A TOXICITY STUDY ON

"PAVALA PARPAM"

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Government Siddha Medical College

Palayamkottai – 627 002

OCTOBER – 2019

GOVT. SIDDHA MEDICAL COLLEGE, PALAYAMKOTTAI

DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled "**A Toxicity Study on PAVALA PARPAM**" is a bonafide and genuine research work carried out by me under the guidance of **Dr. M. P. ABDUL KADER JEYLANI**, **M.D(s).,** Professor, Post Graduate Department of Nanju Noolum Maruthuva Neethi Noolum, Govt.Siddha Medical College, Palayamkottai, and the dissertation has not formed the basis for the award of any Degree, Diploma, Fellowship or other similar title.

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CERTIFICATE OF MINERAL AUTHENTICITY

Certified the following the Mineral (Thathu) drug used in Siddha formulation (Internal) **"PAVAZHA PARPAM"** for **"EELAI & SHAYAM"** taken up for Post-Graduation Dissertation Studies by **Dr.G.KANNAN,** PG Scholar of MD Siddha, Department of Toxicology, have selected the raw drug(Minerals) and have been authenticated through Geological methods(Macro/Micro).

S.NO	DRUG	ENGLISH NAME	SCIENTIFIC NAME
1	Pavazham	Red Coral	Corallium

Station : Palayamkottai

Date:

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CERTIFICATE

Title of the project	:	A TOXICITY STUDY ON PAVALA PARPAM
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ABBREVIATIONS

PP	PAVALA PARPAM
No.	Number
Mg	Milligram
Kg	Kilogram
LD_{50}	Lethal $Dose_{50}$
ED_{50}	Effective $Dose_{50}$
p.o	peros
ML	Milliliter
%	percentage
R&D	Research and Development
EDTA	Ethylene Diamine Tetra Acetic Acid
M	Male
g%	Gram percentage
g	Gram
NOAEL	No-Observed-Adverse-Effect-Level
MLD	Minimum Lethal Dose
MTD	Maximum Tolerated Dose
OECD	Organization of Economic Co-operation and Development
CPCSEA	Committee for the Purpose of Control and Supervision of Experiments on Animals
FTIR	Fourier Transform – Infra Red Spectroscopy
SEM	Scanning Electron Microscopy
ICP-OES	Inductively Coupled Plasma Optical Emission-Spectrometry
LD	Low Dose
MD	Middle Dose
HD	High Dose
BDL	Below Detection Limit

1. INTRODUCTION

Siddha medicine, traditional system of healing that originated in South India and is considered to be one of India's oldest systems of medicine. The Siddha system is based on a combination of ancient medicinal practices and spiritual disciplines as well as alchemy and mysticism. It is thought to have developed during the Indus civilization, which flourished between 2500 and 1700 BCE. According to this theory, it came to South India when the Dravidian people (speakers of Dravidian languages), who may have been the original inhabitants of the Indus valley, migrated southward.

Siddha medicine appears as part of Tamil culture in the earliest Tamil writings (Tamil is one of the principal Dravidian languages). For example, there are references to it in Tamil shangam literature (1st–4th century CE), including mention in the Tolkappiyam ("Ancient Literature"), a treatise on grammar and poetics, and in Tirukkural ("Sacred Couplets"), a work attributed to the Tamil poet-saint Tiruvalluvar.

According to Tamil tradition, there initially were 18 siddhars (the person who has achieved some extra-ordinary powers): Nandi, Agasthiyar, Thirumular, Punnakkeesar, Pulasthiyar, Poonaikannar, Idaikkadar, Bogar, Pulikai isar, Karuvurar, Konkanavar, Kalangi, Sattainathar, Azhuganni, Agappai, Pumbatti, Theraiyar and Kudhambai, but the Agasthiyar (Agastya) was the topmost. He is regarded as the originator of the Siddha medicine and also of the Tamil language. He occupies the same position as Hippocrates in modern western medicine. In the period of Ramayana he seems to have settled in the South. Thus origin of every tradition in the South, including language and culture, is traced back to Agastya. these individuals often are portrayed as having received their knowledge of the Siddha system indirectly from the deity Shiva. Siddhars held that the object of their study was to preserve and prolong life. To do so, they believed, required humans to live according to the laws of nature. They led simple lives themselves and were unconcerned with caste, creed, colour, or nationality. They contributed not only to a system of medicine but also to the knowledge of eternity, alchemy, and Yogic living. Some believe that the siddhars travelled widely to other countries to propagate their system of medicine and enrich the sciences.

Siddhars possessed ashtama siddhi, the eight great supernatural powers. These powers may have been attained at birth (because of one's previous karma), by

chemical means, by the power of words, or through concentration. Meditation on the elements, beginning with the "gross" and ending with the "subtle," enabled the siddhars to gain mastery over the elements. Many of the ancient philosophical tenets of the Siddha system continue to be relevant to modern practitioners.

According to the Siddha system, there are five elements that exist in nature: earth, water, fire, air, and ether, all of which form the original basis of all corporeal things. It is believed that there is an intimate connection between the macrocosm of the external world and the microcosm of the corporeal being. In the human body the element of earth is present in the bone, flesh, nerves, skin, and hair; the element of water is present in bile, blood, semen, glandular secretions, and sweat; the element of fire is present in hunger, thirst, sleep, beauty, and indolence; the element of air is present in contraction, expansion, and motion; and the element of ether is present in the interstices of the stomach, heart, neck, and head.

Three of the elements—air, fire, and water—are emphasized in Siddha medicine because they are believed to form the three fundamental components that make up the human constitution. These three components—vata, pitta, and kapha (representing air, fire, and water, respectively)—are known as humours, and their inharmonious interaction produces various pathological states.

According to the theories of humoral pathology, all diseases are caused by the discordant mixture of vata, pitta, and kapha. Their proportions in the body govern a person's physical and mental disposition. The elements form the connecting link between the microcosm (the human) and the macrocosm (the world). Thus, the external air corresponds to the internal vata, the external heat corresponds to the internal pitta, and the external water corresponds to the internal kapha. Under normal circumstances, according to Siddha theory, vata occupies regions related to the pelvis and the rectum, pitta occupies regions related to the stomach and the viscera, and kapha occupies regions related to breath, the throat, and the head.

Siddhars believed vata to be self-originated and identical to divine energy. Imbalance of vata could be the root cause of all disease. Pitta was believed to represent all the characteristics of fire, such as burning, boiling, heating, and similar sensations. It was the name given to the heat contained in the liquid bile, which causes the expulsion of waste matter in the form of urine and feces, and it was believed to give sight to the eyes, beauty to the skin, and cheerfulness to the mind. Kapha was believed to supply moisture to the body and to give stability, adding to the strength of

the body by increasing the firmness of the limbs and thereby keeping them in harmony with one another. It was also thought to aid in digestion and sensation, such as by imparting taste to the tongue.

The presence and proportion of these humours within the system is indicated by the pulse, which is vital to correct diagnosis.

According to the Siddha medicine various psychological and physiological functions of the body are attributed to the combination of seven elements: first is saram (plasma) responsible for growth, development and nourishment; second is cheneer (blood) responsible for nourishing muscles, imparting colour and improving intellect; the third is oun (muscle) responsible for shape of the body; fourth is kollzuppu (fatty tissue) responsible for oil balance and lubricating joints; fifth is enbu (bone) responsible for body structure and posture and movement; sixth is moolai (nerve) responsible for strength; and the last is sukila (semen) responsible for reproduction. Like in Ayurveda, in Siddha medicine also the physiological components of the human beings are classified as Vatha (air), Pitha (fire) and Kapha (earth and water)

When the normal equilibrium of three humors (vatha, pitha and kapha) is disturbed, disease is caused. The factors, which affect this equilibrium are environment, climatic conditions, diet, physical activities, and stress. Under normal conditions, the ratio between these three humors (vatha, pitha and kapha) is 4:2:1 respectively.

According to the Siddha medicine system diet and life style play a major role not only in health but also in curing diseases. This concept of the Siddha medicine is termed as pathya and apathya, which is essentially a list of do's and dont's.

The treatment in Siddha medicine is aimed at keeping the three humors in equilibrium and maintenance of seven elements. So proper diet, medicine and a disciplined regimen of life are advised for a healthy living and to restore equilibrium of humors in diseased condition. Saint Thiruvalluvar explains four requisites of successful treatment. These are the patient, the attendant, physician and medicine. When the physician is well qualified and the other agents possess the necessary qualities, even severe diseases can be cured easily. The treatment should be commenced as early as possible after assessing the course and cause of the disease. Treatment is classified into three categories: devamaruthuvum (Divine method); manuda maruthuvum (rational method); and asura maruthuvum (surgical method). In Divine method medicines like parpam, chendooram, guru, kuligai made of mercury,

sulphur and pashanams are used. In the rational method, medicines made of herbs like churanam, kudineer, vadagam are used. In surgical method, incision, excision, heat application, blood letting, leech application are used.

According to therapies the treatments of Siddha medicines could be further categorized into following categories such as Purgative therapy, Emetic therapy, Fasting therapy, Steam therapy, Oleation therapy, Physical therapy, Solar therapy and Blood letting therapy, Yoga therapy, etc.

2. AIM AND OBJECTIVE

AIM

The aim of this study is to evaluate the toxicity study on "***PAVALA PARPAM***"

Objective

To collect and purify the raw materials based on literature evidence.

To perform compound analysis for the purified raw drug samples.

To prepare the medicine based on siddha literature evidence.

To estimate the presence of chemical constituents by performing elemental analysis.

To analyse its safety, the medicine is subjected for acute oral toxicity evaluation on rodents when the drug is given in single dose through oral administration at various dose levels.

To analyse its safety, the medicine is subjected for repeated oral dose (subacute) toxicity study evaluation on rodents when the drug is given for 28 days.

Corallium rubrum

பவளம்

3.REVIEW OF LITERATURE

SIDDHA ASPECT

பவளம்

பவளமானது நவமணிகளில் ஒன்றாகவும், கடல்படு திரவியங்கள் ஐந்தனுள் ஒன்றாகவும், குணபாடம் தாது ஜீவ வகுப்பு நூலில் குறிப்பிடப்பட்டுள்ளது.

"ஓர்க்கோலை சங்கமொளிர் பவழம் வெண்முத்த
நீர்ப்படு முப்பினோ டைந்து"

என்ற அடியால் அறியலாம்.

பவளம் சத்திபீஜமாகிய கந்தகத்திற்கும், முத்து சிவபீசமாகிய இரசத்திற்கும் நிகரென்றும், நவரத்தினங்களில் உயர்வு பெற்ற பவளமும், முத்தும் பார்வதியைப் போலும் பரமசிவனைப் போலும் பேர் பெற்று விளங்கும் என்றும் குறிப்பிடப்பட்டுள்ளது. அதனை.

"தேவி யுரத்தைநேர் செப்பலாகுந் துப்பை
யாவி யிரதநே ராகு மவுத்திகந்
தேவியரன் பீசஞ் செப்பு மிவைகளே
தேவர் முதலானோர் தேர்வரிவைகளே"

- திருமூலர்

கிடைக்கும் இடங்கள்:

இந்தியாவில் லட்சத்தீவுகள், மாலத்தீவுகள், இராமேஸ்வரம் முதலிய இடங்களில் கிடைக்கிறது.

மேலும் ஜப்பான், சுமத்ரா தீவுகளில் கிடைப்பதாக T.V.சாம்பசிவம் பிள்ளை அகராதி பாகம் - 5ல் பக்கம் 3223ல் கூறப்பட்டுள்ளது.

பவளத்தின் வேறுபெயர்கள்:

துகிர் - வாருதிக்குள் தண்டு
துப்பு - செந்தண்டுக்கொடி
வித்துருமம் - கன்னிசாத்திய மாலை பிரவாளம்

- போகர் நிகண்டு அட்டவனை பக்கம் 4

"வாருதி தண்டு வளர்த்த செந்தண்டு தான்
நேருதுக் கொடியோன் நேர்பிரவாள மாற்
தூருருவேளை சாத்திய பெண் மாலை
பாருதிநாமம் பவழத்தின் பேருமே"

- சட்டமுனி நிகண்டு 1200 பாடல் 86 பக்கம் 34

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வித்துருமம் துவரம்”

என்று நாமதீபநிகண்டில் (பாடல் 384, பக்கம் 114) பவளத்தின் வேறுபெயர்கள் குறிப்பிடப்பட்டுள்ளது.

“ஊசியாமாமூக்கு அராசிய அந்தமென்றும் பேர
உதிதமாது காமமென்றும் தும்பு என்றும் பேரு
வாசியாய் இலிங்க மென்றிதற்குப் பேரு
வசனித்தோம் வடமென்றிதற்குப் பேரு
ஆதியாம் வங்கத்தூறுசம் என்றும் பேரு
அடவான வக்கண மென்றும் பேரு
பாசியாம் பதிக்கடக மென்றும் பேரு
பாடினோம் இந்தப்படி பவழத்தின்பேரே”

-பஞ்சகாவிய நிகண்டு பக்கம் 111

பவளத்தின் தோற்றம்:

“ஆகின்ற பவளத்தின் பிறப்புக் கேளு
ஆதியான வலாகரன் அரக்கன் என்பான்
தாகின்ற ராட்சசன் தன்தசைக் கோசந்தான்
சண்டையிட்டுத் தேவரெல்லா மிருக்கும்போது
தோகின்ற கடல்தண்ணீல் போய்விழுந்து
சுத்தமான பவழமது தோற்றமாச்சு
ஏகின்ற இந்திரன் வச்சிராயுதத்தால்
ஏற்றமான மலைதன்னின் சிறகைக்கேளே
கேளே வச்சிராயுதத்தால் சிறகைக் கொய்ய
குருதியது கடல்தன்னிலே போய்விழுந்து
ஏளே நீ இவ்விரண்டால் பவளம்தானும்
ஏற்றமான உற்பத்தி தானுமாச்சு
வாளேதான்வலன் தசையில் தோற்றமுற்ற
மாசற்ற பவளத்தின் வன்மைகேளு
வேளேதான் வெளிறினதோர் சிவப்புமாகி
மிக்கினா குன்றிமணி நிறமுமாமே

பவளம் வைப்பு:

பவளத்தை செயற்கையாகத் தயாரிக்கும் முறையும், நம் நூற்களில் குறிப்பிட்டுள்ளது. அதன் விபரம்.

“பருக்கமதே சங்கினுட மாவோர்பங்கு
பாங்கான லிங்கமரைப் பலமும்கூட்டி
தருக்குமதே விளாம்பிசினித் தண்ணீராலே
தாக்கியரை மூன்று நாள் பவளம் போல் செய்து
உருக்குமே தான் உலறவிட்டு மடவைமீனை
உருவயிற்றைக் கீறியதில் வைத்திடாயே
வைத்துமே பாண்டத்தில் கால்வாசிதானும்
வனமான கிளிஞ்சிநீர் அடியில்போட்டு
தைத்துமேதான் மீனதனையதிலே வைத்து
சாங்கமாக கால்வாசி சுண்ணம் போட்டு
நைந்துமே நாலுபடி நல்ல தண்ணீர்விட்டு
நலமாக நீரெல்லாம் வற்றக் காய்ச்சி
வேய்துமதே வெந்நீரில் கழுவிப்போட
மெழுகூர்ந்து பவளம் போலிருக்குந்தானே”

- போகர் 7000, மூன்றாம் காண்டம், பக்கம் 167

பொருள் :

ஊசி, மாமூக்கு, அராசிய அந்தம், உசிதமாது, காமம், தும்பு, இலிங்கம், வடம், வங்கத்தூறுசம், வக்கணம், பதிக்கடகம் என்பன பவழத்தின் வேறுபெயர்கள்,

“பவளத்தின் பேர்தனையே பகரக் கேளு
பாங்கான துகிர்துப்பு விந்துபமாகும்
துவளத்தின் வாருதிக்குள் தண்டுமாகும்
சுயமான செந்தண்டு கொடியுமாகும்
பவளத்தின் விற்பனைக்குளமர்ந்த கன்னி
அழகாக சாற்றியதோர் விற்பவளமாகும்
கூறியதோர் குணமெல்லாங் கூர்ந்துபாரே”

- போகர் நிகண்டு 1200 பாடல் 98 பக்.21

பவளத்தின் செய்கைகள்:

கபஹரகாரி
நரம்புரமாக்கி
சிறுநீர்பெருக்கி
மலமிளக்கி

- குணபாடம் தாதுசீவ வகுப்பு – பக்கம் - 348

Emetic

Antacid

Anti phlegmonous

Anti bilious

- India materia medica - Pg. 157

குற்றங்கள் 6

பிளவு, முடக்கு, திருகல், துளை, கருப்பு, வெளிரல், சிலப்பதிகாரத்தில் கொடிப்பவள வருக்கம் கருப்பத்தே துளைக்கப்படனவும், திருக்கு முறுக்குண்டு எழுந்தனவும் ஆகிய குற்றங்கள் நீங்கியதாய் செம்மைமிக்கு செழிப்புடன் வளர்ந்ததாய் இருக்க வெண்டுமென,

"கருப்பத் துளையவுங் கல்லிடை முடங்கலுந்
திருக்கு நீங்கிய செங்கொடி வல்லியும்
என்ற சிலப்பதிகார அடிகள் உணர்த்துகிறது.

பவளத்தின் பொதுக்குணங்கள்

Pavalam is chiefly used in cough, pthisis, asthma, fever, urinary disease, etc. Dose is 3-12 grains, Thrice a day after meals. It was administered to cases of chronic bronchitis and Pulmonary Tuberculosis and found useful in both classes of disease'.

- Indian Materia Medica – Page 157

"புரையிலாப் பவளத்தன்மை புகலுவேன் குனிதுண்பர்
சுரமொடு வறட்சி மூச்சுத்தொய்வருந் தாகம் சேடல்
இருமலே வாதபித்த மினைபயவை நீக்குமென்று
வரமலிமுனிவர் சித்தர்வகுத்த வாகடங்கள் சொல்லும்"

- பதார்த்த சூடாமணி பக்கம் 100

திறமான முருக்கம்பூ இதழ்போலுந்தான்
திசாசூரன் சதையில் உற்பத்தியான
குரமான பவழநிறம் கனமுமாகும்
குருதியா மலைச்சிறகில் உற்பத்தியான
திறமையான பவழத்தின் குணத்தைக்கேளு
சிவந்துமே தான் கடலினடி தன்னில்தானும்
பிறமான பவழமான கொடியே போன்று
பெரும்நத்தைப் பிரபையுமாய் இருக்குந்தானே

- போகர் 7000 பக்கம் 167

T.V.சாம்பசிவம்பிள்ளை அகராதியில் Vol-V பக்கம் 3223 ல் பவளத்தின் தோற்றம் பற்றி குறிப்பிடப்பட்டுள்ளதாவது

It is hard calcareous substances found in the ocean, secreted by marine polyps for their common support and habitation. The stony .... secreted by them continuously grow like plants which are called “Coral reefs”

பவளத்தின் பண்புகள்

பவளத்திற்கு குணங்கள், குற்றங்கள் என இரு பண்புகள் குணபாடம் தாது சீவம் பக்கம் 346ல் கூறப்பட்டுள்ளது.

குணங்கள் 6

சிந்தூரம், செம்மணி, செங்காய், முசுமுசுக்கை கனி, வீரைக்கனி மாதுளைக்கனி.

பவளத்தின் சுத்திமுறை

பவளத்தை ஒரு நாள் முழுவதும் பழச்சாற்றில் ஊறவைத்து மறுநாள் வெந்நீரில் கழுவி உலர்த்தி எடுத்துக் கொள்ள சுத்தியாகும்.

- குணபாடம் தாதுசீவம் பக்கம் 348
- சிகிச்சாரத்னதீபம் பக்கம் 41

பவளத்தின் வேறு சுத்தி முறைகள்

கொடிபவளத்தை ஒரு துணியில் முடிந்து பசுவின் மோரில் போட்டு மூன்றுநாழிகை ஊறினபின் எடுத்துக் கழுவி உலர்த்தி வைத்துக் கொள்ளவும்

- வீரமாமுனிவர் நசகாண்டவெண்பா பக்கம் 81

பவளத்தை எலுமிச்சை சாற்றில் 1 சாமம் காலம் பாவனை செய்து பிறகு வெந்நீரில் அலம்பி எடுக்க சுத்தியாகும்.

- ரசரத்ன சமுச்சயம் எனும் சித்த ரசாயன நூல் பக்கம்46

பவளத்தை 3 நாள் சம்பீரச்சாற்றில் ஊறப்போட்டு எடுக்கவும்

- 18 சித்தர்கள் ராஜவைத்திய போதினி பக்கம் 103

“துகிரையே அழுக்கொழிந்து துவிதமும் பண்ணாமல்
வகிர்வருவனை யவன்கண் வனிதையே இனிதுகேண்மா
உகிர்முனி இரத்தகாசம் உப்பசம்வல்லை சோபை
நவிர்வதா செய்நீர் பின்னர்நவில்வென வென்றுமுன்னே”

- அகத்தியர் பின் 80-பக்கம் 27.

பவளமானது – சயரோகம், காசம், ரத்தபித்தம், நேத்திர ரோகங்கள் இவைகளைத் தீர்க்கவல்லது.

-ரசரத்ன சமுச்சயம் பக்கம் 46

பவளத்தினால் அதிதாகம், அருசி, ஐயம், காசம், சயரோகம், காமாலை, சுரதோஷம், திரிதோஷம், நாவறட்சி, பித்த பாண்டு, புழுவிஷம், விந்துநட்டம் போகும்.

- குணபாடம் கையேடு பக்கம் 76

பவளத்தினால் சுரதோஷம் ஈளை, காசம், சலக்கட்டு, தாகம், அழலை, நீர்ச்சுருக்கு, பேதி, விந்து நட்டம் முதலியன குணமாகும்.

- அனுபவ வைத்திய தேவரகசியம் பாகம் 1 பக்கம் 95

பவளபற்பத்தின் பொதுக்குணம்

"நற்பவள நீற்றை நயந்தேயுட் கொண்டோர்க்கு
சொற்பசுர மீளை சுகமாகும் - கற்பரசே
தாகமுடன் அழலைத்தட்டு நீர்க் கட்டெரிச்சல்
வேகமுட னோடும் விரைந்து"

- பதார்த்த குணவிளக்கம் - தாதுசீவம் பக்கம் 257

இளைப்பு நோய்க்கு தரப்படும் பவழம் கொண்டு செய்யப்படும் மருந்துகள்

பவள பற்பம்

அளவு - பணஎடை (488 மிகி)

அனுபானம் - வெண்ணெய்

- அகத்தியர் வைத்திய ரத்தின சுருக்கம் பக்கம்-158

பவள பற்பம்

அளவு - 1-3 குன்றி (130-390 மி.கி)

அனுபானம் - தேன், நெய், வெண்ணெய்

ஆதாரம் - தேரையர் காப்பியம், பக்கம் 30

பவள பற்பம்

அளவு - 1 குன்றி (130 மிகி)

அனுபானம் - பால், நெய், தேன்

- யாகோபு வைத்திய சிந்தாமணி 700 பக்கம் 396

பவள பற்பம்

அளவு - 1 குன்றி (130 மிகி)

அனுபானம் - பால், நெய், தேன்

ஆதாரம் - யாகோபு வாத சூத்திரம் பக்கம்.27

பவள பற்பம்

அளவு - 1 குன்றி (130 மிகி)

அனுபானம் - பால், நெய், தேன்

- ஊர்வசி ரசவாத சிட்கா (பஞ்சரத்தினம்) பக்கம்.216

பவள பற்பம்

அளவு - ½ குன்றி (65 மிகி)

அனுபானம் - நெய்

- பதார்த்த குண விளக்கம் தாதுசீவவகுப்பு பக்கம்.258

பவள பற்பம்

அளவு - ¼ குன்றி (33 மிகி)

அனுபானம் - பால், நெய், தேன்

-கண்ணுசாமியம் என்னும் வைத்திய சேகரம் பக்கம்.15

பவள பற்பம்

அளவு - ½ -1 குன்றி (65-130 மிகி)

அனுபானம் - நெய், தேன்

-கண்ணுசாமி பரம்பரை வைத்தியம் பக்கம்.385

பவள பற்பம்

அளவு - ½ குன்றி (65 மிகி)

அனுபானம் - நெய், வெண்ணெய்

- சிகிச்சாரத்தின தீபம் பக்கம். 224

கொடி பவள பற்பம்

அளவு - 65-200 மி.கி

அனுபானம் - நெய், வெண்ணெய்

- பாரத மருந்து செய்முறைகள் பக்கம்.51

பவள பற்பம்

அளவு - ½ குன்றி (65 மிகி)

அனுபானம் - தேன்

- சித்த வைத்திய திரட்டு பக்கம்.150

பவள பற்பம்

அளவு - 3-5 குன்றி (390-650 மிகி)

அனுபானம் - பால், வெண்ணெய், தண்ணீர்

- சரபேந்திரர் சயம் உளமாந்தை ரோக சிகிச்சை பக்கம்.62

பவள பற்பம்

அளவு - 60 மி.கி.

