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Pharmacological Study of Isolated Cardiac Glycoside from Chaksini – Peristrophe bicalyculata Nees on Frog's Heart in Vitro Analysis

Sadrul Hoda
Associate Professor in Saidla
Aligarh unani / Ayurvedic Medical College and acn hospital Aligarh

The drug "Chaksini, Peristrophe bicalyculata Nees" (Family Acanthaceae) is a lesser known drug. It is used for different ailments especially in wound healing in traditional Unani medicine.
We have isolated a glycoside by Charcoal Onto method Peach & Tracy 1955. Now for its confirmatory test we are Studying Pharmacological action on isolated glycoside in Frog heart.

Keywords: Chaksini, Peristrophe bicalyculata, Acanthaceae, Cardiac Glycoside, Isolated Frog Heart, Charcoal.

Introduction

"Peristrophe bicalyculata Nees" is commonly known as "Chaksini". It belongs to the family Acanthaceae. It is a commonly used weed in and around Aligarh, a sub-tropic species of Asia and Africa and also available throughout India (Anonymous, 1959; Kirtikar and Basu, 1987; Bamber, 1916; Hains, 1925; Dalziel, 1948; Duthie, 1960; Prains, 1963). It is a lesser known drug and mentioned in few of the Unani classical books for its medicinal uses. Ghani (1921) and Khan (1882) mentioned that the dried plant is beneficial in psychosomatic disorders and wound healing and possesses anti-venom activity. In medicoethano-botanical literature the plant has been mentioned as a good antidote for snake poison (Mhasker and Caius, 1931; Chopra, 1956). No scientific work is reported so far on this plant, particularly for its wound healing effect. We have isolated a cardiac glycoside from the plant. "Chaksini" for its preliminary test of pharmacological action on Frog heart. The test is carried out with the standard control test. (Peach and Tracy 1955).

Materials and Method:

SEPARATION OF GLYCOSIDE BY ADSORPTION METHOD:

Glycoside may be separated from the "Chaksini Peristrophe – bicalyculata Nees", as crystale form with specific adsorption method. The materials such as fullers earth as a charcoal, have been successfully employed for this purpose (Peachand Tracey 1955).

METHOD:

The fresh plant is grounded in a mortar shade. The powder of Chaksini 150gm

with 200 ml of water containing 5ml concentrated nitric acid is taken in flask. The pulp is pressed though a Lenin cloth and the fluid spun on a centrifuge. The brownish turbid fluid is then filtered through Kisselghur to remove suspended matter. The clean filter treated with activated charcoal at the rate of 3.8 gm/180 gm fresh plant powder. The charcoal is boiled with water for a short time to expel air before addition to the solution. After addition of the charcoal the solution is kept stirred for 20-30 minutes and then charcoal is filtered off on a Buchner funnel using Kisselghur as a filter aid. The filtrate is again treated with a large quantity of charcoal (20gm/ 150gm of plant). This time the charcoal supposed to be adsorbed glycoside onto the charcoal with the 20 minutes. Large bulk of Kisselghur is added and mix thoroughly then filtered off on a Buchner funnel on two paper covered with a thin layer of Kesselghur. The pael is carefully washed with distilled water to remove acid and unabsorbed matter. The glycoside eluted from the charcoal by using with 50% ethanol.

The filtrate is evaporated to a thin syrup under reducing pressure and treated with two volume of methanol. A precipitate containing piction

is filtered off. The clear filtrated evaporated under reduced pressure and finally taken down to a thick syrup on warm bath. Seedling and stirring produced a semi solid mass of crystallization on cooling and stirring with a little methanol more crystals come out glycoside is soluble in warm methanol but almost insoluble in cold methanol. The constant MP (Melting Point) = 220-222^{o}C. Fig. 1, 1.2

Materials and Method :

Bio-assay on Cardio-vascular System

Frogs (Rana Tigrina) weighing about 100-150gm are pitched. The Chest is opened then removes the pectoral girdle and the heart is explored. The peri-cardiac sac is removed and the inferior venacava is cannulated and attached with the Murphys drip to a head reservoir filled with frog's ringer solution. (Appendix) bulbed with oxygen and allow to perfusion at rate of 30/40 drops/ minute. One branch of Aorta lighted and the other branch is connulated with polythene canule to obtain cardiac output. The heart is perfused through the head reservoir ensuring a constant pressure through the experiment. A fine wire hook is connected to a proper magnified starting leveler by a thread and it is passed through the apex of the ventricle. The effects of the drugs is on contractibility of the heart and output is recorded chemo graphically and the

cardiac rate is recorded with the help of stop watch. The effect of the drug on hypodynamic heart studied by using ½ Ca^{++} Ringer's solution.

