting:

2010 Breast Cancer Symposium

Session Type and Session Title:

General Poster Session D

Abstract No:

221

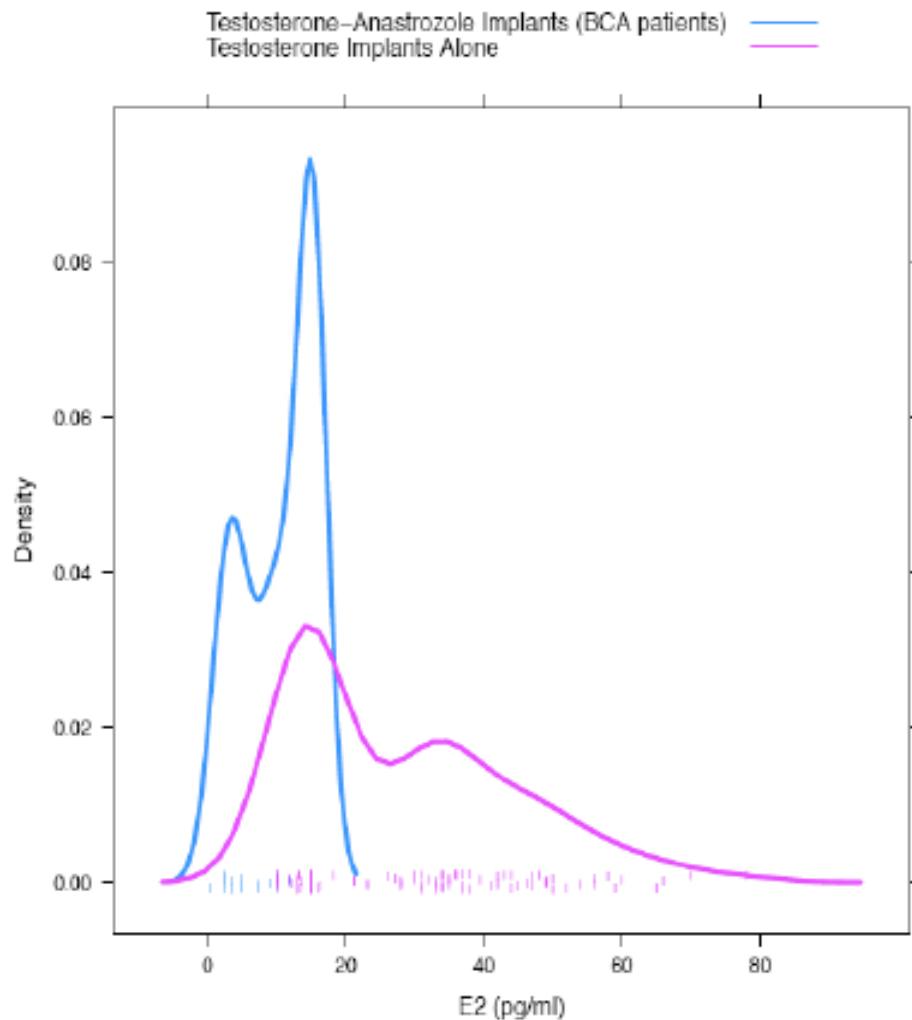
Author(s):

R. L. Glaser; Department of Surgery, Wright State University, Dayton, OH

Abstract:

Background: Breast cancer survivors commonly experience severe symptoms of hormone deficiency that can adversely affect their health and quality of life. Beneficial effects of subcutaneous testosterone therapy include relief of hot flashes, heart discomfort, insomnia, depression, irritability, anxiety, fatigue, memory loss, sexual problems, incontinence, vaginal dryness, and joint and muscular pain. In addition, testosterone protects against bone loss, stimulates bone marrow and enhances immune function. Evidence supports that testosterone is breast protective. However, aromatization of testosterone to estradiol may have adverse effects on breast cancer proliferation. Unpublished data (Glaser) had previously demonstrated that 12 mg of anastrozole, delivered subcutaneously with up to 1,200 mg of testosterone, effectively prevented the conversion of testosterone to estradiol in men with elevated estradiol levels. **Methods:** To evaluate the efficacy of subcutaneous anastrozole in maintaining therapeutic testosterone levels without elevating estradiol in breast cancer survivors, two 3.1 x 6.1 mm implants containing a total of 120 mg testosterone and 8 mg of anastrozole were implanted in the upper gluteal area of 55 patients. Serum levels of testosterone and estradiol were measured 2 weeks following pellet insertion. **Results:** Testosterone-anastrozole therapy was effective in treating symptoms of androgen (hormone) deficiency. In all but 5 of 75 anastrozole/testosterone subcutaneous pellet insertions, serum estradiol levels measured < 30 pg/ml with therapeutic testosterone levels (mean: 281, range: 120-518 ng/dl). A single post-menopausal patient had an estradiol level > 40 pg/ml. A subsequent level measured < 30 pg/ml. There have been no adverse drug events from subcutaneous testosterone-anastrozole therapy in over 110 insertions. No breast cancer survivor treated with testosterone implants has been diagnosed with recurrent disease in up to 3 years of therapy. There has been no progression of disease in 2 patients with metastasis. **Conclusions:** The combination of testosterone with anastrozole, delivered subcutaneously, provides therapeutic levels of testosterone without elevating estradiol levels.

