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Flor-Essence® herbal tonic does not inhibit estrogen receptor negative mammary tumor development in a transgenic mouse model

L. Michelle Bennett¹, Jennifer L. Montgomery², N. Keith Collins¹, Seth M. Steinberg³, and Kristen S. Kulp²

Jennifer L. Montgomery: Montgomery23@lnl.gov; N. Keith Collins: kc@nih.gov; Seth M. Steinberg: steinbes@mail.nih.gov; Kristen S. Kulp: Kulp2@lnl.gov

¹Laboratory of Cancer Prevention, CCR, NCI-Frederick, 567/154, Ft. Detrick, MD 21702

²Chemistry, Materials and Life Sciences Directorate, Lawrence Livermore National Laboratory, Livermore, CA 94550

³Biostatistics and Data Management Section, CCR, NCI, NIH, 6116 Executive Blvd, Bethesda, MD 20892

Abstract

Women who are diagnosed with breast cancer often self-administer complementary and alternative medicines to augment their conventional treatments, improve health, or prevent recurrence. Flor-Essence® herbal tonic is a complex mixture of eight herbal extracts used by cancer patients because of anecdotal evidence that it can treat or prevent disease. In this study four experimental groups of female MMTV-Neu mice were left untreated or treated with 3% Flor-Essence® *in utero*, from birth until 5 weeks of age, or throughout their lifetime. Palpable mammary tumor incidence and body weight was determined weekly for each group. The mice were sacrificed at 28 weeks of age and mammary tumors were enumerated to determine average tumor incidence and multiplicity for each group. Female mice exposed to Flor-Essence® herbal tonic *in utero* weighed significantly more than the control group ($p < 0.001$). The average tumor incidence and tumor multiplicity in the experimental mice treated with Flor-Essence® herbal tonic did not differ from the control animals. Flor-Essence® does not inhibit mammary tumor incidence or mammary tumor multiplicity in MMTV-Neu transgenic mice. Flor-Essence® exposure *in utero* causes increased body weight in experimental animals. This conclusion challenges widely available anecdotal information as well as the hopes of the consumer that this product will inhibit or suppress tumor development.

Lay Abstract—Flor-Essence® herbal tonic is a complex mixture of eight herbal extracts often used by women with breast cancer in hopes that it will help cure disease or prevent recurrence. There is currently very little scientific data to support or refute its self-administration. We tested whether Flor-Essence® would influence tumor development in the mammary glands of a mouse model of Her2/neu breast cancer. The tonic was given at different life stages to determine if timing of the exposure influenced the response to treatment. This report shows that Flor-Essence® did not inhibit mammary tumor development in the treated mice challenging anecdotal information, as well as the hopes of the consumer, that this product will inhibit or suppress tumor development. This report also shows that Flor-Essence® exposure *in utero* causes increased body weight in experimental animals.

Keywords

complementary and alternative medicine; MMTV-Neu; herbal tonic; mammary tumors; transgenic; mouse; dietary supplement; essiac

Introduction

There are reports of herbal tonics being used to treat breast cancer that date back to the 1920's (Kaegi, 1998; Olsen, 1998; Percival, 1994; Snow and Klein, 2001; Tamayo et al., 2000; University of Texas, 2008). There are no epidemiologic data available, and little scientific evidence to support their use as a complementary and alternative medicine (CAM). In addition, there is a serious lack of information about the safety, efficacy, or potential drug interactions of these widely-available tonics. One particular tonic, Flor-Essence® Herbal Tonic, is marketed as a dietary supplement and is widely used by women who have been diagnosed with breast cancer in an effort to treat disease as well as to prevent recurrence (Richardson et al., 2000). Information about product use is largely gained from word of mouth and from the internet. A survey of health food stores reported that twenty-six percent recommended one cup of Flor-Essence® daily to treat breast cancer (Mills et al., 2003).

