PHARMACOLOGICAL STUDIES OF AN AYURVEDIC PREPARATION, **SUKRAMATRIKA VATI.**

A research report submitted in partial fulfillment of the requirements for the degree of Bachelor of Pharmacy

EAST WEST UNIVERSITY

DEPARTMRNT OF PHARMACY

Fall 2009

Submitted by: Shara Khan
ID: 2006-1-70-042

CERTIFICATION

This is to certify that, the thesis "Pharmacological studies of an Ayurvedic preparation, Sukramatrika Vati" submitted to the Department of Pharmacy, East West University, Mohakhali, Dhaka, in partial fulfillment of the requirements for the degree of Bachelor of Pharmacy (B. Pharm) was carried out by Shara Khan (ID# 2006-1-70-042) under our guidance and supervision and that no part of the thesis has been submitted for any other degree. We further certify that all the sources of information and other facilities availed of in this connection is duly acknowledged.

27.12.09.

Mr. Apurba Sarker Apu
Lecturer (Supervisor)
Department of Pharmacy
East West University
43, Mohakhali C/A,
Dhaka-1212

Prof. Dr. Chowdhury Faiz Hossain
Chairperson
Department of Pharmacy
East West University
43, Mohakhali C/A
Dhaka-1212

ACKNOWLEDGEMENT

At the beginning I would like to mention that the success of my entire project was only possible because of the combined effort and cooperation of my respected teachers and group members who showed unperturbed dedication, patience and hard work. For me, this is a great chance to show my gratefulness and admiration.

I want to convey my heartiest gratefulness to my honourable teacher and research guide, Mr Apurba Sarker Apu, Lecturer, East West University, who showed immense dedication throughout the progress of my project and provided me with constant support with his much admired expert qualities.

I would like to take this opportunity to communicate my heartfelt gratitude to Dr. Muniruddin Ahmed, former chairperson of Department of Pharmacy and current Pro-Vice Chancellor, East West University, for giving me the opportunity to conduct such an interesting project in collaboration with Jahanginagar University.

Then, of course, it is quite a privilege for me to express my sincere appreciation to the honourable Chairperson of the Department of Pharmacy, Dr. Chowdhury Faiz Hossain, who facilitated the smooth conduction of my project work.

I would also like to acknowledge Dr. Shahabuddin Kabir Chowdhury, Chairperson of the Department of Pharmacy, Jahangirnagar University, for giving me the great opportunity to work in the Pharmacology Laboratory of Jahangirnagar University without placing any restrictions on laboratory facilities or sharing his knowledge to solve my queries.

Next, would like to thank Ms. Syeda Seraj, Shahed Parvez, Sabrina Nasrin and Mr. Abdul Kader Jilani for their instant and constant guidance during data transformation and arrangement of the paper.

My special thanks and gratefulness are towards my fellow group members Ashfaqur Rahman, Tamzid Zaman, Mehnaz Islam Ferdousi, Tanzina Mollick and Habib Abdullah Md Sakil.

Thank you.

TABLE OF CONTENTS

LIST OF TABLES

	in Hole Board Test
3.2.2.2	Tabular representation of the effect of SMB (200 mg/kg) in ambulation in Hole Board Test
3.2.2.3	Tabular representation of the effect of SMB (400 mg/kg) in ambulation in Hole Board Test
3.2.3.1	Tabular representation of the effect of SMB (100 mg/kg) in head dipping in Hole Board Test
3.2.3.2	Tabular representation of the effect of SMB (200 mg/kg) in head dipping in Hole Board Test
3.2.3.3	Tabular representation of the effect of SMB (400 mg/kg) in head dipping in Hole Board Test
3.3.1.1	Tabular representation of the effect of SMB (100 mg/kg) in ambulation in Open Field Test
3.3.1.2	Tabular representation of the effect of SMB (200 mg/kg) in ambulation in Open Field Test
3.3.1.3	Tabular representation of the effect of SMB (400 mg/kg) in ambulation in Open Field Test
3.3.2.1	Tabular representation of the effect of SMB (100 mg/kg) in center ambulation in Open Field Test
3.3.2.2	Tabular representation of the effect of SMB (200 mg/kg) in center ambulation in Open Field Test
3.3.2.3	Tabular representation of the effect of SMB (400 mg/kg) in center

LIST OF GRAPHS

ABSTRACT

Purpose: The research was carried out in order to study the pharmacological aspects and possible side effects of the marketed cost-effective Ayurvedic formulation, Sukramatrika Vati mainly indicated for the treatment of urolithiasis; based on analgesic, neuropharmacological and gastrointestinal effects on animal models.

