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Original article

# The biochemical effects of nano tamoxifen and some bioactive components in experimental breast cancer

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ABSTRACT

The effect of nano tamoxifen and some bioactive components such as yeast, isoflavone, and silymarin on the level of resistance and prevention of breast cancer progression in experimental animals is the target of this study. Thirty female Sprague-Dawley rats received a single medication dosage of 7,12-dimethylbenz[a]anthracene (DMBA) intragastrically. After fourteen days of DMBA admission, the procedure protocol started out. Finally, all the experimental results evaluated, tabulated and statistically analyzed. The results demonstrated a highly significant elevation in the 8-OHdG level in group 1 (nano yeast) and 3 (nano silymarin) while the results demonstrated a highly significant reduction in group 2 (nano tamoxifen). The apoptosis results demonstrated a significant elevation in group 3 (nano silymarin) where appeared significant reduction in group 4 (nano isoflavone). ErbB-2 results demonstrated a significant elevation in group 2 (nano tamoxifen) and a significant reduction in each of group 3 (nano silymarin) and 4 (nano isoflavone). The lipid peroxide level demonstrated an extremely significant reduction in group 4 (nano isoflavone). And a significant reduction of total antioxidant was observed in group 3 (nano silymarin) in comparison to injected animals control. This may be considered a new vision and strategy to resist breast cancer disease or prevent progression.

## 1. Introduction

Breast carcinoma is recognized as well-known neoplasms in women and also the leading cause of cancer-related deaths [1]. The etiology of breast cancer is multifactorial. Most of risk factors that raise the chance of a woman creating breast cancer have been distinguished by many epidemiologic studies which incorporate early age at menarche, late age of menopause, non-reproduction, overweight, oral contraception, diet, genealogy, female sex, lack of physical exercise, alcohol consumption, hormone replacement therapy amid menopause, ionizing radiation, early age initially monthly cycle, older age and hereditary factors [2,3]. The common denominator for most of these factors is their influence on the particular level and exposure period to endogenous or exogenous estrogens. Breast cancer involves numerous types of cancers that result from the breast tissue. It is the most frequent invasive cancer in women worldwide [4]. Signs or symptoms of breast cancer include a breast lump, an alteration in breast shape and nipple release or a red scaly patch of skin area. There are many lines of treatment of breast cancer rely on the tumor stage. They include hormone blocking therapy, chemotherapy, monoclonal antibodies, and radiotherapy [5].

Tamoxifen (TAM) among the antiestrogens is normally utilized as a first-line endocrine treatment for premenopausal women with breast cancer and levels of resistance improvement [6,7]. The mechanisms of the level of resistance can include pharmacologic mechanisms, reduction or changes in estrogen receptor expression, modification in the regulatory proteins that participate in several cellular processes; inhibition controlled by the Bcl-2 family and modified mRNA expression [8]. These undesirable effects resulted in the utilization of alternative treatments of apoptosis such as complementary and alternative medicine [9]. Development and progression of breast tumors contain a complex series of incidents, including dysregulation of cellular differentiation, extreme proliferation, and level of resistance to apoptosis [10,11].

Only a tiny minority of patients with breast cancer develop the disease because of inheritance of germline mutations in prominent,

*Abbreviations:* DMBA, 7,12-dimethylbenz[a]anthracene; 8-OHdG, rat 8-hydroxy desoxyguanosine; ErbB-2, rat receptor tyrosine-protein kinase; MDA, lipid peroxide (malonaldehyde); Bcl-2, breast cancer genes 1 and 2; ROS, reactive oxygen species; TAM, Tamoxifen; TEM, transmission electron microscope; REs, estrogen receptors

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highly penetrant susceptibility genes such as BRCA1 and BRCA2 [12]. At this point, additional tumor heterogeneity is placed whereby a tiny group of cells within, or may be external, the tumor is both protected to drugs and offer the source of new tumor progress [13,14]. These cells also contribute directly to the seeding of supplementary tumors at distal sites, the primary cause of mortality in breast cancer patients [15]. These drug-resistant cancer initiating cells, also known as breast cancer stem cells (BCSCs) have been exhibited functionally for both human being and mouse mammary tumors and tumor cell lines [16,17]. Experiments on human breast tumors in mouse models, for example, show that whenever these cells were deleted, the rest of the cells were not able to maintain new tumor progress [18]. So, the prevention or cancer level of resistance of the important strategies also focuses on non-progression of the disease, limitation of the body's reaction to the pathogen also one of the main important factors that this research was designed to study them. Numerous studies demonstrated that 7,12-dimethylbenz [a]anthracene (DMBA) may be utilized to induce experimental breast carcinomas in rats and that this process includes disruption of tissue redox balance; subsequently, this shows that biochemical and pathophysiological disruption may derive from oxidative destruction [19,20]. Where, protecting systems can be damaged easily by chemicals, such as DMBA, which disrupt the pro-oxidant–antioxidant balance, resulting in cellular anomalies. Within the breast, DMBA is transformed to epoxides, effective metabolites with a convenience of destroying the DNA molecule, the main event in carcinogenesis initiation. With the higher cellular proliferative index of types 1 and 2 lobules, there is certainly a higher metabolic activity and much more epoxide creation. Rat mammary carcinomas are starting to appear in small mammary ducts [21–23] or from hyperplastic alveolar nodules [24,25].