The nature of action of the unknown drugs on frog's heart is determined at different dose into the heart through tubing. It is unreported glycoside and isolated by the charcoal onto method (Peach & Tracy 1955). The compound is studied in vitro for its cardio vascular action.

Seat of Action:

1. Inject 0.5ml of acctyl choline 0.1% and then injected the test drugs it produce further depression indicate the unknown drug has a sympathomimetic drug and absence of the effect indicate that it has parasympatholytic drug.
2. Inject priscoline (0.5%) 0.5ml to block adrenergic receptors and again inject the unknown drug, if stimulation produce direct action drug.
3. Inject 0.5ml atropine 0.5% repeat the unknown drug if depressed heat returns to normal after atropine there is no further depression of beats after the unknown drug. It indicates the drug is parasympathomimetic drugs. Fig. 2.1
4. If the injection of atropine has no effect on the depressed heart and injection of the unknown further depresses the heart beat. The later is a direct acting drug (Suxcena P.N. 1979) before the administration of the drug (Strace 1931).

The following is a typical record of heart beats on a fast moving drum as shown in plate.

The various parameters of the heart beat are as follows:

1. Amplitude of contraction, the total height AB represents emplitude. Increased AB indicates a mere forceful contraction while decreased AB on Feeble contraction.
2. Tone: The base B rises up with increase in tone and falls with decrease in tone.
3. In tachycardea the rate of contraction increases and it has shown in the contraction and lead coming close together. But in bradycardea, the rate has cleansed and the contraction of the lead become further apart.

Observation of Cardiac Glycoside Isolated from P-bicalyculata Nees on Frog's Heart: in Vitro Study

The atropine has given in the dose of I nμ as a test drug to the frog heart in vitro. It

has shown that it depresses the heart rate the test drug 2 nμ the depressed more as it shows plat 2nd but soon after few second it become normal. The isolated compound is tasted in the dose 1 nμ it decreases the force of contraction and cardiac output soon after the heart became normal. When the actual calculated dose of 2 nμ given it blocks the heart completely and after 4 seconds the heart became normal as plate and experiment shows. It shows the good and forceful power of contraction of the heart.

Discussion:-

The Isolated crystals are hydrolyzed and after the hydrolysis it is tested for the presence of sugar and test of Herbermans's buchard reaching, the presence of this reaction is confirmed to the cardiac glycoside. It has direct effect on the heart . The preliminary test posses good effect on the heart .

The isolated compound is tested in the dose of 1 nμ and 2 nμ on frog's heart. It decreases the force of contraction and cardiac output at 1 nμ and the heart became normal soon after. The actual calculated dose 2 nμ is given which has blocked heart completely with persist cardiac output and after 4 second the heart became normal plate 3. How ever it has shown the direct acting of drug on the heart muscle. The type of mechanism of action of the test compound may be viewed as in the dose of 2 nμ. The atropine depresses the heart but having no prominent action. The structural formula and further study is going on.

