Short Communication

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The Effect of $MnSo_4$ on Angiogenesis in Chick Embryo

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The formation of the blood vessels from pre-existing blood vessels is called angiogenesis. How these blood vessels form is unique and complex process. Blood vessel grows in response to chemicals in their immediate environment. Some chemicals are pro-angiogenic promoting blood vessel growth while other anti-angiogenic, inhibiting blood vessel growth. Depending on which chemicals are present, blood vessels will increase and decrease into a distinct pattern. The dynamic process of angiogenesis continues throughout life. Stimulating angiogenesis can have therapeutic value in several diseases such as ischemic heart disease or in wound healing. Excess or uncontrolled angiogenesis can also be detrimental to the body by contributing to diseases such as retinopathy or by facilitating tumor growth. Drugs that promote or inhibit angiogenesis are currently being developed, particularly in the treatment of cancer [1].

The mechanism of angiogenesis is well studied but its mechanism is not completely understood although it is known that it involves the splitting of existing blood vessels to form new blood vessels. The vessel wall extends into the lumen causing the single vessel to split into two vessels forming artery and vein bifurcations [2]. Angiogenesis plays an important role in cancer as the cancerous cells require a blood supply for the growth of solid tumors. Solid tumor development and metastasis are angiogenesis-dependent processes. The new method of prevention of cancer and cancer therapy is the use of agents having antiangiogenic activity. Chick embryo chorioallantoic membrane is the most popular model to study angiogenesis. Thus, the chorioallantoic membrane (CAM) assay is well-established and widely used as a model to examine angiogenesis and anti-angiogenesis effects.

Manganese sulphate (*$MnSO_4$*)

Manganese sulphate ($MnSO_4$) usually refers to the inorganic compound with the formula $MnSO_4$. It has a role as a nutraceutical. It is a pale pink or white crystal or powder. It is an inorganic compound, and soluble in distilled water. It plays important roles in development, metabolism, and the antioxidant system. It plays an important role in growth, bone development; optimal eggshell quality, and performance of poultry [3]. In the present study, we investigate the effect of $MnSO_4$ on angiogenesis in chick embryos. The objectives of the study were, to study the angiogenesis in chick and to study the effect of Manganese sulphate ($MnSO_4$) on the growth of blood vessels in chick.

Eggs, Incubator, weighing machine, Beakers, Petri dish, watch glass, Insulin syringe, forceps, scissors, cotton, medical tape.

Chemicals: $MnSO_4$ (Manganese sulphate), 70% alcohol

The required numbers of eggs were collected from a backyard poultry farm located at Kaneri, Kolhapur. After collection, healthy and almost same-sized eggs of Rhode Island white were selected by considering parameters such as colour-light brown, shape- small, and oval.

The eggs were placed in an incubator for incubation as per the experimental design.

Preparation of stock solution

Manganese sulphate ($MnSO_4$) is a crystalline substance, white in colour and soluble in water was used as a toxic substance. For the present investigation about 100 mg of manganese sulphate was dissolved in 100 ml of distilled water to prepare a solution of 1mg/ml and injected into the developing embryo in the eggs which were incubated at 37 C for different time period of exposure.

Experimental design and induction of dose

Eggs were selected to start an experiment. For the present experiment, two groups of eggs were prepared first group was kept as a control group, and the second group as an experimental group injected with manganese sulphate solution after 48 h of incubation of eggs. In the experimental group, the eggs were numbered 1, 2, 3, etc. Egg 1 was incubated up to 72 h. Egg 2 was incubated up to 96 h and Egg 3 was incubated up to 120 h. All eggs were kept in the incubator which was sterilized by using 70% alcohol to maintain the aseptic condition and make it free from germs and microorganisms. The incubator was pre-started to maintain 37^0C temperature which is essential for the development of chick embryo. After 48 h of incubation eggs were again cleaned with 70% alcohol and under sterilized and aseptic conditions the eggs were treated with 0.1 ml stock solution of manganese sulphate. After injection eggs with developing embryos were re-sealed with adhesive sterile tapes. Again, experimental eggs were kept in an incubator for further embryonic development [4]. The eggs were dissected and embryos were observed at 72 h (24 h after microinjection), and 96h (48 h after microinjection). After observation of the chick embryo body weight of the embryo was taken, and a comparison done between the control group and the experimental group.

