

**CTRF study:**  
**Early Phase II RCT of *Trametes (Coriolus) versicolor* (1500 mg TID PO)**  
**In women with stage (I/II/III) ER/PR negative breast cancer**  
**DRAFT 11/04/05**

**Design**

Two-arm double-blind randomized placebo controlled efficacy 8-week trial followed by an open label 8-week administration of study compound in both arms.

**Outcomes**

*Primary Outcome*

- Mean rate of change (standard deviation) of natural killer cell activity (NKCA)

*Secondary Outcomes*

- Mean rate of change (standard deviation) of white blood cell count, cytokines (TNF- $\alpha$  and IFN- $\gamma$ ) and phagocytic index in monocytes and granulocytes
- Fatigue and quality of life self-assessment scores measured with the FACIT-Fatigue and FACT-B instruments
- Safety and tolerability as measured by frequency and severity of AEs based on NCI Common Toxicity Criteria v. 3.0 and compliance to study as measured by pill count

**Study Population**

Women with stage I/II/III estrogen/progesterone receptor negative breast cancer aged 21-75 years who have undergone surgery (either mastectomy, excisional biopsy, or lumpectomy), followed by an anthracycline-containing adjuvant chemotherapy regimen, usually doxorubicin (Adriamycin™), and have just completed adjuvant radiotherapy. Subjects will be randomized to verum or placebo within 72 hours after completing radiotherapy.

**Study Compound:**

The verum is a US certified organic GMP-compliant biomass powder made from *Trametes (Coriolus) versicolor* (TCv) mycelium and primordia (young fruiting bodies) in 500 mg tablets supplied by Mycology Research Laboratories, United Kingdom. This formula is commercially sold in the United States as Coriolus MRL. The placebo tablets will consist of inert ingredients, mainly cellulose. The dose is 4,500 mg/day, divided into three 1,500 mg doses before breakfast, lunch and dinner. This dosing lies within the range naturopathic physicians commonly prescribe to breast cancer patients after radiotherapy (3-6 mg/day).

**Rationale for the Open Label Follow-Up**

We are proposing an eight week open label follow up for all women enrolled in the study after completing the 8-week RCT for three reasons: 1) for ethical reasons, 2) to enhance our ability to successfully recruit 50 subjects, and 3) to collect preliminary, albeit, uncontrolled data on the longer term effects of TCv treatment in the verum arm and the impact of delaying TCv treatment for eight weeks in the placebo arm.

Even though a placebo arm is necessary in this design to measure natural recovery of NKCA after radiation and to measure the placebo effect, recruitment for placebo-controlled trials among oncology patients is inherently difficult as most study participants desire what they consider to be the study's active compound (i.e. TCv). We anticipate an

insurmountable problem in recruiting sufficient numbers of study subjects to power a placebo-controlled randomized controlled trial (RCT) that does not provide TCv to the subjects in the placebo group. Moreover, since TCv is a standard of care among NDs for women with breast cancer, withholding it from the women randomly assigned to the placebo arm would seem unethical. Thus, we are proposing an 8-week open label follow-up phase during which both arms receive TCv at 4,500 mg/day. We anticipate successful recruitment using this design.

The primary aim of this study is to evaluate differences in NKCA changes between the verum and placebo arms during the first eight weeks of the study. However, we will also compare the rate of change of NK cell activity from weeks 0-8 and 8-16 for the verum and placebo arms. In the verum arm, we can do a preliminary analysis on whether NK cell activity continues to rise or plateaus with longer duration of administration (16 weeks). In the placebo arm, we can do a preliminary analysis of the impact, if any, of Tv administration that is begun right after radiotherapy versus eight weeks later.

### **Observational sub-study**

An additional time point will be added to a subset of 10 of the 50 study participants within 2 weeks after breast surgery and before chemotherapy to gather preliminary data on the impact of chemotherapy on NK cell activity. These preliminary data will also allow us to explore the possible relationship between pre-chemotherapy NK cell activity levels and response to study (i.e., TCv or placebo administration) during the post-radiation period.

### **Sample size calculations (as initially proposed)**

The primary study outcome for the proposed study is the percent change in NK cell activity (NKCA) associated with oral administration of TCv (4,500 mg/day). For this end point it is important to have adequate numbers of subjects enrolled in order to detect possible differences in NKCA between subjects in the two groups at baseline and at the end of 8 weeks of administration, of either placebo or TCv. Each woman's baseline NKCA will serve as a "within subject" control for testing for change in NKCA over the first 8 weeks of study compound. Estimates of the mean baseline NKCA value and the standard deviation (SD) for the amount of NKCA increase within a subject were based on a study in humans describing mean % cytotoxicity of peripheral blood mononuclear cells from bladder cancer patients against T24 human bladder cancer cells after being treated *in vitro* with TCv extracts (Mizutani and Yoshida 1991). The mean ( $\pm$  standard deviation) pre-intervention control value was  $10\pm 1\%$  cytotoxicity and the post-intervention mean was  $27\pm 1.9\%$  cytotoxicity – a mean increase of 270%. This is an *in vitro* effect size which is difficult to extrapolate to *in vivo* activity. The *in vivo* effect may be smaller than a 270% increase and may have a larger SD. Therefore, we are estimating that a moderate effect size in a human clinical trial with cancer patients would be a change of 75 – 85% in NK cell activity with TCv administration. Dr. Standish has observed increases in NK cell functional activity of 100-200% in her breast cancer patients after 4-12 weeks of TCv following completion of radiotherapy. Assuming this uncontrolled clinical effect size may have been enhanced by a placebo effect, we are estimating that this RCT should be powered to detect a mean difference of at least 75% change in NK cell activity in the subjects in group 1 (placebo) compared to those in group 2 (TCv) after eight weeks on study. Using an alpha level of 0.05 and power of 80%, a two-sided between subjects t-test would require 20 evaluable subjects in each of two groups. With an attrition rate at 20% we will need 25 subjects per group for adequate power. Based on this reasoning we will enroll 50 (25 x 2 groups) women with breast cancer into this placebo controlled RCT.

### **REFERENCES:**

Mizutani, Y. and O. Yoshida (1991). "Activation by the protein-bound polysaccharide PSK (Krestin) of cytotoxic lymphocytes that act on fresh autologous tumor cells and T24 human urinary bladder transitional carcinoma cell line in patients with urinary bladder cancer." J Urol **145**(5): 1082-1087.

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