

Certificate of Achievement

THIS CERTIFICATE IS PROUDLY
PRESENTED FOR HONOURABLE ACHIEVEMENT TO

Practical File of Exp Pharmacology II
M.P. 2 - 2057

AWARDED THIS DAY OF _____



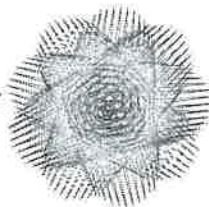
Name of the Student *Reena sharma*

Roll no. _____ Class _____

Examination Centre _____


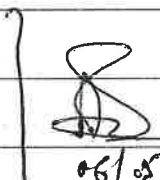
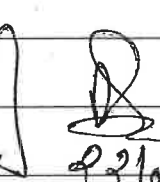
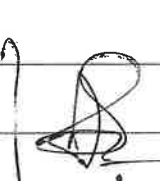
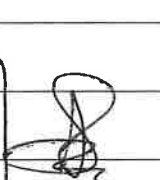
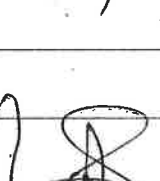
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S.No	Experiment Description	Page No.	Experiment Date	Submission Date	Remarks
01	To study and record the DRC of acetylcholine using frog rectus abdominus muscles.	01-03	10/04/24	22/04/24	 06/05/24
02	To study the bioassay of serotonin by interpolation method using rat fundus.	04-06	22/04/24	06/5/24	 06/5/24
03	To study the bioassay of adrenaline by interpolation method using rabbit jejunum.	07-09	6/05/24	8/5/24	 22/05/24
04	To study the bioassay of oxytocin by interpolation method using rat uterus.	10-12	8/5/24	20/5/24	 22/05/24
05	To study the bioassay of histamine by three point method using guinea pig ileum.	13-15	20/5/24	22/5/24	 22/05/24
06	To study the median LD_{50} of a given drug & demonstrate the acute	16-18	22/05/24	3/6/24	 19/06/24



S.No	Experiment Description	Page No.	Experiment Date	Submission Date	Remarks
07	toxicity (parenteral & oral route) of the drug as per OECD guideline.	19-20	3/6/24	10/06/24	19/06/24
08	To study the acute toxicity in different routes of administration as per OECD guidelines.	20-22	10/06/24	19/06/24	19/06/24
09	To calculate the pD ₂ value for atropine using acetylcholine as agonist template using guinea pig ileum preparation.	23-26	19/06/24	8/07/24	08/07/24
10.	To study drug antagonist using isolated and perfused frog heart.	27-34	8/07/24	8/07/24	08/07/24



Experiment - I

Aim :- To study and record the DRC of acetylcholine using frog rectus abdominus muscles.

Reference :- x-pharmacology software.

Kulkarni S.K, Handbook of Experimental Pharmacology IIIrd edition, Vallabh Prakashan, M.K. Jain 2009.

Dr. Goyal R.K, Patel N.M, Bhatt R.V, Mehta A.A, Prakashan or M.C. Fractiallyn Pharmacology IXth edition, B.S. Shah Prakashan, 2009-2010.

Requirements :- x-pharmacology software, acetylcholine.

Theory :-

Acetylcholine is an organic chemical that functions in brain & body of many types of animals as a neurotransmitter. Its name is derived from its chemical structure. It is an ester of acetic acid & choline parts in the body that use on are affected by acetylcholine are referred to as cholinergic.

Substances that increase or decrease the overall activity of cholinergic system are called cholinergic and anticholinergics.

Acetylcholine is the neurotransmitter used at neurotransmitter junction. It is the chemical that motor neurons of nervous system release in order to activate muscles.

Procedure :-

- (1) The desktop was started and x-pharmacology software already installed was opened.
- (2) After the start-up of the software, appropriate drug that was acetylcholine was selected for recording the DRC curve.
- (3) Conc. of acetylcholine was already taken as 10mg/ml and kymograph speed as 0.12 mm/s.
- (4) After this before the selection of dose a baseline was produced.
- (5) Then, the corresponding doses were selected.
- (6) Minimum dose that was 0.02µg was selected.



and its measurable response was recorded accordingly.

- (7.) After the response, the dose was given a wash and again the baseline was produced.
- (8.) The process was repeated with diff. doses as 0.05, 0.1, 0.2, 0.4, 0.8, 1.6, 3.2, 6.4 μg .
- (9.) The dose was given a wash after each consecutive dose and also baseline was produced at each time.
- (10.) The response of each dose was recorded in geometrically increasing order and was continued until maximum response was achieved that was two consecutive doses giving equal response.

Result :-

The DRC of acetylcholine using frog rectus abdominus muscle was studied and recorded. Maximum response was achieved and the significant doses were found to be 3.2 & 6.4 μg .

Experiment-2

Aim :- To study the bioassay of Serotonin by interpolation method using rat fundus.

Reference :- x- pharmacology software.

Kulkarni S.K, Handbook of Experimental Pharmacology IIIrd edition, Vallabh Prakashan M.K. Jain 2005.

Dr. Goyal R.K, Patel N.M Bhatt R.V, Menta A.A Prakashan M.C, Practicals in pharmacology IXth edition, B.S Shah Prakashan, 2009-2010.

Requirements :- x- Pharmacology software, Serotonin.

Theory :- Serotonin or 5HT is a morphine neurotransmitter. Its biological function is complex & multifaceted modulating mood, cognition, reward, learning, memory & numerous physiological processes such as vomiting & vasoconstriction.

It is primarily found in enteric nervous system located in G.I. However it is also produced in CNS, specifically raphe nuclei located in brain, pulmonary neuroendocrine cells & taste receptor cells in the tongue.

Additionally Serotonin is stored in blood platelets & is released during agitation & vasoconstriction where it acts as an agonist to other platelets.