Method: The research project was conducted using five previously established pharmacological models proposed by various scientists. The experiments were executed at three dose levels of 100 mg/kg, 200 mg/kg and 400 mg/kg on animal models (*Swiss albino*) in order to determine the various degrees of significance on activity resulted by Sukramatrika Vati

Result: Sukramatrika Vati treated mice at dose 100 mg/kg showed a decrease in writhing response in the 2^{nd} minute which was statistically significant ($p = 0.047$). In addition, the overall reduction in pain perception from minutes 1 to 5 was found to be statistically significant ($p = 0.043$). In the hole board test, at the dose of 200mg/kg, the reduction in emotional defaecation at minute 240 was observed to be statistically significant ($p = 0.044$). Apart from these, Sukramatrika Vati treated mice at the dose of 400mg/kg showed an overall decrease in ambulation in the open field test. The results obtained at minute 60 and 120 were statistically highly significant ($p = 0.004$) and ($p = 0.004$). The reading obtained at minute 180 was also statistically significant ($p = 0.010$).

Conclusion: The findings of the experiments on the animal models show that possibly Sukramatrika Vati may act as a useful analgesic for the palliative treatment of urolithiasis. In addition, the results show that the preparation probably also possesses no anxiogenic effects. Further studies using sophisticated models need to be performed to fully fathom the safety of Sukramatrika Vati for Alternative Medical Care.

Keywords: Sukramatrika Vati, urolithiasis, ayurvedic formulation, pharmacological aspects.

Chapter one

Introduction

1.1 Ayurveda

Ayurveda is a Sanskrit word that means "the knowledge of life span." It is considered the most ancient existing medical system and originated around thousands of years ago in India. It is a method of healing that embraces herbal, mineral, and animal therapies, dietary modification, meditation, yoga, aroma therapy, and music therapy (Kieley *et al* in 2008). The central philosophy of Ayurvedic system of medicine is based on the maintenance of health through the even balance of the three vital humors (*doshas*) - *vata* (to move), *pitta* (to heat or burn) and *kapha* (to keep together). Each one has its role to play in the body. For example, *vata*, which combines ether and air, produces movement and relates mainly to the nervous system and the body's energy. *Pitta*, which combines fire and water, relates to the metabolism, digestion, enzymes, acid, and bile. *Kapha*, which combines earth and water, relates to the mucous membranes, phlegm, moisture, fat, and lymphatics. The balance of the three *doshas* depends on a variety of factors, in particular correct diet and exercise, good digestion, healthy elimination of body wastes, and balanced emotional and spiritual health.

1.1.1 Ayurveda over the Millennia

Originally, Ayurveda was an oral tradition that was eventually recorded over 5,000 years ago, according to the University of Minnesota Center for Spirituality and Healing. The four basic texts of historic Ayurveda are called Vedas. They are – Rigveda, Yajurveda, Samaveda and Atharvaveda. Ayurveda is part of this fourth Veda.

Another set of texts that are part of the "great triad," also referred to as the Brhattrayi, are the Caraka Samhita and Sushruta Samhita. These are texts that were recorded more than 2,000 years ago and are the main tenants of modern Ayurvedic medicine that still exist in their entirety and explain the eight branches of Ayurvedic medicine. These sections include:

Internal medicine, Surgery, Organic medicine, Pediatrics, Toxicology, Rejuvenating remedy, Aphrodisiac remedies and Spiritual healing. These eight sections are called "Astanga Ayurveda".

According to many scholars, knowledge of Ayurveda originated from India and influenced the ancient Chinese system of medicine and medical system practiced in Greece. Thus, Ayurveda is also known as the "Mother of all Healing".

Ayurvedic medicine is practiced as the primary medical system in India, as well as in Bangladesh, Sri Lanka, Nepal and Pakistan. In the United States it is considered complementary and alternative medicine (CAM) and more than 200,000 adults in the U.S. used Ayurveda in 2006, according to the National Center for Complementary and Alternative Medicine (NCCAM).

