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Effects Of Retinoic Acid On Survival Percent And Growth Of Swiss Albino Mice

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Abstract: *The development of an organism is a complex process of embryogenesis involved cell proliferation, differentiation, migration and organogenesis. Teratology is the branch of biology that deals with abnormal development . Many agents interfering the development process can cause malformations in the embryo. The study of these congenital abnormalities is called teratology and agents which are responsible for causing these malformations are called teratogens. Susceptibility to teratogens depends on the genotype of the organism, including species as well as strain differences. Etienne Geoffrey Saint Hilaire and his son in 1820 started experiments on chick embryo by disturbing its environment in different ways at different embryonic stages. They found some anomalies like Trioncephally, atrophy of eyes and spina bifida in his experiments (Tuli, 1968). In 1877 Dareste reported found some anomalies like Trioncephally, atrophy of eyes and spina bifida in his Experiments (Tuli, 1968). Retinoids are essential for spermatogenesis oogenesis, placental development, foetal morphogenesis and growth. The role of vitamin A in vision is well known . Active derivative of vitamin A (Retinoids) play an important and multiple role in Mammalian development and homeostasis (Sapin, et.al. 1997, Dup et al 1997). These essential dietary compounds are needed in very small quantities, they are not synthesized by the animals and are obtained from external sources in the form of β carotene (C40H56). In cells of intestine β carotene is converted into vitamin A alcohol and is transported by blood to liver where it is esterfied and stored as vitamin A palmitate. RA is not stored in liver but is derived from retinal and it is biologically the most potent form of vitamin A.Vitamins are organic compounds which are devided into 2 categories i Fat soluble (A,D,E,K) And ii water soluble (B and C).Vitamin A belongs to fat soluble category of vitamins. A fat soluble substance essential for life was first discovered in egg yolk in 1909 by Stepp (Pawson, 1981; Robert and Sporn, 1984). It was confirmed by Mc Collum Devis in 1913 when he found a compound with similar biological activity in butter fats, egg yolk and cod liver oiland was named fat soluble vitamin A. In the present study effects of different doses on Survival rate on different development stages of Swiss albino mice are studied.*

Key words: *Retinoids, Teratological, Swiss albino mice, Survival percentage.*

I. Introduction

Teratology is a branch of Embryology that deals with abnormal development . The development of an organism is a complex process of embryogenesis involved cell proliferation, differentiation, migration and organogenesis. Many agents interfering the development process can cause malformations in the embryo. The study of these congenital abnormalities is called teratology and agents which are responsible for causing these malformations are called teratogens. Susceptibility to teratogens depends on the genotype of the organism, including species as well as strain differences. Teratological experiments was started from 1820s with the studies of Etienne Geoffrey Saint Hilaire and his son on chick embryo by disturbing its environment in different ways at different embryonic stages. They found some anomalies like Trioncephally, atrophy of eyes and spina bifida in his Experiments (Tuli, 1968). In 1877 Dareste reported some congenital malformation in the chick embryo by environmental disturbance.Retinoids are essential for spermatogenesis oogenesis, placental development, foetal morphogenesis and growth. The role of vitamin A in vision is well known . Active derivative of vitamin A (Retinoids) play an important and multiple role in Mammalian development and homeostasis (Sapin, et.al. 1997, Dup et al 1997). These essential dietary compounds are needed in very small quantities, they are not synthesized by the animals and are obtained from external sources in the form of b carotene (C40H56). In cells of intestine β carotene is converted into vitamin A alcohol and is transported by blood to liver where it is esterfied and stored as vitamin A palmitate. RA is not stored in liver but is derived from retinal and it is biologically the most potent form of vitamin A.Vitamins are organic compounds which are devided into 2 categories i Fat soluble

(A,D,E,K) and ii water soluble (B and C).Vitamin A belongs to fat soluble category of vitamins. A fat soluble substance essential for life was first discovered in egg yolk in 1909 by Stepp (Pawson, 1981; Robert and Sporn, 1984). It was confirmed by Mc Collum Devis in 1913 when he found a compound with similar biological activity in butter fats, egg yolk and cod liver oiland was named fat soluble vitamin A. The name of vitamin was given by Dummond In 1920. The natural and synthetically obtained vitamin A is found in several forms of Alcohol (Retinol), aldehyde (Retinol palmitate), acetate and retinoic acid. They were collectively known as retinoids.Retinol or alcoholic form of vitamin A is a unstable organic compound. They are easily oxidized specially in the presence of heat. The ester form of vitamin A are fairly stable. When vitamin A is taken in the form of esters (Palmitate/acetate) , its converted into Alcoholic form (Retinol) in the intestinal cells to be carried to liver, where it is converted to palmitate form for storage. Retinoic acid is not stored in the liver but it is derived from retinol and it is biologically most potent form.In the present study effects of different doses on Survival rate on different development stages
of swiss albino mice are studied.