Procedure :-

- (1) The desktop was started and x-pharmacology software already installed was opened.
- (2) After the start-up of the software, appropriate drug that was Serotonin was selected for recording the curve.
- (3) Before the selection of dose a baseline was produced.
- (4) Then, the corresponding doses were selected.
- (5) Minimum dose that was 0.02 μ g was



selected & its measurable response was recorded accordingly.

(6) After the response, the dose was given a wash and again the baseline was produced.

(7) The process was repeated with diff. doses as 0.05, 0.1, 0.2, 0.4, 0.8, 1.6, 3.2, 6.4 mg.

(8) The dose was given a wash after each consecutive dose and also baseline was produced at each other time.

(9) The response of each dose was recorded in geometrically increasing order & was continued until maximum response was achieved that was two consecutive dose giving equal response.

Result:- Bioassay of Serotonin by interpolation method using rat fundus was studied. Maximum response was achieved and the significant doses were found to be 3.2 & 6.4 μ g.

Experiment - 3

Aim :- To study the bioassay of adrenaline by interpolation method using rabbit jejunum.

Reference :- X- Pharmacology Software.

Kulkarni S.K, Handbooks of Experimental Pharmacology, IIIrd edition. Vallabh Prakashan, M.K Jain, 2005.

Dr. Goyal R.K, Patel N.M, Bhatt R.V, Menta A.A Prabhakar M.C Practicals in Pharmacology, IXth edition, B.S Shan Prakashan 2009-2010.

Requirements :- X- Pharmacology software, adrenaline.

Adrenaline also known as epinephrine, is a hormone & medication which is involved in regulating visceral functions (eg. respiration). Adrenaline is normally produced both by adrenal gland & by a small no. of neurons in medulla oblongata.

It plays an important role in fight or flight response by increasing blood flow to muscle. It is used to treat a no. of conditions including allergic reaction, anaphylaxis, cardiac arrest and superficial bleeding. Inhaled adrenaline may be used to improve the symptoms of Group. It may also be used for asthma when other treatments are not effective.

Procedure :-

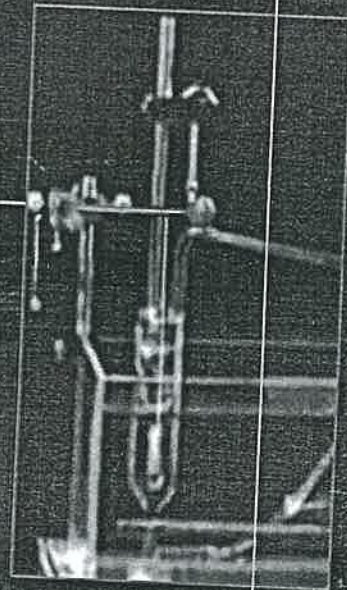
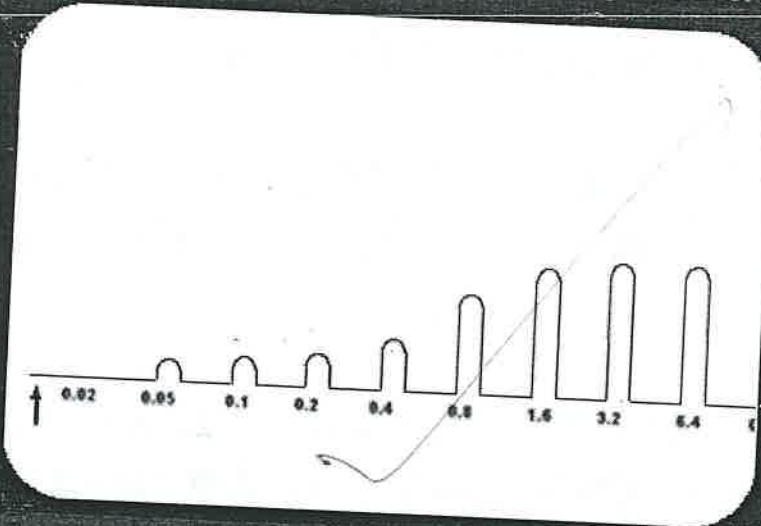
- (1) The desktop was started and x-Pharmacology Software already installed was opened.
- (2) After the start-up of the software, appropriate drug that was adrenaline was selected for recording the curve.
- (3) Before the selection of dose - a baseline was produced.
- (4) Then, the corresponding doses were selected.
- (5) Minimum dose that was 0.02ug was selected & its measurable response was recorded accordingly.

Bioassay of Oxytocin by interpolation method using Rat Uterus.

0:45

Now record the responses of Oxytocin Test solution

Oxytocin Test



Select Dose

Give Wash

Base Line

Kymograph

0.02

0.05

0.1

0.2

0.4

0.8

1.6

3.2

6.4

Dosing Schedule:

Select minimum dose first
Repeat the first dose, which gives measurable response.
Take responses of the doses in geometrically increasing order.
After recording the DRC of standard solution of Oxytocin record responses of test solution.

