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NARINGENIN MITIGATES BERYLLIUM INDUCED BEHAVIORAL ALTERATIONS IN RATS

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ABSTRACT

Beryllium induced neurotoxicity and therapeutic potential of naringenin had been explored for the first time in rats. For this purpose, 30 female albino rats were divided into six groups. Group 1 served as control, group 2 was naringenin *per se* and rest of the four groups were exposed to beryllium nitrate (1mg/kg, i.p.) daily for 28 days. Naringenin was orally administered in group 4, 5 and 6 at different doses (10, 20 and 30 mg/kg) for 5 days after 28 days of neurotoxicity. All the animals were subjected to elevate plus maze, light dark chamber and rotarod experiments. Beryllium exposure decreased body weight, time spent and number of entries in open arm, increased time spent in close arms as compared to control in elevated plus maze whereas decreased % time spent in bright arena, number of entries in bright arena and increased % time spent in the dark arena in light and dark chamber; decreased motar coordination and balance skills on rotarod. Naringenin showed therapeutic potential and brought the studied variables more towards control at 20 mg/kg dose. It can thus, be concluded that naringenin may be an agent of therapeutic choice in case of beryllium induced behavioral alterations.

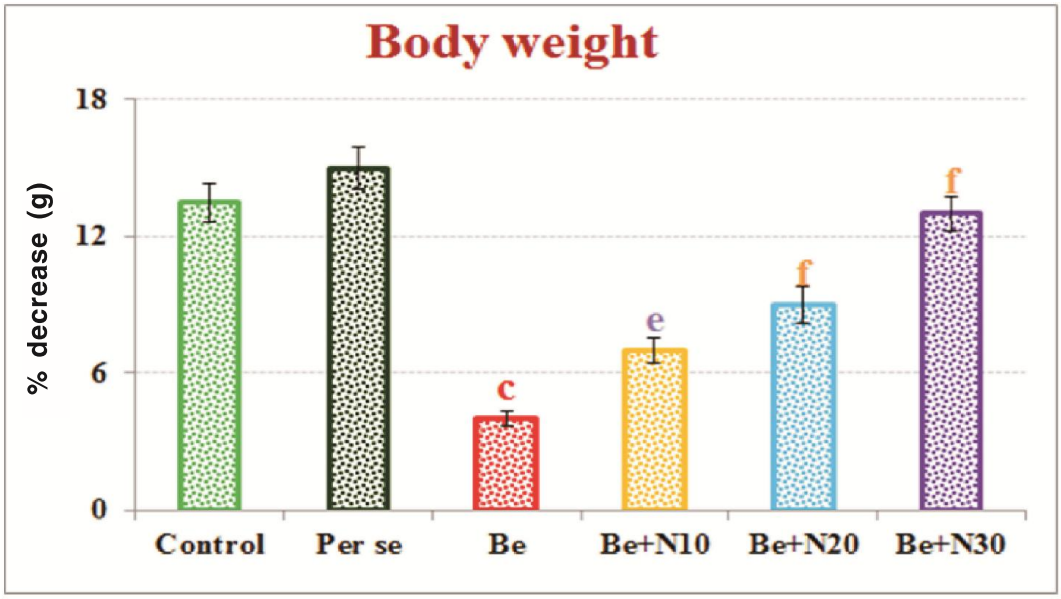
Figures : 03 References : 13 Table : 00
KEY WORDS : Behavioral studies, Beryllium, Naringenin, Rats

Introduction

Industrial pollution is the biggest health risk of mankind. Industrial expansion has led health problems among the workers. A diverse number of metals have been reported to exert serious health effects on human and other organisms. Light metal beryllium and its compounds possess exceptional properties which make them essential in several modern branches of industry. People living in the vicinity of power plant get affected by beryllium from burning of coal and metallurgical processing of beryllium like melting, casting, cutting and electroplating. Other activities like handling of broken florescent tubes, diamond cutting by beryllium knife are also sources of its exposure to toxicity.