Flor-Essence® herbal tonic is a complex mixture of eight herbal extracts: burdock root (*Arctium lappa*), sheep sorrel (*Rumex acetosella*), slippery elm (*Ulmus rubra*), Turkish rhubarb (*Rheum palmatum*), watercress (*Nasturtium officinale*), blessed thistle (*Cnicus benedictus*), red clover (*Trifolium pratense*), and kelp (*Laminaria digitata*) (Richardson et al.; Tamayo et al.). Scientific information to support or refute the claims that the herbal extracts found in Flor-Essence® act synergistically to promote human health is lacking. Understanding the effects of Flor-Essence® through rigorous scientific examination is critical for physicians, patients, and health care providers to make informed decisions about the use of CAMs. Recently several reports have been published in the scientific literature that begin to investigate the impact of Flor-Essence® and Essiac® herbal tonics in mutagenicity assays, using cells in culture, and animal models (Bennett et al., 2004; Felton et al., 2004; Ottenweller et al., 2004; Tai et al., 2004).

We previously published the first report of the effect of Flor-Essence® exposure on the mammary gland in an animal model. We demonstrated that Flor-Essence® herbal tonic promotes DMBA-initiated mammary tumors in the Sprague Dawley rat model, a hormonally sensitive animal model for breast cancer (Bennett et al.). In order to complement and extend this initial investigation, we designed a study to evaluate Flor-Essence® in an animal model that develops mammary tumors that are not hormonally responsive.

In the current study, the effect of Flor-Essence® herbal tonic administration on mammary tumor development in the MMTV-Neu transgenic mouse was investigated. Tumors in the MMTV-Neu mouse strain develop spontaneously due to the presence of Neu oncogene expression in the mammary gland and are estrogen receptor negative (Muller et al., 1988). This model is particularly relevant because approximately 5–55% of human breast tumors over express the Her-2/Neu oncogene (Esteva et al., 2005; Hynes and Stern, 1994; Revillion et al., 1998). Several studies have shown that patients whose breast tumors over express Her-2/Neu have reduced survival and an increased risk of metastasis compared to patients whose tumors do not over express Her-2/Neu (Esteva et al.; Miles, 2001; Olayioye, 2001; Sahin, 2000). Because it has been documented that cancer patients do turn to CAM (Tamayo et al.), understanding the effect of Flor-Essence® on these tumors may help physicians counsel patients with Her-2/Neu over-expressing tumors.

We report here that Flor-Essence® herbal tonic does not inhibit mammary tumor incidence or reduce multiplicity compared to untreated mice in the MMTV-Neu mouse model. Although Flor-Essence® treatment does not influence mammary tumor development in this mouse model, it did have a significant impact on body weight when female mice were exposed *in utero*.

Methods

Animals

Female and male MMTV-Neu mice were purchased from Jackson Laboratory, (Bar Harbor, ME) and a license was obtained from DuPont to breed the mice in the Lawrence Livermore National Laboratory (LLNL) animal care facility to generate the experimental animals. Adult mice were housed four per cage. The animals were maintained in a temperature and humidity controlled room, with a 12-hr dark/light cycle. The rodents were provided an AIN-76A diet (Research Diets, New Brunswick, NJ) as previously described by Lamartiniere (1995). Mice had access to food and liquid *ad libitum*. Flor-Essence® herbal tonic was administered in the drinking water. The care of the animals was in accord with the LLNL Institutional Animal Care and Use Committee (IACUC) committee guidelines.

The male progeny were identified at weaning and euthanized in accordance with the SOP for rodent euthanasia as required by the LLNL IACUC and consistent with the recommendation from the Panel on Euthanasia of the American Veterinary Medical Association for animal euthanasia.

Flor-Essence® administration

Female offspring were randomly assigned to four experimental groups of approximately 40 animals each. On the day of birth, the dams whose offspring were assigned to the control group received only water. The dams of the offspring assigned to the pre-weanling or lifetime exposure treatment groups received 3% Flor-Essence® as their sole drinking source, beginning on the day they gave birth. The offspring were weaned at approximately 24 days. The pre-weanling exposure group was maintained on 3% Flor-Essence® until 5 weeks of age while the lifetime group was maintained on 3% Flor-Essence® as their sole drinking source until the termination of the study (28 weeks of age). The dams that gave rise to the females included in the *in utero* exposure group were administered 3% Flor-Essence® as their sole drinking source until the day of delivery at which time water became the sole drinking source.