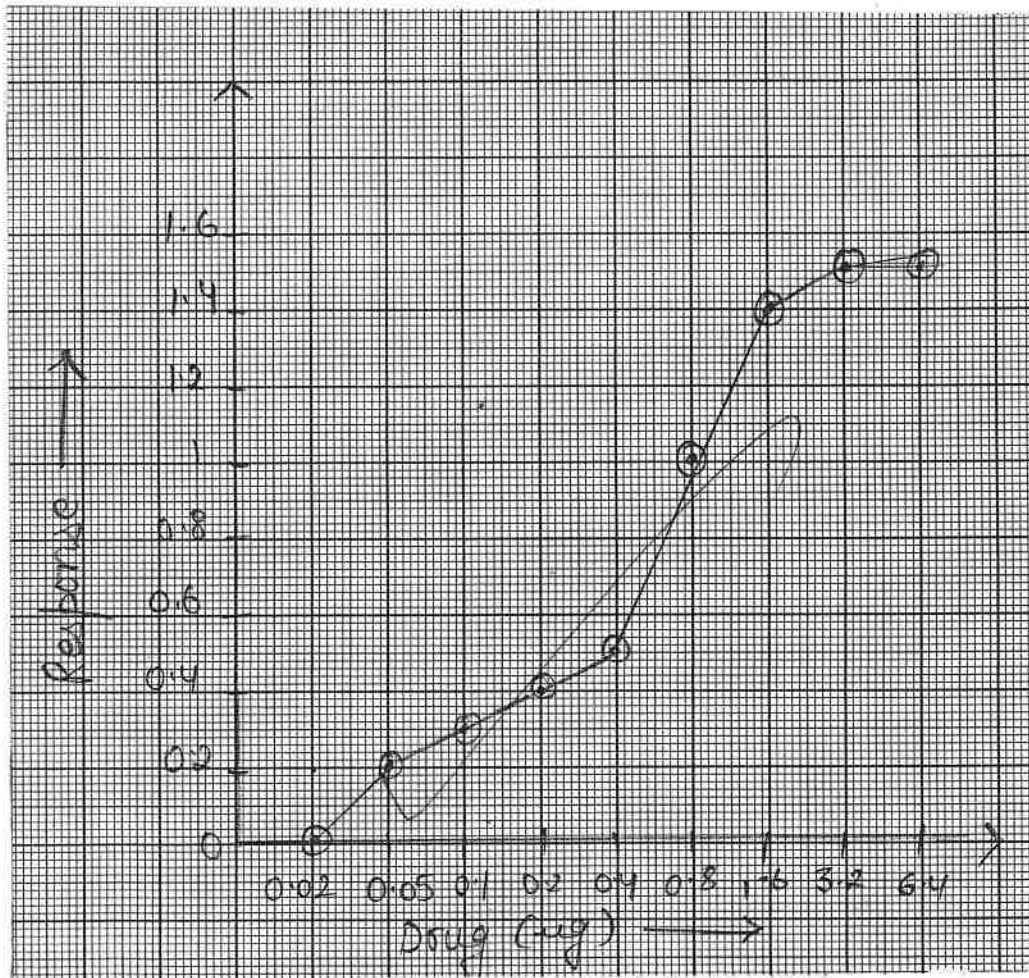
Calculate

Table

Exit

Graph 4.1 - bioassay of oxytocin by interpolation method using rat uterus.





Graph 4.2 - DRC of bioassay of oxytocin by interpolation method using rat uterus.



- (7) The process was repeated with different doses as 0.05, 0.1, 0.2, 0.4, 0.8, 1.6, 3.2, 6.4 μg .
- (8) The dose was given a wash after each consecutive dose & also baseline was produced at each time.
- (9) The response of each dose was recorded in geometrically increasing order and was continued until maximum response was achieved that was two consecutive dose giving equal response.

Result :-

Bioassay of oxytocin by interpolation method using rat uterus was studied. Maximum response was achieved & the significant doses were found to be



Experiment-5

Aim :- To study the bioassay of histamine by three point method using guinea pig ileum.

Reference :- x-pharmacology software.

Kulpani S.K. Handbook of Experimental Pharmacology IIIrd Edition Vallabh Prakashan N.K Jain, 2005

Dr. Goyal R.K, Patel N.M. Bhatt R.V Menta A.A, Prabhakar M.C Practicals. in Pharmacology IXth edition B.S. Shan Prakashan 2009-2010.

Requirements :- x-pharmacology software, Histamine

Theory :- Histamine is an organic nitrogenous compound involved in local immune response as well as regulating physiological functions in gut and acting as neurotransmitter for brain, spinal cord & uterus.

Histamine is involved in inflammatory response has a central role as mediators of itching. As part of immune response

to foreign pathogens. histamine is produced by basophils & mast cells found in nearby connective tissue. It is known to be involved in many physiological functions because of its properties.

Procedure :-

- (1) The desktop was started and x-pharmacology software already installed was opened.
- (2) After startup of the software appropriate drug that was histamine was selected for recording the curve.
- (3) Before the selection of dose a base line was produced.
- (4) Then, the corresponding doses were selected.
- (5) Minimum dose that was 0.02ug was selected & its measurable response was recorded accordingly.

Experiment-6

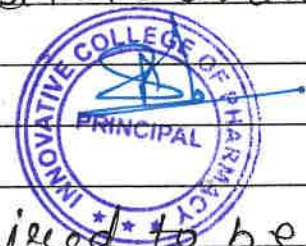
Aim:- To study the median-lethal dose (LD_{50}) of a given drug and demonstrate the acute toxicity (parenteral & oral route) of the drug as per OECD Guidelines.

Reference:- Kulkarni S.K., Handbook of Experimental Pharmacology, IIIrd edition, Vallabh Prakashan, M.K. Jain 2005.

Dr. Goyal R.K, Patel N.M; Bhatt R.V. Menta A.A prabhakar M.c, Practicals in Pharmacology IXth edition, B's shah Prakashan 2009-2010, 147-148.

Requirement - Animal- Mice (20-25 gm)
Drug - Phenobarbitone or drug under study.

Acute toxicity studies are required to be carried out for every new drug before it is to be considered for any "human use" as per schedule "Y" (Drugs & Cosmetic use 1988) acute toxicity should be carried out



In atleast 2 species (Mice & rats) the route of administration should be same as intended for human use.

In addition, one more route of administration should be such that it ensures systemic absorption. It is also recommended that mortality should be looked for upto 72 hrs. after parenteral route of administration and 7 days after oral route of administration.