Earliest recognized reports of beryllium compounds exposure on pulmonary disease were reported[5] in 1930s and 1940s. Hypersensitivity reactions due to beryllium salt was reported in chronic beryllium disease occurred in the 1950s, which lead to the first clinical demonstration of the immunological basis of this disease[9]. Occupational exposure to beryllium causes a number of diseases such as bronchitis, bronchiolitis, chronic pulmonary granulomatosis and pneumonitis[7,10]. However, very less scientific information is available on beryllium induced neurotoxicity. Thus, in the present investigation, naringenin was evaluated for its therapeutic potential against beryllium induced toxic effects in brain.

Naringenin is one of the natural flavonoid compound, abundantly found in the peels of *citrus* fruits[3]. It is reported as powerful antioxidant, anti-inflammatory and immune-modulatory[13] and



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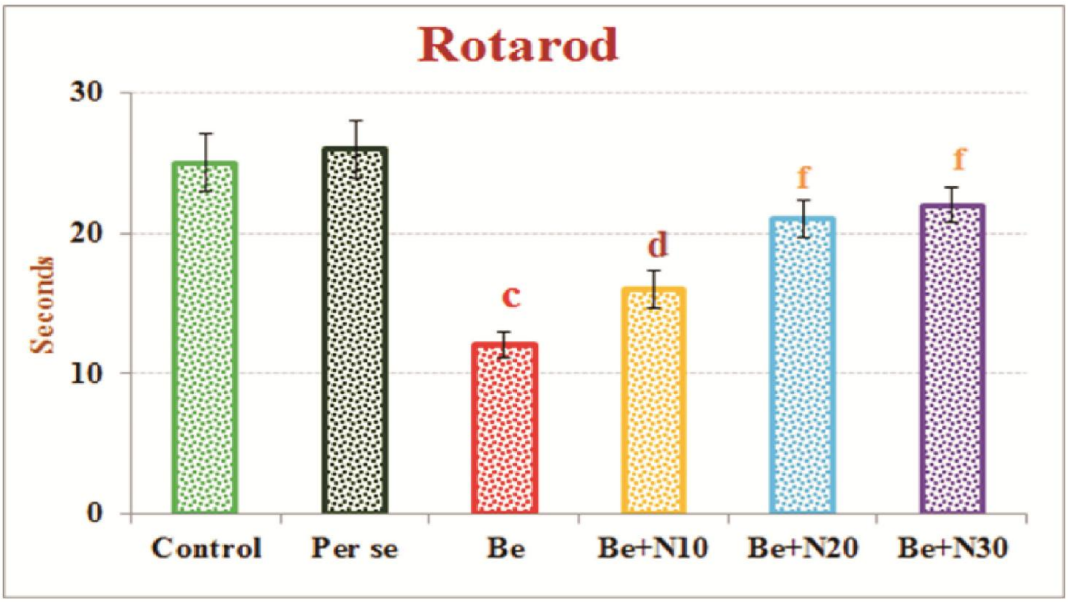

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Fig. 1: Data are mean±S.E of n = 6; [c]Be vs Control at P£0.001,[d]Therapy vs Be at P £ 0.05, [f]Be vs Control at P£0.001, [@]Significant at 5% for ANOVA.

F Variance	Body weight	Rotarod
(At 5% level)	40.01[@]	14.6[@]

Abbreviation: Be= Beryllium, N10= Naringenin 10 mg/kg, N20= Naringenin 20 mg/kg, N30= Naringenin 30 mg/kg.

neuroprotective[8]. It has therapeutic effects against Parkinson's disease, Alzheimer's disease and amnesia related pathology. It protects against oxidative damage in various pathological conditions due to its ability of penetration into the brain[6]. Thus, it was hypothesized that naringenin may also be helpful in mitigating beryllium induced toxic insult to brain and bring behavioral alterations towards normal.

Materials and Methods

Female albino rats of Wistar strain (180± 20 g) were purchased from DRDE, Gwalior Madhya Pradesh, India. Animals were housed in cleaned cages in a proper ventilated house with optimum conditions and acclimatized to animal house condition. Animals were allowed free access to commercial pelleted rat feed and water *ad libitum*. Entire experiment was conducted under the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA) India, guidelines. The study was worked out with the approval of the institutional animal ethics committee (994/Ere/Go/06/CPCSEA).