1.1.2 Ayurveda Today

According to World Health Organization (WHO) report, over 80% of the world population relies on plant-based traditional medicine for their primary healthcare needs and remedies, and the use of traditional medicines is rising in the developed economies such as Europe and America. The professional practice of Ayurveda in the United States began to grow and became more visible in the late 20th century. Recapitulation and adaptation of the older science to modern drug discovery processes can bring renewed interest to the pharmaceutical world and offer unique therapeutic solutions for a wide range of human disorders.

A huge number of Ayurvedic products of different manufacturer are available commercially for various types of diseases. Officially recognized by the government of Bangladesh shortly following independence, Unani and Ayurvedic drugs were brought under a drug control system in 1982 to provide oversight of manufacturing and marketing. Given the success and extensive presence of traditional medicine in Bangladesh, the government is considering

incorporating it in mainstream primary health care services. Such action is considered a cost-effective, comparatively expedient manner of providing health coverage to large segments of the rural population. In order to implement and institutionalize the Ayurvedic Medical System and also to strengthen and widen the range of services in the District hospitals and Thana Health Complexes, the provision of Alternative Medicine in 30 selected District hospitals have began in 1998 under the 1998-2003 plan of HPSP (Health and Population Service Program).The global market for herbal and Ayurvedic medicine is estimated to be more than $60 billion a year and many people in the West are showing growing interests. Allopathic pharma market in Bangladesh is worth around 4,000 crore BDT, while the market size for herbal medicines including Ayurvedic and Unani stands at more than 1,000 crore BDT. Bangladesh government termed herbs and herbal medicine as one of the five priority sectors to diversify the country's export basket. Bangladesh has prospect in making footsteps on the global market for medicinal plant and products as nearly 650 medicinal plant species have been identified to be in use in Bangladesh with around 25 plants having high value.

Though time-tested evidences show immense therapeutic benefits of these Ayurvedic drugs, there are very little pharmacologically established data to support these drugs for using against different diseases and for other benefits. By using Ayurvedic medicine expensive and extensive procedures of clinical investigations can be avoided in many cases and people in these selected areas have the choice to get treatment at a cheaper price depending on their choice (Islam, H.A. 1991)

1.2 Urolithiasis and Ayurveda

Formation of stones in the urinary tract is a global phenomenon and is described in ancient Ayurvedic scriptures as urolithiasis (*mutraashamari).* It is said to be one of the eight most troublesome diseases (*mahaorgas*). Ayurvedic texts have classified the stones according to

doshic profiles, namely, *vata*-, *pitta*-, or *kapha* -related and others. *Doshic* involvement is indicative of the biochemical influences in the formation of stones.

The formation of a stone (calculus) can be at any level in the urinary system. These stones are most frequently formed in the kidney, but they pass farther down the urinary tract toward the bladder. They are intensely painful as they pass along the ureters and out through the urethra. There are useful management and herbal treatments for urolithiasis that have been currently investigated extensively. It is thus important to understand the Ayurvedic concept of urolithiasis and to explore the scientific basis of Ayurvedic therapies (Sridevi,V. *et al* in 2004).

1.2.1 Epidemiology

The overall probability of forming stones varies in different parts of the world. The risk of developing urolithiasis in normal adults appears to be lower in Asia (1 to 5%) than in Europe (5 to 9%) and in North America (12% in Canada, 13% in the U.S.). The highest risk was reported in Saudi Arabia (20.1%). The compositions of stones and their location in the urinary tract, bladder, or kidneys may also significantly differ in different countries. Moreover, in the same region, the clinical and metabolic patterns of stone disease can change over time. In India, bladder stones accounted for 30% of all urinary stones in 1965, but their prevalence had dropped to 5% in 1985. Concurrently, the chemical composition of stones in the upper urinary tract changed; the prevalence of calcium oxalate stones rose from 26 to 82%, and the prevalence of struvite stones fell from 20 to 5%. In Japan, bladder stones decreased from 50 to 5% from 1950 to 1985. In Portugal, over a 20-year period, the prevalence of calcium stones rose from 64 to 82%, struvite stones decreased from 14 to 3%, and uric acid stones decreased from 19 to 12% (Sridevi,V. *et al* in 2004).