Procedures -

- (1) Mice ^{were} kept for over-night fasting.
- (2) Mice are divided into 6-8 groups of 10 mice each. The drug under study was injected by intraperitoneal route (or oral route) in different doses to different groups.
- (3) The animals were then observed for signs of toxicity and the mortality (deaths) for 2 hrs and then at 24 hrs.
- (4) At the end of 24 hrs, no. of animals dead were counted in each group.



(5) Percent death was then calculated and was transferred to probits using "Probit table".

(6) Probits were then plotted (y-axis) against log dose of drug.

Result:-

The median lethal dose (LD₅₀) of a given drug was determined and the acute toxicity of drug was studied successfully.



Experiment - 7

Aim:- To study the acute toxicity in different routes of administration as per OECD guidelines.

Reference :-

Kulkarni S.K "Handbook of Experimental Pharmacology, IIIrd edition, Vallabh Prakashan, M.K Jain, 2005.

Dr. Goyal R.K, Patel N.M., Bhatt R.V, Mehta A.A, Prabhakar M.C, Practicals in pharmacology IXth edition, B.S Shah Prakashan 2009-2010 pg.148-150

Requirements:- OECD Guidelines



Theory :-

Acute toxicity studies gives an idea of toxic effects of a drug after single dose administration. To get the information on toxic effects of a drug after repeated administration, sub-acute toxicity studies are performed.

In this type of study, the drug is administered in 3 types different doses by the

Same route is intended for human use.

The following routes are commonly used -
Oral, Dermal, Inhalation, Intravenous
Intraperitoneal.

(i) OECD 407 - test substance administered once daily by oral route, for 28 days. Animals observed closely each day for sign of toxicity.

(ii) OECD 420 - Test substance administered ~~orally~~ dermally for 28 days. Observed closely each day for sign of toxicity.

(iii) OECD 412 - test substance ~~or~~ inhalation administered for a defined period. Observed closely, each day for sign of toxicity.

Parameters considered for measuring toxic effects :-

The parameters observed during course of treatment & after the completion of treatment can be divide into following graphs :-

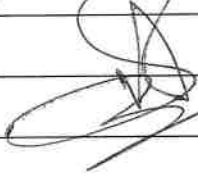
- (1) Mortality & the clinical signs symptoms at the time of death.
 - (2) Change in body weight (loss of body weight by 10% as compared to controls) food & water consumption.
 - (3) Urine - analysis, ^{per vol.} pH, specific gravity, protein content, content of glucose; blood, bile pigments, ketone bodies etc.
 - (4) Haematology parameters like RBCs, WBCs, platelets etc.
 - (5) Clinical biochemistry of blood per glucose, proteins, lipids, creatinine, urea, alkaline phosphatase, glutamic pyruvic transferase etc.
 - (6) Pathophysiology examination of eye, heart etc.
 - (7) Organ weight, their gross pathology & histopathology.
- Any change in parameters should be repeated. while submitting the results



to drug content authorities along with interpretation & comments.

Result:-

The acute toxicity in different routes of administration as per OTC guidelines were studied successfully.



Experiment - 8

Aim :- To study the ADR reporting on design of ADR monitoring protocol.

Reference :- Kulkarni S.K, "Handbook of Experimental pharmacology, IIIrd edition, Vallabh Prakashan, M.K. Jain, 2005.

Dr. Goyal R.K, Patel N.M, Bhatt R.V, Mehta A.A, Prabhakar M.C, Practicals in pharmacology, IXth edition, B.S. Shah Prakashan, 2009-2010.

Requirements-ADR monitoring protocols

Theory :-

Adverse drug reaction reporting tools or monitoring in a process of continuously monitoring of undesirable effect suspected to be associated with use of medical products.

ADR reporting covers all pharmaceutical products, biological, herbal drugs and medical devices.





SUSPECTED ADVERSE DRUG REACTION REPORTING FORM

For VOLUNTARY reporting of Adverse Drug Reactions by Healthcare Professionals

INDIAN PHARMACOPOEIA COMMISSION (National Coordination Centre-Pharmacovigilance Programme of India) Ministry of Health & Family Welfare, Government of India Sector-23, Raj Nagar, Ghaziabad-201002			FOR AMC/NCC USE ONLY		
Report Type <input type="checkbox"/> Initial <input type="checkbox"/> Follow up			AMC Report No. : _____		
A. PATIENT INFORMATION			Worldwide Unique No. : _____		
1. Patient Initials _____	2. Age at time of Event or Date of Birth _____	3. M <input type="checkbox"/> F <input type="checkbox"/> Other <input type="checkbox"/>	12. Relevant tests/ laboratory data with dates _____		
		4. Weight _____ Kgs			
B. SUSPECTED ADVERSE REACTION			13. Relevant medical/ medication history (e.g. pregnancy, smoking, alcohol use, hepatic/renal)		
5. Date of reaction started (dd/mm/yyyy)			14. Seriousness of the reaction: No <input type="checkbox"/> if Yes <input type="checkbox"/> <input type="checkbox"/> Death (dd/mm/yyyy) <input type="checkbox"/> Congenital <input type="checkbox"/> Life threatening <input type="checkbox"/> Required Preventive <input type="checkbox"/> Hospitalization/Prolonged <input type="checkbox"/> Impaired <input type="checkbox"/> Disability <input type="checkbox"/> Other (specify) _____		
6. Date of recovery (dd/mm/yyyy)					
7. Describe reaction or problem					
15. Outcomes <input type="checkbox"/> Recovered <input type="checkbox"/> Recovering <input type="checkbox"/> Fatal <input type="checkbox"/> Recovered with sequelae					

C. SUSPECTED MEDICATION(S)

S.No	8. Name (Brand/Generic)	Manufacturer (if known)	Batch No. / Lot No.	Exp. Date (if known)	Dose used	Route used	Frequency (OD, BD etc.)	Therapy dates		Indication
								Date started	Date stopped	
i										
ii										
iii										
iv										

