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ORIGINAL ARTICLE

Effects of perinatal exposure to Zamzam water on the teratological studies of the mice offspring

Gasem Mohammad Abu-Taweel *

Department of Basic Sciences, College of Education, University of Dammam, P.O. Box 2375, Dammam 31451, Saudi Arabia

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Abstract Zamzam water is well documented for plenty of medicinal value for curing illness. In the present study, the effects of perinatal consumption of Zamzam and normal drinking water by the pregnant mice on their offspring's physical parameters, early sensory motor reflexes, locomotor activities, acetylcholinesterase (AChE) activity in the homogenize brain tissue and blood parameters were compared. To achieve that; Zamzam water was given to female Swiss-Webster strain mice as the only source of drinking fluid and the control animals were administered plain tap water. Treatment started from the first day of pregnancy and continued until the postnatal day fifteen of delivery. All offspring were subjected to various tests. The rate of body weight gain remained relatively unaffected until the second week of weaning period, however; in the last week the offspring exposed to Zamzam water gained significant body weight as compared to their control offspring. Furthermore, the opening of eyes and appearance of body hairs in Zamzam exposed pups remained unaffected as compared to the controls. The sensory motor reflexes in Zamzam exposed pups after birth and during the first two weeks of weaning period were significantly increased. Locomotor Activity Test performed in the male and female offspring after weaning period showed a significant decrease in the male and increase in the female on most of the elements of this test due to Zamzam exposure. AChE activity in the homogenized brain tissue and blood parameters were unaffected as compared to the controls, the present Zamzam effects in the offspring are possibly via in utero action and/or via mother's milk.

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1. Introduction

Water is a basic need and is essential in carrying out various physiological functions in the human body (Cazier and Gekas, 2001). Humans can survive without food for a month, but they can survive without water for only seven days (Vidyasagar, 2007). Only 2.8% of the total water on earth is freshwater; the rest is saltwater that is hard to use (Khalid et al., 2014). Although the world's water sources are limited, and existing sources are depleting rapidly, there is a source

* Mobile: +966 555107301; fax: +966 138269936.

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of water called Zamzam that is providing water to billions of people (Khalid et al., 2014; Naeem et al., 1983).

This source of water is located in the Mecca area, one of the most sacred cities for Muslims. This city is located in the western part of Saudi Arabia about 70 km south of the city of Jeddah on the coast of the Red Sea. Geographically, it can be located at latitude 21° 26' 48" N, longitude 39° 53' 46" E, with an elevation of about 1399 ft. above mean sea level (Khalid et al., 2014).

Brief anecdotes that mention Zamzam are described in the holy books of various religions, including the Torah (Old Testament/Torah), the Bible, and the Quran. As narrated in these holy books, Zamzam is the holy water and is termed as a great gift from God (referred to as Allah in the Quran). It is allegedly an offshoot from a holy spring (currently present in the form of a well) in the barren desert surrounding Mecca. Muslims use Zamzam water to recover from diseases, according to the speech of the Prophet Muhammad, peace be upon him, who said: "Water of Zamzam is good for whatever (purpose) it has been drank." In another Hadith, the Prophet said: "Water of Zamzam is a healer of every disease. It is the place where angel Jibril indicated it and dug it, and it is the water of Allah that made Ismail (to be the first) to drink it" (Ahmad and Ibrahim, 1996).

Khalid et al. (2014) pointed in their review article that Zamzam water can be used to recover from some diseases. Also, it has unique analytic properties and has a strong anti-inflammatory effect. Zamzam water has a strong antitumor necrosis factor (TNF α) and effect on interleukin 1 (IL1). Zamzam water has analytic action through an indirect influence on endocrine immunology and the growth system of the body (Ali et al., 2009c). Clinical examination revealed that, in the mixed dentition group, no statistically significant differences were detected, whereas among the permanent dentition group, the mean decayed, missing, filled teeth (DMFT) score was the lowest in all the children using Zamzam water (Al-Zuhair and Khounganian, 2006).

It was used in the treatment of implantation failure, for stimulation of endometrial prolactin, defense, luteinizing hormone (LH), endometrial vascular endothelial growth factor (VEGF), and angiopoietin receptors (Ali et al., 2009b). Furthermore, Zamzam water causes upregulation of gap junctional inter-cellular communication and connexin 43 antibodies in endometrium. Ali et al. (2009a) demonstrated that Zamzam water stimulates stem cells' differentiation in the endometrium. This phenomenon is triggered due to the high calcium and magnesium content of Zamzam. Also, the water provides more support to many other biochemical processes in the endometrium, including its vital role as a coenzyme during the formation of immunoglobulin (Ali et al., 2009b,c).

Furthermore, Zamzam water may be used in planting. Mutwally et al. (2015) reported that sole use of Zamzam water or a combination of Zamzam, with either treated water or tap water resulted in pronounced increases in the percentage of seed germination, shoot length, and the fresh and dry weights of the shoots. Similarly, the percentage of flowers in broad bean plants watered with Zamzam was considerably higher in comparison with other water treatments.

Researches that have examined the effect of drinking Zamzam water by mother during pregnancy and its impact on its offspring's growth and behavior was very little or almost

non-existent. Therefore, the current study was designed to investigate the effect of prenatal exposure to Zamzam water on the growth, behavior, some blood indicators and enzyme of acetylcholinesterase in laboratory mice offspring.

2. Materials and methods

2.1. Experimental animals

Male and female Swiss-Webster strain mice (8–9 weeks old) were housed in opaque plastic cages (three females to one male in each cage) measuring 30 × 12 × 11 cm, in the animal facility of the Zoology Department, King Saud University, Riyadh, Saudi Arabia. Animals were kept under reversed lighting conditions with white lights on from 22.30 to 10.30 h local time. The ambient temperature was regulated between 18 and *noe*. After pregnancy (appearance of vaginal plug was considered as day one of pregnancy), the males were removed from the cages and the females were subjected to experimental treatments. Food (Pillsbury's Diet) and water were available *ad libitum*, unless otherwise indicated.

2.2. Zamzam water administration

The animals were divided on the first day of pregnancy (the appearance of vaginal plug) into two groups, the first is the control which was given tap water, while the second group was given Zamzam water (purchased from the local market). Treatment of mothers was started after the delivery of offspring from day 1 (PD1) and was continued until postnatal day 15 (PD15) and thereafter the mothers were switched to tap water.

2.3. Physical assessment during weaning period

On the day of birth (postnatal day 0, PDO) the pups were culled to only eight per dam and were left with their mothers until PD22. During this weaning period, three pups in each litter were color marked from the others and were subjected to various behavioral tests (described below) under dim lighting (ca 8 lux). In all, 21 pups belonging to seven litters from each treatment category were considered. All observations were recorded on PD 1 and repeated every other day until PD21 in the same three color marked pups of each litter. These observations were used to measure the early development of sensory motor coordination reflexes together with morphological development in the pups. For statistical analysis, the mean of all three color marked pups per litter was considered as a single score. Thus, seven replicates from each treatment category were considered in these observations.

2.3.1. Body weight

Weight is a useful indicator of development. Thus, the pups were weighed every alternate day from PD 1 to PD21.

2.3.2. Eye opening and hair appearance

The day at which the body hair fuzz appeared, and the eyes opened was also recorded. These two parameters are also useful morphological indicators of development.

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