Beryllium as beryllium nitrate [Be $(NO_3)_2$] purchased from Sigma Aldrich and administered intraperitoneally for the induction of toxicity. To evaluate therapeutic potential of naringenin, thirty six female rats were divided into six groups of six animals each and received beryllium nitrate for 28 days followed by naringenin dose as follows:

Group 1: Normal

Group 2: Naringenin *per se* (20 mg/kg, po)

Group 3: Beryllium (1 mg/kg. ip)

Group 4: Be (1 mg/kg. ip) + Naringenin (10 mg/kg, po)

Group 5: Be (1 mg/kg. ip) + Naringenin (20 mg/kg, po)

Group 6: Be (1mg/kg. ip) + Naringenin (30 mg/kg, po)

Animals were weighed before dose administration and after 24 hours of last animal dose. The % difference (increase) was calculated to evaluate naringenin against beryllium induced behavioral alterations. Elevated plus maze and light and dark chamber are two experimental model of anxiety, based on the assumption that unfamiliar, non-protective and brightly light environmental stress provokes inhibition of normal behavior. This normal behavior suppression was further augmented in the presence of fear of anxiety like state.

Elevated plus maze:

The elevated plus maze consists of two open arms (length 50 cm X breadth 10 cm) and enclosed arms of the same size (Height 40 cm), an open roof arranged in such a manner that the two open arms remain opposite to each other. Rats were individually placed on the centre of the maze facing an open arm, and the number of entries and the time spent in closed and open arms were recorded during 5 minutes observation period[12].

Light dark chamber:

This apparatus consist of two distinct chambers, a dark chamber (20 X 30 X 35 cm) painted black and a bright chamber (30X 30 X35 cm) painted white and brightly illuminated with 100 W white light sources[11,14]. By observing the time duration an animal spends in light or dark arena in light dark chamber, one can predict the anxiety status of that animal.

Motar coordination:

Rotarod was used to monitor the motar coordination status or balance skill of the animals. In this, rats were placed on a rotating rod that can be accelerated for desired rpm to analyze the effects of beryllium on motor coordination and balance skills[2].

Statistical analysis

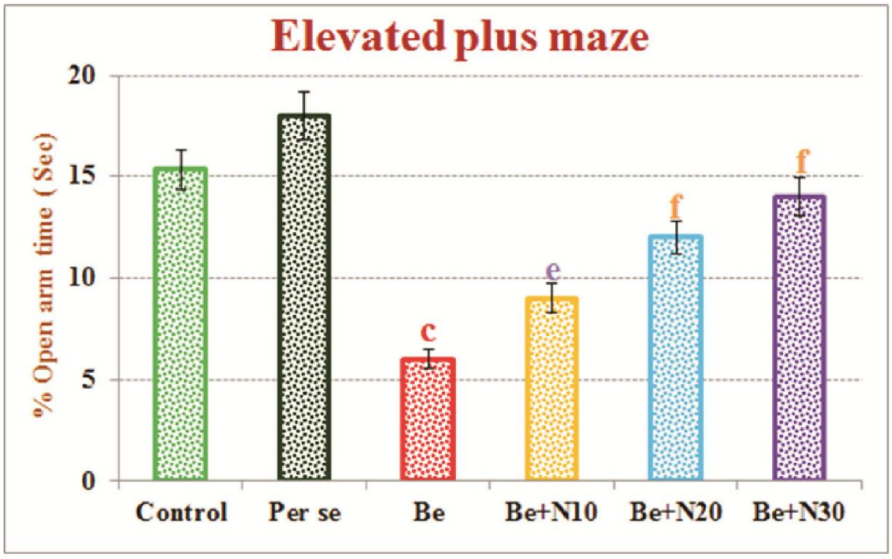
The data was expressed as Mean ± S.E.M. The data was analyzed by student's t-test and compared with controlled group at p≥0.001, p0.01and p0.05.

Results and Discussion

Beryllium administration at 1 mg/kg dose for 4 weeks decreased body weight significantly (Fig. 1A, p0.001). Significant restoration was seen in body weight gain with treatment of naringenin at doses *viz* 20 and 30 mg/kg, po and at 10 mg/kg dose when compared with beryllium (p0.001 and p0.01). Maximum recovery in body weight was observed at 30 mg/kg dose of naringenin.