S.No as per C	9. Action Taken (please tick)						10. Reaction reappeared after reintroduction (please tick)		
	Drug withdrawn	Dose increased	Dose reduced	Dose not changed	Not applicable	Unknown	Yes	No	Effect unknown
i									
ii									
iii									
iv									

11. Concomitant medical product including self-medication and herbal remedies with therapy dates (Exclude those used to treat the reaction)

S.No	Name (Brand/Generic)	Dose used	Route used	Frequency (OD, BD, etc.)	Therapy dates	
					Date started	Date stopped
i						
ii						
iii						



Healthcare Professionals → Peripheral Centres
 ↓
 NPC/CDSIO ← Zonal Centres ← Regional Centres
 ↳ WHO/UMC

Objectives of ADR reporting :-

- (i) To detect the nature & frequency of ADRs.
- (ii) To assist the drug regulatory authority, Public Health Programme, Scientists & Consumer Society to minimize ADRs.
- (iii) Providing updated Drug Safety Information to health care professionals.
- (iv) Dissemination of information by designing proper education program to consumers.
- (v) To identify risk of factors that may predispose, induce or influence the development, severity and incidence of ADRs.

Information required for ADRs monitoring :-
 Patient information - ADRs description
 Information related to suspected drug;
 Information on management



National Coordination Centre
Pharmacovigilance Programme of India
Ministry of Health & Family Welfare
Government of India
Sector-23, Raj Nagar, Ghaziabad-201002
Tel.: 0120-2783400, 2783401, 2783392
Fax: 0120-2783311
www.ipc.nic.in

**Pharmacovigilance
Programme of India for
Assuring Drug Safety**

ADVICE ABOUT REPORTING

A. What to report

- Report serious adverse drug reactions. A reaction is serious when the patient outcome is:
 - Death
 - Life-threatening
 - Hospitalization (initial or prolonged)
 - Disability (significant, persistent or permanent)
 - Congenital anomaly
 - Required intervention to prevent permanent impairment or damage
- Report non-serious, known or unknown, frequent or rare adverse drug reactions due to Medicines, Vaccines and Herbal products.

B. Who can report

- All healthcare professionals (Clinicians, Dentists, Pharmacists and Nurses) can report adverse drug reactions

C. Where to report

- Duly filled Suspected Adverse Drug Reaction Reporting Form can be send to the nearest Adverse Drug Reaction Monitoring Centre (AMC) or directly to the National Coordination Centre (NCC).
- Call on Helpline (Toll Free) 1800 180 3024 to report ADRs.
- Or can directly mail this filled form to pvpi@ipcindia.net or pvpi.ipcindia@gmail.com
- A list of nationwide AMCs is available at:
<http://www.ipc.gov.in>, http://www.ipc.gov.in/PvPI/pv_home.html

D. What happens to the submitted information

- Information provided in this form is handled in strict confidence. The causality assessment is carried out at AMCs by using WHO-UMC scale. The analyzed forms are forwarded to the NCC through ADR database. Finally the data is analyzed and forwarded to the Global Pharmacovigilance Database managed by WHO Uppsala Monitoring Centre in Sweden.
- The reports are periodically reviewed by the NCC-PvPI. The information generated on the basis of these reports helps in continuous assessment of the benefit-risk ratio of medicines.
- The information is submitted to the Steering committee of PvPI constituted by the Ministry of Health & Family Welfare. The Committee is entrusted with the responsibility to review the data and suggest any interventions that may be required.

E. Mandatory field for suspected ADR reporting form

- Patient initials, age at onset of reaction, reaction term(s), date of onset of reaction, suspected medication(s) and reporter information.



Information about reporter.

What to Report :- ?

- (i) All ADRs as a result of prescribing & non-prescribing medicinal products.
- (ii) All suspected ADRs regardless of product information provided by company.
- (iii) Unexpected reaction with product regardless of product information provided by company.
- (iv) A serious reaction whether expected or not.
- (v) All suspected ADRs associated with drug-drug, drug-food or drug-food & supplement interactions.
- (vi) ADRs occurring from overdose or medication error.
- (vii) unusual lack of efficacy or when suspected pharmaceutical defects are observed.

Who should report

Health care professionals, providers, manufacturers of product, Health care centres

When to report

ADR should be reported as soon as possible



(ii) Delay in reporting make report inaccurate & unreliable.

How to report :-

(i) Should be on standardized ADR reporting form.

(ii) Duly filled ADR in form when ADR is encountered.

(iii) use separate form for each patient.

(iv) Complete ADR form is then returned to AMC/ACC.

(v) Any follow up information for ADR case can be sent on another ADR form or communicated by fax etc.

(vi) follow up reports should be identifiable and following should be indicated on report.

- follow-up information
- Date of original report
- Patient identify.

Result :-

ADR monitoring / reporting, design of ADR monitoring protocol was studied ~~fully~~ fully



Experiment - 09

Aims - To calculate the P_{A_2} value for atropine using Acetylcholine as agonist employing guinea pig ileum preparations.

References:- Kulkarni S.K, Handbook of Experimental Pharmacology, IIIrd edition, Vallabh Prakashan, M.K Jain, 2005, 95-97

Dr. Goyal R.K, Patel N.M, Bhatt R.V, Mehta A.A, Parbhakar M.C, Practicals in Pharmacology IXth edition, B.S Shah Prakashan 2009-2010

Requirements:- Animals - Guinea pig (400-600g overnight fasted) :- Drugs - Acetylcholine stock solution (1mg/ml) Atropine stock sol (1mg/ml) physiological solution - Tylenol.