அனுபானம் - பனந்தொங்கிநீர் + தேன்

- அனுபோக வைத்திய நவநீதம் பாகம் -3 பக்கம்.127

கொடிபவள சுண்ணம்

அளவு - 2-3 குன்றி (260-390 மிகி)

அனுபானம் - வெண்ணெய், நெய், தேன்

- அனுபோக வைத்திய நவநீதம் பாகம் -3 பக்கம்.139

பவள சிற்பி பற்பம்

அளவு - 3-5 Grain

அனுபானம் - வெண்ணெய், நெய், தேன்

- The pharmacopoeia of Siddha Research of Medicine Pg.No.29

பவள சிருங்கி பற்பம்

அளவு - 2-4 Grain

அனுபானம் - வெண்ணெய், நெய், தேன்

- The pharmacopoeia of Siddha Research of Medicine Pg.No.31

பவள பற்பம்

அளவு - 2-3 அரிசி எடை (130-195 மிகி)

அனுபானம் - வெண்ணெய், நெய்

- நம்நாட்டு வைத்தியம் பக்கம் .261

பவள செந்தூரம்

அளவு - ½ -1 குன்றி (65-130 மிகி)

அனுபானம் - நெய், தேன்

-கண்ணுசாமி பரம்பரை வைத்தியம் பக்கம்.121

அனுபானம்	**தீரும்நோய்கள்**
வெந்நீர்	இரத்த மூலக்கூறு
சர்க்கரை	பவுத்திரம்
வெங்காய சாறு	சத்தி குன்மம்
அதிமதுரம்	சூதகசன்னி
மாவிலங்கப்பட்டை சாறு	கிராணி

பவழச்சுண்ணம்

சுத்தி செய்த பவழத்தை ஒரு சட்டியில் போட்டு எலுமிச்சம் பழச்சாற்றையும் புளித்த மோரையும் சமமாக கலந்து அதில் விடவேண்டும். பவழமானது 4 அங்குலம் ஆழ்ந்திருக்கவேண்டும். இதனை அடுப்பேற்றி சிறுதீயாக எரித்து நீர் சுண்டின பின்பு ஒரு நாள் வெயிலில் வைத்து முயல் இரத்தத்தை விட்டு பிசறி வெய்யிலில் வைத்து மறுநாள் ஓட்டிலிட்டு மேலோடு மூடி சிலைமண் செய்யாமல் 300 பலம் வறட்டியில் புடமிட்டு ஆறினபின் எடுக்க கடுங்காரச் சுண்ணமாய் இருக்கும்.

அளவு : 2-3 குன்றியெடை (260-490 மிகி)

துணைமருந்து : வெண்ணெய், பசுவின் நெய், தேன்

தீரும் நோய்கள் : சயம், இருமல், சுவாசகாசம்

- அனுபோக வைத்திய நவநீதம் பாகம் 3 பக்.139

இப்பற்பத்தினைக் கொள்ளும் காலம், கார்த்திகை, மார்கழி, பங்குனி நீங்கிய மற்றைய ஒன்பது மாதங்களுமாகும்.

ஏற்ற நிலம்:

மருதம்

பற்பத்தின் மகிமை

இது விகாரத்துடன் பாடக்கூடிய தொனியை நீக்கிச் சுத்த சுரங்களையும், நரை, திரை, மூப்பு சாக்காடாகிய குற்றங்களை நீக்கிக் காயசித்தியையும் தேக வன்மையையும் கொடுக்குமென்பதை,

“சிந்தூரத்துப் பாடடலை சேர்காய வண்மையுண்டாம்

சிந்தூரத்துப் பாடடலை செய்யுமுன்னே”

பத்தியம்

இதனை உண்ணுங்காலத்துப் புகையிலையையும் பெண் போகத்தையும் தவிர்க்க வேண்டுவதே பத்தியத்தின் இயல்பு. இதை, “பிரம்கத்திரமும் இளம்பிடியும் வர்ச்சிதமே” என்னும் தேரன் பாடலால் அறியலாம்.

செந்தூரம்

ஒரு பலம் பவழத்தை பனஞ்சாறு, பருத்திச்சாறு, ஓரிதழ் தாமரைச்சாறு, உத்தாமணிச்சாறு, கமுகஞ்சாறு, கடம்பின்சாறு இவைகள் ஒவ்வொன்றாலும் தனித்தனியாக அரைத்து புடமிட வேண்டும். பற்பத்திற்கு சொன்ன விதிமுறைகளை இதற்கு அனுசரிக்கவும்.

அளவு

மிளகில் ஒன்று, இரண்டு, மூன்று, நான்கு, ஐந்து கூறுகளாம். இவை முறையே உத்தமம், மத்திமம், அதமம், துணிபு அனாமத்தாகும்.

அனுபானம்	**தீரும் நோய்கள்**
நெய்	கயரோகத்தை சேர்ந்த உப்பு வாந்தி
பசுந்தயிர்	கடுப்பு வாதம்
பசுமோர்	நடுக்குவாதம்
பசும்பால்	கபசுரம்
பனைநுங்கு	கபகாசம்
துளசிச்சாறு	கல்லீரல் வீக்கம்
நீர்	பிடிப்பு வாதம்
இளநீர்	பித்தகாசம்

Cleome gyandra

தைவேளை

தைவேளை

வேறுபெயர்கள்:

தமிழ் : அசுகண்டர், பாலையம் விவிதம்

தெலுங்கு : Vaminta

மலையாளம் : Karvela

கன்னடம் : Shrikala

Sans. : Ajaganda

Hindi : Hurhur

இது வேளையில் ஒரு வகை.

பயன்படும் உறுப்பு:

இலை, பூ, விதை, வேர்

சுவை :

கார்ப்பு

தன்மை :

வெப்பம்

பிரிவு:

கார்ப்பு

செய்கை

இலை : தடிப்புண்டாக்கி

விதை : புழுக்கொல்லி, இசிவகற்றி, அகட்டுவாய்வகற்றி, வியர்வை பெருக்கி

"வேளை" என்கிற பெயரின்கீழ் காய்வேளை, முட்காவேளை, நல்வேளை, நாய்வேளை, தைவேளை என்கிற பெயரால் வழங்கும் பூண்டுகளில் மூன்றுக்கு நான்குக்கும் பொருத்தம் மிகவுண்டு. மூன்றாவதன் பூ வெண்மை, நான்காவதன் பூ

மஞ்சள் (இதற்கே காட்டுக்கடுகு என்றும் பெயர்). ஐந்தாவது சிறிது இலையிலும், பூ நிறத்திலும், உருவத்திலும் சிறிது வேறுபடும். பூ வெண்மை கலந்த நீலநிறமாயும், பெரிதாகவும் இருக்கும். இதன் சமூலச் சாற்றுடன் பசுவின் பாலும் சர்க்கரையும் கலந்து உட்கொண்டுவர இரத்த பீநசம் நீங்கும்.

- குணபாடம் மூலிகை வகுப்பு

Ghee

நெய்

நெய்

நெய் பொதுக்குணம்

“நெய்யுண வுண்டவை நேர்வுறச் செய்துமேன்

மெய்யையுந் திண்ணிய மேருவெனச் செய்யும்.”

நெய்யை வேண்டிய அளவாய் உணவுகளிற் சேர்த்து உட்கொள்ளின், அ.்.து உண்ட உணவைச் சரிப்படுத்திச் சரீரத்திற்கு மிகுந்த பலத்தையும் புஷ்டியையும் உண்டாக்கும்.

பசுவின் நெய்க்குணம்

“தாகமுழ லைசுட்கம் வாந்தி பித்தம் வாயுபிர

மேகம் வயிற்றெரிவு விக்கலழல் - மாகாசங்

குன்மம் வறட்சி குடற்புரட்ட லஸ்திசுட்கஞ்

சொன்மூலம் போக்குநிறைத் துப்பு”.

பசுவினது நெய்யானது தாகம், உழலைப்பிணி, அதிசுட்கரோகம், வாந்தி, பித்தாதிக்கம், வாத விஷம், விரணப் பிரமேகம், வயிற்றிலெரிவு, பித்த விக்கல், இருமல், வயிற்றுவலி, வறட்சி, சினைப்பு, குடல்நெளிதல், அஸ்தி சூம்பல், மூல ரோகம் ஆகியவற்றை நீக்கும்.

பசுநெய்

இதனால் தாகம், உழலைப்பிணி, அதிசுட்கரோகம், வாந்தி, பித்தாதிக்கம், வாதம், பிரமேகம், வயிற்றெரிவு, பித்த விக்கல், இருமல், வயிற்றுவலி, குடற்புரட்டல், அஸ்தி, சோம்பல், மூலரோகம், வறட்சி இவை நீங்கும்.

காராம்பசு நெய்

இதனால் நேத்திரத்திற்கு ஒளியும், சரீர புஷ்டியும் பொற்சாயலும் உண்டாகும். கண், புருவம், நெற்றி, சிரசு இவைகளைப் பற்றிய நோய்கள் நீங்கும்.

எருமை நெய்

இதனால் அறிவு, அழகு, கண்ணொளி இவைகள் மத்திபமாம். வாதபித்த தோஷம் கரப்பான் இவை உண்டாகும்.

வெள்ளாட்டு நெய்

இது, கபாதிக்கத்தையும், வாதகோபத்தையும் நீக்கும். சரீரத்தை வளர்ப்பதுமன்றி, கண்ணுக்கு ஒளியையும் உண்டாக்கும். பத்தியத்திற்கும் உதவும்.

செம்மறியாட்டு நெய்

இதனால் கபநோய் அதிகரிக்கும்.

கலப்புநெய்

எருமை, வெள்ளாடு, பசு இவைகளின் நெய் சேர்ந்த கலப்பு நெய்யால், சுக்கிலமும் தீபனமும் உண்டாகும். உடல் வலுக்கும், பித்ததோஷம், தேகக் கொதிப்பாலாகிய பித்த சுரம் போம்.

MODERN ASPECT

RED CORAL

Zoological aspects

Classification:

Kingdom	:	animals
Sub kingdom	:	radiates
Infra kingdom	:	coelenterate
Phylam	:	cnidaria
Class	:	Anthozoa
Sub- class	:	Alcyonaria (octocorallia)
Order	:	Gorgonacea
Family	:	corallidae
Genus	:	corallium
Species	:	rubrum
Zoological name	:	Corallium rubrum (Linn. 1758)
Synonyms	:	Madrepora rubra (Linn.1758)
		Isis nobilis
		Gorgonia nobilis (Linn. 1758)

- www.sn2000.taxonomy.nl/main/classified

Vernacular Names

Tamil	:	pavalam
English	:	Red coral, noble coral
Sanskrit	:	pravalam

Hindi	:	parvara
Gujarati	:	parvalc
Kannadam	:	havala
Telugu	:	pagadamu
Malayalam	:	poalam
Germany	:	koralian
Italy	:	coralio
French	:	corali rouge
Arab	:	bussud
Persian	:	marjan
Spanish	:	coral rojo
Singalam	:	bubalo

- Nadkarni, Indian material medica vol 2, page 1

Varieties of corals

There are varieties of corals like red corals black colrals, white coralls, blue corals, stony corals, horny corals, soft corals and organ pipe corals.Here the red coral [or] precious coral has been taken for my dissertation.

Distribution

The red coral inhabits the rocky bottoms of seas, chiefly in the western and eastern Mediterranean [from Greece and Tunisia to the straits of Gibraltar including Corsica] , red sea, Persia and Arabian gulf, atlanic ocean.

It has been reported in the malasian waters, islands of japan , Sumatra and Maldives . red corals flourish at depths of 30-200m.

- Wealth of Indian part 2 , page 323

Habitat and biology

A rocky bottom spiecies inhabiting depth with weak luminosity mainly between 20-200m depth. At their shallowed depth distribution . it is characteristic of the corallium rubrum faicies in the bio cenosis of semi dark caves. Coral exploitation is progressively restricting their occurrence at the deeper range of their bathymetric distribution. C.rubrum is slow growing a few centimeters per year and long living species . Diet based on small zooplankton organisms captured with the help of the polyps tentacles.

C. rubrum population are infested by a variety of endobiotic boring sponges.

Size

Some colonies attains 20cm height. The base of the largest specimens can attain to 3cm of diameter. An intensive exploitation reduced, however, the maximum size of colonies.

Charateristics features

Rigid colonies may branch in all directions, given a bushy shapes. Calyces short and hemispherical are distributed on the branches. Expanded polyps in living colonies are white transparent and eight tentacles having fine pinnules without sclerties.

Polyps are retracted within calices once corals have been captured. Calcareous spicules (seleries) from the crust, radically symmetrical are warty with a short stick shape

Colour

Often dark red, sometimes and rarely pink or even white

- www.fao.org/figts/servle/species

In smell it resembles frankincense it easily break with crackling sound.

-Indian material medical vol.2, page 157

Growth of corals

Corals are calcareous skeletons the minute marine organism possessing a more or less cylindrical body, a mouth surrounded by a large number of tentacles and a cavity in which the food is digested. The body wall consists of an outer ectoderm, an endoderm and mesogloe. In the simplest form the skeleton is in the form of minute irregular deposits of calcium carbonate called spicules, lodged within the mesogloea.

In some forms the spicules become fused together and form a continuous tube inside the polyp, whie in some other a hard rod like axis through the boby is formed. In the true (or) stony corals, the principal builders of coral reef and islands, the skeleton is secreted externally at the base and lower part of the body.

Coral reefs

The true or study coral is most colonial and is provided with a hard calcareous exoskeleton secreted by the epidermis and lying wholly outside the polyp body. The skeleton of the colony as a whole is termed corallium and of each polyp, corallite. As the colony grows, the polyps on the outer edges multiply, smothering out the lower ones and growing upon their skeletons, continuously with drawing calcareous material from the water. Under favorable conditions the growth of coral reefs is sufficiently rapid, requiring a reversion of navigation charts every 20 years.

Among the organisms playing a role in the formation of coral reefs are: coralline alage, fleshy alcyonaceae, millipores and sponges. Coral formation are inhabited by a vast number of marine creatures such as sea urchins, star fish, crabs and number of brightly coloured fish.

Reefs corals are limited in their vertical distribution. They don't grow below 50m and flourish best above 30m. exposure to sun, excessive turbidity of sea water and impact of fresh water during heavy rains are unfavourable for coral growth. Coral reefs are usually classified as,

a) Fringing reefs.
b) Barrier reefs.
c) Atolls.

Corals reefs are valued as a source of lime.

Parts used

The calcareous shell or skeleton.

Composition of red corals

The pigments of red corals are unidentified organic compounds of iron.

Calcium carbonate – 86.974%

Magnesium carbonate – 6.804%

Calcium sulphate - 1.271%

Ferrous oxide – 1.720%

Organic matter – 1.350%

Water – 0.550%

Phosphoric acid (p2o5), silica etc. – 1.331%

-wealth of india part 2, page 325

Animal (or) organic matter -8pc

Carbonate of lime – 8.3pc

Magnesium carbonate -3.5pc

Oxide of iron – 4.5pc

- Nadkarni, Indian material medica, vol 2, page 157.

Specific gravity

The specific gravity of the precious corals ranges from 2.6 to 2.7 and the hardness 3.75.

CLEOME GYNANDRA

THAIVELAI

Cleome gynandra is used as a medicinal plant and can be found in all over world .It grows as a weed in paddy fields and also in road sides and in open grass lands. Cleome gynandra L. commonly known as spider plant belongs to the Capparaceae family and subfamily Cleomoideae.Cleome is a largest genus includes 601 plant species. Cleome is a major group of angiosperms, comprising many species found in tropical and sub-tropical areas of the globe. Of more than 600 plants, 206 (34.3%) plants are having accepted species.

BOTANICAL DESCRIPTION:

MORPHOLOGY:

It is an erect, annual herb up to 250-600 mm tall; much branched and sometimes becomes woody with age. Stem- The stem is sticky with glandular hairs and marked with longitudinal parallel lines. Leaves-Leaves are palmately compound, with 3-5 leaflets. The leaf stalk is 20-50 mm long with glandular hairs. The leaflets radiate from the tip of the leaf stalk, are 20-100 x 8-40 mm, smooth or with glands, and taper toward the base; on the under surface, are smooth to finely glandular, and often with scattered multi cellular hairs on the main nerves.

INFLORESCENCE

The inflorescence is a terminal raceme, many-flowered, elongating in fruit; the bract is 3-foliolate to simple above, resembling the leaves but smaller and sessile. The flower stalk is 10-20 mm long with glandular hairs. Petals are white, sometimes fading to rose pink, 10-20 x 3-5 mm, rounded at the apex, abruptly narrowed to a basal claw.

FLOWERS

bisexual, bracteate, white or tinged with purple Fruits-The fruits are in capsule form. The capsule is linear, sub-erect to spreading, 30150 x 2.5-5 mm; the persistent style is 2 mm long and the valve is thin-textured, glandular with hairs. The

seeds are brown, circular in outline, 1.5 mm in diameter, with an obscurely netted surface.

MICROSCOPIC STRUCTURE:

Dark brown, oily; under microscope shows a number of fragments of epidermis of testa consisting of thin-walled, polygonal cells; groups of cells, resembling like stone cells, reddish-brown with non-lignified walls; a large number of oval, rounded or irregularly shaped protein bodies; starch and crystals absent1 Leaf thickness ranges from 112-398 µm. Upper epidermis single layered, large, slightly deep, tubular cell contains thick lamellar cuticle. Multiseriate glandular hairs embedded in both surfaces, foot 2-3 celled embedded in epidermis, bi-celled stalk, large columnar, head is about 3-5 tiered clavate. Mesophyll consists of palisade and spongy parenchyma; palisade cells are adaxial hypodermal, single layered, long rectangular with little inter cellular spaces, chloroplast abundant: spongy parenchyma 2-3 layered with large, intercellular spaces. Vascular bundles large, collateral and arc-shaped in primary veins, small and round in secondary veins. Xylem towards adaxial side, phloem in abaxial, bundle sheath large, parenchymatous cells distinct, barrel shaped, bundles of tertiary veins buried between mesophyll cells. Lower epidermis single layered, large, thick walled; guard cells large, thick walled, vertically embedded to subsidiary cells thick-cuticle, lamellar, forming very minute outer ledges over guard cells.

Epidermis in surface view

The costal epidermal cells are large axially oriented 5-10 times longer than broad, rectangular to rhomboidal in shape, thick walled and straight. Intercostal cells are large and in variously shaped, thin walled, slightly to deeply sinuous. Evidently, three types of glandular hairs, namely uniseriate clavate, multiseriate-spherical and multiseriate-clavate were found in both costal and intercostal regions.

Vernacular names in India:

Sanskrit : Pasugandhi,Ajagandha

Assamese : Bhutmulla

Bengali : Hurhuria, Shulte

English : Dog Mustard

Gujarat : Talvani, Dhelitalavan

Hindi : Hulhul, Hurhur, Kavalia

Kannada : Naram bele Soppu, Nayeetulasi

Kashmiri : Gandi Buti

Malayalam : Atunari vela

Marathi : Tilvan, Bhatvan, Mabli, Tilavana, Tilvant

Oriya : Anasorisia, anasorisa,Hulhulia

Punjabi : Bugra

Tamil : Thai valai, Thal velai .

Telugu : Vaminta, Vayinta

Taxonomic position of Cleome gynandra L :

Kingdom: Plantae

Division: Angiosperms

Class: Dicotyledones

Order: Capparidales (Capparales)

Family: Cleomaceae

Genus: Cleome

Species: Gynandra.

Phytochemical importance of C. gynandra:

Qualitative phytochemical screening of the powdered leaf revealed the presence of following class of compounds. A good number of phytochemicals have been isolated from different parts of white mutant C. gynandra which confirms its current understanding of nutritional claims and pharmacological evidence, whereas a

few compounds, namely, clenbuterol, stearin compound, bicyclohexyl derivatives,and (5Z,8Z)-3-hydroxypropyl dodeca-5,8-dienoate only been isolated from the pink mutant variety, only available in N.E states. Further summarizes the isolated phytochemicals from both the mutant varieties of C. gynandra with respective citations.

Pharmacological importance of C. gynandra:

The pharmacological importance of C. gynandra is referred in Ayurveda; Gulma (tumor, irregularity, or diverticulosis), Krmiroga (worm infection), Asthila (Prostate enlargement), Kandu (pruritus), and Karnaroga (ear infections). The indigenous information s traditional medicine has been figured, reported, and eventually wind up noticeably with composed frameworks of the drug.

The following are some therapeutic investigation reported by various researchers from India and from other nations as well:

- Sap from leaves utilized as pain relieving agent, especially in cerebral pain.
- Sap from pounded leaves is pressed into ears, nostrils, and eyes to treat epileptic fits and ear infection.
- A decoction or mixture of bubbled leaves and/or roots is regulatedto:
- Encourage labor pain in pregnant ladies.
- Treat stomach-throb and constipation.
- Treat conjunctivitis.
- Treat serious thread worm disease.
- Relieve burning chest pains.
- The leaves have anti-inflamatory activities and are utilized to treat joint inflammation.
- Leaves are rubefacient and vesicant and used to treat neuralgia, otalgia, rheumatism, and stiffness. The leaves are rubbed on the affected parts.
- In Taiwan, it is utilized to treat looseness of the bowels, gonorrhea, fever, and rheumatoid arthritis.

In India, the plant has been usually utilized as a rubefacient and anthelmintic agents. Leaves are applied directly over the injuries to prevent sepsis. The plant also

used to treat piles, different stomach aches and in tumor. The juice of the root is utilized to treat fevers.

- Bruised leaves are applied to boils to stop pus discharge.
- Infusion from leaves is utilized to treat iron deficiency.
- Sap from the leaves used to cure intermittent intestinal sickness.
- Leaves are rubbed onto the skin to relieve pneumonia.
- An infusion of the leaves utilized as an eyewash.
- Seeds are anthelmintic and rubefacient and are consumed for the removal of roundworms, or a mixture is applied externally on the stomach as a painkiller.
- Seeds are blended with oil and applied to the scalp to treat headache.
- Mixture of seed controls coughing.
- Seeds are also utilized for veterinarians to treat stomach pains.
- Leaves and the plant have anti-ticks and fleas preventive properties.
- A decoctionof roots is accounted for to have gentle febrifugal properties

High in vitamins and micronutrients:

Spider plant is nutritious. It is known to contain high levels of beta-carotene, vitamin C, and moderate levels of calcium, magnesium, and iron. Regarding vitamin A content, an analysis carried out in Tanzania showed that the in vitro accessibility of all-trans-β-carotene in spider plant was the highest (26%) compared with cowpea, amaranth, sweet potato leaves, pumpkin, or combinations of these vegetables. The study also showed that when spider plant leaves were cooked with oil, in vitro accessibility increased to 53%. However, the total amount of all 9cis β-carotene did not significantly increase with the addition of oil. The analysis showed that spider plant could contribute 72% (without addition of cooking oil) to 477% (with addition of cooking oil) of the daily vitamin A requirement. The daily requirement for children is set at 400 μg RE with an assumption that 50% of all accessible β-carotene will be converted to retinol in mucosa. The weight of a vegetable portion consumed varied from 52 to 157 g, with a median weight of 84 g used as the basis for calculations.