Estradiol density plot: Female patients treated with TE implant alone (non-cancer patients) or the combination TE-Anastrozole implant (Breast Cancer survivors).



The level of Estradiol (E2) in the group with the aromatase inhibitor is significantly less than in the group without it (2-sample Wilcoxon rank sum test, $P < 0.0001$).

The separation of E2 in both groups is almost disjoint as illustrated by the kernel density plot.

Estradiol levels after testosterone implant alone (pink) vs. the combination testosterone-anastrozole implant (blue). Estradiol levels of < 30 pg/ml were given a value of 15 pg/ml.
The combination TE-A implant provided therapeutic levels of testosterone without elevating estradiol.
No patient has had recurrent disease on the combination implant with up to 2 years follow up.

See ASCO Breast Cancer Symposium poster presentation and/or power-point presentation for additional information.

DRUG SAFETY AND ADVERSE EVENTS

No major adverse drug events (ADE) in over 1,200 insertions.

CONTRAINDICATIONS

Prior allergic reaction to anastrozole.

DOSSAGE AND ADMINISTRATION

Dosing Males

- In androgen deficient males, 4 implants of 60 mg TE, 4 mg Anastrozole (Total dose: 240 mg TE, 16 mg A) are used in combination with (additional) therapeutic doses of testosterone implants.
 - July 2009- Dec. 2010, the majority of men were treated with 3, 60 mg TE, 4 mg A implants for a total dose of 12 mg anastrozole. (In addition to male doses of TE implants)
- In symptomatic males with elevated estrogen levels and mid-range testosterone levels, 4 implants (240 mg TE, 16 mg A) may be used alone or with lower doses of TE (i.e. < 600 mg total TE).

Dosing Females

- 2 implants of 60 mg TE, 4 mg of Anastrozole. Total dose: 120 mg TE, 8 mg A (Feb 2010-current)
 - Additional testosterone in non cancer patients, based on weight
 - July 2009- February 2010 dosing: T120-A 7.0 mg.

CLINICAL EXPERIENCE

1408 TE-A pellet insertions performed through **July 2011**

Inserts in Male patients: 401

Over two-thirds (69.34%) of hypogonadal male patients on testosterone replacement therapy had indications for, and were treated with the combinations TE-A implants.

Indications for TE-A therapy in men

1. **86% had elevated estradiol levels** (baseline or on TE therapy)
 - a. Symptoms of elevated estradiol
 - i. Weight gain, increased abdominal fat
 - ii. Lack of effect from TE
 - iii. Anxiety, irritability, aggression
 - iv. Breast discomfort or enlargement, gynecomastia
 - v. Prostate problems
 - vi. Elevated HbA1c or diabetes, usually in association with weight
 - vii. Fluid retention
2. **8%** had prostate issues (BPH, elevated PSA, prostate ca) and a normal estradiol level
3. **6%** Other
 - a. Breast pain, obesity, abdominal fat without significant elevation of estradiol (≤ 30 pg/ml) at baseline

Inserts in Breast Cancer survivors: 272 through April 2011

Indications for TE-A therapy in breast cancer survivors

1. **100% of patients were treated for symptoms of androgen (hormone) deficiency including bone loss**
 - Testosterone therapy is effective in treating symptoms of androgen (hormone) deficiency* including:
 - Hot flashes, sweating
 - Breast pain, fibrocystic disease
 - Heart discomfort (heart skipping, racing, chest tightness)
 - Sleep problems (insomnia, difficulty falling asleep, waking)
 - Depressive mood, feeling sad, down, lack of drive, mood swings

- Irritability, feeling nervous, inner tension, feeling aggressive
- Anxiety, inner restlessness, feeling panicky
- Physical exhaustion, decrease in performance
- Mental exhaustion, impaired memory, decrease in concentration, forgetfulness
- Sexual problems (change in desire, activity and satisfaction)
- Bladder problems (difficulty urinating, frequency, bladder incontinence)
- Dryness of vagina (burning, difficulty with intercourse)
- Joint and muscular discomfort (pain in joints, rheumatoid complaints)
- Muscle weakness
- Bone loss
- The combination of testosterone with anastrozole delivered subcutaneously provides therapeutic levels of testosterone without elevating estradiol levels.