Motar coordination:

Therapeutic effect of naringenin against beryllium was assessed by behavioral alterations that include motar coordination by rotarod, level of anxiety by elevated plus maze and light and dark chamber. Administration of 1 mg/kg beryllium for 4



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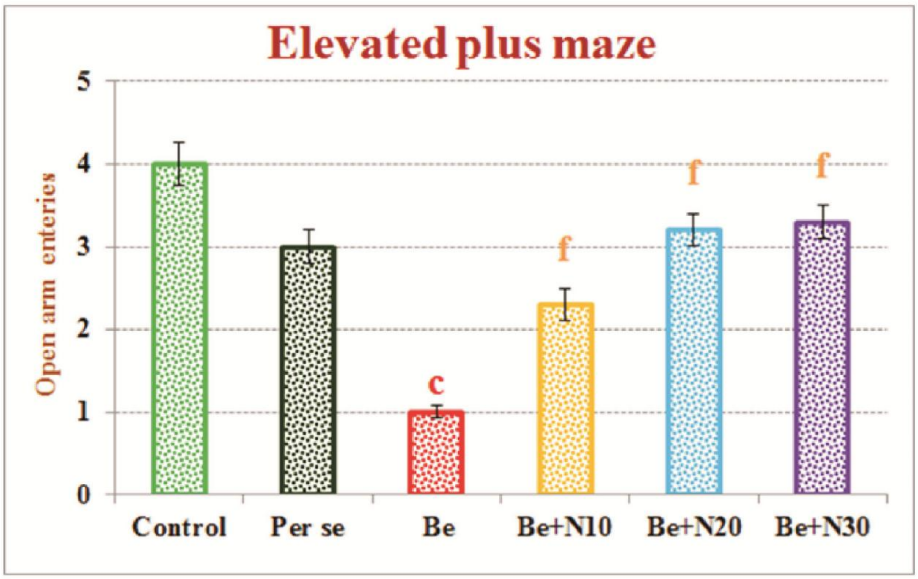

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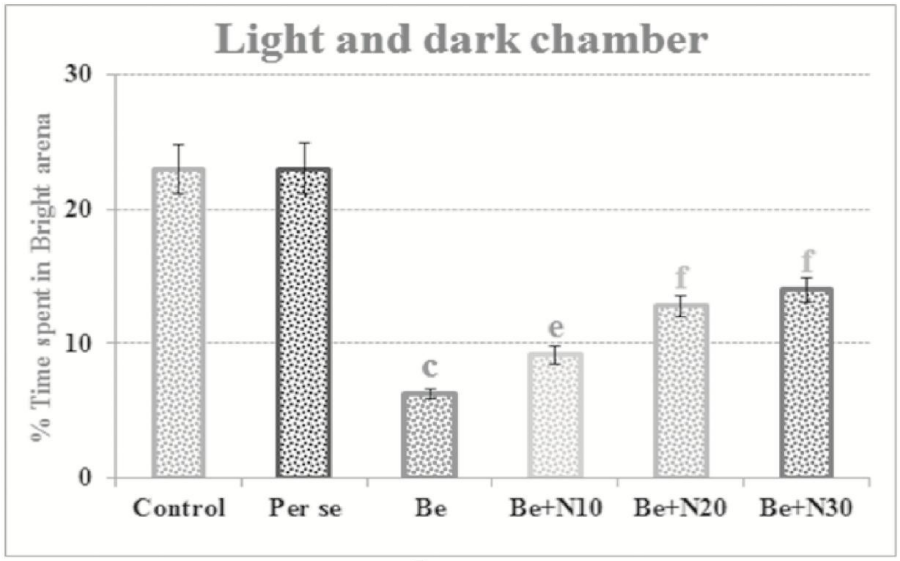
Fig. 2: Data are mean±S.E of n = 6; [c]Be vs Control at P£0.001, [e]Therapy vs Be at P£0.01, [f]Be vs Control at P£0.001, [@]Significant at 5% for ANOVA.