The plant contains high crude protein, lipids, and phenolic compounds. The amino acid profile in spider plant is better than groundnut, as all amino acid contents are higher.

Protein consumption can be compromised by consuming food containing elevated levels of trypsin, which inhibits proteases activities. A study showed that trypsin inhibitor activity in spider plant was low (0.45 and 0.32 μg/mg dry weight of plant respectively before and after boiling for 5 min) compared with the soybean reference (1.32 and 1.03 μg/mg dry weight of plant respectively before and after boiling for 5 min).

Vegetables can lose their vitamin C content after cooking, but showed that spider plant best retains vitamin C compared with other vegetables. In fact, when 20 g of spider plant are cooked in 100 mL (very little) or 400 mL (excess) water, the losses were 5.3% and 18.3% respectively. By comparison, losses for amaranths A. graecizans were 86.2% and 46.4%, and A. spinosus 96.5% and 67.0%; for Ethiopian mustard (Brassica juncea), 51.4% and 86.1%; Moringa (Moringa oleifera) 85.4% and 98.5%; and bitter lettuce (Launaea cornuta) 93.5% and 94.5%. Understanding why spider plant retains most of its vitamin C after cooking would help indigenous vegetable breeders improve the nutritive value of this vegetable.

Free radicals are responsible for "oxidative stress" and often are implicated in the expression of several human diseases including diabetes, cancer, coronary heart diseases, neurodegenerative ailments, rheumatoid arthritis, etc. The human body has an antioxidant defense system that is believed to be strengthened by antioxidant-rich diets. Antioxidants include β-carotene (pro-vitamin A caretonoids) and vitamin C, which are present in fruits and vegetables. Analyzed antioxidant activity in 35 Ugandan fruits and vegetables and found that spider plant had an antioxidant activity of 0.53 to 2.92 mmol/100 g and the derived food was a major contributor to the total dietary antioxidant capacity in the Ugandan diet.

High oil content in seed Seed: of spider plant has high levels of polyunsaturated oils that can reach up to 29.6%. The oil can be extracted by simple pressing and does not require refining. The seed cake can be used for animal feed, and the seed itself for feeding birds.

CHEMICAL CONSTITUENTS OF CLEOME:

The isolation of oleic acid, linolic acid, palmitic acid, stearic acid, arachidic acid and a phytosterul from seeds oil of Cleome pentaphylla Linn.

The root consists of two glyco-flavonones as narringenin-4-galactoside-1 and dihydrokaemferol-4f galactoside-2. Identification of a new glycoside-7, 3r-4-trihydroxyflavonone-5-0-Lrhamnopyranoside-3 were reported from the whole plant of Cleome viscosa . Continuing their studies same workers in the same year reported the isolation of a new glycoside- the Cleome prenols isolated from Cleome spinosa L. These were identified as nonaprenol, decaprenol and undecaprenol, which are composed of a U terminal isoprene, three internal E-isoprene and the remaining Z-isoprene residues respectively. S.B. Mahota and co-workers in 1979 reported the isolation of a novel diterpene lactone Cleomeolide -6 from Cleome icosandra Linn (syn Cleome viscosa Linn). From Cleome viscosa, S.K. Srivastava reported in 1980, the isolation of a new saponin identified as stigma-5, 24-(28) - diene-3B-0-a-L-rhamnoside-7. Isolation and identification of p-amyrin, lupeol, and a new glycoside from the roots of Cleome viscosa are also reported by the same author in the same year. In 1982 a Coumarinolignoid (Cleomiscosin-B) was isolated from Cleome viscosa seeds (by Anil L. Ray and S.K.Chattopadhyaya . In 1984 reported the isolation of kaempferol and luteolin-7-O-Glucoside. Some studies have been conducted to investigate the nutritional composition of the raw leaves of C. gynandra.

PROPERTIES AND ACTION ACCORDING TO SIDDHA CONCEPT - THAIVELAI:

Rasa : Katu

Guna : Laghu, Ruksa

Virya : Sita

Vipaka : Katu

MEDICINAL USES:

According to ethno-pharmacological surveys, spider plant has a number of medicinal uses. In Uganda, the plant is used to induce labor during childbirth. After giving birth, some women consume spider plant to increase lactation and blood formation. Spider plant remedies are used to alleviate migraine, vomiting, diphtheria, vertigo, headache, pneumonia, septic ears, and stomach ailments; the plant also is used as an eyewash and fed to boys after circumcision and 43 plants in Tanzania claimed to have medicinal properties and found that 37% of them had antimalarial

activity and some had an IC50 as low as 10 μg/mL. For spider plant, the ethyl acetate extract was the most effective with an IC50 obtained with 14 μg/mL. In that methanolic extracts of spider plant could inhibit Candida albicans and Mycobacterium smegmatis at 50 mg/mL and Staphylococcus aureus and Bacillus subtilis were susceptible to inhibition to methanolic extracts.

Experimental rats suffering from arthritis were administered ethanolic spider plant leaf extracts at a dose of 150 mg/kg of body weight for 30 days. Analysis of enzymes involved in the expression of arthritis showed that the rats had recovered from the disease and their status was comparable to the healthy control rats. The control of the disease was related to substances present in the leaf extracts, including saponins, glycosides, lectins, steroids, flavonoids, tannins, triterpens, resins, phenolic compounds, and arthroquinones. However, the individual involvement of these compounds in the control of the disease needs to be investigated. In another experiment administering spider plant leaf extracts to rats expressing severe arthritis, the analysis of lipid peroxidases, catalases, glutathione peroxidase (enzymes involved in the scavenging of free radicals) showed that these enzymatic activities had increased significantly in the diseased rats compared with the control diseased rats that were not fed the leaf extract treatment. On the other hand, the level of enzymes generating free radicals (glutathione and superoxide dismutase) was reduced significantly in the treated rats.

Free radicals also are cited as involved in the expression of plant diseases. Examining whether spider plant is effectively less susceptible to diseases than other plants (by using spider plant mutants that do not synthesize enzymes involved in the scavenging of free radicals) would be a useful model for plant pathologists.

GHEE

GHEE:

Traditional system of medicine, evolved over the ages, had been completely looking after the healthcare of the world. Ghee is widely considered as the Indian name for clarified butterfat, usually prepared from cow's milk, buffalo milk or mixed milk. Cow Ghee is a semi-liquid form of butter without water content, lactose and other milk solids. It is prepared by gently heating butter until it becomes a clear golden liquid. Cow Ghee is light, pure and does not become rancid for a long time. Cow Ghee is sweet in taste, cold in nature and has a sweet after taste. It is considered smoothing, soft, and oily.

APPEARANCE OF GHEE:

The standard specifies ghee to have 96% minimum milk fat, 0.3% maximum moisture, 0.3% maximum free fatty acids (FFA) (expressed as oleic acid), and a Peroxide Value (PV) less than 1.0. Its physical structure should consist of a mixture of higher softening point fats in crystalline form dispersed in the liquid lower softening point fats and this gives the ghee a somewhat granular appearance.

NUTRITIONAL ASPECTS OF GHEE:

Ghee is a source of lipid nutrients, fat-soluble vitamins and essential fatty acids. About 70% of the fatty acids in milk are saturated, of which about 60% are long-chain fatty acids. The monoenes mainly 18:1, constitute most of the remainder, with thedienes and trienes together only accounting for about 3%. Although medical criticism has been directed at milk fat on the basis of its saturated nature, it is more accurate to consider it as lacking in polyunsaturates because ofitshigh18:1content. Debate on the role of milk fat in human health is still continuing. It suffices to highlight here, some of the alleged positive and negative aspects relevant to ghee.

ABSORPTION OF COW GHEE:

Cow ghee is oil that can bond with lipid-soluble nutrients and herbs to penetrate the lipid-based cell walls of the body. Thus, it increases the potency of certain herbs by carrying the active components to the interior of the cells, which helps to increase Marrow, Semen & Ojas (Immunity). It is also used as a carrier of

nutrients to be absorbed across the cell membrane. The potency and efficiency of a drug is usually dependent on its

- Ionization
- Solubility in body fluids
- Blood flow changes on administration

Sometimes a water based drug will not be able to diffuse properly in the cerebrospinal fluid (CSF) or other body parts. However, with Cow ghee as a solvent, an Ayurvedic formulation would reach the targeted areas with more efficiently. Cow Ghee based medicines are digested and absorbed more easily. The antioxidant properties of ghee help prevent damage of nervous and brain tissues besides retarding the progress of degenerative diseases. The cholesterol problem does not raise its ugly head in the administration of ghee as it is found that absorption of Cow ghee increases only the 'good' (HDL) and not the 'bad' cholesterol (LDL) level. This is because Cow ghee is capable of increasing the range of vitamins soluble in fat, like Vitamin E and thereby prevents the oxidation of LDL. Due to this there is no conceivable change in the lipid profile. All this leads to higher chances of prevention of atherosclerosis, stroke or heart attack.

MEDICINAL USES OF GHEE:

Cow Ghee is known to be digested 96% which is highest as compared to all other vegetable or animal source fats. It contains antioxidants like Vitamin E and beta carotene (600IU) besides other nutrients like phospholipids, diglycerides and triglycerides. It is either used as a part of a formulation as a nourishing, extracting, assimilating and/or absorbing agent. Cow Ghee is an integral part of the siddha health science. A food that helps maintains good health, vitality and longevity. Cow Ghee is excellent for balancing Vata (air) and Pitta (fire) related doshas (humors). It is satvic (healthy) food, which has a pure influence on mind, body and spirit. Vata type people can enjoy more ghee than Pitta (fire) type who in turn can enjoy more ghee than Kapha (Water) types. Cow Ghee also brings out the aroma and flavor of many foods. Cow Ghee contains no water, so it does not spoil easily and hence preserves the original freshness and potency of herbs and foods. Therefore, no refrigeration is required for Cow ghee. It is ancient siddha and folk assumption that as Cow ghee

becomes aged, though its taste becomes slightly bitter its effectiveness and healing properties increases.

The Cow ghee should not be administered

- In Tuberculosis
- Upper respiratory tract infections and bronchitis when mucous secretion is more
- Indigestion, constipation, fever, diabetes.
- Those with obesity should be very frugal in their use of ghee and those with high ama should not take ghee at all.

It used for numerous medical applications, including the treatment of allergy, skin and respiratory diseases. Cow Ghee is also known to retard the undesirable effects of drugs besides cancelling the effect of toxins in the body.

It aimed at treating ailments related to the nervous system, digestive system and for psychological ailments too. As per siddha Cow ghee is very much benifical to human beings It loosen up and liquefy toxins and pacify humors (Doshas) in the skin and blood (called the outer disease pathway).It opens the small channels for dislodging and removing heavy toxins. Thus, toxins begin to drain from deeper tissues and start to flow in the gastro intestinal tract for elimination. It also lubricates and moistens the membranes and tissues. It protects tissues from damage, helps in the proper flowing of wastes and toxins from the body. People with a strong digestive power can use more ghee than those with a weak digestive system. As per siddha the colon is related to all other organs and tissues. When the colon is cleansed and toned successfully, the entire body receives healing and rejuvenation. The colon is the main organ through which the body absorbs nutrients thus, a proper functioning colon is imperative for efficient assimilation of nutrients. Cow Ghee is beneficial & its benefits are mentioned in the siddha For Example, it imparts the benefits of the best essential fatty acids without the problems of oxidized cholesterol, trans-fatty acids or hydrogenated fats. It is also resistant to free radical damage and is both salt and lactose free. It contains butyric acid, a fatty acid with antiviral and anti-cancer properties. Digestion- The siddha texts say that Cow ghee lubricates the digestive system and improves the digestive power because it helps enhance digestion without irritating the stomach and balance the stomach acids to maintain and repair the mucus

lining of the stomach. Cow ghee aids proper digestion and nutrient assimilation. People who are lactose intolerant can generally consume Cow ghee. Cow ghee is said to promote all three aspects of mental functioning-- learning, memory and recall. The traditional texts also designate. Cow ghee - (Healthy and balanced fat in the body), beneficial for mental alertness and memory. It supports healthy vision, voice, intelligence and brain function.

Cow Ghee & oil are widely used in siddha system of medicine as mediums to administer herbal preparations. Herbs, pastes of herbs or decoction of herbs are infused in oil or Cow ghee and later administered to patients after diagnosing the condition of diseases. Cow ghee, when churned from yogurt or butter milk, it is not only a perfect & healthy cooking medium, but also a wonderful medicine.

In siddha, Cow ghee is known as 'amrita'(nectar) & considered the natural oil for all internal body mechanisms.

OTHER USES:

- for body massage- apply Cow ghee all over the body, rubbing into head, chest, limbs, joints & orifices. It will bypass the digestive system & allow the qualities of Cow ghee to penetrate directly into the deeper tissues. Massaging the skin creates endorphins or peptides, which enhances the body immune system. Regular slows the aging process.
- Cow Ghee is used in purvakarma (early panchkarma), where the small amount of Cow ghee is taken first thing in the morning by the practitioner to oleate the internal organs & dissolve the ama or toxic wastes in the tissues, allowing them to be to be carried to the digestive tract for elimination.
- Cow Ghee is used as a carrier for herbs and its supreme penetrating qualities and thus ability to carry these substances deep into the tissues.
- One or two teaspoons first thing in the morning followed immediately with hot water produce a bowel movement. Two spoon full of Cow ghee in warm milk before bedtime soothing to the nerves and lubricates the intestines and facilitates the bowel movement in the morning.
- In ancient India Cow ghee used for recovery from wounds. It can be applied on broken bones & bruises & effective against skin rashes.

- Cow Ghee is excellent for scrapes & both type of burns i.e. fire or chemicals.
- Cow Ghee is excellent for a gargle, to improve the health of the teeth & gums.
- Cow Ghee can be used as a bath oil & an exquisite facial moisturizer.

DRUG PAVALAPARPAM PROCESS

PAVALA PARPAM

4. MATERIALS AND METHODS

PREPARATION OF MEDICINE:

Selection of Drug

'***PAVALA PARPAM***' as mentioned in the text Kannusamy Parambarai vaithiyam, Pg.385,386 was choosen.

Ingredients:

- Purified PAVAZHAM - 2palam (70gm)
- Thai Velai Leaf Karkam

PURIFICATION OF RAW DRUG :

Pavazham is soaked in lime juice for one day and it is washed with hot water.

METHOD OF PREPARATION:

Step 1 :

Thai velai leaf is made into karkam, and then it is kept on sunlight.

Step 2 :

2 Palam of purified pavazham is kept inside the karkam and closed.

Step 3 :

And then it is closed with earthern saucer with mud and dried. Then it is cupellated with 40-50 cow dung cake to get the parpam.

DOSE :

½ - 1 Kuntri Edai

Adjuvant :

Ghee or Honey

Indication:

It is being used for **Eelai, Shayam, Megasoodu, Dega varatchi, Erumal.**

QUALITATIVE AND QUANTITATIVE ANALYSIS
PHYSICOCHEMICAL ANALYSIS

Sample Description	:	***PAVALA PARPAM***
Equipment used	:	Atomic Absorption Spectrometer (AAS)

Colour:

About 50gm of ***PAVALA PARPAM*** was taken in a clean glass beaker and tested for its colour by viewing again a water opaque background under direct sunlight.

pH:

The pH of ***PAVALA PARPAM*** was estimated as per the method prescribed in Indian Standard (IS) – 6940 (1982). One gram of the ***PAVALA PARPAM*** was taken into a 100ml graduated cylinder containing about 50ml of water and filled up to the mark with water. The cylinder was stopped and shaken vigorously for two minutes and the suspension was allowed to settle for an hour at 25^0 to $27^{0.}$ About 25ml of the clear aqueous solution was transferred into a 50ml breaker and tested for pH using DIGISUN digital pH meter (DIGISUN Electronics, Hyderabad, India)

Determination of Ash Value:

Weighed accurately 2 grams of ***PAVALA PARPAM*** in tarred platinum or silica dish and incinerate at a temperature not be exceeding 450^0C until free from carbon, cooled and weighed. Calculate the percentage of ash with reference to the air dried drug.

Water Soluble Ash:

To the gooch crucible containing to the total ash, added 25ml of water and boiled for 5 minutes. Collected the insoluble matter in a sintered glass crucible or on ash less filter paper. Wash with hot water and ignite in a crucible for 15 minutes at a temperature nor exceeding 450^0 C subtract the weight of the insoluble matter from the weight of the ash the difference of the weight represents the water soluble ash. Calculate the percentage of water soluble ash with reference to the air dried drug.

Acid Insoluble Ash:

Boiled the ash 5 minutes with 25ml of 1:1 dil HCL. Collect the insoluble matter in gooch crucible on an ash less filter paper wash with hot water and ignite. Cooled in a desiccators and weighted calculated the percentage of acid insoluble ash with reference to the air dried drug.

Loss on Drying:

Five grams of ***PAVALA PARPAM*** is heated in a hot oven at 105^0C to constant weight and the percentage of loss of weight has calculated there from.

FOURIER TRANSFORM – INFRA RED SPECTROSCOPY

PERKIN ELMER – SPECFTRUM ONE

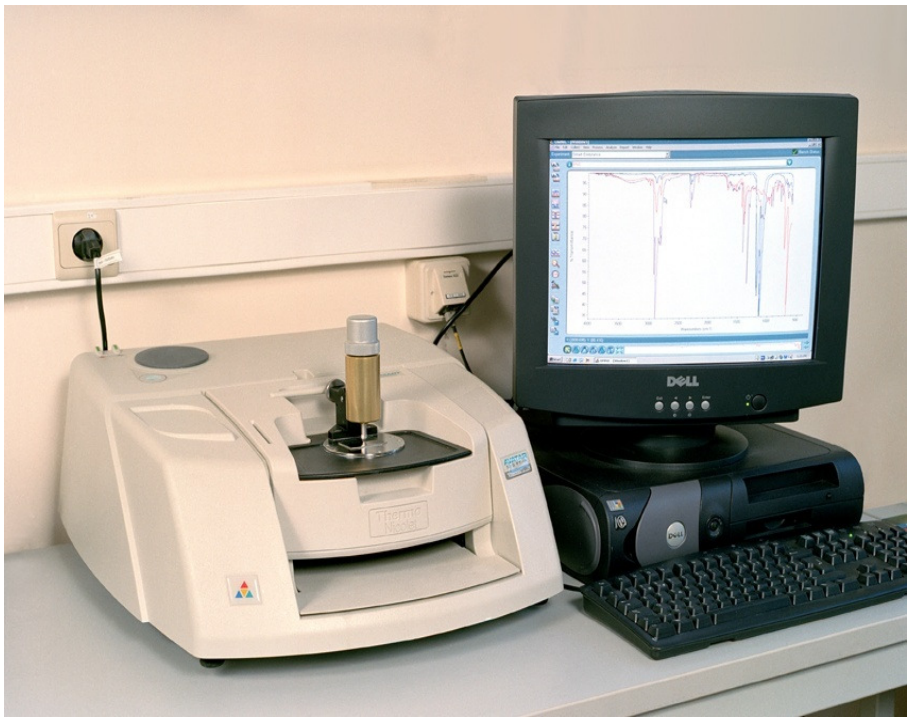
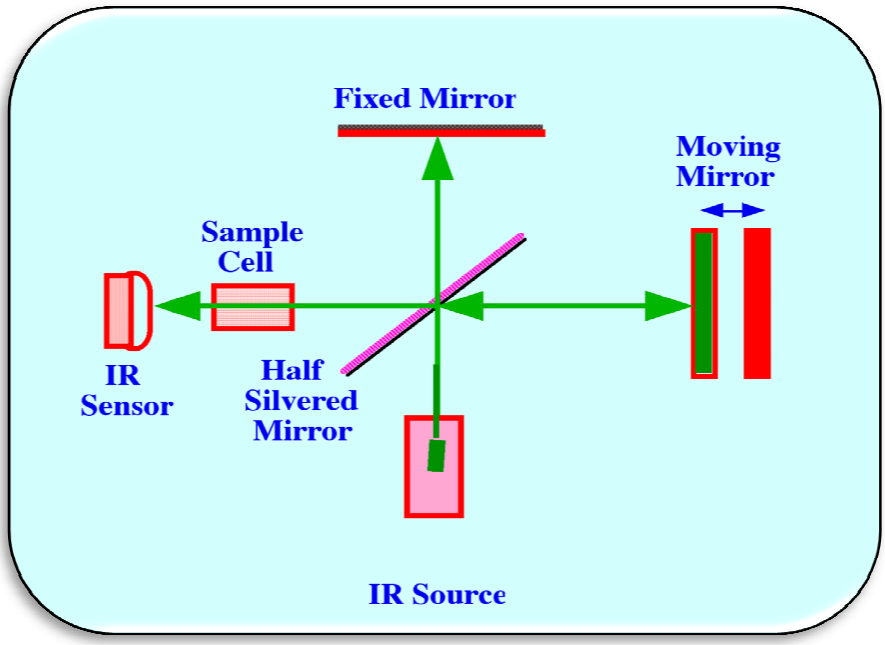


Fig. 3: FTIR Apparatus

FTIR-Mechanism



FOURIER TRANSFORM – INFRA RED SPECTROSCOPY

PERKIN ELMER – SPECFTRUM ONE

Introduction

Vibrational spectroscopy is an extremely useful tool in the elucidations of molecular structure. The spectral bands can be assigned to different vibrational modes of the molecule. The various functional groups present in the molecule can be assigned by a comparison of the spectra with characteristic functional group frequencies. As the positions of the bands are directly related to the strength of the chemical bond, a large number of investigations including intermolecular interactions, phase transitions and chemical kinetics can be carried out using this branch of spectroscopy.

In IR spectroscopy, the resonance absorption in made possible by the change in dipole moment accompanying the vibrational transition. The infrared spectrum originates from the vibrational motion of the molecule. The vibrational frequencies are a kind of fingerprint of the compounds. This property is used for characterization organic, inorganic and biological compounds. The band intensities are proportional to the concentration of the compound and hence qualitative estimations are possible. The IR spectroscopy is carried out by using Fourier transform technique.

Principle

Infra red spectroscopy involves study of the interaction of electromagnetic radiation with matter. Due to this interaction,l electromagnetic radiation characteristic of the interacting system may be absorbed (or emitted). The experimental data consist of the nature (frequency of wave length) and the amount (intensity) of the characteristic radiation absorbed or emitted. These data are correlated with the molecurlar and electronic structure of the substance and with intra – and inter molecular interactions.

FT-IR spectroscopy is used primarily for qualitative and quantitative analysis of organic compounds, and also for determining the chemical structure of inorganic materials. The region between 500-4000 wave number is referred to as the finger print region. Absorption bands in this region are generally due to intra molecular phenomena and are highly specific for each material. The specificity if these bands allow computerized data searches to be performed against reference libraries to identify a material.