Inserts in Females without breast cancer: 425

20% of females (without breast cancer) are treated with the combination (TE-A) implant

Indications

1. Weight gain, inability to lose weight or belly fat
 - a. With or without elevated estradiol levels
 - b. Pre and post menopausal females
2. Gynecologic disease associated with excess estrogenic stimulation
 - a. Uterine fibroids
 - b. Dysfunctional Uterine Bleeding (DUB), Menorrhagia.
 - c. Endometriosis
3. Women at increased risk for breast cancer who desire hormone therapy
 - a. Family history of breast cancer (one or more first degree relatives)
 - b. Atypical ductal hyperplasia
 - c. Atypical lobular hyperplasia
 - d. Multiple breast biopsies
4. Women treated with testosterone therapy with symptoms of estradiol excess, with or without elevated serum estradiol levels (Serum estradiol levels do not always reflect cellular aromatase activity).
 - a. Fluid retention, weight gain, belly fat, elevated blood sugar
 - b. Menstrual or migraine headaches
 - c. Breast pain, fibrocystic breast disease
 - d. PMS
 - e. Irritability
 - f. Anxiety

Currently treating approximately 110 patients per month with the combination implant

Clinical Experience UPDATE: **July 2011, n=1408** TE-A insertion procedures

- Male patients
Total, **502** insertions from 7/13/09-7/12/11
223 insertions in 170 male patients 1/4/11-7/12/11
- Breast Cancer patients
***314** insertions in 89 patients 7/15/09-7/8/11
- Female patients without BrCa, total **592** insertions 9/9/09-7/8/11
316 inserts in 197 patients **1/4/11-7/8/11**

***UPDATE: 430** insertions in breast cancer patients 7/15/09-9/2/12

Testosterone-Anastrozole Combination Implant
 Number of patient insertions July 2009-July 2011 (n=1408)
 Percent of patients treated with combination January-July 2011

■ 70% of male patients ■ 95% of breast cancer patients ■ 30% of females w/o BrCa

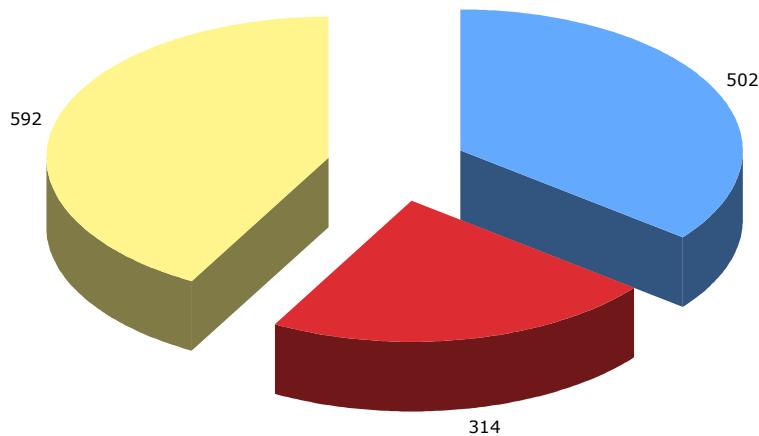


Fig. 1 Number of insertions (July 2009-2011) in male patients, breast cancer patients, and female patients without breast cancer, treated with the Testosterone-Anastrozole combination subcutaneous implant.

Legend: Percent of patients treated with the combination implant; 70% of male patients (170/240), 95% of breast cancer patients (60/63) and 30% of female patients without breast cancer (197/655) were treated with the combination implant 4 January-8 July 2011.

UPDATE INSERTIONS
 24 September 2011, n=1760

REFERENCES SUPPORTING CONCEPT

- There is an age related increase in aromatase activity, particularly in fatty tissue (Jordan 07, Leder 04, Nelson 01)
- Local increased aromatase activity, elevated estradiol levels and lowered testosterone levels (i.e. elevated E2/T ratio) adversely affect the prostate gland; BPH and Prostate Cancer (Ellem 10, Ricke 09, Bosland 06)
- Increased aromatase activity in gynecomastia (Rhoden 04, Czajka-Ofinec 08)
- Aromatase inhibitors as therapy in male breast cancer (Doyen 10, Carmona-Bayonas 07)
- Testosterone and aromatase inhibitors in gynecologic disease (Bulun 01, 05, Morales 05)
- Obesity and increased aromatase activity (Cohen 08, Loves 08)
- Non adherence to *oral* aromatase inhibitor therapy (Sedjo 10)
- Aromatase inhibitors and testosterone neuroprotective (Saldanha 09, Herzog 10, Fargo 07, 09)
- Increased body fat (bmi), decreased muscle mass (lean body mass) with decreased testosterone, increased aromatase and subsequent increased estradiol (Cohen 01)
- See FOLDER 'Androgens and the breast' and 'Aromatase'