Recent innovations in biomedical and nanotechnology point out to the role of Nutrition research areas that might benefit to making use nanotechnology include research that seeks to improve food composition. There is certainly little information about the potential health threats of nanoparticles.

Nanotechnology has the capacity to enhance the nutrition science and that through helping in the detection, development, and delivery of many intervention strategies to enhance health insurance and limit the chance of risk and difficulties of many diseases. Those prepared to advance the systems which are at present being utilized or potentially changed for nutrition research. It is trusted that by highlighting these advances the potential advantage of nanomaterials to reform nutrition research is known [26].

Over the previous years, Decuzzi and Ferrari [27] had demonstrated how the action of particulate systems can be calibrated by the physicochemical properties additionally controlling their geometrical properties, including size and shape. These three designing parameters (size, shape, and physico-science) assume a pivotal part in particulate, transportation inside the blood circulation and in the tissue; identification of vascular and extravascular targets; collaboration with specific cells and the immune system cells [27].

Breast malignancy is the medical field with the best existence of nanotechnological therapeutic agents in the medical clinic. Breast cancer experts in the medical clinic, the pharmaceutical and the essential biological laboratory and nanotechnologists, technical engineers, physicists, chemists are the major necessity for success in the improvement of new therapeutic strategies and enhance their capacity to work in the close coordinated effort [28].

Worldwide, the breast cancer progression has already been increasing. Treatments of breast cancer, such as chemotherapy, radiotherapy, and other treatments cause serious adverse side effects. Lately, attention is focused on distinguishing nutrients and food additives which have the capability to prevent the carcinogenesis processes. So, we should seek out novel anticancer agents that having a different mode and low side effects.

This work is designed to study converting tamoxifen drug and some bioactive component sources such as yeast, isoflavone, and silymarin to nanoparticles and studies their influence on reducing the progress and prevalence of cancer. As well as finding novel methods of treatment. Also, this present study had a need to focus on the impact of the utilization of nanoparticles in nutrition among the new therapeutic methods aimed at trying to resistance breast cancer progression.

## 2. Materials and methods

The carcinogen 7,12-dimethylbenz[a]anthracene (DMBA) with a chemical formula $C_{20}H_{16}$, molecular weight 256.35 g/mol, was purchased from Sigma Chemical Company (Sigma Chemicals, Sigma–Aldrich, St. Louis, MO, USA). The DMBA was used to induce mammary carcinoma in rats by a single dose (25 mg/kg body weight) [29].

### 2.1. Preparation of nano-materials

All of the nano-materials which were used in the experiment were prepared in my laboratory in the National Research Centre which classified as the following:

- Nutrient as yeast.
- Drug as tamoxifen.
- Bioactive components include silymarin and isoflavone.

#### 2.1.1. Measurement techniques for nano-nutrient, drug, and bioactive components

*2.1.1.1. Transmission electron microscopy*. Transmission electron micrograph of the studied samples was performed using TEM (JEM-1234), operating voltage at 120 kV energy, magnification power is 600,000×, resolving power is 0.3 nm, CCD-camera equipped, and programmable heating/cooling facility is from −190 °C up to 1000 °C. Note: samples were held on the carbon-coated copper grid in the Central Lab in the National Research Center.

*2.1.1.2. Mass spectroscopy*. All graphs of the studied samples were performed using mass spectroscopy JEOL (JMS-AX500) in the Central Lab in the National Research Centre.

*2.1.1.3. Zetasizer Nano ZS*. The *z*-average hydrodynamic diameter was determined the studied samples at 25 ± 0.1 °C by photon correlation spectroscopy using a Zetasizer Nano ZS (Malvern Instruments Inc., Southborough, MA) in the Central Lab in the National Research Centre.

### 2.2. Experimental animals

Thirty female Sprague-Dawley rats, which induced breast cancer received a single dose (25 mg/kg body weight) of 7,12-dimethylbenz[a] anthracene (DMBA) intragastrically by gavage, which was described by [29]. Two weeks after DMBA treatment, a time by which the animals had recovered from DMBA-induced toxicity, the rats were divided into 5 groups (6 for each). A group of injected animals fed on the basal synthetic diet that served as control. Injected animals (4 groups) fed on the basal synthetic diet supplemented with nanoparticles (yeast, tamoxifen, isoflavone, and silymarin) respectively, as illustrated in Table 1.

The composition of salt and vitamin mixture is prepared according to the methods of Briggs and William [30] and Morcos [31] respectively.

### 2.3. Plasma and serum biochemistry

At the end of the experiments (six months), a blood sample of each animal collected where serum and plasma are separated by centrifugation at 3000 rpm for 15 min and stored in −20 °C to measure the biochemical parameters including apoptosis, 8-OHdG, ErbB-2, plasma