Table of Characteristic IR Absorptions

Frequency, cm-1	**Bond**	**Functional group**
3640 - 3610 (s, sh)	O-H stretch	Free hydroxyl alcohols phenols
3500 - 3200 (s,b)	O-H stretch, H – bonded	Alcohols, phenols
3400 – 3250 (m)	N – H stretch	Primary, secondary, amines, amides
3300 – 2500 (m)	O – H stretch	Carboxylic acids
3330 - 3270 (n, s)	–C (triple bond) C – H : C - H stretch	Alkynes (terminal)
3100 – 3000 (s)	C – H stretch	Aromatics
3100 – 3000 (m)	= C – H stretch	Alkenes
3000 – 2850 (m)	C – H stretch	Alkenes
2830 – 2695 (m)	H – C = 0; C –H stretch	Aldehydes
2260 - 2210 (v)	C (triple bond) N stretch	Nitriles
2260 – 2100 (w)	C (triple bond) C- stretch	Alkynes
1760 – 1665 (s)	C = 0 stretch	Carbonyls (general)
1760 – 1690 (s)	C = 0 stretch	Carboxylic acids
1750- 1735 (s)	C = 0 stretch	Esters, saturated aliphatic
1740 – 1720 (s)	C = 0 stretch	Aldehydes, saturated aliphatic
1730 – 1715 (a)	C = 0 stretch	Alpha, beta – unsaturated esters
1715 (s)	C = 0 stretch	Ketones, saturated aliphatic
1710 – 1665 (s)	C = 0 stretch	Alpha, beta – unsaturat aldehydes, ketones
1680 – 1640 (m)	-C = C -	Alkenes
1650 – 1580 (m)	N – H bend	Primary amines
1600 – 1585 (m)	C-C stretch (in – ring)	Aromatics
1550 – 1475 (s)	N – 0 asymmetric stretch	Nitro compounds
1500 – 1400 (m)	C –C stretch (in – ring)	Aromatics
1470 – 1450 (m)	N – 0 asymmetric stretch	Nitro compounds
1370 – 1350 (m)	C – H bend	Alkanes
1360 – 1290 (m)	C – H rock	Alkanes

1335 – 1250 (s)	C – N stretch	Aromatic amines
1320 – 1000 (s)	C – 0 stretch	Alcohols, carboxylic acids, esters, ethers
1300 – 1150 (m)	C – H wag (- CH2X)	Alkyl halides
1250 – 1020 (m)	C – N stretch	Aliphatic amines
1000 – 650 (s)	=C – H bend	Alkynes
950 – 910 (m)	O – H bend	Carboxylic acids
910 – 665 (s, b)	N – H wag	Primary, secondary amines
900 – 675 (s)	C – H "oop"	Aromatics
850 – 550 (m)	C – CI stretch	Alkyl halides
725 – 720 (m)	- C (triple bond) C-H : C- H bend	Alkynes
690 – 515 (m)	C - Br stretch	Alkyl halides

M = medium, w = weak, s=strong, n = narrow, b = broad, sh = sharp

Sampling techniques:

There are a variety of techniques for sample preparation depending on the physical form of the sample to be analyzed.

Solid : KBr or Nujol mull method

Liquid : CsI / TIBr Cells

Gas : Gas Cells

Experimental Procedure: Done at SAIF, IIT Madras, Chennai – 36KBr Method

- The Sample was grounded using – an agate mortar and pestle to give a very fine powder.
- The finely powder sample was mixed with about 100 mg dried KBr salt.
- The mixture was then pressed under hydraulic press using a dye to yield a transparent disc (measure about 13 mm diameter and 0.3mm in thickness), through which the beam of spectrometer passed.

HR SEM - METHODOLOGY

HR SEM-Methodology:

An SEM is essentially a high magnification microscope, which used a focused scanned collection beam to produce images of the sample, both top-down and, with the necessary sample preparation, cross-sections. The primary electron beam interacts with the sample in a number of key ways:-

Primary electrons generate low energy secondary electrons, which tend to emphasize the topographic nature of the specimen.

Primary electrons can be backscattered which produces images with a high degree of atomic number (Z) contrast.

Ionized atoms can relax by electron shell-to-shell transitions. Which lead to either X-ray emission or Auger electron ejection. The X-rays emitted are characteristic of the elements in the top few urn of the sample.

Sample Preparation:

Sample preparation can be minimal or elaborate for SEM analysis depending on the nature of the samples and the data required. Minimal preparation includes acquisition of a ***PAVALA PARPAM*** that will fit into the SEM chamber. And it should be analyzed.

INDUCTIVELY COUPLED PLASMA OPTICAL EMISSION SPECTROMETRY (ICP-OES),

Fig. 5: ICPOES Apparatus

ICP OES METHODOLOGY:

ICP, abbreviation for Inductively Coupled Plasma, is one method of optical emission spectrometry, When plasma energy is given to an analysis sample from outside, the component elements (atoms) is excited. When the excited atoms return to low energy position, emission rays that correspond to the photon wavelength are measured. The element type is determined based on the position of the photon rays, and the content of each element is determined based on the ray's intensity.

To generate plasma, first, argon gas is supplied to torch coil, and high frequency electric current is applied to the work coil at the tip of the torch tube. Using the electromagnetic field created in the torch tube by the high frequency current, argon gas in ionized and plasma is generated. This plasma has high electron density and temperature (10000K) and this energy is used in the excitation –emission of the sample. Solution samples are introduced into the plasma in an atomized state through the narrow tube in the center of the torch tube.

Sample preparation:

Solids cannot be analyzed directly. Such samples should be made into clear aqueous medium quantitatively. When acids are used to prepare solutions care should be taken. The concentration of the acids in the final provided solution should not be more than 2% v/v. highly acidic and organic solutions cannot be analyzed. As a guide line weigh exactly, around 200mg of substance and dissolve in 5mL of 5% of water or aquaregia or whatever acid to make 100mL of final solution. Make proper dilutions, if necessary. Free HF should not present in the final solution to be aspirated.

Ideal concentration is around 100 ppm of the element of interest. Total dissolved solids should be not more than 0.2% w/v in the final solution Very dilute solution may not give reliable results. Each element has a detection limit. A minimum solution volume of 25 ml is necessary for analysis.

In ICP intensity of light emitted when the sample "sprayed or aspirated into an argon plasma" is measured at different wavelengths. The intensity of light at a given wavelength will be proportional to a particular elemental ion concentration. The intensity is calibrated with known standard concentration. For accurate quantitative results It is necessary to simulate the sample matrix condition with that of the standard. Each element generally will have many emission lines and the sensitivity is different for each of this wave length. When more than one element is present it is quite common that some emission lines interfere due to overlapping.

It is preferable to use plastic containers for sample handling and preserving samples for **ICP-OES** analysis. Glass containers can give problems especially when analyzing certain metal ions at low concentration.

The samples of ***PAVALA PARPAM*** was prepared.

6. PRECLINICAL STUDY

ACUTE TOXICITY STUDY IN FEMALE WISTAR ALBINO RATS TO EVALUATE TOXICITY PROFILE OF *PAVALA PARPAM*

OBJECTIVES

The aim of this study is to evaluate the toxicity of the test substance ***PAVALA PARPAM*** , when administered orally to Female Wistar Albino Rats with different doses, so as to provide a rational base for the evaluation of the toxicological risk to man and indicate potential target organs.

Guidelines followed:

OECD Guidelines No.423

The experimental protocol was approved by Institutional Animal Ethical Committee as per the guidance of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Environment and Forest, Government of India.

Study design and Controls:

1) Female Wistar Albino Rats in controlled age and body weight were selected.
2) ***PAVALA PARPAM*** was administered at 5mg/kg, 50mg/kg, 300mg/kg, 2000 mg/kg body weight as (Water) as suspension along with blank.
3) The results were recorded on the day of drug administration approximately 1^{st}, 3^{rd} , 4^{th} and 24^{th} hour in post dosing and further made into observation upto 14 days.

EXPERIMENTAL PROCEDURE

1.Animals

1.1.Supply

A total of 15 Female Wistar Albino Rats with an approximate age of 6 weeks are purchased from JAWAHARLAL INSTITUTE OF POSTGRADUATE MEDICAL EDUCATION AND RESEARCH, PUDUCHERRY-6. On their arrival, a sample of animals was chosen at random and weighted to ensure compliance with the age requested. The mean weights of Female Wistar Albino Rats were 100 – 150g respectively. The animals were housed in metabolic cages (55 X 32.7 X 19 cm), with

sawdust litter, in such a way that each cage contained a maximum of 3 rats of the same sex.

All animals underwent a period of 14 days of observation and acclimatization between the date of arrival and the start of treatment. During the course of this period, the animals were inspected by a veterinary surgeon to ensure that they fulfilled the health requirements necessary for initiation of the study.

1.2. Housing

The Female Wistar Albino Rats were housed in metabolic cages (55 X 32.7 X 19 cm), placed on racks. From the week before initiation of the treatment, each cage contained a maximum of 3 rats of the same sex and treatment group.

Each cage was identified by a card, color coded according to the dose level. This card stated the cage number, number and sex of the animals it contained, Study number, test substance code, administration route, dose level and study drugs name, date of the arrival of the animals and initiation of treatment.

The temperature and relative humidity were continuously monitored. Lighting was controlled to supply 12 hours of light (7:00 to 19:00 hours) and 12 hours of dark for each 24 hour period.

The animals were kept in a clean environment with 12 hour light and 12 hour dark cycles. The air was conditioned at 22±3°C and the relative humidity was maintained between 30 – 70% with 100% exhaust facility.

The cages corresponding to each experimental group were distributed on racks in such a manner that external factors, such as environmental conditions, were balanced as far as possible.

2.Diet

All the rats had free access to a pelleted rat diet. The diet was analyzed by the manufacturer to check its composition and to detect possible contaminants.

2.1. Water

The water was offered ad libitum in bottles.

3.Administration Route and Procedure

The test substance was administered orally. The Female Wistar Albino Rats belonging to the control group were treated with the vehicle(Water) at the same administration volume as the rest of the treatment groups.

The animals were marked on body with picric acid solution prepared in water.

The marking within the cage was as below.

Table-1 Grouping and Marking of Animals

Group No	Animal Marking
1	Head
2	Body
3	Tail

The group number, cage number, sex of the animal and animal number were identified as indicated below using cage label and body marking on the animals.

Table – 2 Numbering and Identification

Cage No	Group No	Animal marking	Sex
1	I	H,B,T	Female
2	II	H,B,T	Female
3	III	H,B,T	Female
4	IV	H,B,T	Female
5	V	H,B,T	Female

3.1. Doses:

The doses for the study were selected based on literature search and range finding study. Following the period of fasting, the animals were weighted and then drug was administered orally as single dose using a needle fitted onto a disposable syringe of approximate size at the following different doses.

Table – 3. Doses

GROUP	DOSE
Group –I	Control
Group – II	5mg/kg
Group – III	50mg/kg
Group – IV	300mg/kg
Group – V	2000mg/kg

The test substance was administered as single dose. After single dose administration period, all animals were observed for 14 days.

3.2. Dose Preparation

PAVALA PARPAM was added in distilled water and completely dissolved to oral form for administration. The dose was prepared for a required concentration before dosing by dissolving in distilled water. It was mixed well. The preparation for different doses was vary in concentrations to allow a constant dosage volume.

3.3. Administration

The test substance was administered orally to each female Wistar Albino rats as single dose using a needle [straight, 10guage, 5.9 inches (15.2cm) length, 6.4mm tip] fitted onto a disposable syringe of appropriate size at the following different doses. The concentration was adjusted according to its body weight. The volume was not exceeding 10ml/kg bodyweight. Variability in test volume was minimized by adjusting the concentration to ensure a constant volume at all dose levels.

3.4. Observation period

All animals were observed for any abnormal clinical signs and behavioral changes. The appearance, change and disappearance of these clinical signs, if any, were recorded for approximately 1.0, 3.0, 4.0 and 24.0 hours post-dose on day of dosing and once daily thereafter for 14 days. Animals in pain or showing severe signs of distress were humanely killed. The cageside observation includes changes in skin, fur, eyes and mucous membranes, occurrence of secretions and excretions. Autonomic activity like lacrimation, piloerection, pupil size and unusual respiratory pattern, changes in gait, posture, response to handling, presence of clonic or tonic movements, stereotypes like excessive grooming and repetitive circling or bizarre behavior like self – mutilation, walking backwards etc were observed. At the 14^{th} day, sensory reactivity to stimuli of different types (e.g: auditory, visual and proprioceptive stimuli) was conducted. Auditory stimuli responses were measured by clicker sound from approximately 30cm to the rats; Visual stimuli response were measured with the help of shining pen light in the eye of rats and placing a blunt object near to the eye of rats. Response to proprioceptive stimuli was measured by placing anterior/dorsal surface of animals paw to the table edge. The responses of reactions for these three exercises were normal in animals belonging to both the controls as well as drug treatment dose groups.

4.0 Mortality and Morbidity

All animals were observed daily once for mortality and morbidity at approximately 1.0, 3.0, 4.0 and 24.0 hours post dose on day of dosing and twice daily (morning and afternoon) thereafter for 14 days.

SUB-ACUTE TOXICITY STUDY IN WISTAR ALBINO RATS TO EVALUATE TOXICITY PROFILE OF *PAVALA PARPAM*

1.Objective

The objective of this 'Sub-acute toxicity study of ***PAVALA PARPAM*** on Wistar Albino Rats' was to assess the toxicological profile of the test item when treated as a single dose. Animals should be observed for 28 days of drug administration. This study provides information on the possible health hazards likely to arise from exposure over a relatively limited period of time.

2. Test Guideline followed

OECD 407 Method – Sub-Acute Toxic Class Method (Repeated Dose 28-Day Oral Toxicity Study in Rodents)

3. Test Substance Detail

Name : ***PAVALA PARPAM***

4. Test System Detail

The study was conducted on 3 male and 3 female Wistar Albino Rats. These animals were selected because of the recommended rodent species for oral studies as per followed guideline and availability of animals 6 weeks old male and female rats were selected after physical and behavioral examination. The body weight range was fallen within ± 20% of the mean body weight at the time of randomization and grouping. The rats were housed in standard laboratory condition in Polypropylene cages, provided with food and water *ad libitum*. The experimental protocol was approved by Institutional Animal Ethical Committee as per the guidance of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Environment and Forest, Government of India.

5.Acclimatization

The animals were selected after veterinary examination by the Veterinarian. All the selected animals were kept under acclimatization for a week.

6.Randomization & grouping

One day before the initiation of treatment (days 0- last day of acclimatization), the selected animals were randomly grouped into four different groups containing minimum 6 animals (3 Male + 3 Female) per group.

7. Numbering and Identification

The animals were marked on body with picric acid solution prepared in water. The marking within the cage was as below.

Table-4 Animal Identification

Group No	Animal marking
Control	H,B,T, E, L, NM
Low dose	H,B,T, E, L, NM
Mid dose	H,B,T, E, L, NM
High dose	H,B,T, E, L, NM

The group no., cage no., sex of the animal and animal no. were identified as indicated below using cage label and body marking on the animals.

Table-5 Animal Marking

Cage no	Group no	Animal marking	Sex
1	Control	H, B,T	Male
		E, L, NM	Female
2	Low dose	H, B,T	Male
		E, L, NM	Female
3	Mid dose	H, B,T	Male
		E, L, NM	Female
4	High dose	H, B,T	Male
		E, L, NM	Female

H – Head, B – Body, T – Tail, E- Earlobe, L – Limb, NM – No Marking

8. Husbandry

8.1. Housing

The Wistar Albino Rats were housed in standard polypropylene cages with stainless steel top grill. Paddy husk was used as bedding. The paddy husk was changed at least twice in a week. From the week before initiation of the treatment, each cage contained a maximum of 3 rats of the same sex and treatment group.

8.2. Environmental conditions

The animals were kept in a clean environment with 12 hour light and 12 hour dark cycles. The air was conditioned at 22±3°C and the relative humidity was maintained between 30 – 70% with 100% exhaust facility. The cages corresponding to each experimental group were distributed on racks in such a manner that external factors such as environmental conditions were balanced as far as possible.

8.3 Feed & feeding schedule

Feed was provided ad libitum throughout the study period, except over night fasting (18-20 hours) prior to dose administration. After the substance has been administered, food was with held for a further 3-4 hours.

8.4 Water

The water was offered ad libitum in bottles. They were periodically analyzed to detect the presence of possible contaminants.

8.5 Doses

The doses for the study were selected based on literature search and range finding study. Following the period of fasting, the animals were weighed and then test substance was administered orally as single dose using a needle fitted onto a disposable syringe of approximate size at the following different doses.

Table – 6 Dose level

TEST GROUP	DOSE TO ANIMALS (mg/kg body – weight/day)	NUMBER OF ANIMALS
Group – I	Control	6(3male and 3female)
Group – II	Low dose of ***PAVALA PARPAM*** (5mg/ Kg)	6(3male and 3female)
Group – III	Mid dose of ***PAVALA PARPAM*** (10mg/ Kg)	6(3male and 3female)
Group – IV	High dose of ***PAVALA PARPAM*** (20mg/ Kg)	6(3male and 3female)

The test substance was administered as single dose for 28 days and all animals are observed .

8.5 a. Dose preparation

PAVALA PARPAM was added in distilled water and completely dissolved to for oral for administration. The dose was prepared of a required concentration before dosing by dissolving ***PAVALA PARPAM*** in distilled water. It was mixed well. The preparation for different doses was vary in concentrations to allow a constant dosage volume.

8.6. Administration

The test substance was administered orally to each rat as single dose using a needle fitted onto a disposable syringe of appropriate size at the following different doses. The concentration was adjusted according to its body weight. The volume was not exceeding 10ml/kg body weight. Variability in test volume was minimized by adjusting the concentration to ensure a constant volume at all dose levels.

9. Observation

These observations were also performed on week – ends. The observations included but were not limited to changes in skin, fur, eyes, mucous membranes, and in the respiratory, circulatory, central nervous, autonomous systems, somatomotor activity and behavior.

9.1. Clinical signs of toxicity

All the rats were observed atleast twice daily with the purpose of recording any symptoms of ill-health, behavioral changes and clinical signs of toxicity daily for 28 days.

9.2. Food intake

Prior to the beginning of treatment and daily food intake of each cage was recorded for period of 28 days and the mean weekly intake per rats was calculated.

9.3 Water intake

Water intake was checked by visual observation during the study. In addition, the water consumption in each cage was measured daily for a period of 28days.

9.4 Body weight

The body weight of each rat was recorded one week before the start of treatment, and during the course of the treatment on the day of initial, 7^{th}, 14^{th} , 21^{st}

and 28th day. The mean weight for the different groups and sexes were calculated from the individual weight.

Blood collection

Blood was collected through retro – orbital sinus from all the animals of different groups on 29th day. The blood was collected in tubes containing Heparin/EDTA as an anticoagulant. Animals were fasted over night prior to the blood collection.

LABORATORY STUDIES

After the 4th week of treatment, samples of blood were withdrawn from the orbital sinus from each group, under weak ether anesthesia after fasting for 16 hours. The blood samples are used to evaluate Hematological parameters like RBC, WBC, Hb% and DC. The collected blood samples also centrifuged 10000rpm in 10 minutes to separate the serum. The separated serum used to evaluate biochemical parameters like SGOT, SGPT, ALP, UREA and CREATININE etc.

Hematology

The following hematological parameters were analysed using Autoanalyser

Hb : Haemoglobin(gm%)

WBC : White Blood Corpuscles ($x10^3$/cu.mm)

RBC : Red Blood Corpuscles ($x10^6$/cu.mm)

Differential count:

N : Neutrophils (%)

L : Lymphocytes (%)

M : Monocytes (%)

E : Eosinophils (%)

B : Basophils (%)

Clinical biochemistry:

The following clinical Bio-chemical parameters were analysed using Auto analyser

ALT/SGPT : Alanine amino transferase (U/L)

AST/SGOT : Aspartate amino transferase (U/L)

ALP : Alkaline serum phosphatase (U/L)

SERUM UREA (mg/dl)

SERUM CREATININE (mg/dl)

Terminal studies

Sacrifice and macroscopic examination

On completion of the 28 days of treatment, Wistar Albino Rats were sacrificed by ether inhalation. A full autopsy was performed on all animals which included examination of the external surface of the body, all orifices, cranial, thoracic and abdominal cavities and their contents both in situ and after evisceration. As the number of animals exceeded the number that could be sacrificed in one day, the autopsies were carried out over three consecutive days at the end of the treatment period.

Organ weights:

After the macroscopic examination the following organs were weighed after separating the superficial fat: Heart, Kidneys and Liver.

HISTOPATHOLOGICAL STUDIES

Anatomy of the liver was studied immediately after sacrificing the animals. A small portion was fixed in 10% neutral buffered formalin as described by Luna 14. Thin sections of 4-5µm were taken, stained with Haematoxylin and Eosin and histology was studied.