One in every six people in Bangladesh suffers from kidney diseases and 40,000 die annually from kidney-related complications, according to the health experts and doctors. About 20 million people are now suffering from kidney ailments as against 10 million a decade ago. The number of kidney patients is increasing in the country at an alarming rate as about 1.8 crore people are suffering from kidney disease. Around 40,000 people die of kidney failure each year in the country as about 95% people cannot afford the expensive kidney treatment. (Alam, M. 2009)

1.2.2 Definition

Urolithiasis is defined as a stone or stone-like hard substance formed in the urinary tract. The definition is consistent with current knowledge of urolithiasis — the accretion of hard, solid, nonmetallic minerals in the urinary tract consisting of a nucleus of organic material around which urinary salts are deposited in concentric layers.

1.2.3 Clinical Description

A renal calculus is similar to that of the kadamba flower. It is three-layered, resembles a stone, and is either hard or smooth in texture. The prodromal signs and symptoms described in Ayurvedic texts consist of severe pain around or near the urinary bladder region, suprapubic region, internal urethral orifice, testicles, and in the penis. Common symptoms include distension of urinary bladder, fever, anorexia, dense and turbid urine, dysuria, fatigue, and odor of urine resembling the smell of a sheep.

The major clinical features described are pain in the umbilical and suprapubic regions and in the penis; obstructed urinary flow; split voiding of urine; hematuria; honey colored or yellowish red urine, turbid urine; sandlike particles passing along with urine; pain aggravated by jumping, swimming, riding a horse or camel, climbing in upward direction; and polyurea.

All these clinical features are very much similar to those currently known. In conventional medicine, clinical features of stones vary according to their size, shape, and location of the stone and the nature of underlying condition. The most common complaint is intermittent dull pain in the loin or back increased by movement. Proteins, red cells, or leucocytes may appear in the urine (Sridevi,V. *et al* in 2004).

1.2.4 Clinical Features According to the Location of the Calculi

Renal Calculus

Pain is characterized by a fixed dull ache in the angle between the lower border of the last rib and lateral border of sacro spinalis. Pain is also felt anteriorly in the corresponding hypochondriac region. Pain worsens with movement like running, jumping, and climbing up stairs and eases with rest. Sudden gripping pain is felt in the loin and tends to radiate toward the groin. Patients may experience fitful sleep because of pain. Pain may be associated with hematuria and may be complained of either during or after an attack.

Ureteric Calculus

Ureteric colic starts as soon as the stone enters into the pelviureteric junction and recurs at shorter or longer intervals as long as the stone remains in the ureter. Ureteric colic ceases when the stone is ejected into the bladder or impacted in the ureter. When the stone is present in the upper one third of the ureter, pain starts in the loin or near the renal angle and gradually radiates to the groin. Pain is gripping and starts suddenly. The patient experiences fitful sleep because of pain, which is often associated with hematuria and may be complained of either during or after an attack. When the stone is at a lower level, pain commences rather anteriorly just above the iliac crest and is referred along the two branches of the genitofemoral nerve to

the testis in males, labia majora females, and anteromedial aspect of the thigh in both sexes. When the stones enter into the intramural part of the ureter in males, pain is referred to the tip of the penis, and the patient complains of strangury.

When the stone is impacted, colic ceases; a dull ache arises according to the site of impaction. Such pain varies in intensity, and it increases with exercise and is relieved by rest.

Vesical Calculus

With increased frequency of micturition, pain is often referred to the tip of penis or the labia majora and becomes aggravated by running and jolting. Children may scream and pull the prepuce for pain after micturition and experience hematuria at end of the micturition (Sridevi, V. *et al* in 2004).

1.2.5 Etiology

In Ayurveda the causes of urinary calculi are mainly nonadoption of the purificatory measures such as emesis, purgation, and medicated enemas in order to eliminate the vitiated *doshas* (toxic materials) and practice of unhealthy diets and lifestyles. These factors are responsible for the formation of calculi. They are primarily classified into two categories: unhealthy diet or excessive physical activity.