Flor-Essence® herbal tonic (Flora Manufacturing and Distributing Ltd., B.C., Canada) was purchased in 500 ml bottles as a pre-prepared and sterile formulation from a local health food store. Flor-Essence® herbal tonic was administered to rodents in their drinking water as 3% solutions and was readily accepted (Bennett et al.). The dose of 3% was originally selected based on the recommendation of a popular natural health proponent for cancer patients to consume 2 fl oz of Flor-Essence® three times per day (Whitaker, 1995). This was also the dose used in the Sprague Dawley rat study (Bennett et al.), making cross-study comparison possible.

Tumor development and body weight

All mice were uniquely identified. Beginning at weaning, the mice were examined for palpable mammary tumors and were weighed weekly. The mice were sacrificed at 28 weeks of age, when tumors had grown larger than 1 cm, or when moribund. At the time of terminal sacrifice, the animals were weighed and the mammary tumors enumerated. The tumors were dissected from their pelts; their locations and measurements recorded. Any tumor measuring

greater than 0.5 cm in diameter was weighed after dissection. To confirm tumor type, routine histopathology was performed on tumors from control and treated groups and a veterinary pathologist diagnosed the tumors.

Statistical Analysis

The Kaplan-Meier method was used to compare the probability of developing a mammary tumor over time for the four experimental groups (Kaplan and Meier, 1958). The statistical significance among Kaplan-Meier curves was determined by the Mantel-Haenszel method (Mantel, 1966). The fractions of mice with palpable mammary tumor development were compared among the four groups, overall, at each weekly time point, using Mehta's modification of Fisher's exact test (Mehta and Patel, 1983). Mammary tumor incidence and multiplicity were determined from the data collected at sacrifice. The number of tumors at sacrifice was compared among the four experimental groups using a Kruskal-Wallis test. In addition, mammary tumor multiplicity was compared between each of the three Flor-Essence® treated groups and the untreated group using a Wilcoxon rank sum test. The incidences overall were compared using Mehta's version of Fisher's exact test (Mehta and Patel), while individual comparisons of incidence between controls and each of the other three treatment arms were done using Fisher's exact test. To determine if there were differences in overall weight, a repeated measures analysis of variance model, with polynomial transformation, was used.

Results

Mammary tumor development

Palpable mammary tumor incidence was determined weekly for each experimental group beginning at weaning until the end of the study (Figure 1). The first palpable mammary tumors were detected in the control mice at 23 weeks of age. For those groups receiving Flor-Essence®, the lifetime group, pre-weanling group and group treated *in utero* developed their first palpable mammary tumors at 20, 19 and 22 weeks, respectively. There was no significant difference in time to palpable tumor ($p = 0.47$ for global comparison), and no statistical evidence for a difference at each week in the probability of having mammary tumors among the four groups (all $p > 0.20$, except for $p = 0.044$ at 27 weeks, which would not be considered statistically significant in view of the multiple comparisons performed).

Mammary tumor incidence and multiplicity

The terminal necropsy was performed when the mice were 28 weeks of age or when the tumor burden became too great. The mammary tumor incidence values observed at the final necropsy in the control, pre-weanling, lifetime, and *in utero* exposure Flor-Essence® groups were 95%, 94%, 100%, and 97%, respectively and differed from palpable mammary tumor incidence because not all tumors visible at necropsy could be palpated (Table 1). There was no difference in tumor incidence between the untreated and Flor-Essence® treated mice ($p = 0.68$ among all 4 groups; $p = 0.50$ for comparison of untreated vs. each group of Flor-Essence® treated mice).

At the end of the study, mammary tumor multiplicity in the control and experimental groups consuming Flor-Essence® was determined (Table 1). Whereas the control group had a mean ($\pm$ SEM) of $6.6 (\pm 0.8)$ tumors per animal, the lifetime, pre-weanling, and *in utero* treated groups had means of $5.2 (\pm 0.7)$, $6.7 (\pm 0.8)$, and $7.6 (\pm 0.8)$, respectively. Mammary tumor multiplicity among the four groups was not statistically different ($p = 0.18$ comparing all 4 groups; $p > 0.20$ for comparison of untreated vs. each group of Flor-Essence® treated mice).