RESULTS

QUALITATIVE AND QUANTITATIVE ANALYSIS

Table -7

Colour characters of *PAVALA PARPAM*

No	Nature of drug	Nature of colour
1	***PAVALA PARPAM***	White

Table 8— Physicochemical analysis of samples of *PAVALA PARPAM*

[Values are mean of three determinations ±SEM]

Parameters	Total ash	Values
Ash value	Water soluble ash	7.75±0.011
	Acid insoluble ash	0.95±0.011
Extractive value	Water soluble extractive value	8.20±0.310
Loss on drying	Loss on drying at 70°C	8.30±0.240

SEM- singularity expansion method

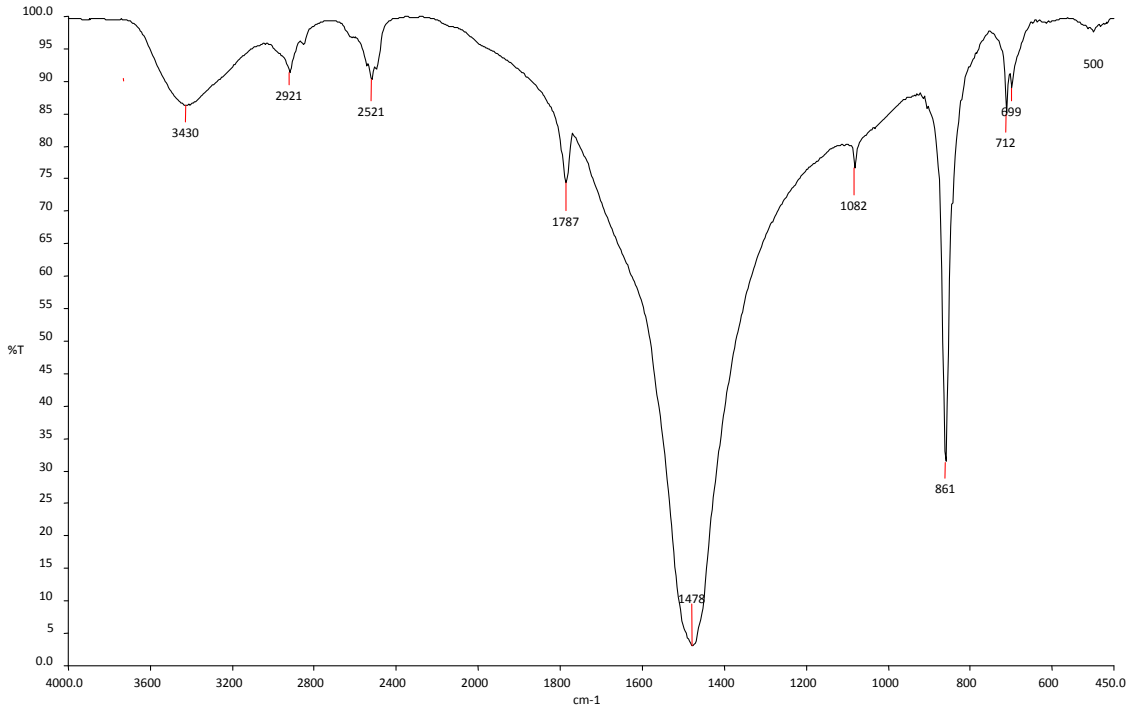
Table-9

Particle size and pH of *PAVALA PARPAM*

S.No	Parameters	Values obtained
1	Particle size by SEM	2-1 µ
2	pH	8.751

PAVALA PARPAM (FTIR METHOD)

Pavala Parpam



PAVALA PARPAM

Table – 10 of characteristic IR Absorptions

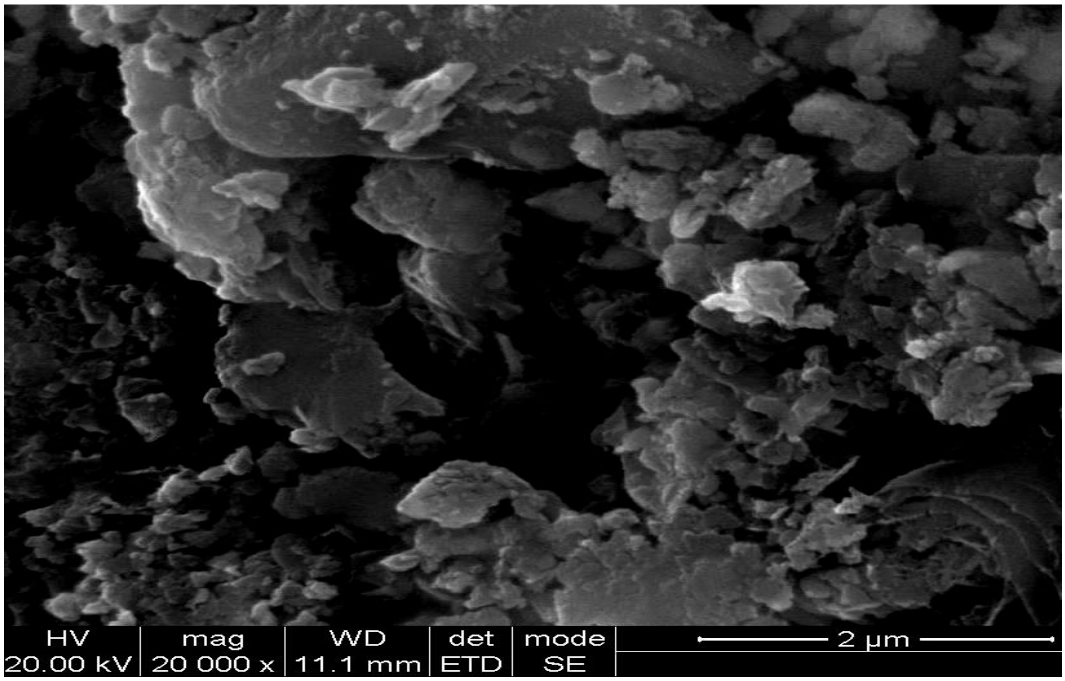
Functional Group	Frequency (cm-1)	intensity
water OH Stretch	3700-3100	strong
alcohol OH stretch	3600-3200	strong
carboxylic acid OH stretch	3600-2500	strong
N-H stretch	3500-3350	strong
≡C–H stretch	~3300	strong
=C-H stretch	3100-3000	weak
-C-H stretch	2950-2840	weak
-C-H aldehydic	2900-2800	variable
stretch	~2250	strong
C≡C stretch	2260-2100	variable
C=O aldehyde	1740-1720	strong
C=O anhydride	1840-1800, 1780-1740	weak, strong
C=O ester	1750-1720	strong
C=O ketone	1745-1715	strong
C=O amide	1700-1500	strong
C=C alkene	1680-1600	weak
C=C aromatic	1600-1400	weak
CH_2 bend	1480-1440	medium
CH_3 bend	1465-1440, 1390-1365	medium
C-O-C stretch	1250-1050 several	strong
C-OH stretch	1200-1020	strong
NO_2 stretch	1600-1500 and 1400-1300	strong
C-F	1400-1000	strong
C-Cl	800-600	strong
C-Br	750-500	strong
C-I	~500	strong

Inference :

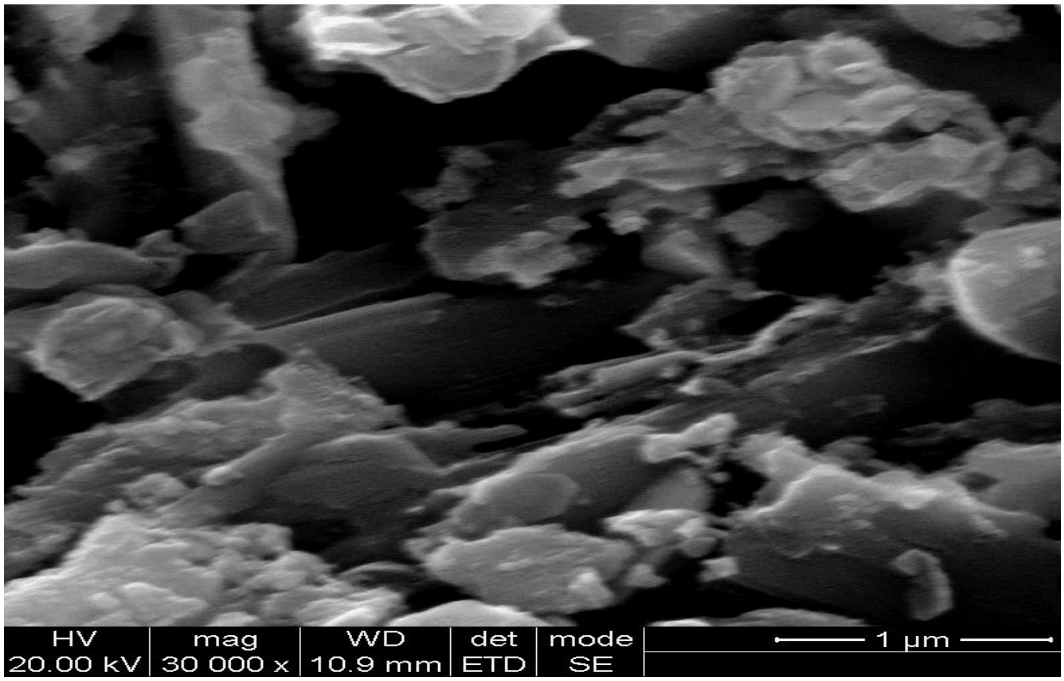
The ***PAVALA PARPAM*** contains, Aromatic compounds, Alkynes, Alkene, Alkyl halide, Alcohal, Acyl chloride, Esters, Carboxylic acids, Amides, Amines.

SEM ANALYSIS

Scanning Electron Microscope (SEM)



SEM -20000 Magnification



SEM -30000 Magnification

Results and Interpretation of SEM analysis:

The morphology of the ***PAVALA PARPAM*** sample can be determined by SEM (FEI Quanta). A representative portion of each sample must be sprinkled onto a double side carbon tape and mounted on aluminium stubs, in order to get a higher

quality secondary electron image for SEM examination. We have observed from SEM photographs that particles are spherical in shapes and sizes are in the range from 2 - 1 micron. Although the particle sizes of different batches showed similarity, it seems that these particles are aggregates of much smaller particles. When dispersed in an aqueous medium, these preparations form a negatively charged hydrophobic particle suspension. This hydrophobicity gives these particles a tendency to aggregate together to form larger particles. PAVALA PARPAM exhibited larger sizes and agglomeration of the particles. Therefore, the comparatively larger size may be due to the agglomeration of the particles by repeated cycles of calcinations involved in preparation.

SOPHISTICATED ANALYTICAL INSTRUMENT FACILITY

IITM, CHENNAI-36

PERKIN ELMER OPTIMA 5300 DV ICP-OES

Sample ID	**Elements Symbol Wavelength (nm)**	**Concentration**
Pavala Parpam (wt:0.41210g)		
Al	396.152	BDL
As	188.979	BDL
Ba	455.403	05.221 mg/L
Ca	315.807	881.100 mg/L
Cd	228.802	BDL
Cu	327.393	BDL
Hg	253.652	BDL
K	766.491	03.171 mg/L
Mg	285.213	01.784 mg/L
Na	589.592	02.280 mg/L
Ni	231.604	BDL
Pb	220.353	BDL
P	213.617	126.227 mg/L
Sr	421.502	03.654 mg/L
S	180.731	11.324 mg/L

BDL- Below detection limit

PAVALA PARPAM contains Aluminium, Arsenic, Cadmium, Copper, Mercury, Nickel, Lead are in below detection limit.

RESULTS:

PH range : **8.751**

FTIR data : ***PAVALA PARPAM*** contains Aromatic compounds, Alkynes, Alkene, Alkyl halide, Alcohal, Acyl chloride, Esters, Carboxylic acids, amides and amines.

HR SEM : ***PAVALA PARPAM*** has the particle size in the range of **2-1 micron**

ICP-OES data : ***PAVALA PARPAM*** shows Aluminium, Arsenic, Cadmium, Copper, Mercury, Nickel and Lead are in below detection limit.

BIO-CHEMICAL ANALYSIS OF *PAVALA PARPAM*

Preparation of the extract:

100mgs of parpam is weighted accurately and placed into a clean beaker and added a few drops of Conc. Hydrochloric acid and evaporated it well. After evaporation cooled the content and added a few drops of conc. nitric acid and evaporated it well. After cooling the content add 20ml of distilled water and dissolved it well. Then it is transferred to 100ml volumetric flask and made up to 100ml with distilled water, mix well, filter it. Then it is taken for analysis.

Table - 11

BIO-CHEMICAL ANALYSIS

S.NO	EXPERIMENT	OBSERVATION	INFERENCE
1.	**TEST FOR CALCIUM** 2ml of the above prepared extract is taken in a clean test tube. To this add 2ml of 4% Ammonium oxalate solution	A white precipitate is formed	**Indicates the presence of calcium**
2.	**TEST FOR SULPHATE** 2ml of the extract is added to 5% Barium chloride solution.	A white precipitate is formed	**Indicates the presence of sulphate**
3.	**TEST FOR CHLORIDE** The extract is treated with silver nitrate solution	A white precipitate is formed	**Indicates the presence of chloride**
4.	**TEST FOR CARBONATE** The substance is treated with concentrated Hcl.	A Brisk effervessence is formed	**Indicates the presence of carbonate**
5.	**TEST FOR STARCH** The extract is added with weak iodine solution	No Blue colour is formed	Absence of starch
6.	**TEST FOR FERRIC IRON** The extract is acidified with Glacial acetic acid and potassium ferro cyanide.	No blue colour is formed	Absence of ferric iron

7.	**TEST OF FERROUS IRON** The extract is treated with concentrated Nitric acid and Ammonium thio cyanate solution	Blood red colour is formed	**Indicates the presence of ferrous iron**
8.	**TEST FOR PHOSPHATE** The extract is treated with Ammonium Molybdate and concentrated nitric acid	No yellow precipitate is formed	Absence of phosphate
9.	**TEST FOR ALBUMIN** The extract is treated with Esbach's reagent	No Yellow precipitate is formed	Absence of Albumin
10.	**TEST FOR TANNIC ACID** The extract is treated with ferric chloride.	No Blue black precipitate is formed	Absence tannic acid
11.	**TEST FOR UNSATURATION** Potassium permanganate solution is added to the extract	It doesnot gets decolourised.	Absence of unsaturated compound
12.	**TEST FOR THE REDUCING SUGAR** 5ml of Benedict's qualitative solution is taken in a test tube and allowed to boil for 2 mts and add 8-10 drops of the extract and again boil it for 2 mts.	No colour change occurs.	Absence of Reducing sugar
13.	**TEST FOR AMINO ACID** One or two drops of the extract is placed on a filter paper and dried well. After drying, 1% Ninnhydrin is sprayed over the same and dried it well.	No Violet colour is formed	Absence of Amino acid
14.	**TEST FOR ZINC** The extract is treated with Potassium Ferrocyanide.	No white precipitate is formed	Absence of Zinc.

Inference:

Analysis reveals the presence of **Calcium, Sulphate, Chloride, Carbonate, Ferrous iron** in ***PAVALA PARPAM* .**

Biochemical Analyis report was given by **Mrs. N.Nagaprema, M.Sc., H.O.D, Bio Chemical Department, Government Siddha Medical College, Palayamkottai.**

EFFECT OF ACUTE TOXICITY (14 DAYS) OF *PAVALA PARPAM*

Table - 12 Physical and behavioral examinations.

Group no.	Dose(mg/kg)	Observation sign	No. of animal affected.
Group-I	Control Distilled water (1ml/kg)	Normal	0 of 3
Group- II	5mg/kg	Normal	0 of 3
Group-III	50mg/kg	Normal	0 of 3
Group-IV	300mg/kg	Normal	0 of 3
Group-V	2000mg/kg	Normal	0 of 3

Table- 13 Showed the effect of Control – Distilled water (1ml/kg) on general behavior after single oral administration in Rat.

SI.NO	General Behavior	Time of observation after Control - Distilled water (1ml/kg) administration			
		1st Hr	3rd Hr	4th Hr	24th Hr
1	Sedation	-	-	-	-
2	Hypnosis	-	-	-	-
3	Convulsion	-	-	-	-
4	Ptosis	-	-	-	-
5	Analgesia	-	-	-	-
6	Stupor reaction	-	-	-	-
7	Motor activity	-	-	-	-
8	Muscle relaxant	-	-	-	-
9	CNS stimulant	-	-	-	-
10	CNS depressant	-	-	-	-
11	Pilo erection	-	-	-	-
12	Skin colour changes	-	-	-	-
13	Lacrimation	-	-	-	-
14	Stool consistency	-	-	-	-

'+' PRESENT & '-' ABSENT

Table - 14 Showed the effect of *PAVALA PARPAM* (5mg/kg) on general behavior after single oral administration in Rat.

SI.NO	General Behavior	Time of observation after *PAVALA PARPAM* (5mg/kg) administration			
		1st Hr	3rd Hr	4th Hr	24th Hr
1	Sedation	-	-	-	-
2	Hypnosis	-	-	-	-
3	Convulsion	-	-	-	-
4	Ptosis	-	-	-	-
5	Analgesia	-	-	-	-
6	Stupor reaction	-	-	-	-
7	Motor activity	-	-	-	-
8	Muscle relaxant	-	-	-	-
9	CNS stimulant	-	-	-	-
10	CNS depressant	-	-	-	-
11	Pilo erection	-	-	-	-
12	Skin colour changes	-	-	-	-
13	Lacrimation	-	-	-	-
14	Stool consistency	-	-	-	-

'+' PRESENT & '-' ABSENT

Table - 15 Showed the effect of *PAVALA PARPAM* (50mg/kg) on general behavior after single oral administration in Rat.

SI.NO	General Behavior	Time of observation after *PAVALA PARPAM* (50mg/kg) administration			
		1st Hr	3rd Hr	4th Hr	24th Hr
1	Sedation	-	-	-	-
2	Hypnosis	-	-	-	-
3	Convulsion	-	-	-	-
4	Ptosis	-	-	-	-
5	Analgesia	-	-	-	-
6	Stupor reaction	-	-	-	-
7	Motor activity	-	-	-	-
8	Muscle relaxant	-	-	-	-
9	CNS stimulant	-	-	-	-
10	CNS depressant	-	-	-	-
11	Pilo erection	-	-	-	-
12	Skin colour changes	-	-	-	-
13	Lacrimation	-	-	-	-
14	Stool consistency	-	-	-	-

'+' PRESENT & '-' ABSENT

Table - 16 Showed the effect of *PAVALA PARPAM* (300mg/kg) on general behavior after single oral administration in Rat.

SI.NO	General Behavior	Time of observation after *PAVALA PARPAM* (300mg/kg) administration			
		1st Hr	3rd Hr	4th Hr	24th Hr
1	Sedation	-	-	-	-
2	Hypnosis	-	-	-	-
3	Convulsion	-	-	-	-
4	Ptosis	-	-	-	-
5	Analgesia	-	-	-	-
6	Stupor reaction	-	-	-	-
7	Motor activity	-	-	-	-
8	Muscle relaxant	-	-	-	-
9	CNS stimulant	-	-	-	-
10	CNS depressant	-	-	-	-
11	Pilo erection	-	-	-	-
12	Skin colour changes	-	-	-	-
13	Lacrimation	-	-	-	-
14	Stool consistency	-	-	-	-

'+' PRESENT & '-' ABSENT

Table - 17 Showed the effect of *PAVALA PARPAM* (2000mg/kg) on general behavior after single oral administration in Rat.

SI.NO	General Behavior	Time of observation after *PAVALA PARPAM* (2000mg/kg) administration			
		1st Hr	3rd Hr	4th Hr	24th Hr
1	Sedation	-	-	-	-
2	Hypnosis	-	-	-	-
3	Convulsion	-	-	-	-
4	Ptosis	-	-	-	-
5	Analgesia	-	-	-	-
6	Stupor reaction	-	-	-	-
7	Motor activity	+	-	-	++
8	Muscle relaxant	-	-	-	-
9	CNS stimulant	-	-	-	-
10	CNS depressant	-	-	-	-
11	Pilo erection	-	-	-	-
12	Skin colour changes	-	-	-	-
13	Lacrimation	-	-	-	-
14	Stool consistency	-	-	-	-

'+' PRESENT & '-' ABSENT

Table - 18 Home cage activity

Functional and Behavioral observation	Observation	Control Distilled water (1ml/kg)	5mg/kg Group (G-II)	50mg/kg (G-III)	300mg/kg (G-IV)	2000mg/kg (G-V)
		Female n=3	Female n=3	Female n=3	Female n=3	Female n=3
Body position	Normal	3	3	3	3	3
Respiration	Normal	3	3	3	3	3
Clonic involuntary Movement	Normal	3	3	3	3	3
Tonic involuntary Movement	Normal	3	3	3	3	3
Palpebral closure	Normal	3	3	3	3	3
Approach response	Normal	3	3	3	3	3
Touch response	Normal	3	3	3	3	3
Pinna reflex	Normal	3	3	3	3	3
Tailpinch response	Normal	3	3	3	3	3

Table - 19 Hand held observation

Functional and Behavioral observation	Observation	Control Distilled water (1ml/kg)	5 mg/ kg (G-II)	50 mg/kg (G-II)	300 mg/kg (G-III)	2000 mg/kg (G-V)
		Female n=3	Female n=3	Female n=3	Female n=3	Female n=3
Reactivity	Normal	3	3	3	3	3
Handling	Normal	3	3	3	3	3
Palpebral closure	Normal	3	3	3	3	3
Lacrimation	Normal	3	3	3	3	3
Salivation	Normal	3	3	3	3	3
Piloerection	Normal	3	3	3	3	3
Pupillary reflex	Normal	3	3	3	3	3
Abdominal tone	Normal	3	3	3	3	3
Limb tone	Normal	3	3	3	3	3

Table - 20 Mortality

Group no	Dose level (mg/kg)	Mortality
Group-I	Control [Distilled water (1ml/kg)]	0 of 3
Group-II	5(mg/kg)	0 of 3
Group-III	50(mg/kg)	0 of 3
Group-IV	300(mg/kg)	0 of 3
Group-V	2000(mg/kg)	0 of 3

Result:

From acute toxicity study it was observed that the administration of ***PAVALA PARPAM*** up to the dose of 2000mg/kg to the rats do not produce drug-related toxicity and mortality. So No-Observed-Adverse-Effect-Level(NOAEL) of ***PAVALA PARPAM*** is 2000mg/kg.

DISCUSSION

PAVALA PARPAM was administered single time at the dose of 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg to rats and observed for consecutive 14 days after administration. Doses were selected based on the pilot study and literature review. All animals were observed daily once for any abnormal clinical signs. Weekly body weight and food consumption were recorded. No mortality was observed during the entire period of the study. Data obtained in this study indicated no significance physical and behavioral signs of any toxicity due to administration of ***PAVALA PARPAM*** at the doses of 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg to rat.

At the 14^{th} day, all animals were observed for functional and general behavioral examination. In functional and general behavioral examination, home cage activity, hand held activity were observed. Home cage activities like Body position, Respiration, Clonic involuntary movement, Tonic involuntary movement, Palpebral closure, Approach response, Touch response, Pinna reflex, Sound responses, Tail pinch response were observed. Handheld activities like reactivity, Handling, Palpebral closure, Lacrimation, Salivation, Piloerection, Papillary reflex, abdominal tone, Limb tone were observed. Functional and general behavioral examination was normal in all

treated groups. Food consumption of all treated animals was found normal as compared to normal group.

Body weight at weekly interval was measured to find out effect of ***PAVALA PARPAM*** on the growth rate. Body weight change in drug treated animals was found normal.

SUMMARY & CONCLUSION

Summary:

The present study was conducted to know single dose toxicity of ***PAVALA PARPAM*** on female Wistar Albino Rats. The study was conducted using 15 female Wistar Albino Rats. The female animals were selected for study of 8-12 weeks old with weight range of within ±20% of mean body weight at the time of randomization. The groups were numbered as group I, II, III, IV and V and dose with 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg of ***PAVALA PARPAM***. The drug was administered by oral route single time and observed for 14 days. Daily the animals were observed for clinical signs and mortality. Body weight of animals was recorded once in a week.

There were no physical and general behavioral changes observed in Wistar Albino Rats of 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg to rats during 14 days.

Body weight of all animals did not reveal any significant change as compared to vehicle control group.

Food consumption of all group animals was normal.

Mortality was not observed in all treatment groups.

Conclusion:

The study shows that ***PAVALA PARPAM*** did not produce any toxic effect and mortality at dose of 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg to rats. So NO-Observed-Adverse-Effect-Level(NOAEL) of ***PAVALA PARPAM*** is 2000mg/kg.