II. Materials And Methods

The mice were obtained from mice breeding center , Department of Zoology M.D.S.U. Ajmer. Animals were fed synthetic diet mice feed pallets (Brook Bond Lipton India Ltd.) Supplemented with germinated grains, seasonal green vegetables, multivitamin drops and water ad libitum. Tetracycline mixed in water in water was regularly given.There are four virgin female mice 25+-1gm were caged with fertile healthy male in the evening and these were examined for presence of vaginal plug, the next morning. The day appearance of vaginal plug was considered as day 0 of pregnancy.

The veginal plug

After copulation a secretion of seminal vesicles in the ejaculate of the male coagulate to form a plug in the vegina extending from the cervix to the valve , where it is ordinarily Visible and is a convenient external sign that mating has occurred (Green , 1996).

Duration of pregnancy

The gestation period in this species is 19 days and occasionally 20 days. The youngs are most frequently born in the early hours of the morning between midnight and 4 AM.

Objectives

Investigation of the effects of retinoic acid on different developmental stages (5^{th}, 8^{th}, 11^{th}, and 14^{th}) of mouse embryo.
To study effects of RA on organogenesis of skin, liver, heart .
Effect of RA on skeletal elements.

Experimental design

The pregnant females were devided into following groups :
(6 animals per group)
Group A : untreated
Group B : Treated

Doses

Suitable (non lethal or sublethal) doses of RA were screened on mice embryos on different
stages of developments. The following doses were found to produce various teratological defects on developing mice embryos
1. 2mg RA /pregnant female.
2. 4 mg RA /pregnant female.

III. Results And Discussion

Teratological effects of RA on different development stages of mice embryo :
Survival percent (S p) and growth retardation(Weight and length). Survival percentage Studies on the effect of RA on percent survival of mice embryos were carried out by Treating the pregnant female at 5^{th}, 8^{th} , 11^{th}, 14^{th} day gestation with different doses of RA.For each dose of RA (2mg and 4 mg) 3 pregnant female were selected and normal litter of per female was 8-10 embryos . Therefore percent survival was based on 24 to 26 embryos per dose of RA.
Untreated The survival percentage (mean) was observed to be 90% in the embryos of untreated Pregnant females.

Dose 2 mg RA

When 2mg RA was administered on 5^{th},8^{th},11^{th}, and 14^{th} day gestation the survival % was
85%, 59.2%, 69.4% and 79% respectively.(Table 1)

Dose 4 mg RA

When pregnant female of day 5^{th},8^{th},11^{th} and 14^{th} day gestation were treated with 4mg dose of RA the percent survival was 75%, 50%, 55% and 705 respectively. (Table 2).

Growth Retardation

The growth retardation effects of RA on mouse embryos was calculated in terms of weight and length (snout to base of tail) of the embryos.

Control (Untreated)

The average weight of control embryos was observed to be 2.5 gm and average length was found to be 30mm.

Dose 2mg RA

The weight of embryos of females treated with 2mg conc. Of RA was found to be 2.3gm, 1.3gm, 1.52gm and 1.7 gm respectively on the 5^{th}, 8^{th}, 11^{th}, and 14^{th} day gestation period (Table 3).

The average length of 2mg RA treated embryos of pregnant females of 5^{th}, 8^{th}, 11^{th}, and 14^{th} day gestation found to 27mm, 22mm, 23mm, and 24mm respectively 9Table 3 fig 1).

Dose 4 mg RA

The next higher dose of RA has found to retarded growth of embryos severly. Pregnant female treated on 5^{th},8^{th},11^{th}, and 14^{th} day gestation showed 2.0 gm, 1.05 gm, 1.32gm, and 1.4gm weight embryos and length of 25mm, 22mm, 23mm and 24mm of the embryos of treated female (Table 4 fig 2).

Table 1

TABLE - 1

Survival percent of the embryos of pregnant mice treated with 2 mg RA at different gestation days.