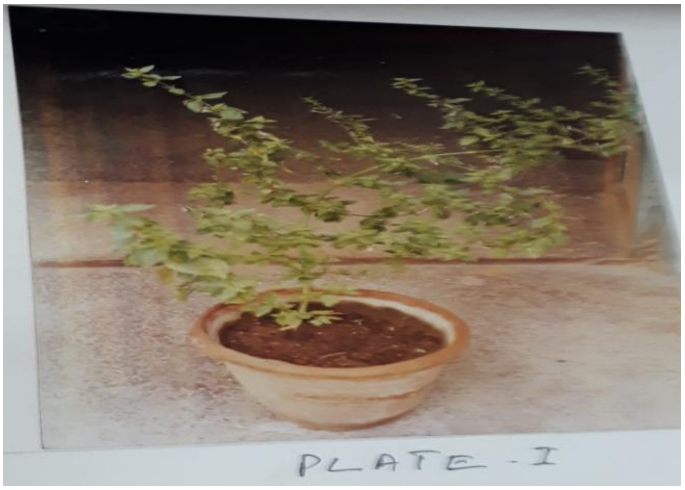

Fig. 1 – Peristrophe bicalyculata Nees

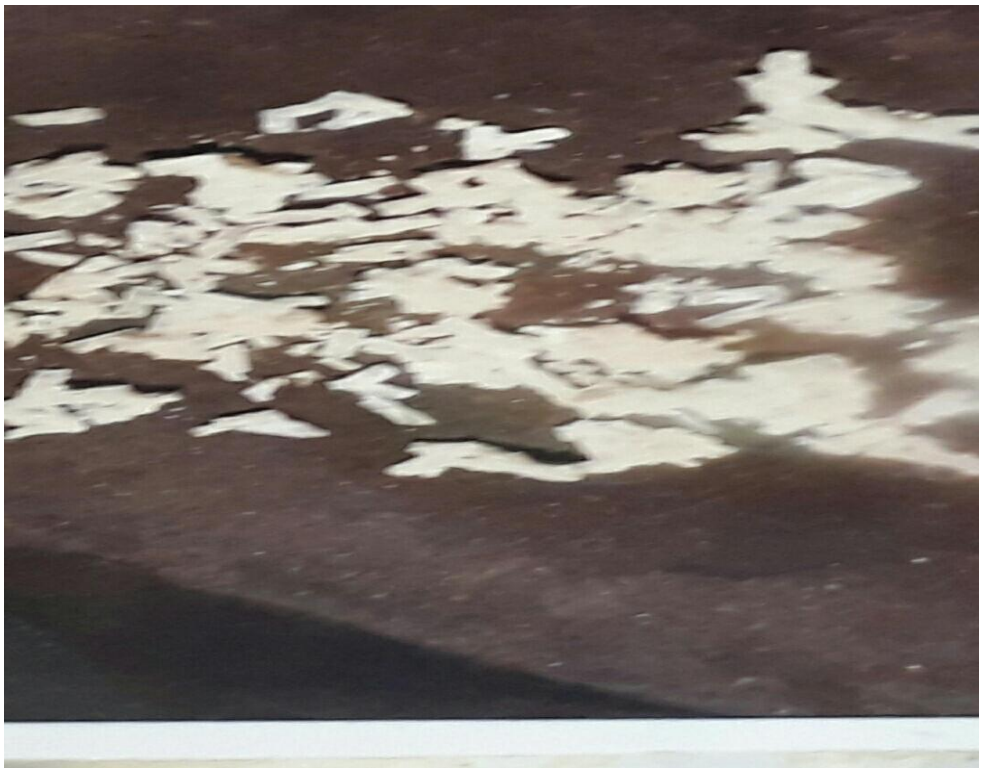

Fig. 1.2 Isolated Cardiac Glycoside