SUB-ACUTE TOXICITY STUDY

TABLE 21. EFFECT OF PAVALA PARPAM ON BODY WEIGHT DURING 28 DAYS TREATMENT IN RATS

Groups	Drug Treatment	Body Weight (gms)				
		1st Day	7th Day	14th Day	21st Day	28th Day
I	Control - Ghee (1ml/kg, p.o)	168.73± 3.42	176.22± 4.97	196.34± 4.77	210.54± 6.04	220.22± 4.40
II	**Pavala Parpam** (5mg/kg, p.o)	162.72± 5.50	175.89± 4.03	189.32± 6.43	208.22± 3.72	217.22± 5.97
III	**Pavala Parpam** (10mg/kg, p.o)	159.55± 6.80	169.33± 3.05	182.55± 4.25	200.53± 5.07	216.22± 5.44
IV	**Pavala Parpam** (20mg/kg, p.o)	163.77± 4.80	175.52± 5.45	185.42± 4.66	205.68± 5.30	218.62± 5.51

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 1. Effect of Pavala Parpam on body weight during 28 days treatment in rats

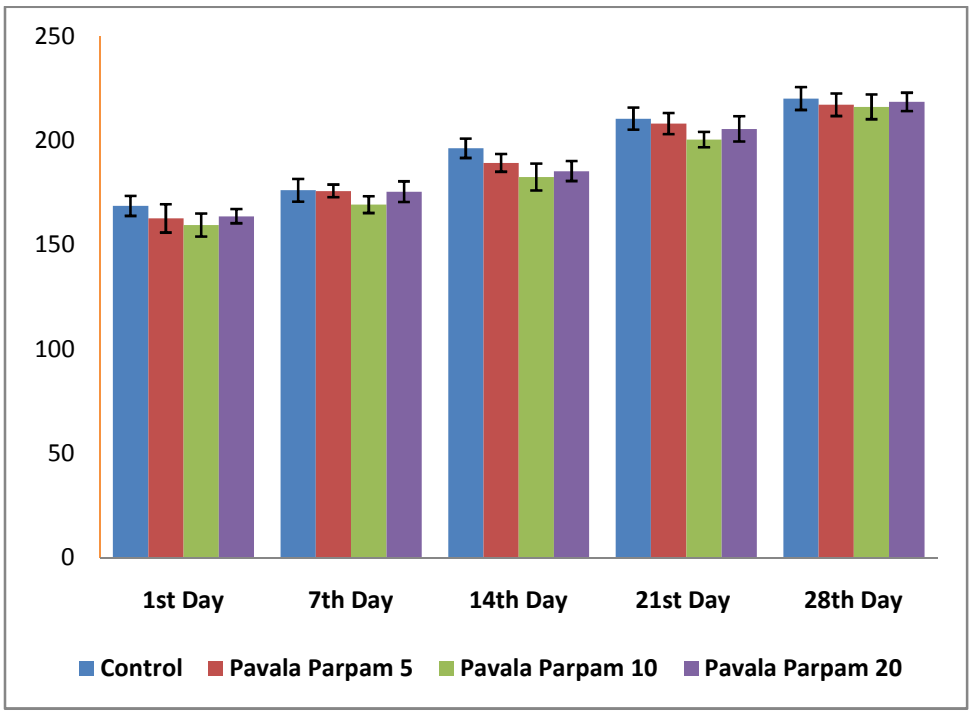


TABLE 22. EFFECT OF PAVALA PARPAM ON FOOD INTAKE DURING 28 DAYS TREATMENT IN RATS

Groups	Drug Treatment	Food Intake (gms)				
		1st Day	7th Day	14th Day	21st Day	28th Day
I	Control - Ghee (1ml/kg, p.o)	38.42±1.32	42.56±2.86	39.30±2.20	53.75±3.19	56.90±2.72
II	**Pavala Parpam** (5mg/kg, p.o)	45.66±2.15	58.73±1.97	54.00±3.98	47.74±3.28	58.25±4.45
III	**Pavala Parpam** (10mg/kg, p.o)	45.50±2.78	52.26±3.46	68.64±4.81	63.22±3.80	63.75±4.53
IV	**Pavala Parpam** (20mg/kg, p.o)	45.28±3.22	53.86±3.25	64.70±3.75	62.23±3.84	70.28±2.22

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 2. Effect of Pavala Parpam on food intake during 28 days treatment in rats

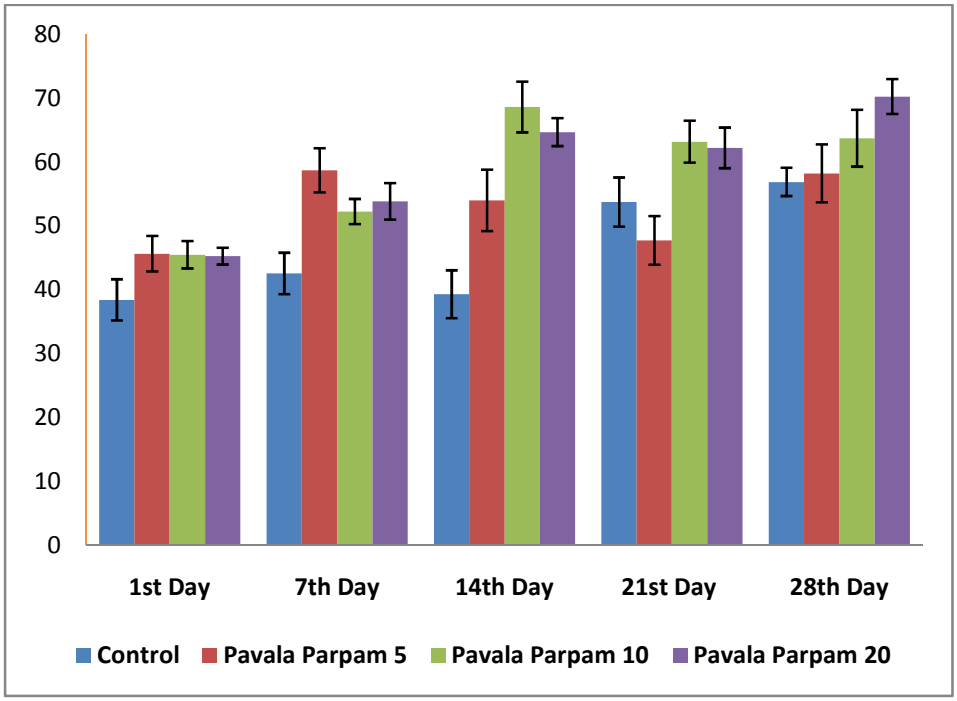


TABLE 23. EFFECT OF PAVALA PARPAM ON WATER INTAKE DURING 28 DAYS TREATMENT IN RATS

Groups	Drug Treatment	Water Intake (ml)				
		1st Day	7th Day	14th Day	21st Day	28th Day
I	Control - Ghee (1ml/kg, p.o)	51.66±2.02	58.76±4.22	64.00±3.90	70.34±4.73	65.76±4.80
II	**Pavala Parpam** (5mg/kg, p.o)	50.84±3.46	78.31±4.87	60.24±4.00	72.28±5.42	66.26±5.96
III	**Pavala Parpam** (10mg/kg, p.o)	60.90±3.55	70.00±3.90	70.48±5.46	68.87±5.87	76.23±5.32
IV	**Pavala Parpam** (20mg/kg, p.o)	75.26±4.76	73.23±5.84	70.54±4.62	80.76±5.62	82.64±4.66

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 3. Effect of Pavala Parpam on water intake during 28 days treatment in rats

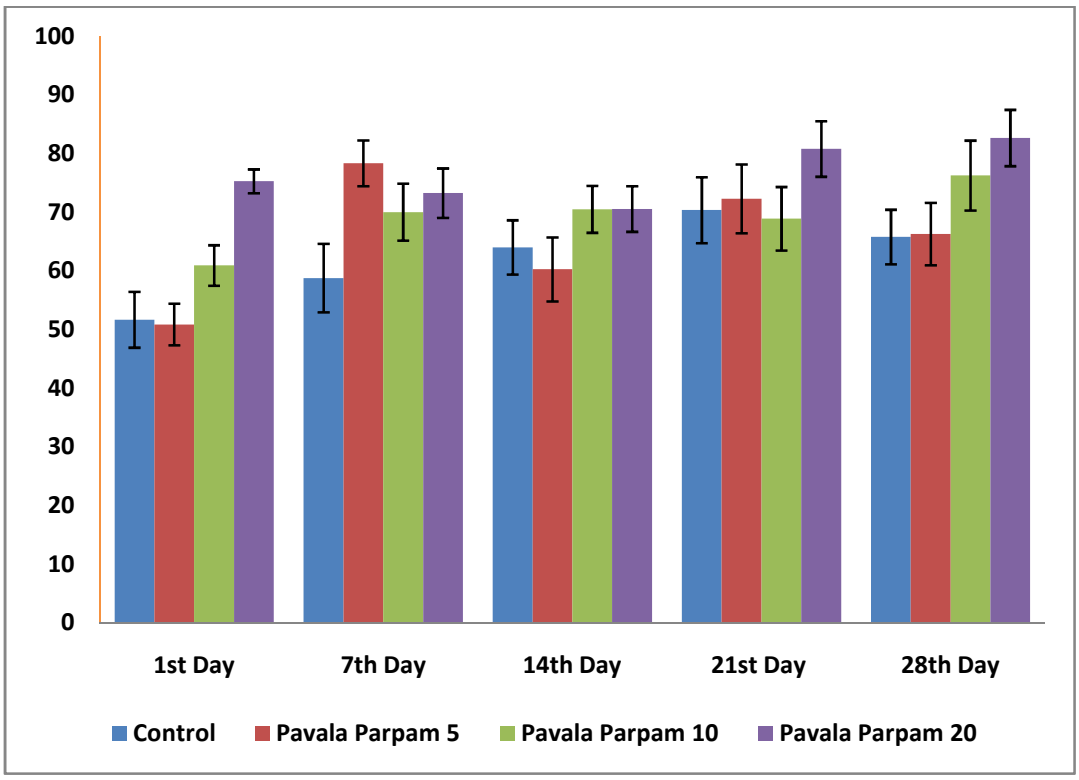


TABLE 24. SHOWS THE EFFECT OF PAVALA PARPAM ON RBC, WBC AND HB IN RATS AFTER 28 DAYS TREATMENT

Groups	Drug Treatment	RBC million cells/cmm	WBC cells/cmm	Haemoglobin gm %
I	Control - Ghee (1ml/kg, p.o)	4.57 ±0.16	8638.52±87.66	11.77±0.28
II	**Pavala Parpam** (5mg/kg, p.o)	4.42 ±0.16	8627.37±107.88	12.14±0.25
III	**Pavala Parpam** (10mg/kg, p.o)	4.40 ±0.14	8536.87±154.34	11.18±0.37
IV	**Pavala Parpam** (20mg/kg, p.o)	4.33 ±0.10	8694.20±16.19	11.31±0.43

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 4. Shows the effect of Pavala Parpam on RBC and Hb in rats after 28 days treatment

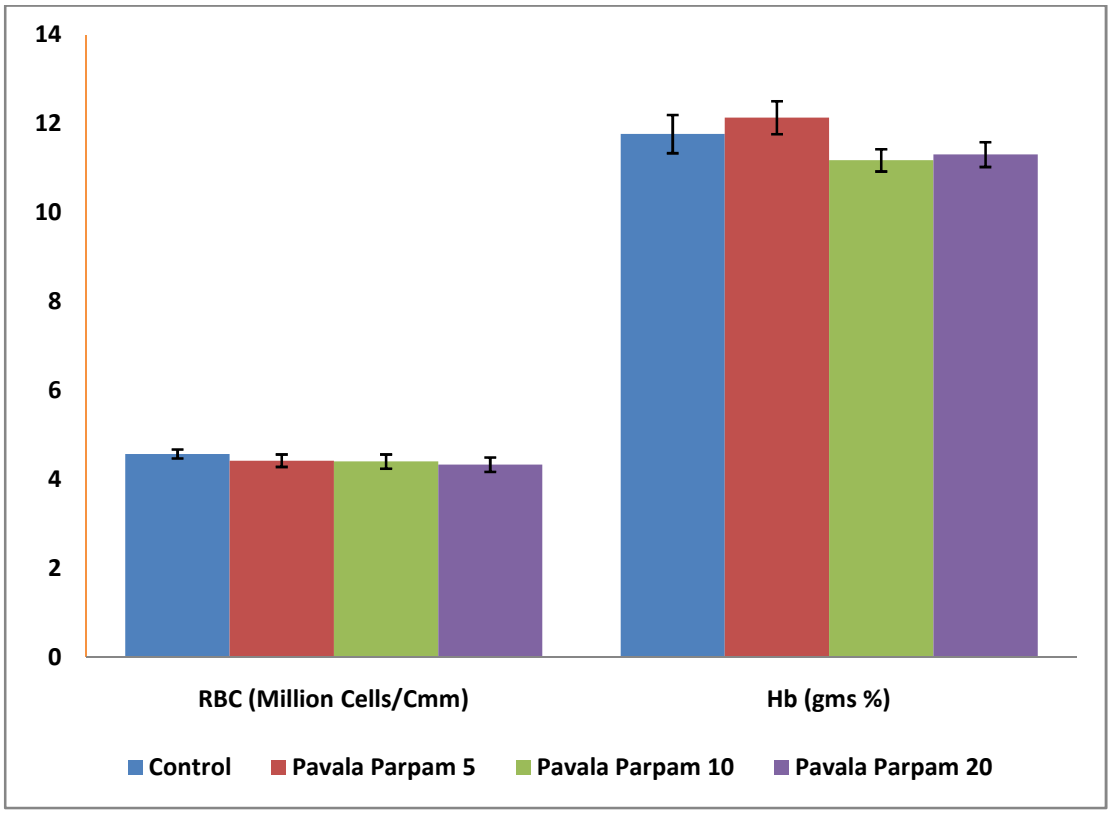


Figure 5. Shows the effect of Pavala parpam on WBC in rats after 28 days treatment

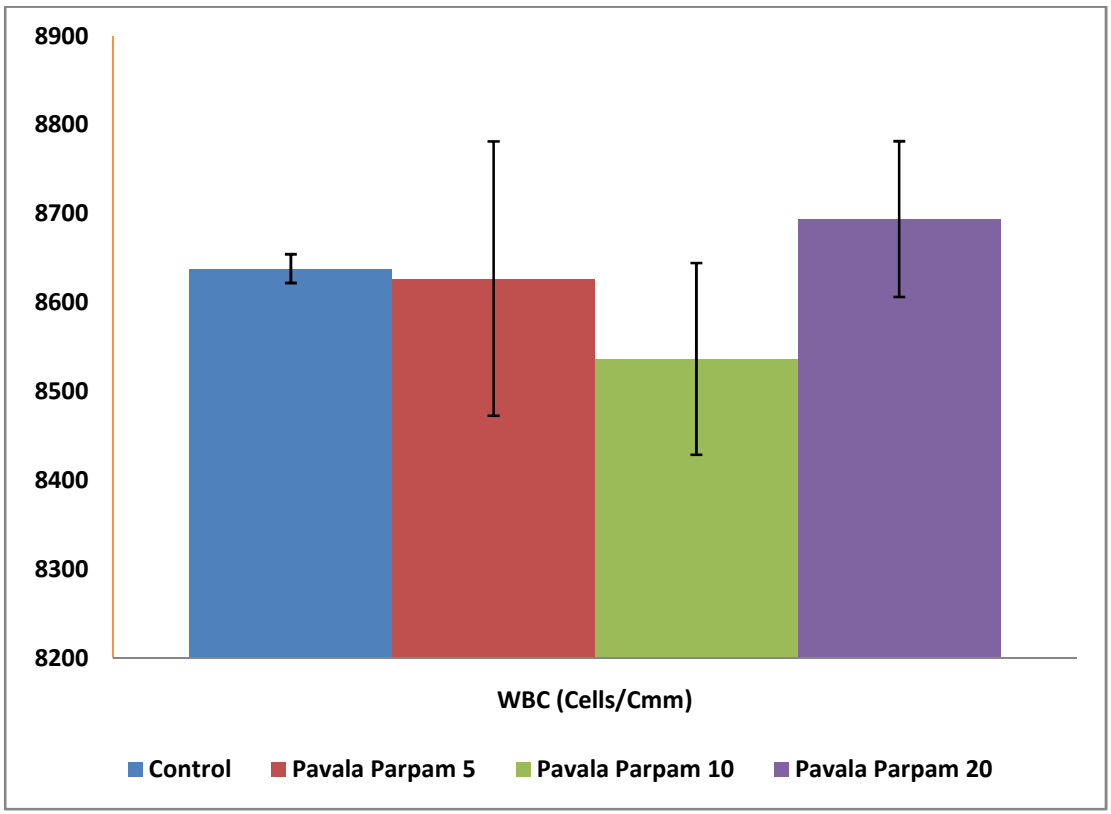


TABLE. SHOWS THE EFFECT OF PAVALA PARPAM ON DIFFERENTIAL COUNT IN RATS AFTER 28 DAYS TREATMENT

Groups	Drug Treatment	Differential Count %			
		Neutophils	***Eosinophils***	***Monocyte***	***Lympocyte***
I	Control - Ghee (1ml/kg, p.o)	30.00±1.79	1.17±0.31	3.33±0.42	65.83±1.64
II	**Pavala Parpam** (5mg/kg, p.o)	29.50±0.22	1.00±0.05	2.83±0.70	66.17±1.35
III	**Pavala Parpam** (10mg/kg, p.o)	28.00±0.63	1.20±0.06	3.20±0.49	67.20±1.46
IV	**Pavala Parpam** (20mg/kg, p.o)	26.40±0.51	1.40±0.09	3.00±0.71	69.00±0.71

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 6. Shows the effect of Pavala Parpam on Differential Counts in rats after 28 days treatment

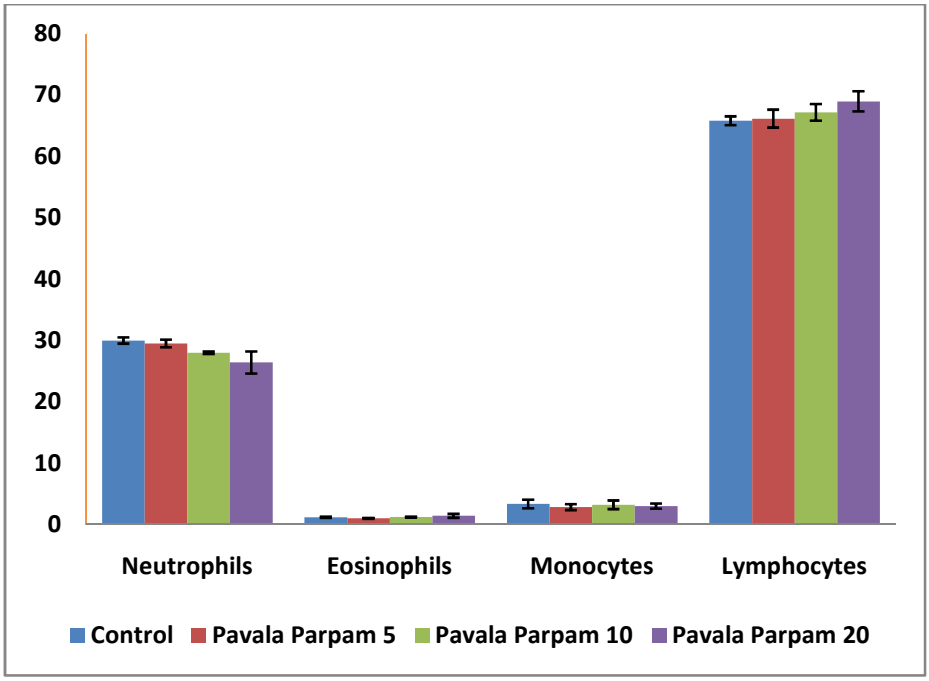


TABLE. 25 SHOWS THE EFFECT OF PAVALA PARPAM ON HEPATIC FUNCTIONS (SGPT, SGOT AND ALP) IN RATS AFTER 28 DAYS TREATMENT.

Groups	Drug Treatment	SGPT (IU/L)	SGOT (IU/L)	ALP (IU/L)
I	Control - Ghee (1ml/kg, p.o)	83.83±1.42	150.17±4.59	270.83±4.17
II	**Pavala Parpam** (5mg/kg, p.o)	82.00±2.02	153.00±2.02	279.50±3.82
III	**Pavala Parpam** (10mg/kg, p.o)	88.40±2.66	166.60±3.53	287.40±2.77
IV	**Pavala Parpam** (20mg/kg, p.o)	108.20±2.08*	185.00±2.12*	302.20±2.27*

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 7. Shows the effect of Pavala Parpam on Hepatic Functions in rats after 28 days treatment

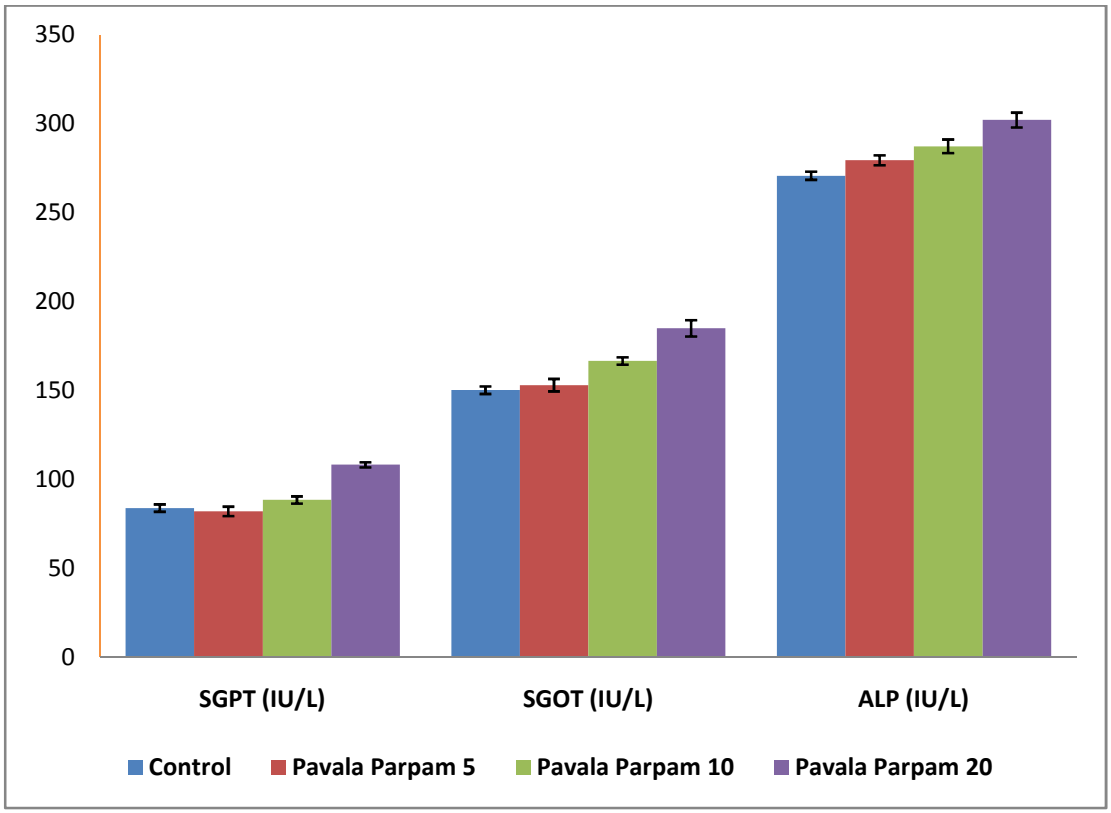


TABLE. SHOWS THE EFFECT OF PAVALA PARPAM ON KIDNEY FUNCTIONS IN RATS AFTER 28 DAYS TREATMENT

Groups	Drug Treatment	Urea (mg/dl)	Creatinine (mg/dl)
I	Control - Ghee (1ml/kg, p.o)	34.67±1.12	0.84±0.07
II	**Pavala Parpam** (5mg/kg, p.o)	35.17±1.58	0.85±0.09
III	**Pavala Parpam** (10mg/kg, p.o)	47.00±2.65	1.15±0.16
IV	**Pavala Parpam** (20mg/kg, p.o)	64.60±1.36**	2.12±0.04**

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 8. Shows the effect of Pavala Parpam on Kidney Functions (Blood Urea and Creatinine) in rats after 28 days treatment