In conventional medicine, there are three primary factors considered responsible for stone formation. They are the supersaturation of stone-forming compounds in urine, the presence of chemical or physical stimuli in urine that promote stone formation, and the inadequate amount of compounds in urine that inhibit stone formation (e.g., magnesium, citrate).

Categories of specific risk factors for stone formation are listed below.

- ✓ Diet associated with stone formation — Vitamin A deficiency; a high-oxalate diet rich in purine levels; a diet high in protein from animal sources, glucose, or sucrose; etc.

- ✓ Medication associated with stone formation — Calcium supplements, vitamin D supplements, ascorbic acid in megadoses (4 g/day), sulfonamides, triamterene, indinavir, etc.
- ✓ Diseases associated with stone formation — Hyperparathyroidism, renal tubular acidosis (complete or partial), jejunoileal bypass, Crohn's disease, intestinal resection, malabsorptive conditions, sarcoidosis, hyperthyroidism, etc.
- ✓ Anatomical abnormalities associated with stone formation — Tubular ectasia, pelviureteral junction obstruction, calix diverticulum or calyceal cyst, ureteral stricture, vesicoureteral reflux, horseshoe kidney, etc.

The additional risk factors include habitually low urine volume, high urine excretion of calcium, uric acid and oxalate, low urine pH (uric acid and cystine are less soluble in acid urine), and high urine pH (struvite and calcium phosphate are less soluble in alkaline urine). Some of the biochemical processes not only become relevant here, but also lay the basis for the drug therapy.

The stone is the outcome of accretion of inorganic material around an organic nidus not soluble in its own solution. Urine, which is an end-point excreta in a liquid form, represents the biochemical status of a person's metabolism. For example, in normal urine, nephrocalcin is an acidic glycoprotein, rich in γ-carboxy glutamic acid, which inhibits calcium oxalate crystal growth. The nephrocalcin present in the organic matrix of calcium oxalate kidney stones resembles the nephrocalcin present in the urine of the patient from whom the stone was removed, but it differs from the nephrocalcin in normal urine. The stone's former nephrocalcin lacks γ-carboxy glutamic acid, which reduces to air-water interfacial films that are less stable than those formed by nephrocalcin from normal urine. It is safe to presume that the alteration of the biochemical quality of urine can help in the prevention of stone formation. The biochemical quality of urine can change with the quality and quantity of fluid

inputs, the type of diets, and the constitutional factors. This, in turn, can play a great role in the formation or nonformation of stones in the urine. Ayurveda suggests a number of herbs and herbal preparations for stone breakdown. It is possible that the therapeutic agents are capable of altering the chemical composition of the urine and its pH.

1.2.6 Pathology

In Ayurveda, the concept of renal calculi pathogenesis is indicated as when the *kapha dosha* is vitiated because of the etiological factors; *kapha* reaches to the urinary system and, with the help of *vata* and *pitta doshas*, dries up and forms the calculus. There is another similar opinion regarding the pathogenesis of urolithiasis.

Urinary concretions may vary greatly in size. There may be particles like sand anywhere in the urinary tract or large stones in the bladder. Staghorn calculi fill the whole pelvis and branch into the calyces and are usually associated with pyelonephritis. Deposits may also be present throughout the renal parenchyma, giving rise to nephrocalcinosis (Sridevi,V. *et al* in 2004).

1.2.7 Classification of Renal Calculi

Ayurvedic texts have described four types of urinary calculi: *sleshmaashmari, pittaashmari, vataashmari,* and *sukraashmari.*

- ✓ *Sleshmaashmari*— In *sleshmaashmari*, stones are white, unctuous, and as big as a hen's egg. They produce symptoms such as dysuria, cutting, incising, pricking pain, heaviness, and a cold sensation over area the bladder region.
- ✓ *Pittaashmari*— In *pittaashmari*, stones are reddish, yellowish, and blackish and resemble the color of honey. They produce a sucking type of pain, burning sensation, a warm feeling over the bladder region, and *ushnavata.*

- ✓ *Vataashmari*— In *vataashmari*, stones are dusty in color, hard, irregular, rough and nodular or spiny like the kadamba flower. Patients experience severe pain (and may scream); pull out the prepuce; and have difficulty when passing flatus, urine, and stools.
- ✓ *Sukraashmari*— *Sukraashmari* occurs in adults only. It is due to suppression of ejaculation for months or years and frequent coitus or coitus interruption. The semen to be ejaculated will be obstructed, condensed, and brought in-between the scrotum and penis (prostatic part of the urethra) by *vata*. This causes dysuria, scrotal swelling, and lower abdominal pain. The special characteristic of *sukraashmari* is that handling can dissolve it.