F Variance	%Open arm time	open arm entry
(At 5% level)	29.7[@]	33.8[@]

Abbreviation: Be = Beryllium, N10 = Naringenin 10 mg/kg, N20 = Naringenin 20 mg/kg, N30 = Naringenin 30 mg/kg.



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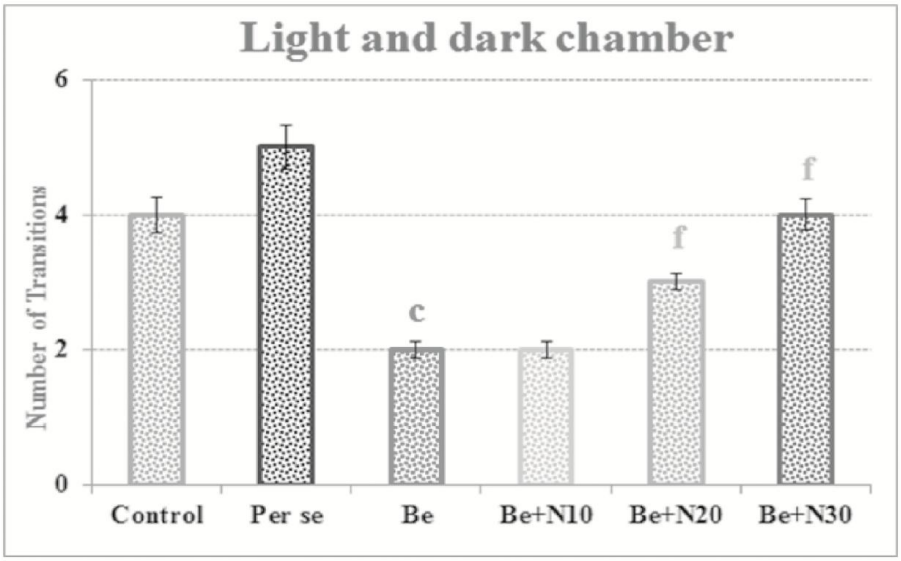

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Fig. 3: Data are mean±S.E of n = 6; [c]Be vs Control at P£0.001,[d]Therapy vs Be at P £ 0.05, [e]Therapy vs Be at P£0.01, [f]Be vs Control at P£0.001, [@]Significant at 5% for ANOVA.

F Variance	Number of transition	%TSBA
(At 5% level)	38.4[@]	39.3[@]

Abbreviation: Be= Beryllium, %TSBA = % Time spent in bright arena, N10= Naringenin 10 mg/kg, N20= Naringenin 20 mg/kg, N30= Naringenin 30 mg/kg.

weeks decreased (p0.001) coordination was found (Fig. 1B). Significant recovery at 0.1% was found in motar coordination when animals were exposed to 20 and 30 mg/kg doses of naringenin. Naringenin dose at 10 mg/kg was restored motar coordination at 1% level of significant.

The neurological problem was indicated by inability of rat to coordinate on rotating rod for 3 min in each trial[1]. Beryllium exposure significantly decreased motor coordination or balance skill that was restored toward control in the experimental rats with 20 mg/kg dose of naringenin.

Elevated plus maze:

Beryllium administration (1 mg/kg, ip) for 4 weeks enhanced (p0.001) anxiety level (decreased % Open arm time and decreased open arm entries) (Fig. 2A & 2B). Restoration (p0.001) was found in open arm entries at all therapeutic doses. Therapeutic doses of naringenin 20 and 30 mg/kg were increased (p0.001) open arm time and 10 mg/kg dose of naringenin increased % open arm time at 1% level when compared with beryllium induced group.

Anxiolytic drug enhances the number of entries and time spent in open arms[2,14]. In the present study, beryllium exposure to experimental animals decreased number of entries and %time spent in open arms showing anxiogenic effect, so 20 mg/kg, naringenin worked as anxiolytic drug against beryllium.

