Original Article

Neurobehavioral Assessment of Rats Exposed to Yttrium Nitrate during Development*

LI Chen Xi[1,2], MA Chuan[2], FANG Hai Qin[2], ZHI Yuan[2], YU Zhou[2], XU Hai Bin[2,#], and JIA Xu Dong[2,#]

1. National Institute for Nutrition and Food Safety, Chinese Center for Disease Control and Prevention, Beijing 100021, China; 2. Key Laboratory of Food Safety Risk Assessment of Ministry of Health, China National Center for Food Safety Risk Assessment, Beijing 100021, China

Abstract

Objective The aim of this study was to assess the effects of yttrium nitrate on neurobehavioral development in Sprague-Dawley rats.

Methods Dams were orally exposed to 0, 5, 15, or 45 mg/kg daily of yttrium nitrate from gestation day (GD) 6 to postnatal day (PND) 21. Body weight and food consumption were monitored weekly. Neurobehavior was assessed by developmental landmarks and reflexes, motor activity, hot plate, Rota-rod and cognitive tests. Additionally, brain weights were measured on PND 21 and 70.

Results No significant difference was noted among all groups for maternal body weight and food consumption. All yttrium-exposed offspring showed an increase in body weight on PND 21; however, no significant difference in body weight for exposed pups versus controls was observed 2 weeks or more after the yttrium solution was discontinued. The groups given 5 mg/kg daily decreased significantly in the duration of female forelime grip strength and ambulation on PND 13. There was no significant difference between yttrium-exposed offspring and controls with respect to other behavioral ontogeny parameters and postnatal behavioral test results.

Conclusion Exposure of rats to yttrium nitrate in concentrations up to 45 mg/kg daily had no adverse effects on their neurobehavioral development.

Key words: Rare earth elements; Yttrium; Developmental neurotoxicity; Neurobehavior

Biomed Environ Sci, 2015; 28(4): 281-290 *doi: 10.3967/bes2015.039* *ISSN: 0895-3988*

www.besjournal.com (full text) *CN: 11-2816/Q*

INTRODUCTION

Rare earth elements (REEs) consist of 17 transition metals in Group III of the Periodic Table. Their widespread use in agriculture and industry has resulted in increasing amounts of REEs being released into the environment, thereby increasing the risk of its accumulation in humans. Studies of the biological effects of REEs and their underlying mechanisms of action have attracted considerable attention. Thus far, it is known that REEs, like many other heavy metals, affect tissues of the lung, liver, kidney, spleen, and other organs[1]. REEs may also affect brain development in animals and human beings. Briner and colleagues believed that lanthanum (La) has teratogenic effects on behavior that include subtle but detectable effects on mouse development[2]. Feng and colleagues found that subchronic exposure of rats to La can alter the concentration of DNA and the ratio of protein to DNA in the brain, thereby significantly impairing

*This research was financially supported by the National Science and Technology Support Program (2012BAK01B00).

#Correspondence should be addressed to XU Hai Bin, Tel: 13910636893, E-mail: hbxu1231602@vip.sina.com; JIA Xu Dong, Tel: 13520375960, E-mail: jiaxudong@cfsa.net.cn

Biographical note of the first author: LI Chen Xi, male, born in 1984, master degree, assistant researcher, majoring in nutrition and food hygiene.

memory and learning abilities[3]. He and colleagues came to a similar conclusion based on their finding of changes in homeostasis for Ca^{2+}/Ca^{2+}-ATPase and in the inhibitory activity of certain antioxidant enzymes[4]. An epidemiological investigation by Zhu and colleagues revealed that children living in REE-rich regions (especially regions rich in heavy REEs) had lower intelligence quotients (IQ) and worse memory than those living in areas with normal REE levels. This suggests that long-term exposure to REEs can affect brain function[5].

Developmental neurotoxicity studies are developed to collect data on the functional disturbances and morphological hazards to the nervous system arising in the offspring from exposure of the mother during pregnancy and lactation, and to provide dose-response characterizations of these exposures[6]. Because of the limitations involved in carrying out developmental neurotoxicity studies in humans, there is a strong need for animal experiments to gather such data[7]. For example, rat pups randomly chosen from among control and exposed litters could be tested for gross neurological and behavioral abnormalities using assessments of physical development, behavioral ontogeny, motor activity, motor and sensory function, learning, and memory. Additionally, brain weight and neuropathology could be monitored throughout postnatal development and adulthood[8].

It has been reported that human exposure to compounds containing yttrium, a heavy REE widely used in industry and medical, may cause lung disease. Experiments in rats and hamsters also have shown that this silvery-metallic transition metal, which is chemically related to the lanthanides, can translocate to the skeleton and liver[1]. However, studies of the developmental neurotoxicity of yttrium are lacking, no official no observed adverse effect level (NOAEL) or acceptable daily intake (ADI) value for yttrium either. In the present study, we investigated the effects of yttrium on the postnatal development of rat pups, with a particular interest in neurobehavioral development, which was evaluated using current international test guidelines[8]. Our results could provide specific scientific data for the food safety risk assessment of yttrium or rare earth elements.

MATERIALS AND METHODS

Test Substance

A yttrium nitrate $Y(NO_3)_3$ solution was prepared by dissolving yttrium oxide (purity >99.99%; Beijing Research Institute of Nonferrous Metals, Beijing, China) in nitric acid and diluting it to a pH of 6 using 1 mol/L sodium hydroxide and deionized water.

Animals and Method of Administration

Sexually mature virgin female and male Sprague-Dawley (SD) rats were obtained from the Laboratory Animal Center of the Academy of Military Medical Sciences (Beijing, China). After 2 weeks of acclimatization, each female rat was mated with a resident male rat of the same strain. Evidence of a vaginal plug was used to indicate successful mating and Day 0 of pregnancy. Throughout the experiment, the animals were kept in a room with controlled illumination (a 12-h light/dark cycle), temperature (20-25 °C), and humidity (40%-70%) and were fed regular rat chow and water *ad libitum.* The procedures were carried out according to ethics guidelines for the care and use of animals in research formulated by the China National Center for Food Safety Risk Assessment.

Mated female rats were randomly divided into four groups of 20 animals each. From gestation day (GD) 6 to postnatal day (PND) 21, the pregnant rats were housed singly and received one of four doses (0, 5, 15, or 45 mg/kg) of the $Y(NO_3)_3$ solution by gavage each day. Mean maternal weight and food consumption were calculated for each group on GD 0, 3, 7, 10, 14, 17, and 21 and on PND 1, 4, 7, 10, 14, 17, and 21. Pregnant females were allowed to deliver their pups spontaneously and nurse them. On PND21, the dams were sacrificed and subjected to a gross examination. Pregnancy status was determined for those rats that failed to deliver.

Each litter was observed daily for signs of toxicity. Body weight was recorded on PND 1, 4, 7, 10, 14, 17, and 21 and weekly thereafter until PND 70. Food consumption was calculated weekly from PND 21 to PND 70. On PND 1, the number of live and dead pups was determined and the sex ratio of the pups was recorded. On PND 4, the number of pups in each litter was reduced to 8 (4 males and 4 females). The pattern of pup assignment, which appears in Table 1, is based on OECD 426 guidelines, with slight modifications.

The first set of pups (20 per sex and dose level; 1 male and 1 female per litter) was evaluated for pre-weaning behavior using behavioral ontogeny tests and for histopathology. Following weaning on PND 21, half of the pups (10 per sex and dose level)