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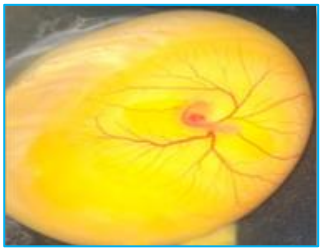
After the incubation control and experimental group eggs were observed and compared. It is observed in the experimental group that there is an insignificant reduction in the weight of chick embryos at 72 h and 96 h after treatment with manganese sulphate as compared to the weight of chick embryos in the control group [5]. The difference between the weight of the control egg embryo and the experimental egg embryo is given below:

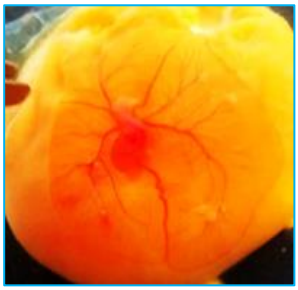
Control		Treated	
Incubation hours	Weight of embryo	Incubation hours	Weight of embryo
72	0.448gm	72	0.332 gm
96	0.781gm	96	0.465gm



72 hours (control embryo)

72 hours (treated embryo)

96 h (control embryo)



96 h (treated embryo)

Changes in angiogenesis

After 72 h (i.e. 24 h after manganese sulfate exposure) and 96 h (i.e. 48 hours after manganese sulfate exposure) of incubation the treated embryo showed reduced number of blood vessels as compare to the control embryo also reduction in size of blood vessel [6]. Reduced in size of heart and reduced in movement of heart.

SUMMARY

The present investigation is about the effect of $MnSO_4$ on angiogenesis in chick embryos, Angiogenesis is the development of new blood vessels from the preceding small blood vessels about cell proliferation, the required numbers of eggs were collected from a backyard poultry farm located at Kaneri, Tal- Karveer. The eggs were placed in an incubator for incubation as per experimental design. The stock solution $MnSO_4$ was prepared in 1mg/ml concentration. To study the effect eggs were kept in two groups, the first group kept as a control group and the second group as an experimental group. The experimental group was injected with manganese sulphate solution after 48 h of incubation of eggs. In the experimental group, the eggs were numbered 1, 2, 3, etc. Egg 1 was incubated up to 72 h. Egg 2 was incubated up to 96 h and Egg 3 was incubated up to 120 h. After the incubation control and experimental group eggs were observed and compared. It is observed in the experimental group that there was an insignificant reduction in the weight of the chick embryo at 72 h and 96 h after treatment with manganese sulphate as compared to the weight of the chick embryo in the control group. The $MnSO_4$ can induce toxic interaction which can highly reduce the angiogenesis in the chick embryo. It can be concluded that $MnSO_4$ can induce toxic interaction which can highly reduce the viability of the embryo. Though it is used in Poultry feed as important nutrition it may causes toxic effects in the embryonic development of chick.

LITERATURE CITED

1. Folkman J. 2007. Angiogenesis: an organizing principle for drug discovery? *Nat. Rev. Drug Discovery* 6: 273-286.
2. Adair TM, Montani J. 2011. Colloquium series on integrated systems physiology: from molecule to function to disease: Angiogenesis. *Morgan and Claypool Life Sciences* 10: 1-71.
3. Olgum O. 2016. Manganese in poultry nutrition and its effect on performance and eggshell quality. *Worlds Poultry Science Journal* 73: 45-56.
4. Tendulkar SS, Kamble NA. 2020. Copper sulphate induced embryological anomalies in avian species *Gallus gallus*. *International Journal of Zoological Investigations* 6(2): 233-239.
5. Richards MP, Stock MK, Metcalfe J. 1991. Effects of brief hypoxia and hyperoxia on tissue trace element levels in the developing chick embryo. *Magnes. Trace Elements* 10(5/6): 305-320.
6. Cao Y, Linden P, Farnebo J, Cao R, Eriksson A, Kumar V, Qi JH, Claesson-Welsh L, Alitalo K. 1998. Vascular endothelial growth factor C induces angiogenesis in vivo. *Proceedings of National Academy of Sciences USA* 95(24): 14389-14394.