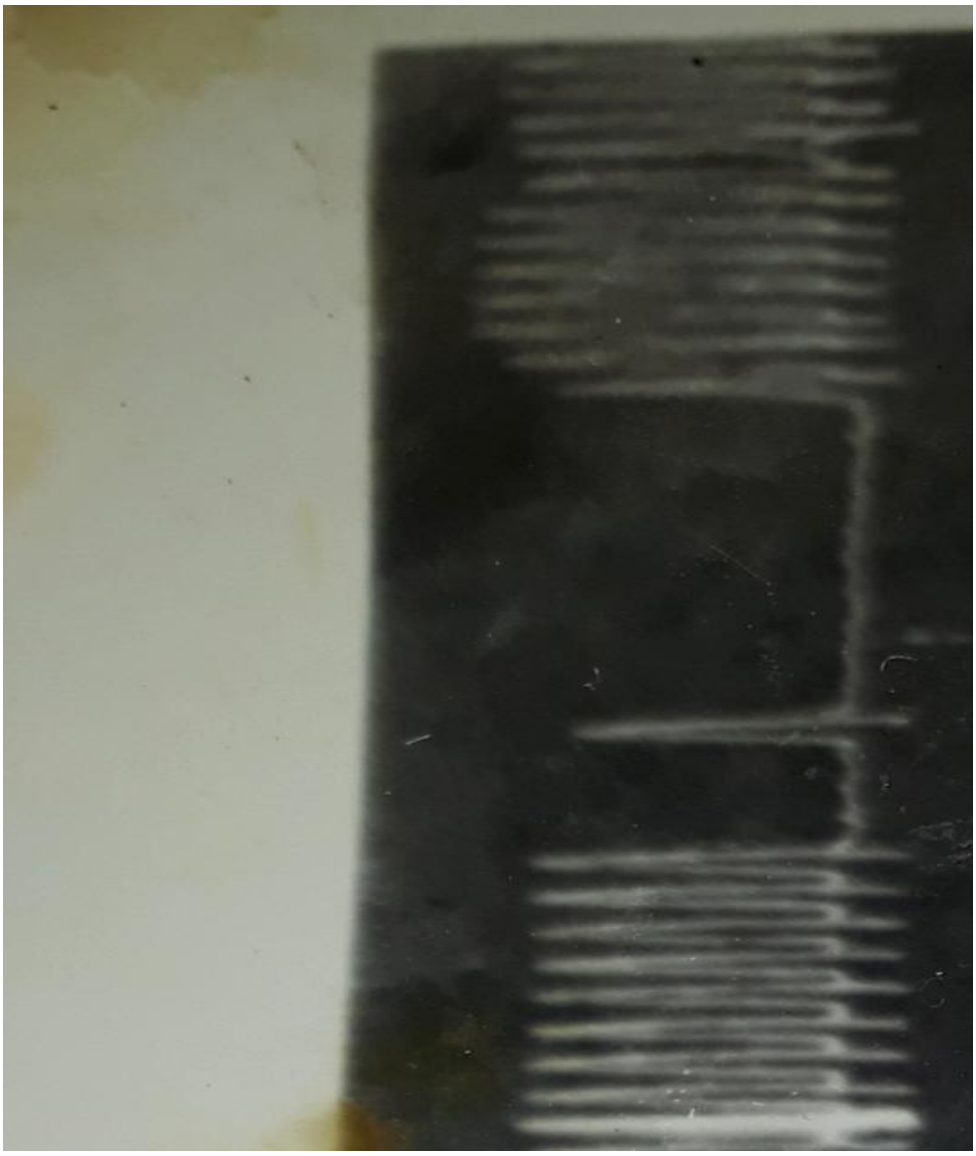

***Fig. 2.1 – Dose 1* mμ on Frog Heart**

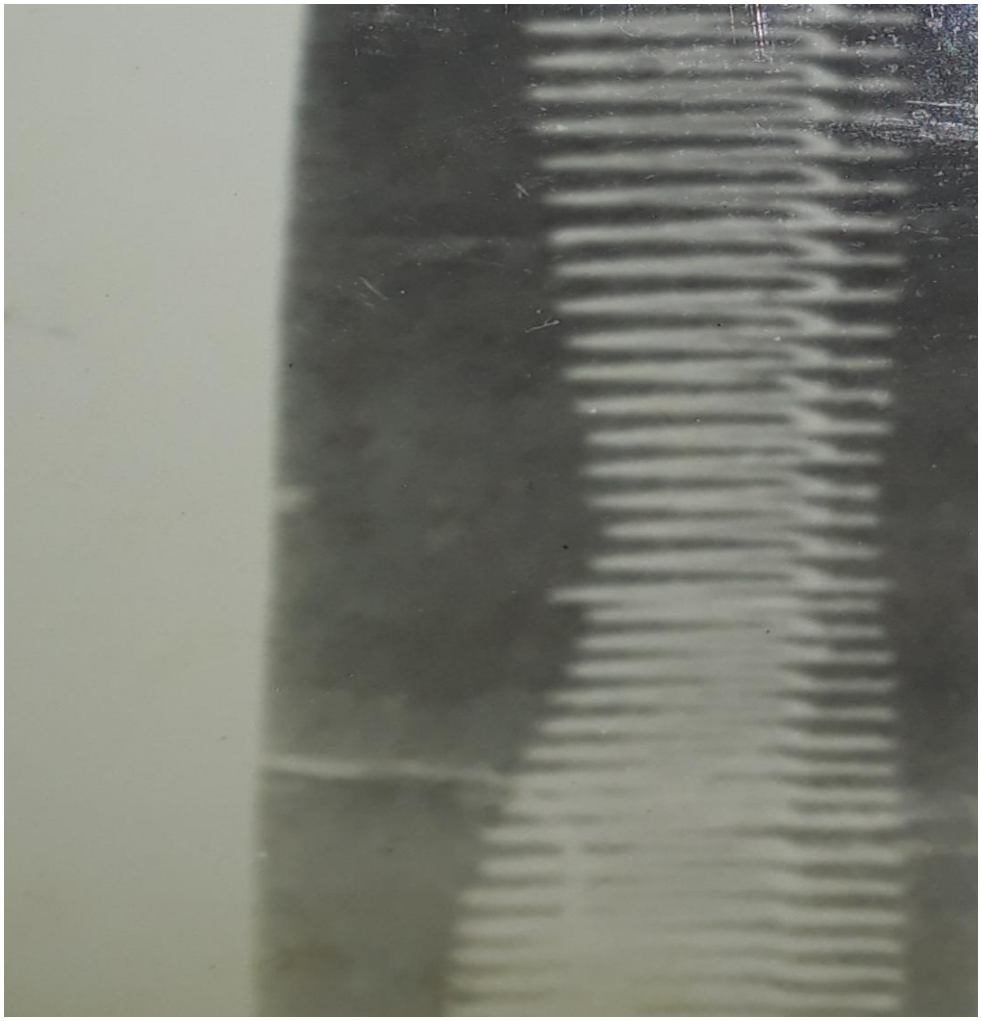