Per female dose of RA	Gestation period	Survival percent (mean)
CONTROL	—	90 %
2mg RA/pregnant female	5th	85.0%
2mg RA/pregnant female	8th	59.2%
2mg RA/pregnant female	11th	69.4%
2mg RA/pregnant female	14th	79.0%

Table 2

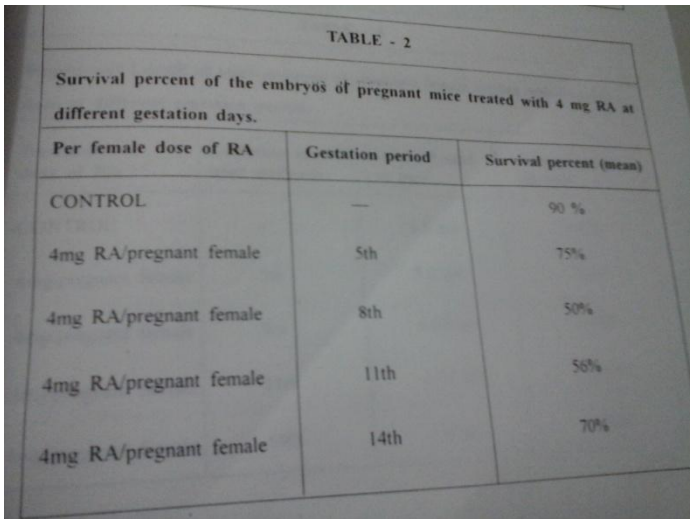

TABLE - 2

Survival percent of the embryos of pregnant mice treated with 4 mg RA at different gestation days.

Per female dose of RA	Gestation period	Survival percent (mean)
CONTROL	—	90 %
4mg RA/pregnant female	5th	75%
4mg RA/pregnant female	8th	50%
4mg RA/pregnant female	11th	56%
4mg RA/pregnant female	14th	70%

Table 3

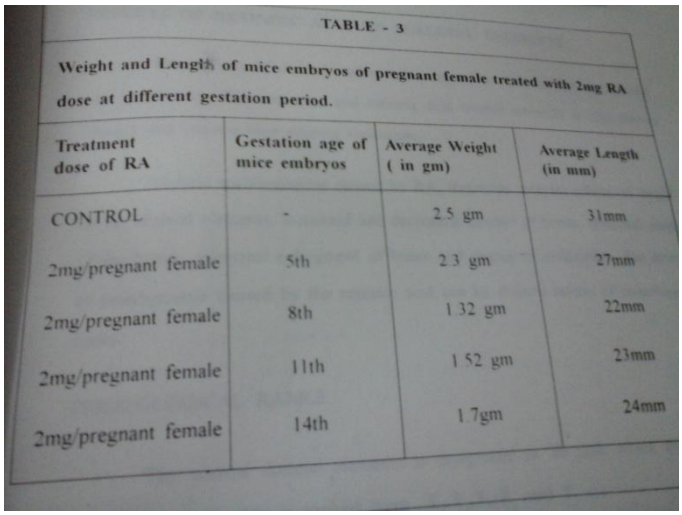

TABLE - 3

Weight and Length of mice embryos of pregnant female treated with 2mg RA dose at different gestation period.

Treatment dose of RA	Gestation age of mice embryos	Average Weight (in gm)	Average Length (in mm)
CONTROL	-	2.5 gm	31mm
2mg/pregnant female	5th	2.3 gm	27mm
2mg/pregnant female	8th	1.32 gm	22mm
2mg/pregnant female	11th	1.52 gm	23mm
2mg/pregnant female	14th	1.7gm	24mm

Table 4

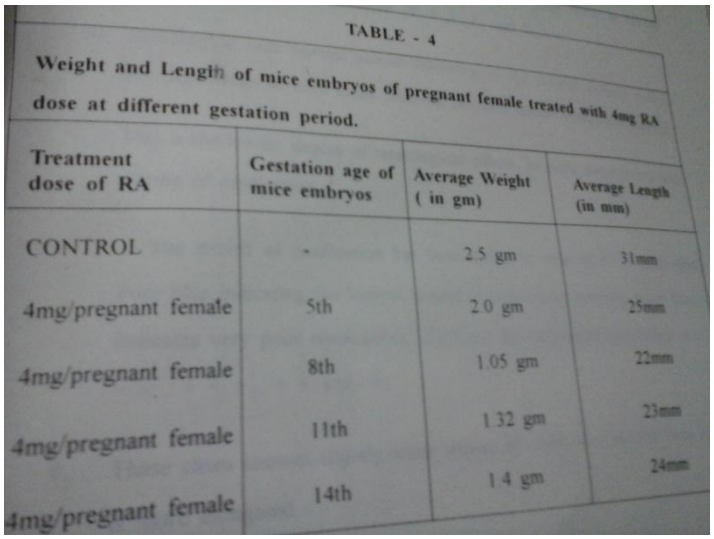

TABLE - 4

Weight and Length of mice embryos of pregnant female treated with 4mg RA dose at different gestation period.