Mammary tumors were randomly selected from the experimental groups for histological analysis. As expected, all mammary tumors examined from the four groups were diagnosed as adenocarcinomas.

Body weight

The body weight of all mice after weaning was documented weekly throughout the study. The mean body weight for the animals treated with Flor-Essence® *in utero* was greater at every time point examined compared to the control and the groups treated with herbal tonic throughout their lifetime or as preweanlings (Figure 2). The group exposed to Flor-Essence® *in utero* was significantly heavier than the animals in the control group ($p = 0.0008$). The groups that were treated throughout their lifetime or as preweanling were not statistically different from the control animals (each $p > 0.50$).

Conclusions

This study demonstrates that Flor-Essence® herbal tonic does not inhibit the development of mammary tumors in the MMTV-Neu transgenic mouse. This finding is contrary to the hopes of many individuals who are purchasing this product, largely on advice from friends and family, to treat their own cancer (Tamayo et al.). The results presented in this report represent new science-based information that will contribute to a very meager scientific literature base for this tonic. This is the first report describing the effect of this tonic in a Her-2/Neu animal model for human breast cancer. Over expression of Her-2/Neu occurs in approximately 5–55% of human breast cancer cases (Esteva et al.; Hynes and Stern; Revillion et al.). The conclusions from this study will contribute science based information that can be used to make informed decisions about the use of CAM for cancer treatment and prevention.

The MMTV-Neu mouse was developed as a model for human Her2/Neu breast cancer and the mammary tumors that spontaneously develop in this mouse are estrogen receptor negative (Guy et al., 1992; Hewitt et al., 2002; Muller et al.). Our results suggest that Flor-Essence® does not promote the estrogen receptor negative tumors that develop in this model system. These results contrast our previous study using the Sprague Dawley rat where lifetime exposure to Flor-Essence® resulted in the promotion of DMBA-induced mammary tumors (Bennett et al.). Mammary tumors that develop in DMBA treated rats are estrogen sensitive. This molecular phenotype may play a role in the difference in effect observed between these studies.

The effect of Flor-Essence® and Essiac® on the proliferation of cells *in vitro* has been studied by us and by others. Tai et al. demonstrated that these tonics induce proliferation at the lower doses of 1% and 2% in the estrogen receptor positive human breast cell line MCF-7 while at higher concentrations of 5% and 10% they have an anti-proliferative effect. When they treated the estrogen receptor-negative cell line MDA-MB-468 in a similar manner, they observed weak growth inhibition at all concentrations of EssiacR and 1% and 2% concentrations of Flor-Essence®. The higher concentrations (5% and 10%) of Flor-Essence® caused a marked growth inhibition. Ottenweller et al. exposed the prostate cell line LNCaP to concentrations of Essiac® that ranged from 5% to 30% and observed growth inhibition at each condition tested compared to controls. We have performed a series of *in vitro* studies wherein we exposed several different human breast cell lines that varied in estrogen receptor status to Flor-Essence® or Essiac® (Kulp et al., (2006). Using a different methodology than those described for the studies of Ottenweller et al. and Tai et al., we depleted our systems of estrogen prior to experimentation. Using this approach, we observed that concentrations of Flor-Essence® and Essiac® ranging from 1% to 8% were capable of inducing proliferation of both estrogen receptor positive and estrogen receptor negative cell

lines whereas 16% was growth inhibitory and 32% was cytotoxic. In addition we performed transcriptional activation assays and demonstrated that both tonics induced reporter activity by direct interaction with the estrogen receptor (Kulp et al.).