Theory - Principle :-
 P_{A_2} value is calculated to compare the potency of antagonists acting on same reaction. It is defined as negative logarithm of molar concentration of antagonist required to reduce the effect of multiple dose (x) of agonist to that of single dose in absence of antagonist. Fuzer

Tissue	Atropine - Acetylcholine	Mepyramine → histamine
Guinea pig ileum	8.8	9.3

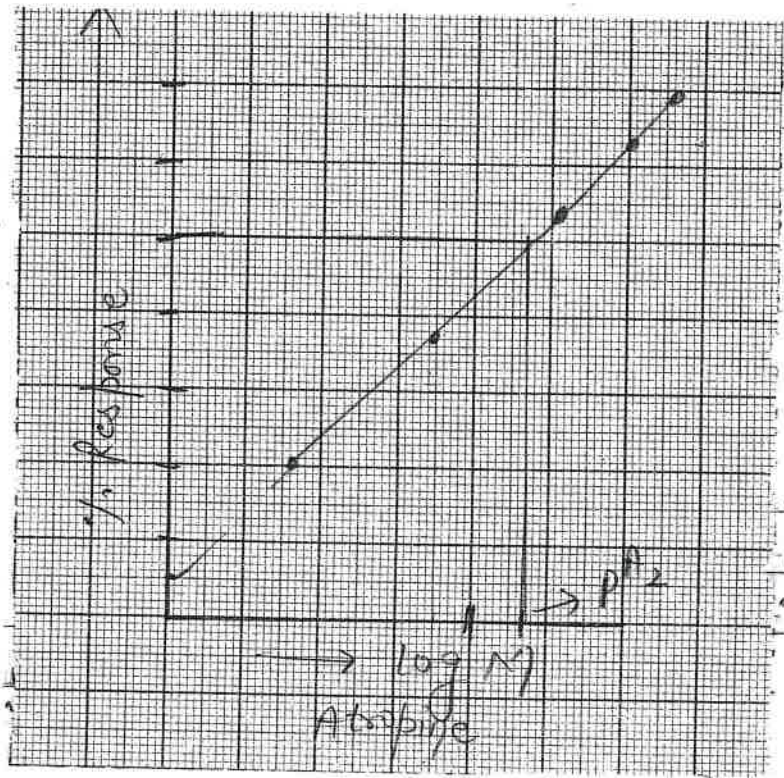
Table 1.1 - pA₂ value of Atropine & Mepyramine
in Guinea pig ileum



pA_x value, more potent is antagonist the determination of pA_2 ($x-2$) & pA_0 values have wider applications. (22/10)

Procedure :-

- (1) The guinea pig was sacrificed by blow on head & carotid bleeding.
- (2) The abdomen was cut and opened. The caecum was lifted to trace the ileocaecal junction. few centimeter long of ileal portion was cut and removed and immediately placed in watch glass containing Tyrode solution. trimmed the mesentery and with gentle care cleaned the contents of ileum by pushing Tyrode solⁿ into lumen of ileum. utmost care was taken to avoid any damage to gut muscle. The ileum was cut into small segments of 2-3 cm long.
- (3) Took one piece of ileum of 2-3 cm long & tied that thread to top & the bottom end without closing lumen, mounted the tissue in organ bath containing tyrode solⁿ maintained at $32-35^\circ\text{C}$ & bubbled with O_2 / air. A tension of 0.5g was applied and tissue was allowed to equilibrate for 30 mins. before adding drugs to organ bath.

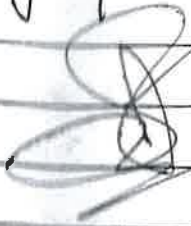


- (4) Recorded Concentration dependent Concentration due to Acetylcholine until a peak response was obtained.
- (5) Selected 2 doses bearing 1:2 dose ratio & eliciting submaximal response (A, 2A) for pA_2 determination.
- (6) Standardized the tissue with selected dose of Acetylcholine.
- (7) Record the Concentration due to double dose of Acetylcholine (2A) in presence of varying Concentration (B, B₂, B₃) of Atropine.
- (8) Considered the response due to double the dose of Acetylcholine (2A) i.e. before adding Atropine, as 100% response. Determined the corresponding (%) response to this dose of Ach (2A) in presence of varying Concentration of Atropine.
- (9) Plotted a graph representing negative log of molar Concentration of Atropine employed along x-axis & response along y-axis.

(10) Readed out the pA_2 value for atropine from graph directly. It corresponded to % response obtained with half the dose of Ach.

Result :-

The pA_2 value of atropine against acetylcholine was found to be 8.76 in guinea pig ileum.



Experiment - 10

Aim :- To Study Drug antagonist using isolated and perfused frog heart.

Reference :- Kulkarni S.K, Handbook of Experimental pharmacology, IIIrd edition, Vallabh Prakashan M.K Jain 2005.

Dr. Goyal R.K Patel N.M, Bhatt R.V Menta A.A, Prabha par M.C Practicals in Pharmacology, IXth edition B.S Shah Prakashan, 2009-2010.

α - pharmacology Software

Requirements :- α - pharmacology Software, Drugs - KCl, $CaCl_2$, Acetylcholine, Adrenaline, atropine, Propranolol.

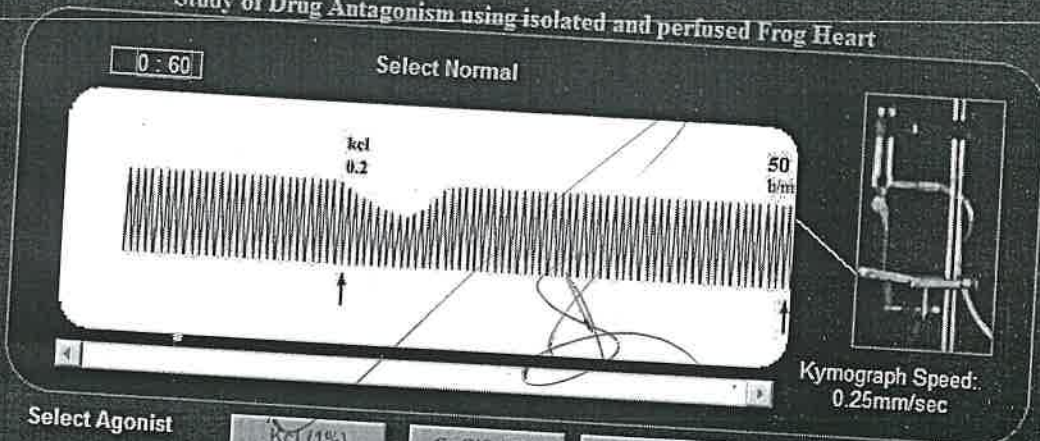
Theory :- KCl is a metal halide salt composed of potassium and chlorine. It is used to prevent or treat low blood levels of potassium (hypokalaemia)