Fig. Dose 1 nμ Atropine

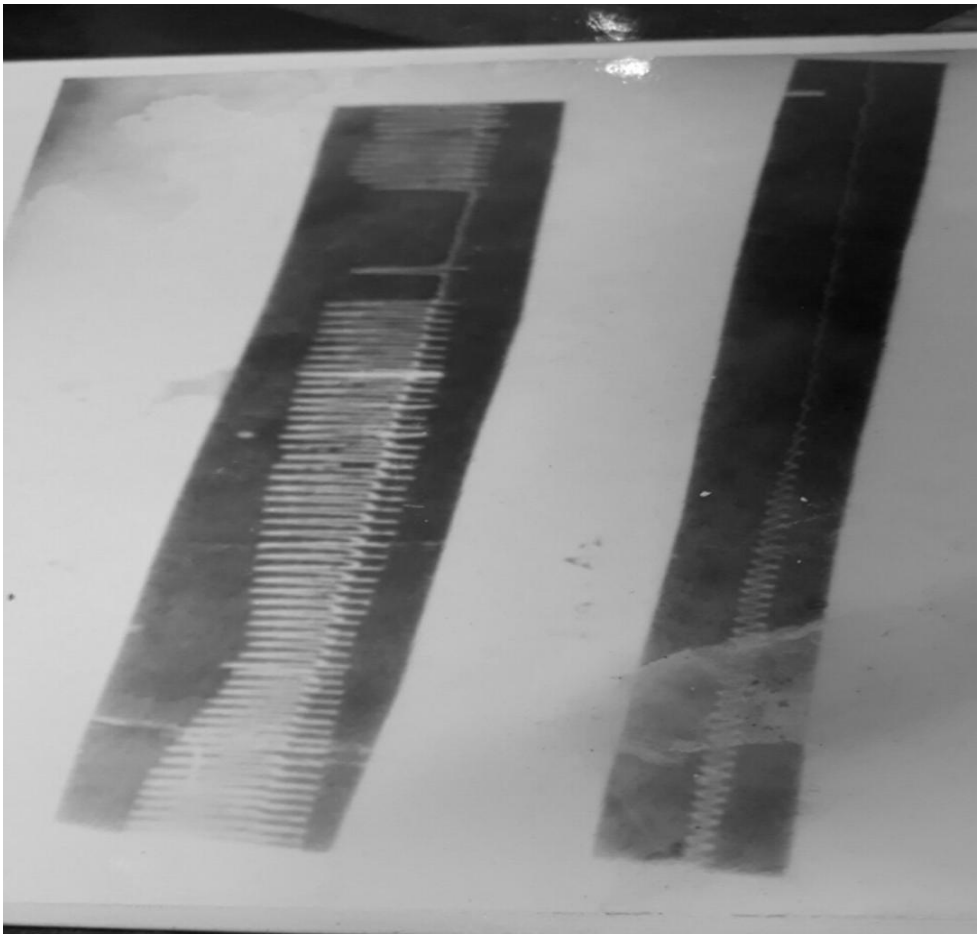

***Fig. Dose 2* nμ on Frog Heart**

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