The *in vivo* results presented here, in combination with the *in vitro* results provide valuable insight into the possible mechanisms by which Flor-Essence® may be affecting tumor development. Our results suggest that the estrogen-like components of Flor-Essence®, detected in the *in vitro* studies, play a role in the promotion of hormone-responsive mammary tumors in the Sprague Dawley rat. Several of the herbs in this tonic have estrogenic activity including *red clover* (*Trifolium pratense*), *sheep sorrel* (*Rumex acetosella*), and *burdock root* (*Arctium lappa*) (Burdette et al., 2002; Tamayo et al.). More specifically, genistein, present in several of the tonic's constituent components, has been shown to modulate mammary tumor development in several studies (Bouker and Hilakivi-Clarke, 2000; Constantinou et al., 1998; Fritz et al., 1998; Hilakivi-Clarke et al., 1999a; Hilakivi-Clarke et al., 1999b; Lamartiniere, 1995; Lamartiniere, 2000; Murrill et al., 1996). In contrast, the estrogen-independent growth promoting effects we observed *in vitro* do not contribute to significant changes in tumor cell growth in the MMTV-Neu mouse model (Kulp et al.).

In this study Flor-Essence® exposure *in utero* resulted in a significant increase in average body weight relative to controls. There were no observable differences in overall health or behavior of the adults in the *in utero* exposed group compared to controls. In a study suggesting that genistein acts as an estrogen *in utero*, the offspring of CD-1 outbred mice exposed to genistein during gestation demonstrated an accelerated weight gain (Nikaido et al., 2004). In these same mice, offspring of mice exposed during gestation to resveratrol (RES), zearalenone (ZEA), bisphenol (BPA), or diethylstilbestrol (DES), other compounds known to have estrogenic activity, were also shown to have an accelerated weight gain by 16 weeks of age. Offspring of Sprague Dawley rats exposed during gestation to BPA were observed to have an increased body weight soon after birth (Rubin et al., 2001). Kaijser et al. (2000) reported a strong positive association in women between total maternal estriol levels during pregnancy and newborn birth weight. Given the shown estrogenic effects of Flor-Essence®, our results are consistent with the results seen with *in utero* exposure to estrogenic compounds.

The combination of these *in vitro* and *in vivo* results provide valuable information for the individual considering self-administering Flor-Essence® herbal tonic. The results from several assay systems suggest that Flor-Essence® does not inhibit mammary tumor development. It is critical that scientific information exist and be made available about the effect of herbal tonics on normal and neoplastic mammary gland growth and development. This information will contribute to a scientific peer-reviewed literature base that will support informed decision-making regarding use.

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References

- Bennett LM, Montgomery JL, Steinberg SM, Kulp KS. Flor-Essence herbal tonic does not inhibit mammary tumor development in Sprague Dawley rats. *Breast Cancer Res Treat.* 2004; 88(1):87–93. [PubMed: 15538049]