$CaCl_2$ is an inorganic compound, a salt with the high stability in water. It is indicated in immediate treatment of



X-COLOGY EXPERIMENTAL PHARMACOLOGY

Study of Drug Antagonism using isolated and perfused Frog Heart



Select Agonist

KCl (1%)

CaCl₂ (1%)

Acetylcholine

Adrenaline

Select Antagonist

Atropine

Propranolol

Normal

Stop

Time Cycle:

For each dose,
Record the normal response for 30 seconds.
Record the response to a dose of drug for 60 seconds.
Repeat this cycle through out the experiment.

Table

Exit

Dosing Schedule:

Take responses of the Agonists as in earlier experiments.
Take responses of both antagonists.
Select any one antagonist and take its response. Before the antagonist is washed out of cannula (i.e. before 60 seconds are over) add the selected agonist. Take response of agonist for 60 seconds.
Study effect of prior addition of antagonist on the responses of all the agonists.



hypocalcemic tetany.

Ach is organic chemical that functions in brain as neurotransmitter. It mainly acts on muscular system by activating muscle contraction after release.

Adrenaline also known as epinephrine is a hormone & medication which is involved in regulating visceral functions.

Atropine is a tropane alkaloid used to treat symptoms of low heart rate (bradycardia) reduce salivation & bronchial secretions before surgery.

propranolol is β -adrenergic receptor antagonist used to treat hypertension.

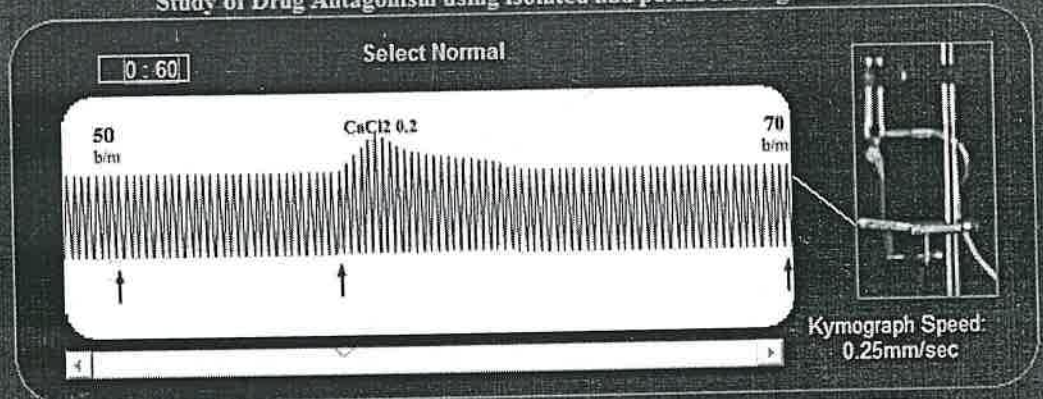
Procedure :-

(1) The desktop was started and x-pharmacology software already installed was opened.

(2) After the start-up of software, any one agonist was selected.

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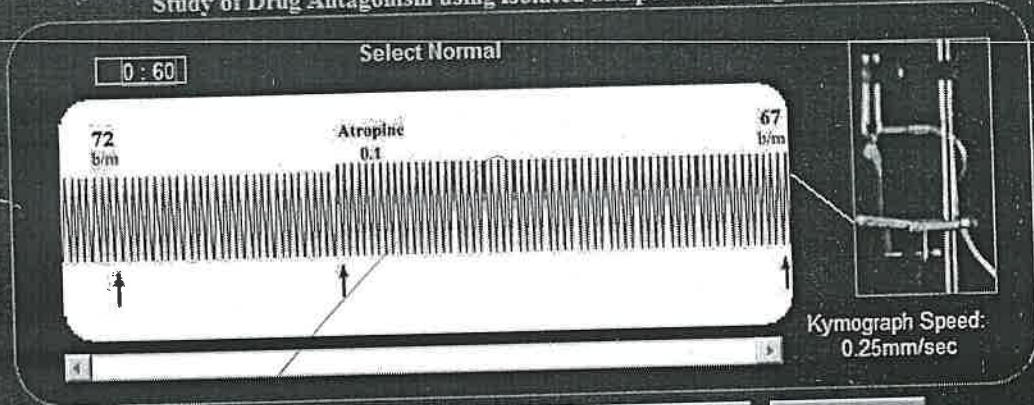
- (3) Kymograph speed was taken as 0.25 mm/sec.
- (4) Amongst all the agonist firstly KCl (1%) was selected and the normal response was recorded for 30 seconds for each dose.
- (5) The response to a dose of drug was recorded for 60 seconds and then it was selected to normal.
- (6) The cycle was repeated with diff. agonists like $CaCl_2$ (1%), Acetylcholine & adrenaline.
- (7) After this, response of antagonist were recorded.
- (8) first antagonist atropine was selected and its response was recorded.
- (9) Before the antagonist was washed out of cannula (i.e. before 60 seconds were over) added the selected agonist & response of agonist for 60 second was recorded.
- (10) Process was repeated with another antagonist propranolol.
- (11) The effect of prior adding of antagonist on the response of all antagonists were studied.