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APPENDIX XVII: ACCUPEN™ DISPENSING DEVICE APPLICATION DIRECTIONS

AccuPen™ Dispensing Device

Measured Dose for Measured Results

Our pharmacy is proud to introduce you to the patient-friendly AccuPen, an accurate dispensing device for your topical compounded preparations. The AccuPen is engineered to be an airless metered dose pump, with a unique design that allows you to conveniently transport your medication. Unique to the AccuPen is a clear window that shows the amount of preparation remaining in the pen.

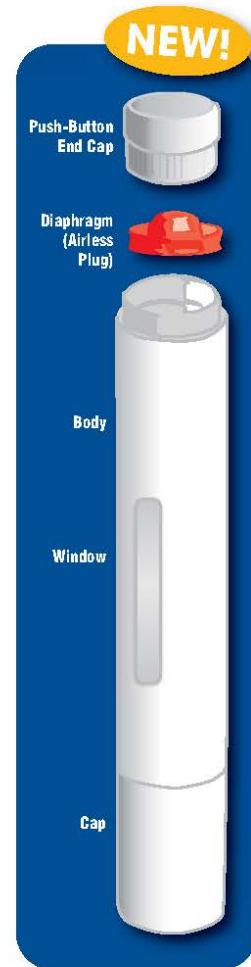
Highlights and Key Characteristics

Unique Locking Feature! Turn the End Cap clockwise to ‘lock’ the device – ideal for traveling, carrying in bags, purses, etc.!



Simple to Use: First, remove the cap. To use the AccuPen, simply turn the Push-Button End Cap counter-clockwise to unlock the actuator, and then push the Push-Button End Cap in the same manner you would push an ink pen. A metered volume of the topical preparation will then be pumped out of the tip! It's that easy!

How It's Made: Sturdy design featuring a polypropylene (PP) body, a low density polyethylene (LDPE) diaphragm, and a PP, LDPE, stainless steel pump.



distributed by
 PCCA

Ask your pharmacist for the AccuPen for easy and convenient dispensing of your topical medications. Have questions? Ask the pharmacy staff – we're here to help!

General Directions for Applying Testosterone Transdermal Gel**Directions:**

- Apply at the same time every day, preferably in the morning. For best absorption, do not apply right before bedtime.
- Apply to non-fat pad areas. These areas include:
 - tops of the feet
 - top of the chest
 - behind the knees
 - inner forearms.

The site should be changed with each application. Never apply to open sores, wounds, or irritated skin.

- Completely clean and dry skin.
- Apply the prescribed amount on the appropriate area. Using a circular motion, rub in the medication until it is absorbed, and the area feels dry.
- There is a slight risk of transferring the medication to other people, surfaces, and pets that you come in contact. To avoid this follow these precautions:
 - Wash your hands thoroughly with soap and water immediately after application to avoid transferring the medication to other people.
 - Let the medicated area dry for a few minutes before getting dressed to prevent your clothes from wiping it off your skin.
 - Allow at least 4 hours for the medication to absorb fully.
 - The application may be washed after 4 hours have elapsed.
 - Use separate hand towels from other members of the family.
 - Wipe off faucet handles after use.
 - Wait at least an hour (preferably 4 hours) after application before bathing or swimming to ensure the best absorption.

Storage:

Store gel at room temperature.

Additional Instructions (if applicable):

Occasionally, the red plunger within the AccuPen Dispensing Device can get stuck and lose contact with the gel. If this happens, the AccuPen device may not deliver an accurate dose. Therefore, please follow the instructions below should this issue happen to your device.

1. Remove the Push-Button End Cap by gently twisting and pulling on the Push-Button End Cap.
2. Once the End-Cap is removed, push the red diaphragm slowly down into the body of the device with a thin utensil (such as a pen or thin object) until it comes into contact with the gel.

Note: There should not be any air between the red diaphragm and the gel preparation for the dispensing device to work properly.

3. Push the end cap down where the lines match up and make sure the cap "clicks" on. You should hear an audible "click" noise.
4. Twist the cap back and forth to ensure it becomes locked and unlocked.
5. Once the cap is in place, turn the Push-Button-End Cap counter-clockwise to unlock the actuator.
6. Then pump the pen twice to re-prime the pen. The pen should now be ready to use once again.