- Bouker KB, Hilakivi-Clarke L. Genistein: does it prevent or promote breast cancer? *Environ Health Perspect.* 2000; 108(8):701–708. [PubMed: 10964789]
- Burdette JE, Liu J, Lantvit D, Lim E, Booth N, Bhat KP, Hedayat S, Van Breemen RB, Constantinou AI, Pezzuto JM, Farnsworth NR, Bolton JL. *Trifolium pratense* (red clover) exhibits estrogenic effects in vivo in ovariectomized Sprague-Dawley rats. *J Nutr.* 2002; 132(1):27–30. [PubMed: 11773503]
- Constantinou AI, Krygier AE, Mehta RR. Genistein induces maturation of cultured human breast cancer cells and prevents tumor growth in nude mice. *Am J Clin Nutr.* 1998; 68:1426S–1430S.
- Esteva FJ, Cheli CD, Fritsche H, Fornier M, Slamon D, Thiel RP, Luftner D, Ghani F. Clinical utility of serum HER2/neu in monitoring and prediction of progression-free survival in metastatic breast cancer patients treated with trastuzumab-based therapies. *Breast Cancer Res.* 2005; 7(4):R436–443. [PubMed: 15987448]
- Felton JS, Knize MG, Bennett LM, Malfatti MA, Colvin ME, Kulp KS. Impact of environmental exposures on the mutagenicity/carcinogenicity of heterocyclic amines. *Toxicology.* 2004; 198(1–3): 135–145. [PubMed: 15138037]
- Fritz WA, Coward L, Wang J, Lamartiniere CA. Dietary genistein: perinatal mammary cancer prevention, bioavailability, and toxicity testing in the rat. *Carcinogenesis.* 1998; 19:2151–2158. [PubMed: 9886571]
- Guy CT, Webster MA, Schaller M, Parsons TJ, Cardiff RD, Muller WJ. Expression of the neu protooncogene in the mammary epithelium of transgenic mice induces metastatic disease. *Proc Natl Acad Sci U S A.* 1992; 89(22):10578–10582. [PubMed: 1359541]
- Hewitt SC, Bocchinfuso WP, Zhai J, Harrell C, Koonce L, Clark J, Myers PI, Korach KS. Lack of ductal development in the absence of functional estrogen receptor alpha delays mammary tumor formation induced by transgenic expression of ErbB2/neu. *Cancer Res.* 2002; 62(10):2798–2805. [PubMed: 12019156]
- Hilakivi-Clarke L, Cho E, Onojafe I, Raygada M, Clarke R. Maternal exposure to genistein during pregnancy increases carcinogen-induced mammary tumorigenesis in female rat offspring. *Oncol Reports.* 1999a; 6:1089–1095.
- Hilakivi-Clarke L, Onojafe I, Raygada M, Cho E, Skaar T, Russo I, Clarke R. Prepubertal exposure to zearalenone or genistein reduces mammary tumorigenesis. *Br J Cancer.* 1999b; 80(11):1682–1688. [PubMed: 10468283]
- Hynes NE, Stern DF. The biology of erbB-2/neu/HER-2 and its role in cancer. *Biochim Biophys Acta.* 1994; 1198(2–3):165–184. [PubMed: 7819273]
- Kaegi E. Unconventional therapies for cancer: 1. Essiac. The Task Force on Alternative Therapies of the Canadian Breast Cancer Research Initiative. *Cmaj.* 1998; 158(7):897–902. [PubMed: 9559016]
- Kaijser M, Granath F, Jacobsen G, Cnattingius S, Ekblom A. Maternal pregnancy estriol levels in relation to anamnestic and fetal anthropometric data. *Epidemiology.* 2000; 11(3):315–319. [PubMed: 10784250]
- Kaplan E, Meier P. Non-Parametric estimation from incomplete observations. *J Am Stat Assoc.* 1958; 53:457–481.
- Kulp KS, Montgomery JL, Nelson DO, Cutter B, Latham ER, Shattuck DL, Klotz DM, Bennett LM. Essiac and Flor-Essence herbal tonics stimulate the in vitro growth of human breast cancer cells. *Breast Cancer Res Treat.* 2006; 98(3):249–259. [PubMed: 16541326]
- Lamartiniere CA. Neonatal genistein chemoprevents mammary carcinogenesis. *Proc Soc Exp Biol Med.* 1995; 208:120–123. [PubMed: 7892285]
- Lamartiniere CA. Protection against breast cancer with genistein: a component of soy. *Am J Clin Nutr.* 2000; 71(6 Suppl):1705S–1707S. discussion 1708S–1709S. [PubMed: 10837323]
- Mantel N. Evaluation of survival data and two new rank order statistics arising in its consideration. *Cancer Chemother Rep.* 1966; 50(3):163–170. [PubMed: 5910392]
- Mehta CR, Patel NR. A network algorithm for performing Fisher's exact test in r x c contingency tables. *J Am Stat Assoc.* 1983; 78:427–434.
- Miles DW. Update on HER-2 as a target for cancer therapy: herceptin in the clinical setting. *Breast Cancer Res.* 2001; 3(6):380–384. [PubMed: 11737889]