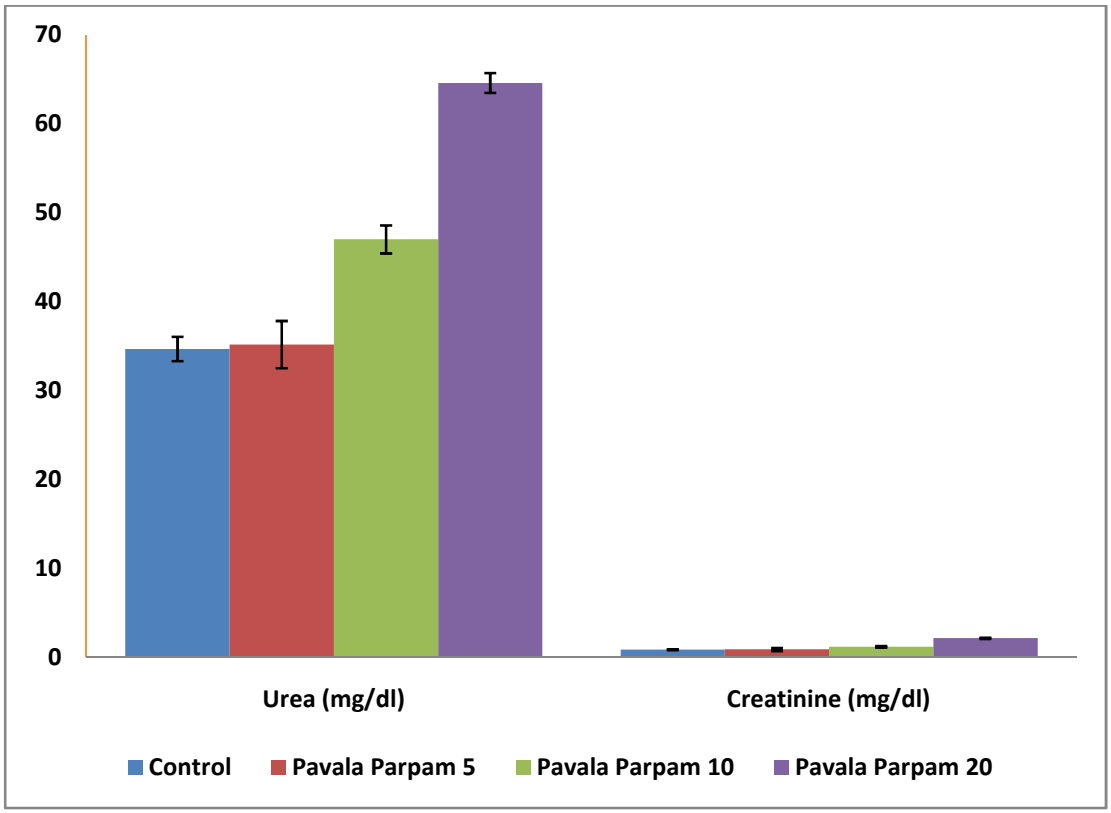


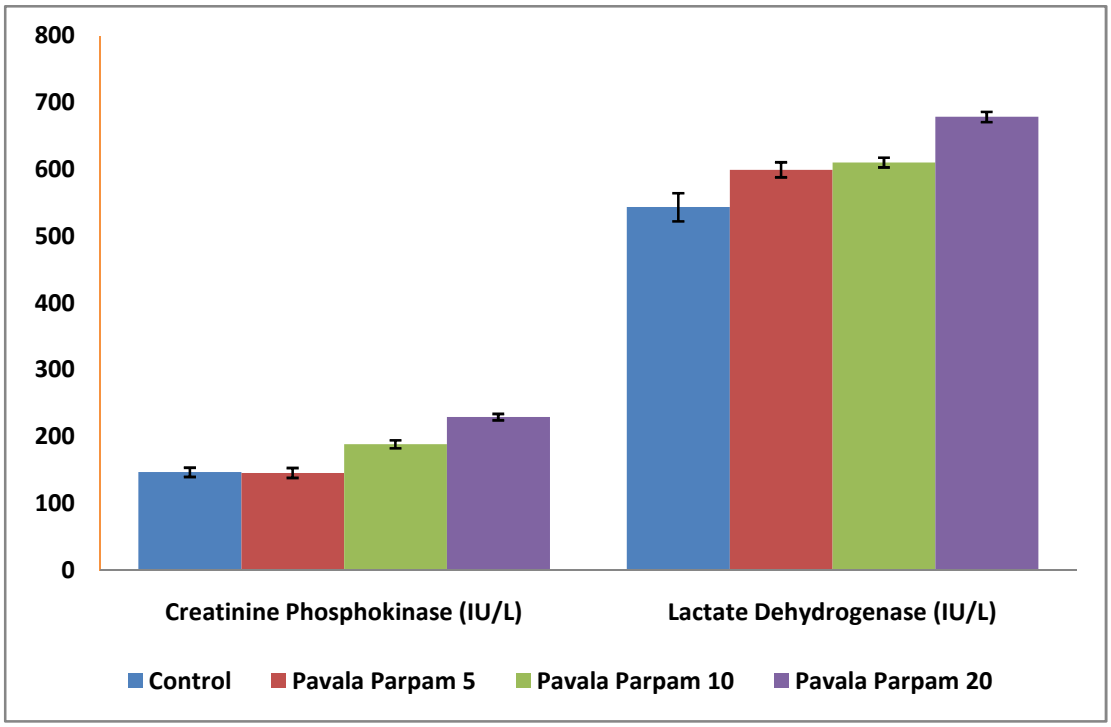
TABLE . SHOWS THE EFFECT OF PAVALA PARPAM ON CARDIAC FUNCTIONS IN RATS AFTER 28 DAYS TREATMENT

Groups	Drug Treatment	Creatinine Phosphokinase (IU/L)	Lactate Dehydrogenase (IU/L)
I	Control - Ghee (1ml/kg, p.o)	146.83±4.79	543.50±7.72
II	**Pavala Parpam** (5mg/kg, p.o)	145.83±6.03	599.50±7.36
III	**Pavala Parpam** (10mg/kg, p.o)	188.60±7.37*	610.40±11.29
IV	**Pavala Parpam** (20mg/kg, p.o)	229.40±6.96**	678.60±20.99*

Values are in mean ± SEM (n=6)

*P<0.05, **P<0.01, ***P<0.001 Vs Control

Figure 9. Shows the effect of Pavala Parpam on Cardiac Functions (Creatinie Phosphokinase and Lactate Dehydrogenase and in rats after 28 days treatment

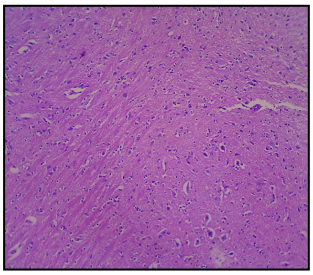


Name : **Ref. No. : [H0 373A/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

SPECIMEN **:** BRAIN.

Group – I -A : PP (High Dose)

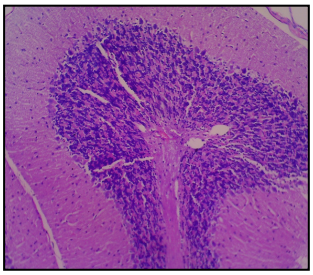


10x shows cortex_brain

GROSS APPEARANCE:

Received a specimen of brain measuring 3.2x2.5x1.8cms.

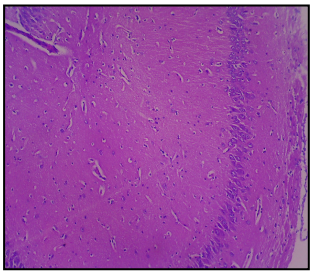
(PE): One bit – One block.



10x shows cerebellum

MICROSCOPIC APPEARANCE:

Section from brain with cerebral cortex, Cerebellum and Hippo campus shows no significant pathology. There is no evidence of toxicity.



10x shows hippocampus

Dr.C.R.Ajeeth kumar. M.D. (Path),

Consultant pathologists:
Dr.S.Kalyani.M.D. (Path), Dr. C.R.Ajeeth kumar.M.D. (Path),

Checked

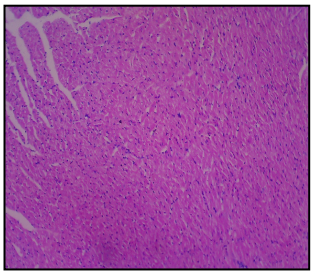
Name : **Ref. No. : [H0 373B/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

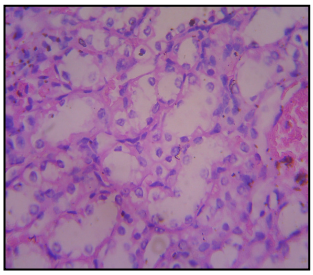
Toxicity study

SPECIMEN : Heart.

Group – I-A : PP (High Dose).



10x shows myocardium with myocytes_heart



10x shows with blood vessels shows normal

GROSS APPEARANCE:

Received a specimen of heart measuring 1.4 x1.0x0.5cms (PE): One bit – One block.

MICROSCOPIC APPEARANCE:

Section studied from the heart shows myocardium with myocytes showing no significant pathology. The blood vessel shows normal.

Dr.C.R.Ajeeth kumar. M.D. (Path),

Consultant pathologists:
Dr.S.Kalyani.M.D. (Path), Dr. C.R.Ajeeth kumar.M.D. (Path),

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Name : **Ref. No. : [H0 373C/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Lungs.

Group – I-A : PP (High Dose)

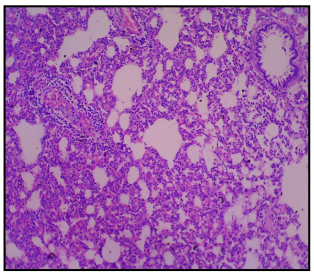
GROSS APPEARANCE:

Received a specimen of lung measuring 2.5x2.2x1.6cms.

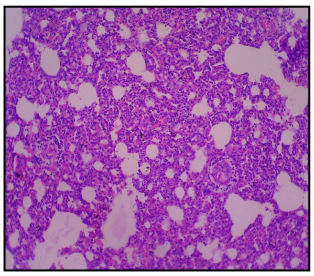
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

Section from lung shows normal alveoli, bronchioles.
Blood vessels show congestion.
Septal wall shows chronic inflammatory infiltrates.



10x shows alveoli with bronchioles_ lungs



10x shows chronic inflammatory infiltrates

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Dr.S.Kalyani.M.D. (Path), Dr. C.R.Ajeeth kumar.M.D. (Path),
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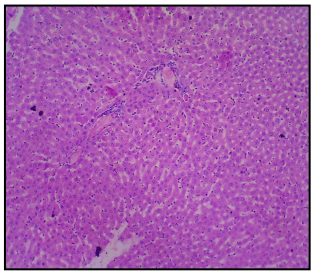
Name : **Ref. No. : [H0 373D/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Liver.

Group – I-A : PP (High Dose)

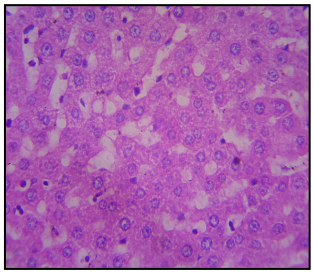


10x shows lobular architecture

GROSS APPEARANCE:

Received a specimen of liver measuring 3.8x2.4x1.5cms.

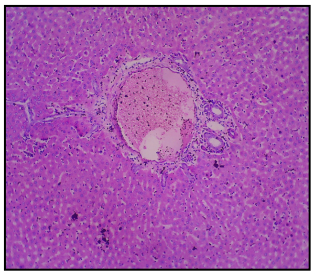
(PE): Two bits – One block.



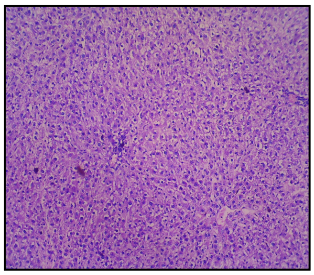
10x shows individual hepatocytes

MICROSCOPIC APPEARANCE:

The sections from the liver shows normal lobular architecture. Individual hepatocytes show no significant pathology. Portal traid shows mild periportal inflammation and bile duct hyperplasia. Sinusoids show mild dilatation. Central vein shows congestion.



10x shows central vein congestion with periportal inflammation



10x shows sinusoidal dilation

Dr.C.R.Ajeeth kumar. M.D. (Path),

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Name : **Ref. No. : [H0 373E/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Spleen.

Group – I-A : PP (High Dose)

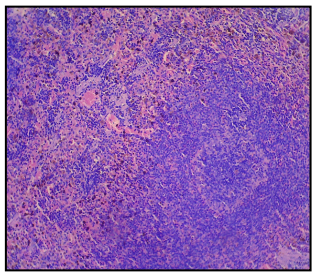
GROSS APPEARANCE :

Received a Specimen of spleen measuring 3.3cms in length.

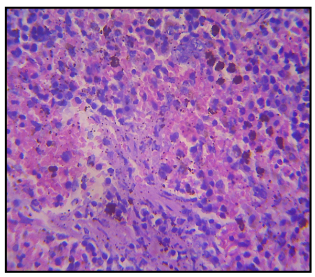
AE: Two bits – One block.

MICROSCOPIC APPEARANCE:

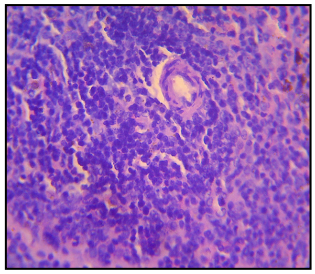
Section studied from the spleen shows normal red pulp and hemosiderin laden macrophages. White pulp shows prominent lymphoid aggregates with germinal centre formation. The pencillar artery shows normal. No evidence of toxic changes.



10x shows normal red and white pulp spleen



10x hsows normal red pulp with hemosiderin laden macrophages



10x shows normal white pulp with pencillar artery _ testis

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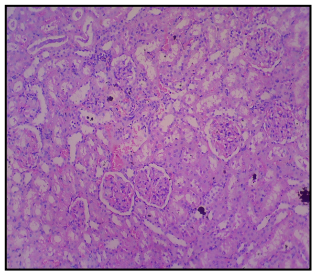
Name : **Ref. No. : [H0 373F/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Kidneys

Group – I-A : PP (High Dose)

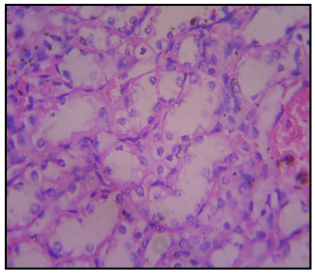


10x shows normal glomeruli and tubules kidney

GROSS APPEARANCE:

Received a specimen of kidneys each measuring 1.2x0.7x0.6cms and 1.2x0.6x0.5cms.

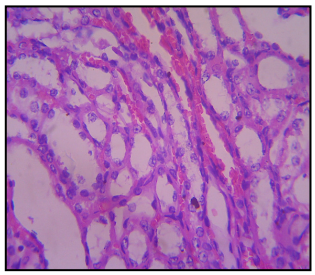
(PE): Two bits – One block.



10x shows normaltubules with blood vessels congestion

MICROSCOPIC APPEARANCE:

Section studied from the kidney shows both cortex and medulla. The Glomeruli show normal morphology. The tubules show no significant pathology. Interstitium shows unremarkable. Blood vessels show congestion.



10x shows interstitium with blod vessels congestion

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Name : **Ref. No. : [H0 373G/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Testis.

Group – I-A : PP (High Dose)

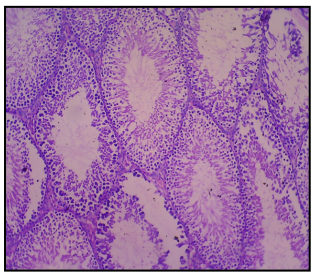
GROSS APPEARANCE:

Received a specimen of testis each measuring 2.3x1.0x0.6cms and 2.1x1.0x0.5cms.

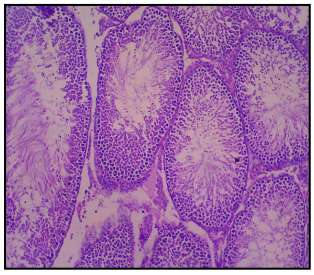
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

Section from testis with seminiferous tubules showing normal spermatogenesis with varying stages of maturation. Leydig cells are normal in number and distribution. There is no evidence of toxic changes.



10x shows step wise maturation testis



10x shows normal tubules testis

Dr.C.R.Ajeeth kumar. M.D. (Path),

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Name : **Ref. No. : [H0 374A/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

SPECIMEN **:** BRAIN.

Group – II-A : PP (Mid Dose)

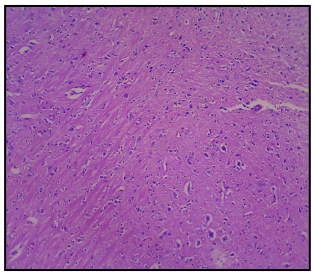
GROSS APPEARANCE:

Received a specimen of brain measuring 3.5x2.5x1.6cms.

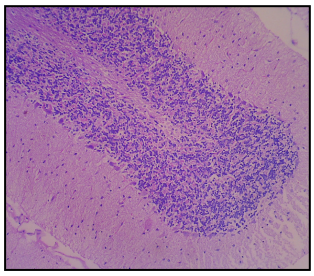
(PE): One bit – One block.

MICROSCOPIC APPEARANCE:

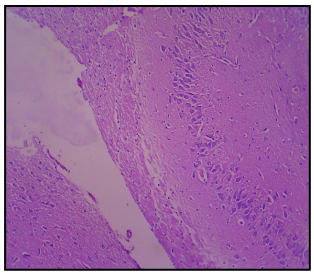
Section from brain shows normal cerebral cortex, cerebellum and hippo campus showing normal morphology. There is no evidence of toxicity.



10x shows cortex_brain



10x shows cerebellum



10x shows hippocampus

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Name : **Ref. No. : [H0 374B/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Heart.

Group – II-A : PP (Mid Dose)

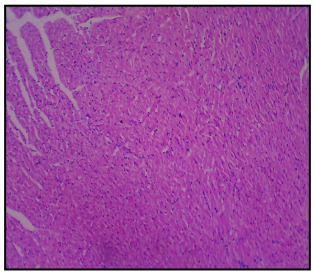
GROSS APPEARANCE:

Received a specimen of heart measuring 1.5 x0.8x0.6cms

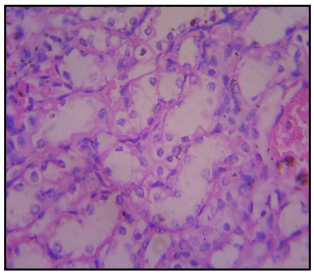
(PE): One bit – One block.

MICROSCOPIC APPEARANCE:

Section studied from the heart shows myocardium with myocytes showing no significant pathology. The blood vessel shows congestion.



10x shows myocardium with myocytes_heart



10x shows with blood vessels shows normal

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Name : **Ref. No. : [H0 374C/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

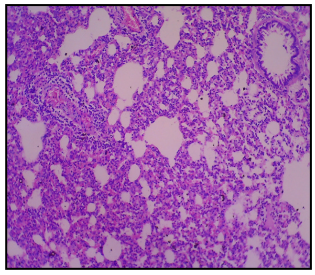
SPECIMEN : Lungs.

Group – II-A : PP (Mid Dose)

GROSS APPEARANCE:

Received a specimen of lung measuring 3.0x2.2x1.4cms.

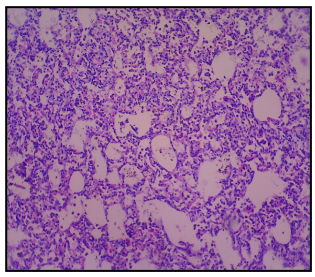
(PE): Two bits – One block.



10x shows alveoli with bronchioles lungs

MICROSCOPIC APPEARANCE:

Section from lung shows normal alveoli, bronchioles. Blood vessels show congestion. There is presence of interstitial inflammation.



10x shows scattered inflammatory infiltrates and septal wall congestion

Dr.C.R.Ajeeth kumar. M.D. (Path),

Consultant pathologists:
Dr.S.Kalyani.M.D. (Path), Dr. C.R.Ajeeth kumar.M.D. (Path),

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Name : **Ref. No. : [H0 374D/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Liver.

Group – II-A : PP (Mid Dose)

GROSS APPEARANCE:

Received a specimen of liver measuring 4.2x2.8x1.6cms.

(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

The sections from the liver shows mild altered lobular architecture. Individual hepatocytes show cytoplasmic vacuolation. Portal traid shows mild periportal inflammation and bile duct hyperplasia. Sinusoids show mild dilatation. Central vein shows no significant pathology.

Dr.C.R.Ajeeth kumar. M.D. (Path),

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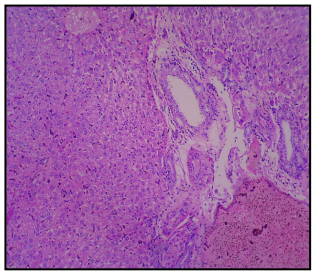
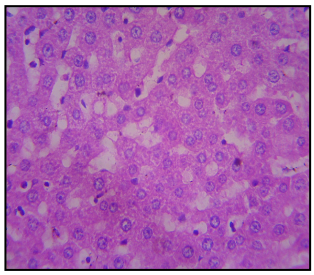
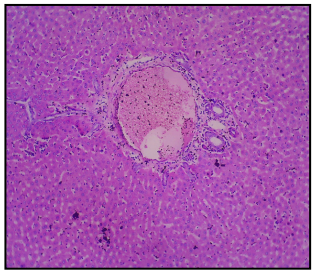
Checked



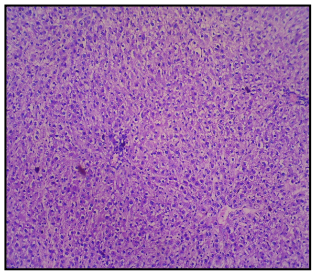
10x shows mild altered lobular architecture



10x shows individual hepatocytes



10x shows central vein congestion with periportal inflammation



10x shows sinusoidal dilation

Name : **Ref. No. : [H0 374E/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Spleen.

Group – II-A : PP (Mid Dose)

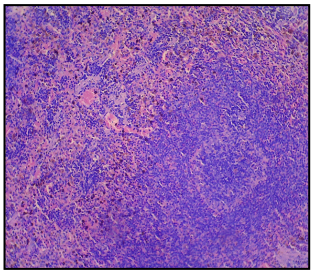
GROSS APPEARANCE :

Received a Specimen of spleen measuring 3.6cms in length.

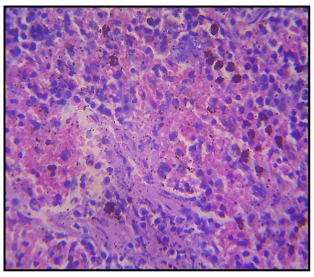
AE: Two bits – One block.

MICROSCOPIC APPEARANCE:

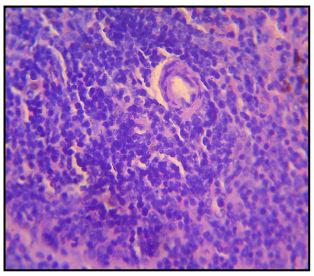
Section studied from the spleen shows normal red pulp and hemosiderin laden macrophages. White pulp shows prominent lymphoid aggregates with germinal centre formation. The pencillar artery shows normal. No evidence of toxic changes.



10x shows normal red and white pulp spleen



10x hsows normal red pulp with hemosiderin laden macrophages



10x shows normal white pulp with pencillar artery _ testis

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Name : **Ref. No. : [H0 374F/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Kidneys

Group – II-A : PP (Mid Dose)

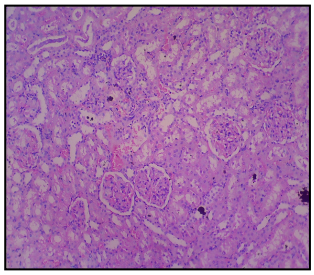
GROSS APPEARANCE:

Received a specimen of kidneys each measuring 1.4x0.8x0.5cms and 1.3x0.7x0.5cms.

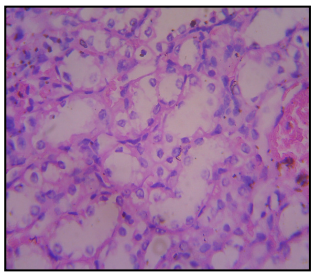
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

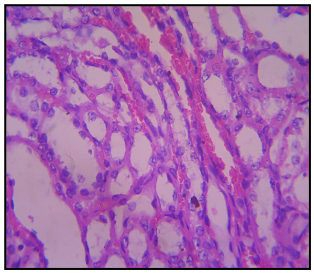
Section studied from the kidney shows both cortex and medulla. The Glomeruli show normal morphology. The tubules show no significant pathology. Interstitium shows unremarkable. Blood vessels show unremarkable.