Light and dark chamber:

Administration of beryllium dose 1mg/kg for 4 weeks enhanced (p0.001) anxiety level (Number of transition and % time spent in bright arena) (Fig. 3A and 3B). Number of transition and time spent in bright arena were restored in dose dependent manner of naringenin. Number of transition were increased (p0.001) at 20, 30 mg/kg but 10 mg/kg dose of naringenin was not showed any significant effect; therapeutic doses of naringenin 20 and 30mg/kg were increased (p0.001). %time spent in bright arena and 10 mg/kg dose of naringenin increased time spent in bright arena at 1% level when compared with beryllium induced group.

Reduction in time spent and number of entries in the light chamber is regarded as markers of anxiety[2,14]. This study showed that beryllium exposure decreased % time spent and number of entries in bright arena the light chamber which confirmed anxiogenic effects of beryllium that was decreased by naringenin dose at 20 mg/kg.

Beryllium administration for 28 days altered behavior of the experimental animals. Naringenin showed therapeutic potential in a dose dependent manner, however, 20 mg/kg dose was found to be most effective. It can thus, be concluded that naringenin may be an agent of therapeutic choice in case of beryllium induced behavioral alterations.

References

1. DUNHAM, N.W. AND MIYA, T.S. (1957) A note on a simple apparatus for detecting neurological deficit in rats and mice. *J. Am. Pharm. Assoc.* **46** : 208-209.
2. GOPAL, KRISHNA, H.N., SANGHA, R.B., MISHRA, N. AND MRSM, PAI. (2006) Antianxiety activity of NR-ANX-C, a polyherbal preparation in rats. *Indian J. Pharmacol,* **38** : 330-335.
3. HEO, H.J., KIM, D.O., SHIN, S.C., KIM, B.G. AND SHIN, D.H. (2004) Effect of antioxidant flavonone, naringenin, from *Citrus junos* on neuroprotection. *Journal of Agricultural and Food Chemistry* **52** :1520-1525.
4. HOLMES, A. (2003) Mouse Behavioral Models of Anxiety and Depression. *Mouse Behavioral Phenotyping*, 42-47.
5. JAMESON, C.W. (1996) Introduction to the conference on beryllium related disease. *Environmental Health Perspective* **104** : 935.
6. KHAN, M.B., KHAN, M.M. AND KHAN, A. (2012) Naringenin ameliorates Alzheimer's disease (AD)-type neurodegeneration with cognitive impairment (AD-TNDCI) caused by the intra cerebroventricular streptozotocin in rat model. *Neurochemtry International* **61** : 1081-1093.

7. MATHUR, S., PRAKASH, A.O. AND MATHUR, R. (1990) Therapeutic use of Liv-52 in beryllium induced reproductive toxicity in rats. *Probe* **4** (XXIX) : 266.

8. MUTHAIAH, V.P., VENKITASAM,Y L., MICHAEL, F.M., CHANDRASEKAR, K. AND VENKATACHALAM, S. (2013) Neuroprotective role of naringenin on carbaryl induced neurotoxicity in mouse neuroblastoma cells. Journal of *Pharmacology and Pharmacotherapeutics* **4** (3):192-197.

9. NEWMAN, L.S., LLOYD, J. AND DANIOFF, E. (1996) The National history of beryllium sensitization and chronic beryllium disease. *Environmental Health Perspective* **104** : 937.

10. NIRALA, S.K., BHADAURIA, M., MATHUR, R. AND MATHUR, A. (2008) Influence of á-tocopherol, propolis and piperine on therapeutic potential of tiferon against beryllium induced toxic manifestations. *Journal of Applied Toxicology* **28** : 44-54.

11. PARMAR, N.S. AND PRAKASH, SHIV (2006) Screening methods in Pharmacology. *Published by N.K. Mehra for Narosa publishing House pvt. Ltd,* **73**, 239.

12. SHABANI, M., PARSANIA, S., ASADY, SHEKAARI, M. AND SHAHROKHI, N. (2012) Profound, destructive effects of adolescent exposure to Vincristine accompanied with some sex differences in motor and memory performance. *Can. J. Physiol. Pharmacol.* **90** (4) : 379–86.

13. YILMA, A.N., SINGH, S.R., MORICI, L. AND DENNIS, V.A. (2013) Flavonoid naringenin: a potential immunomodulator for *Chlamydia trachomatis* inflammation. Mediators of Inflammation. *Mediators of inflamation* 1-13.