- Mills E, Ernst E, Singh R, Ross C, Wilson K. Health food store recommendations: implications for breast cancer patients. *Breast Cancer Res.* 2003; 5(6):R170–174. [PubMed: 14580252]
- Muller WJ, Sinn E, Pattengale PK, Wallace R, Leder P. Single-step induction of mammary adenocarcinoma in transgenic mice bearing the activated c-neu oncogene. *Cell.* 1988; 54(1):105–115. [PubMed: 2898299]
- Murrill WB, Brown NM, Zhang JX, Manzillo PA, Barnes S, Lamartiniere CA. Prepubertal genistein exposure suppresses mammary cancer and enhances gland differentiation in rats. *Carcinogenesis.* 1996; 17(7):1451–1457. [PubMed: 8706248]
- Nikaido Y, Yoshizawa K, Danbara N, Tsujita-Kyutoku M, Yuri T, Uehara N, Tsubura A. Effects of maternal xenoestrogen exposure on development of the reproductive tract and mammary gland in female CD-1 mouse offspring. *Reprod Toxicol.* 2004; 18(6):803–811. [PubMed: 15279878]
- Olayioye MA. Update on HER-2 as a target for cancer therapy: intracellular signaling pathways of ErbB2/HER-2 and family members. *Breast Cancer Res.* 2001; 3(6):385–389. [PubMed: 11737890]
- Olsen, C. *Essiac: A Native Herbal Cancer Remedy.* Twin Lakes, WI: Lotus Press; 1998.
- Otteweller J, Putt K, Blumenthal EJ, Dhawale S, Dhawale SW. Inhibition of prostate cancer-cell proliferation by Essiac. *J Altern Complement Med.* 2004; 10(4):687–691. [PubMed: 15353028]
- Percival, J. *The Essiac Handbook.* 2. Orlando, FL: Rideout Publishing Company; 1994.
- Revillion F, Bonnetterre J, Peyrat JP. ERBB2 oncogene in human breast cancer and its clinical significance. *Eur J Cancer.* 1998; 34(6):791–808. [PubMed: 9797688]
- Richardson M, Sanders T, Tamayo C, Perez C, Palmer J. Flor-Essence® herbal tonic use in North America: A profile of general consumers and cancer patients. *Herbalgram.* 2000; 50:40–46.
- Rubin BS, Murray MK, Damassa DA, King JC, Soto AM. Perinatal exposure to low doses of bisphenol A affects body weight, patterns of estrous cyclicity, and plasma LH levels. *Environ Health Perspect.* 2001; 109(7):675–680. [PubMed: 11485865]
- Sahin AA. Biologic and clinical significance of HER-2/neu (cerbB-2) in breast cancer. *Advances in Anatomic Pathology.* 2000; 7(3):158–166. [PubMed: 10809222]
- Snow, S.; Klein, M. *Essiac: The Secrets of Rene Caisse's Herbal Pharmacy.* Dublin: Newleaf; 2001.
- Tai J, Cheung S, Wong S, Lowe C. In vitro comparison of Essiac and Flor-Essence on human tumor cell lines. *Oncol Rep.* 2004; 11(2):471–476. [PubMed: 14719086]
- Tamayo C, Richardson MA, Diamond S, Skoda I. The chemistry and biological activity of herbs used in Flor-Essence herbal tonic and Essiac. *Phytother Res.* 2000; 14(1):1–14. [PubMed: 10641040]
- University of Texas, M. D. A. C. C. *Herbal/Plant Therapies: Essiac Detailed Scientific Review.* 2008. Retrieved May 15, 2008, from <http://www.mdanderson.org/departments/CIMER/display.cfm?id=03F2CD11-EE0F-11D4-810200508B603A14&method=displayFull&pn=6EB86A59-EBD9-11D4-810100508B603A14>
- Whitaker, J. Dr Whitaker's Newsletter. 1995. Should you take Essiac tea against cancer?; p. 5

Figure 1.

Palpable mammary tumor incidence in control compared to Flor-Essence® treated mice. Control (diamond), lifetime treatment (circle), preweanling treatment (triangle), and *in utero* treatment (square) were palpated weekly, beginning at weaning, for the presence of mammary tumors. All mice were uniquely identified.

Figure 2. Comparison of average body weight among control and Flor-Essence® treated MMTV-Neu mice. Average body weight was determined weekly for control mice (diamond) and lifetime (circle), preweanling (triangle), and *in utero* (square) Flor-Essence® treated groups.

Table 1

Mammary tumor multiplicity in control and Flor-Essence® treated mice at sacrifice.

Group	Number	Mean Incidence (%)	Multiplicity			
			Mean (SEM)	Min.	Med.	Max.
Control	41	95	6.6 (0.8)	0	6	29
Lifetime	32	94	5.2 (0.7)	0	4	19
Pre-weanling	32	100	6.7 (0.8)	1	5.5	21
<i>in utero</i>	36	97	7.6 (0.8)	0	7	19