10x shows normal glomeruli and tubules kidney



10x shows normaltubules with blood vessels congestion



10x shows interstitium with blood vessels unremarkable

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Name : **Ref. No. : [H0 374G/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN **:** Ovary.

Group – II-A : PP (Mid Dose)

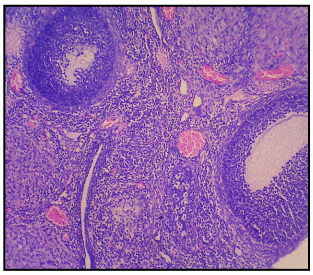
GROSS APPEARANCE:

Received a specimen of ovaries each measuring 0.4cms and 0.3cms.

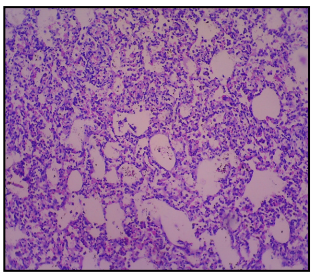
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

Section from ovary shows varying stages of ovarian follicle showing normal development with oocyte in the center. Stroma shows focal mild inflammatory infiltrates. Blood vessels show congestion.



10x shows normal ovarian follicles ovary



10x shows scattered inflammatory infiltrates and septal wall congestion

Dr.C.R.Ajeeth kumar. M.D. (Path),

Consultant pathologists:
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Name : **Ref. No. : [H0 375A/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

SPECIMEN **:** BRAIN.

Group – III-A : PP (Low Dose)

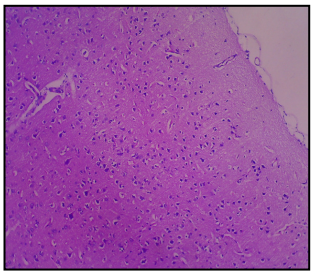
GROSS APPEARANCE:

Received a specimen of brain measuring 3.8x2.6x1.5cms.

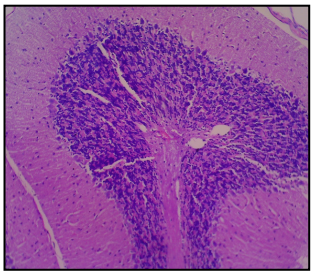
(PE): One bit – One block.

MICROSCOPIC APPEARANCE:

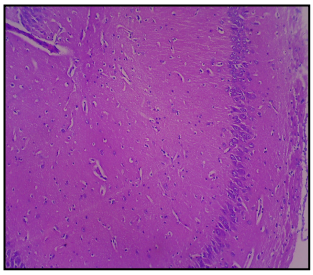
Section from brain with cerebral cortex, cerebellum hippo campus shows no significant pathology. There is no evidence of toxicity.



10x shows cortex_brain



10x shows cerebellum



10x shows hippocampus

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Name : **Ref. No. : [H0 375B/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Heart.

Group – III-A : PP (Low Dose)

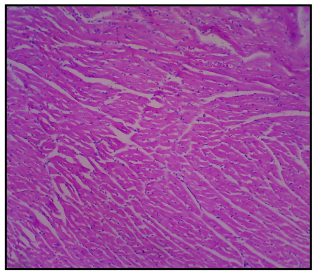
GROSS APPEARANCE:

Received a specimen of heart measuring 1.5 x1.0x0.5cms

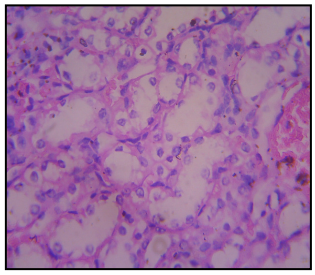
(PE): One bit – One block.

MICROSCOPIC APPEARANCE:

Section studied from the heart myocardium shows mild myocyte degeneration extravasated rbc's. The blood vessel shows normal.



40x shows mild degenration with extravasated rbcs



10x shows with blood vessels shows normal

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Name : **Ref. No. : [H0 375C/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Lungs.

Group – III-A : PP (Low Dose)

GROSS APPEARANCE:

Received a specimen of lung measuring 3.0x2.0x1.4cms.

(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

Section from lung shows normal alveoli. Bronchioles show mild peribronchiole inflammation. There is presence of septal wall congestion and scattered lymphocytic infiltrates.

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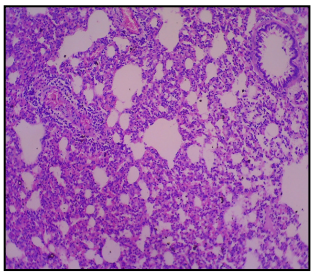
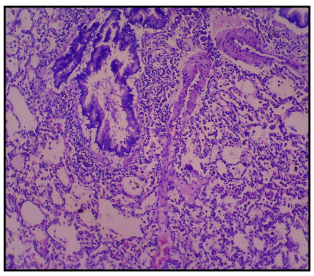
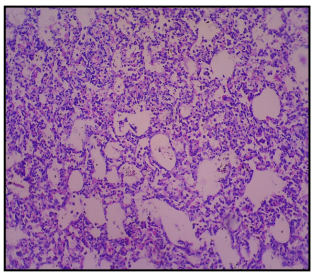
Checked



10x shows alveoli with bronchioles lungs



10x hows peri bronchial region shows inflammatory infiltrates



10x shows scattered inflammatory infiltrates and septal wall congestion

Name : **Ref. No. : [H0 375D/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Liver.

Group – III-A : PP (Low Dose)

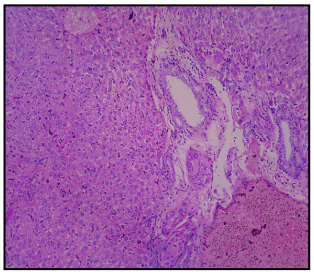
GROSS APPEARANCE:

Received a specimen of liver measuring 4.0x2.5x1.3cms.

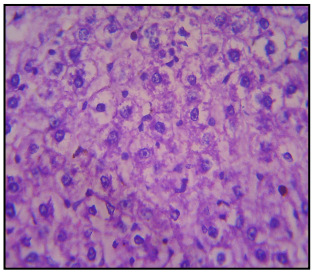
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

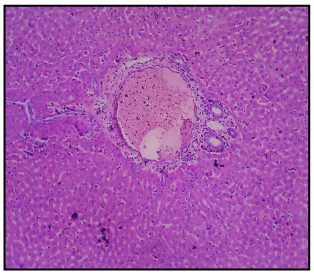
The sections from the liver shows lobular architecture. Individual hepatocytes show cytoplasmic vacuolation. Portal traid shows mild periportal inflammation. Sinusoids show mild dilatation. Central vein shows no significant pathology.



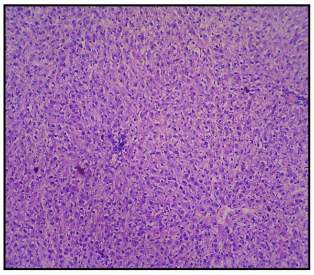
10x shows mild altered lobular architecture



shows cytoplasmic vacuolation



10x shows central vein congestion with periportal inflammation



10x shows sinusoidal dilation

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Name : **Ref. No. : [H0 375E/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Spleen.

Group – III-A : PP (Low Dose)

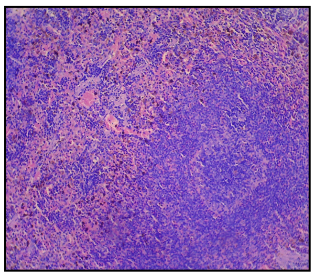
GROSS APPEARANCE :

Received a Specimen of spleen measuring 3.7cms in length.

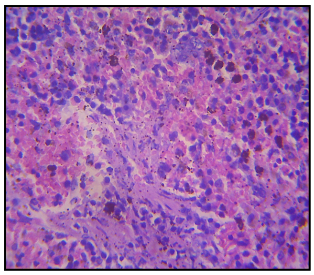
AE: Two bits – One block.

MICROSCOPIC APPEARANCE:

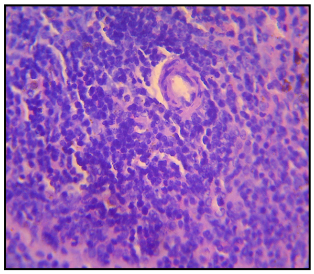
Section studied from the spleen shows normal red pulp and hemosiderin laden macrophages. White pulp shows prominent lymphoid aggregates with germinal centre formation. The pencillar artery shows normal. No evidence of toxic changes.



10x shows normal red and white pulp spleen



10x shows normal red pulp with hemosiderin laden macrophages



10x shows normal white pulp with pencillar artery _ testis

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Name : **Ref. No. : [H0 375F/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

SPECIMEN : Kidneys

Group – III-A : PP (Low Dose)

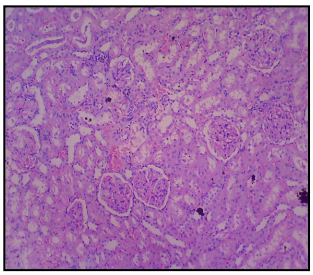
GROSS APPEARANCE:

Received a specimen of kidneys each measuring 1.4x0.8x0.6cms and 1.3x0.6x0.6cms.

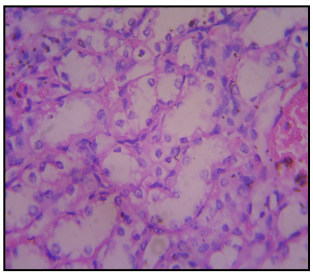
(PE): Two bits – One block.

MICROSCOPIC APPEARANCE:

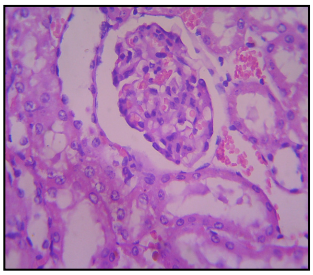
Section studied from the kidney shows both cortex and medulla. The Glomeruli show normal morphology. The tubules show no significant pathology. Interstitium shows unremarkable. Blood vessels show congestion.



10x shows normal glomeruli and tubules kidney



10x shows normaltubules with blood vessels congestion



40x shows normal glomeruli and tubules with blood vessels

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Name : **Ref. No. : [H0 375G/19]**	**Rec.On : 13/04/2019** **Rep.On : 29/04/2019**

HISTOPATHOLOGY

Toxicity study

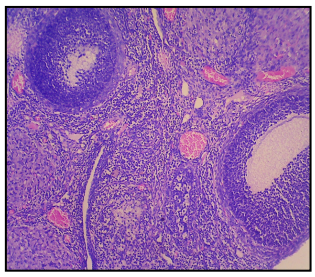
SPECIMEN : Ovary.

Group – III-A : PP (Low Dose)

GROSS APPEARANCE:

Received a specimen of ovaries each measuring 0.4cms and 0.3cms.

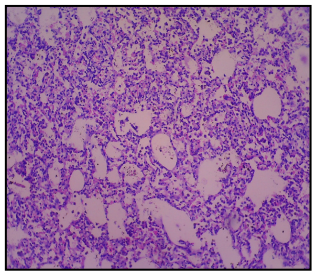
(PE): Two bits – One block.



10x shows normal ovarian follicles ovary

MICROSCOPIC APPEARANCE:

Section from ovary shows varying stages of ovarian follicle showing normal development with oocyte in the center. Stroma shows focal mild inflammatory infiltrates. Blood vessels show congestion.



10x shows scattered inflammatory infiltrates and septal wall congestion

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Checked

6.0 RESULTS:

CLINICAL SIGNS:

All animals in this study were free of toxic signs throughout the dosing period of 28 days.

Mortality:

All animals in control and in all the treated dose groups survived throughout the dosing period of 28 days.

Body weight:

The effect of ***PAVALA PARPAM*** on body weight during 28 days treatment in rats was given in table 21. There was no significant change in the body weight compared to control with both the doses of ***PAVALA PARPAM*** during 28 days treatment.

Food consumption:

The effect of ***PAVALA PARPAM*** on food intake during 28 days treatment in rats was given in table 22. ***PAVALA PARPAM*** did not alter the food intake at both the dose levels as compared to control during the 28 days treatment. It indicates that it does not influence food intake.

Water consumption:

The effect of ***PAVALA PARPAM*** on water intake during 28 days treatment in rats was given in table 23. ***PAVALA PARPAM*** did not alter the water intake at both the dose levels as compared to control during the 28 days treatment. There was no significant change in water intake as compared to control.

Organ Weight:

Group Mean Relative Organ Weights (% of body weight) are recorded in Table No.24 Comparison of organ weights of treated animals with respective control animals on day 28 was found to be comparable similarly.

Hematological investigations:

The results of hematological investigation (Table.25) conducted on 28th day revealed following significant changes in the values of different parameters investigated when compared with those of respective controls; however, the increase or decrease in the values obtained was within normal biological and laboratory limits or the effect was not dose dependent.

Biochemical Investigations:

Results of Biochemical investigations conducted on 28th day and recorded in revealed the following significant changes in the values of hepatic serum enzymes studied. When compared with those of respective control. However, the increase or decrease in the values obtained was within normal biological and laboratory limits.

Histopathology:

In histopathological examination, revealed normal architecture in comparison with control and treated animal.

DISCUSSION

The results of acute toxicity study of PAVALA PARPAM were shown on table 3. There was no mortality with the Pavala Parpam after 1hr & 24 hrs even at higher dose of 2000mg/kg. The Pavala Parpam did not alter the general behavior after 1hour of oral administration with mild increase in motor activity. After 24hrs of PAVALA PARPAM oral administration, it showed significant increase in motor activity without altering other general behavior. It did not show any lethality or toxic reactions during and after the study. From the dose mention for human in literatures, 5, 10 and 20mg/kg were selected for further sub-acute toxicity study.

In sub-acute toxicity study, body weight, food intake and water intake were observed on 1st, 7th, 14th 21st and 28th day of Pavala Parpam administration. The effect of Pavala Parpam on body weight during 28 days treatment in rats was given in table 4 and figure 1. There was no significant change in the body weight compared to control with all the three doses of Pavala Parpam during 28 days treatment.

The effect of Pavala Parpam on food intake during 28 days treatment in rats was given in table 5 and figure 2. Pavala Parpam did not alter the food intake at both the dose

levels as compared to control during the 28 days treatment. It indicates that it does not influence food intake.

The effect of Pavala Parpam on water intake during 28 days treatment in rats was given in table 6 and figure 3. Pavala Parpam did not alter the water intake at both the dose levels as compared to control during the 28 days treatment. There was no significant change in water intake as compared to control.

Table 7, figure 4 and 5, shows the effect of Pavala Parpam on haematological parameters like RBC, WBC and Hb in rats after 28 days treatment. Both the doses of Pavala Parpam did not produce any significant change in RBC, WBC and Hb compared to control.

The effect of Pavala Parpam on Differential Count in rats after 28 days treatment was shown on table 8 and figure 6. All the three doses of Pavala Parpam did not show any significant change in differential counts like Neutrophils, Eosinpophils, Monocyte and Lympocytes. From the effect of Pavala Parpam on hematological parameters it was found that it does not produce any toxicity in haemopoietic system.

The effect of Pavala Parpam on hepatic functions in rats after 28 days treatment was shown on table 9 and figure 7. The hepatic enzymes (SGPT, SGOT and ALP) were remaining normal with 5 and 10mg/kg of Pavala Parpam. But with the higher dose of Pavala Parpam i.e. 20mg/kg significantly ($P<0.05$) increase the levels of SGPT, SGOT and ALP.

The effect of Pavala Parpam on renal functions in rats after 28 days treatment was shown on table 10 and figure 8. Low and intermediate dose i.e. 5 and 10mg/kg of Pavala Parpam does not showed any significant change in urea and creatinine after 28 days treatment compared to control. But the higher dose 20mg/kg of Pavala Parpam significantly ($P<0.01$) increased the urea and creatinine compare to control.

The effect of Pavala Parpam on Cardiac functions in rats after 28 days treatment was shown on table 11 and figure 9. Low dose of Pavala Parpam (5mg/kg) did not alter the cardiac biomarker enzymes as compared to control animals. Pavala parpam at 10 mg/kg significnaly ($P<0.01$) increase the level of Creatinine Phosphokinase and did not alter the levels of Lactate Dehydrogenase compare to control. But the higher dose 20mg/kg of Pavala parpam significantly increase the levels of both Creatinie Phosphokinase ($P<0.01$) and Lactate Dehydrogenase ($P<0.05$) after 28 days administration, compared to control.

CONCLUSION

Pavala Parpam was studied for its acute and sub-acue toxicity studies using laboratory animals. In acute toxicity study, Pavala Parpam did not produce any specific toxicity or mortality even at the dose of 2000mg/kg in mice, but it slightly enhance the motor activity. In sub-acute toxicity study, 5, 10 and 20mg/kg of Pavala Parpam was used and it was administered once daily for 28 days through oral route. Pavala Parpam did not alter the body weight, food intake and water intake during the study period. After 28 days the blood was subjected to Hematological, liver, kidney and cardiac function test. All the three doses (5, 10 and 20mg/kg) of Paval parpam did not showed and change in hematological parameter. The higher dose of Pavala parpam (20mg/kg) showed mild toxic effect in Liver, kidney and cardiac functions by elevating its specific biomarkers like SGPT, SGOT, ALP, Urea, Creatinine, CK phosphokinase and Lactate Dehydrogenase. Form the study it was concluded that, Low and moderate dose of Pavala parpam was found to be safe in laboratory animals.

BIOSTATISTICAL ASPECTS

Biological assay refers to assessment of the potency of vitamins, hormones, toxicants and drugs of all types by means of the responses produced when doses are given to experimental animals. In every dose response situation, two components must be considered; the Stimulus and the Subject.

The stimulus is applied to the Subject as a stated dose namely concentration, weight, time or appropriate measure. The subject manifest a response, the level of intensity below which the response does not occur & above which the response occur, such a value has often been called threshold. But the term tolerance is now widely accepted.

MEDIAN EFFECTIVE DOSE (ED50)

It is the dose which produces the desired response in half the animal population tested.

MEDIAN LETHAL DOSE (LD 50)

It is the dose which kills half the population of the animal tested.

LD50 Measurment(Toxicity)

- If the test compound shows any pharmacological activity then the LD50 of the drug is determined.
- By determining the LD50, we can justify whether to proceed with the drug or not.

Table - 26

Acute Toxicity Study Analysis

Group	Dose in mg / kg	No. of rats	No. of rats died
I	Distilled water (1ml/kg)	3	-
II	5	3	-
III	50	3	-
IV	300	3	-
V	2000	3	-

Since there was no mortality of the animal in acute toxicity study, lethal dose of drug could not be calculated.

Table - 27

Subacute Toxicity Study Analysis

Group	Dose (mgs / kg)	No. of rats	Days	No. of rats died
I	Control	6 (3 M + 3F)	28	-
II	5mg	6 (3 M + 3F)	28	-
III	10mg	6 (3 M + 3F)	28	-
IV	20mg	6 (3 M + 3F)	28	-

In case of Subacute Toxicity Study, with the help of physiological parameters such as Hematological investigations and with the histopathological studies the drug reaction within the animal can be assessed and are being tabulated respectively.

Lethal dose of the drug ***"PAVALA PARPAM "*** can be calculated with higher dose level of the drug which can be done in further studies.

From the above biostatistical measures ***"PAVALA PARPAM"*** is safe upto the dose level 20mg/kg body weight of the animal.

8. DISCUSSION

- ✓ The present study with ***PAVALA PARPAM*** was conducted with an objective to find out whether this drug has got any side effects or adverse reactions in short and Long term administration
- ✓ ICP OES analysis indicates that Aluminium, Arsenic, Cadmium, Copper, Mercury, Nickel and Lead are in below detection Limit.
- ✓ FTIR analysis of ***PAVALA PARPAM*** indicates the presence of Aromatic compounds, Alkynes, Alkene, Alkyl halide, Alcohol, Acyl chloride, Esters, Carboxylic acids, Amides and Amines.
- ✓ SEM analysis indicated that the particle size were in Range 2 - 1 micron.
- ✓ Biochemical Analysis of ***PAVALA PARPAM*** indicates the presence of Calcium, Sulphate, Chloride, Carbonate and Ferrous iron.
- ✓ On the basis of acute toxicity study results the study shows that ***PAVALA PARPAM*** did not produce any toxic up ot the dose of 2000mg/kg to rats.
- ✓ On the basis of subacute toxicity Results, the study reveals that ***PAVALA PARPAM*** did not cause either any lethality or adverse changes with general behaviour in rats and also there were no observable detrimental effects (5 -20 mg/kg body weight) over a period of 28 day.
- ✓ In histopathological examination revealed normal architecture in comparison with control and treated animal.
- ✓ Haematological analysis revealed no abnormalities attributable to the treatment.
- ✓ These results indicate that ***PAVALA PARPAM*** upto 5 - 20mg/kg body weight did not produce any toxicity effects in long term administration.

9. SUMMARY

- ✓ The medicine ***PAVALA PARPAM*** was taken for the dissertation work based on ***Kannu Sammy, Parambarai Vaithiyam, Page No :385, 386***
- ✓ The aim of this dissertation is to study the acute and sub-acute toxicity of the medicine ***PAVALA PARPAM*** administered at various presumed moderate dosage, in the experimental animals.
- ✓ The Ingredients of ***PAVALA PARPAM*** are Pavalam and Thaivelai. The pavalam were purchased from fishermen of Tiruchendur Seashore and Thaivelai collected from Moolikulam region.
- ✓ The raw samples were taken for purification and the test medicine was prepared, as per the method narrated in the literature.
- ✓ The drug was analysed for its physicochemical properties and contents by using qualitative biochemical analysis and modern techniques such as inductively couped plasma-optical emission spectrometry.
- ✓ Depending upon the result of these analysis the contents of test sample was identified.
- ✓ By scanning electron microscope (SEM), the size of the particles about 2-1 micron, were analyzed.
- ✓ The study was done at Department of Pharmacology, Nandha College of Pharmacy, Erode District.
- ✓ To evaluate the acute toxicity study 15 rats were selected and divided into 5 groups (Group I,II,III,IV,V) and they were administered with the drug with different graded doses ranging from Control, 5mg/kg, 50mg/kg, 300mg/kg and 2000mg/kg body weight of animal orally with control group. Daily the animals were observed for clinical signs and mortality. The drug did not produce any mortality and is safe upto 2000mg/kg body weight.
- ✓ Sub acute Toxicity was conducted for about 28 days duration. No signs of toxicity was observed in animals from different dose groups during the dosing period.
- ✓ The haematological index shows no significant changes

- ✓ During long term administration of the drugs at both low dose and high dose SGOT, SGPT, Serum Urea, Serum Creatinine level found to be within the normal range.
- ✓ Biostatistical measures to the acute and subacute toxicity studies shows the drugs ***"PAVALA PARPAM*** " found to be safe up to 2000mg/kg body weight of the animal in acute toxicity study and found to be safe upto 20mg/kg body weight of the animal in sub-acute toxicity study.

In this study since there is no mortality, the lethal dose of drug could not be calculated

10. CONCLUSION

From acute toxicity study it was observed that the administration of ***PAVALA PARPAM*** up to the dose of 2000 mg/kg to the Wistar Albino Rats did not produce drug-related toxicity and mortality. So No-Observed-Adverse-Effect- Level (NOAEL) of ***PAVALA PARPAM*** is 2000 mg/kg.

The subacute toxicity studies also reveals that the drug ***"PAVALA PARPAM"*** can be considered safe, as it did not produce either any lethality or adverse changes with general behaviour of rats and also there were not observable determental effects in the doses (5 to 20mg/kg body weight) over a period of 28 days. It is concluded that the "***PAVALA PARPAM*** " is relatively safe in long administration upto the dose of 20mg/kg.

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