Treatment dose of RA	Gestation age of mice embryos	Average Weight (in gm)	Average Length (in mm)
CONTROL	-	2.5 gm	31mm
4mg/pregnant female	5th	2.0 gm	25mm
4mg/pregnant female	8th	1.05 gm	22mm
4mg/pregnant female	11th	1.32 gm	23mm
4mg/pregnant female	14th	1.4 gm	24mm

IV. Conclusion and Summary

The results of the present study clearly demonstrated that RA effect the morphogenetic Pathway of various organs in specific manners during development of mouse embryos. The effects are dose dependent and also depend on the stage of development of mouse embryos. The results showed growth retarding effect of RA and survival percentage in early and late embryos. Similarly the results of present study related to teratogenic effects of RA on mouse embryo further confirm by investigations of some others, who also observed teratogenic and toxic effect of RA on dev embryos of vertebrates.(Hardy, 1968, Thompson 1969, Kochhar, 1973, Lotal 1980, Sulik and de hart, 1988, Tickle 1989, Jhonson and scadding, 1991, Dersch and Zile, 1993, Singh, 1995, Dickman and Smith 1996, Colbert, 1997, Dickman et.al. 1997, Shobhawat ,1998. 2 and 4mg conc RA on 5^{th}, 8^{th}, 11^{th}, and 14^{th} day ges females was found that survival was higher on at 2mg dose level as compared to 4mg dose. Further stage 8^{th},and 11^{th} were most crucial development stage when survival percent was very low when compared with 14^{th} day gestation stages. This clearly indicate that the dose dependent effect of RA on mouse embryo. Stage 5^{th} was found to be an exception because % survival was higher at this stage as compared to 8^{th} or 11^{th} day gestation stages.The decrease in percent survival of mouse embryo treated with RA is due to its toxic effects. Hypervitaminosis has been found to be toxic in many other mammalian system.
(Lammer, 1985).

References

[1]. Coblan , S.Q.(1953):Excessive intake of Vit A as a cause of congenital anomalies in Rat. Science 117:535-536.
[2]. Coblan , S.Q. (1954):Congenital anomalies in rat produced by excessive intake of vit A during pregnancy. Pediatrics. 13 556-557.
[3]. Dickman , E.D. and Smith, S.M. (1996) Selective regulation of cardiomyocyte expression and cardiac morphogenesis by RA dev dynamics 206(1):39-48.
[4]. Effendy. I., (1996): Differential irritant skin responses to topical RA and sodium layryl
[5]. sulphate alone and in crossover design British J. of Dermatology 134: 420-430. (Abstract).
[6]. Elmzar, M.M.A.(1996). Pattern of Retinoid induced teratogenic effect. Possible relationship with relative selectivity for nuclear retinoid receptors RAR-alpha,RAR-beta and RAR gama. Teratology 53(3):158-167.(Abstract).
[7]. Fisher, G.J. and Voiegwwa, J.J. (1996): molecular mechanisms of retinoid actions in skin,FASEB J.10(9).1002-1013.
[8]. Giroud, R. and Martinet,(1954): Fertes du Polasis chezl embryos de rat per hypervitaminose A., Comp.Rend.Soc.Biol.148:1742-1743.
[9]. Goodman, A.B. (1996) : Congenital anomalis in relative of Schizophrenic probands may indicate a retinoid pathology. Schizophrenia Research 19: 2-3:169-170.(Abstract).
[10]. Griffiths. C. E. M. (1996): Tropical RA changes the epidermal cell surface glycosylation
[11]. pattern towards that of a mucosa epithelium. British Journal of Dermatology. 134; 431-436.
[12]. Leelaprute. L .; Boonpucknaving. V.; Bharmar apratuati. N., and Weerapradist. W.(1973). Hypervitaminosis A in rat Arch. Sci 85:42-55.
[13]. Kalter, H.C.(1960): The teratogenic effect of hypervitaminosis upon the face and mice of inbred mice. Ana. N.Y. Acad. Sci.85:42-55.
[14]. Kalter, H.C. and Warkany J. (1961): Experimental production of congenital malformation
[15]. In strains of inbred mice by maternal treatment with hypervitaminosis. Amer J. path,381-21.
[16]. Kochhar, D.M. (1967): Teratogenic activity of Retinoic Acid. Acta Pathol Micro. Biol. Scand.70:398-404.
[17]. Moore, T. (1957) : " Vitamin A " Am. Elsevier, New Delhi.
[18]. Niazi, I.A. : Pescitelli M.J.; and Stocum, D.L., (1985) ; Stage dependent effects of RA on regenerating urodele limbs.