Result :-

Drug antagonist using isolated and perfused frog heart was studied successfully.



Study of Drug Antagonism using isolated and perfused Frog Heart



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Adrenaline

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Atropine

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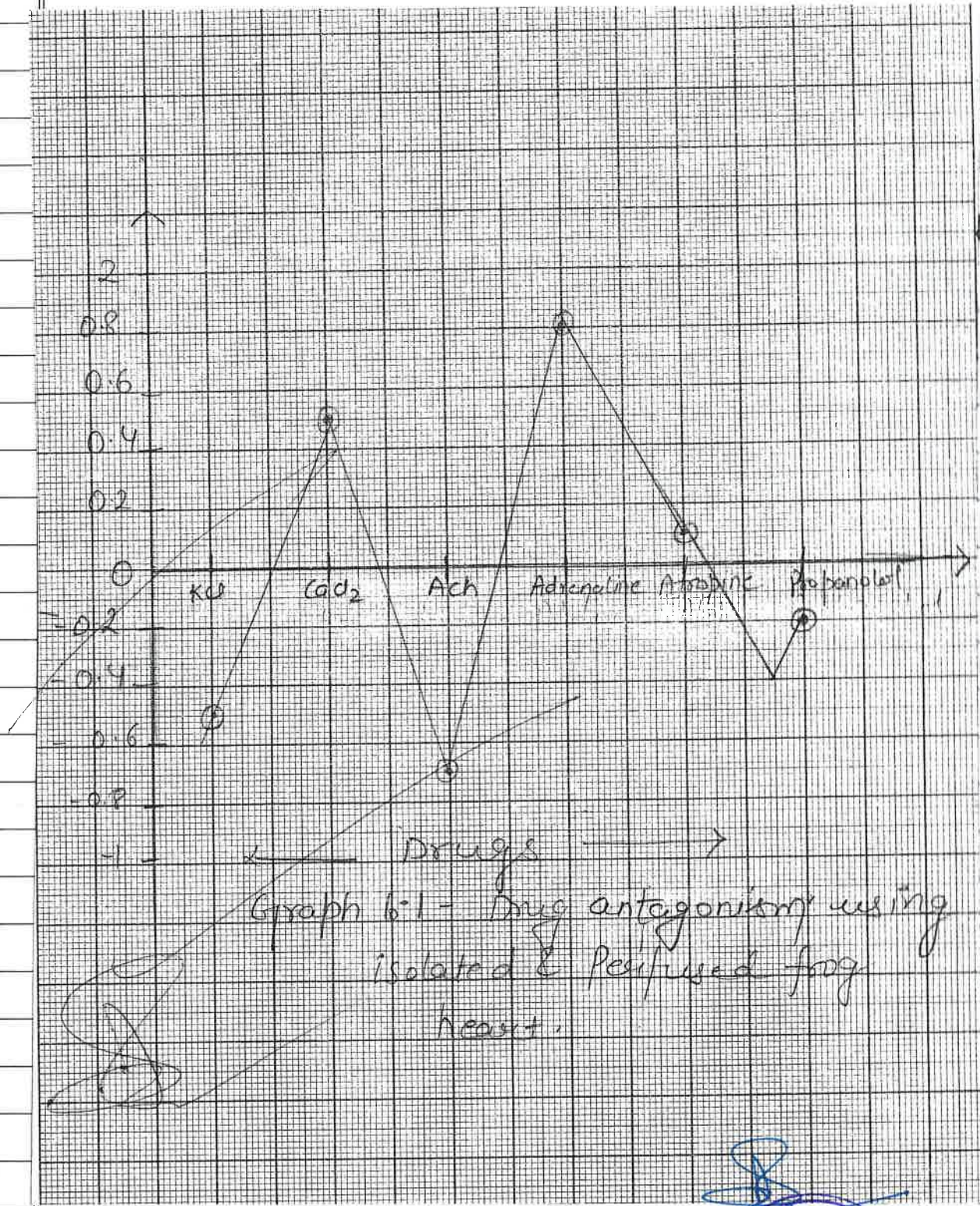
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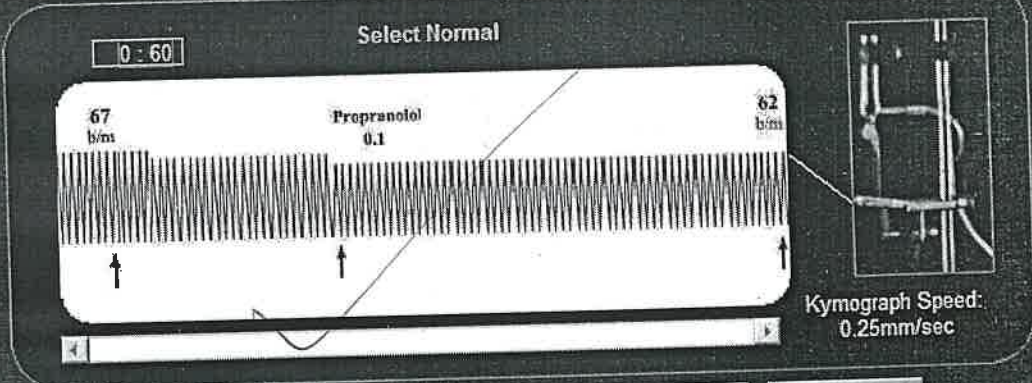




Graph 6.1 - Drug antagonism using isolated & perfused frog heart.



Study of Drug Antagonism using isolated and perfused Frog Heart



Select Agonist

Kcl (1%)	CaCl ₂ (1%)	Acetylcholine	Adrenaline
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Select Antagonist

Atropine	Propranolol	Normal	Stop
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Time Cycle:

Table	Exit
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