In conventional medicine, urinary calculi are classified according to their chemical components. Examples include uric acid and urates, calcium oxalates, calcium and ammonio-magnesium phosphate (Struvite), cystine, combinations of the preceding items, and drugs or their metabolites (e.g., phenytoin, triamterene) (Sridevi,V. *et al* in 2004).

1.2.8 Treatment

The management of urolithiasis in Ayurveda basically includes herbal formulas, alkaline liquid and surgical procedures. Newly formed calculi can be cured by herbal formulas, but chronic calculi have to be treated surgically.

1.2.9 Types of Management

Shamana Therapy

Palliative treatment includes administration of herbal drugs and herbal formulas orally. The palliative drugs used to treat renal calculi are analgesic, diuretic, and linthnotriptic agent and are able to balance *vata*. One such preparation is Sukramatrika Vati.

Shodhana Therapy

The idea here is that transmucosal fluxes are encouraged away from the kidney for removal of unwanted metabolites, thereby reducing the ionic load on the kidney filtration system. This may be considered as a type of dialysis procedure.

Alkali Therapy

Most of the alkaline materials (*kshara*) act as diuretics, lithotriptic, alkalizer, and antispasmodic agents. These pharmacological activities are shown to be effective in the management of different symptoms of urolithiasis (Sridevi,V. *et al* in 2004).

1.3 Sukramatrika Vati

It is a small tablet like form (vati) of the formulation that contains a number of herbs. It alleviates painful micturation and urinary calculus. (web)

Sukramatrka Vati is included (page 314) in the Bangladesh National Formulary of Ayurvedic Medicine 1992 (Approved by the Government of Bangladesh vide Ministry of Health and Family Welfare Memo No. Health-1/Unani-2/89/(Part-1) 116 dated 3-6-1991). Bangladesh National Formulary of Ayurvedic Medicine is compiled by the National Unani and Ayurvedic Formulary Committee and published by the Bangladesh Board of Unani and Ayurvedic Systems of Medicine, 38, Bangabandhu Avenue, Dhaka-1000 under the authority vested in the Board vide section 13(j) of the Bangladesh Unani and Ayurvedic practitioners Ordinance, 1983 in collaboration with the World Health Organization. Directorate of Drug Administration has issued Notification DA/Admin/1-10/96/6212 dated 19th October 1996 has issued license under Drug Act, 1940 and Rules there under and Drug (Control) Ordinance 1982 for local manufacture and sale in Bangladesh. (Published Bangladesh Gazette #24 Part

VI dated Thursday, June 11th 1998.) At present a good number of Ayurvedic manufacturers are manufacturing and marketing the Classical Ayurvedic Medicinal Preparation.

1.3.1 Drug Composition and Administration

The formulation of Sukramatrika Vati (SMB) in Ayurveda is given in Table 1.3.1.

1.3.1 Table of formulation of Sukramatika Vati.

Ingredients (in Sangkrit)	***Ingredients (in English)***	***Amount***
Goksura bija	Puncture vine fruit	24 g
Haritaki	Chebulic myrobalan	24 g
Bibhitaka	Beleric myrobalan	24 g
Amalaki	Indian gooseberry	24 g
Tejpatra	Cinnamon	24 g
Ela	Lesser cardomon	24 g
Rasanjana	Rasont	24 g
Dhanyaka	Coriander	24 g

Cavya	Gajj Pippal Herb	24 g
Jiraka	Cumin seed	24 g
Talisa patra	Himalyan silver fir	24 g
Tankana-shuddha	Borax (purified)	24 g
Dadima	Pomegranate	24 g
Guggulu-shuddha	Goo-gall(purified)	12 g
Rasa (parada)-shuddha	Mercury(purified)	48 g
Gandhaka-shuddha	Sulphur(purified)	48 g
Abhraka bhasma	Mica	48 g
Lauha bhasma	Iron	48 g

SMB is available in the form of vati or pills and it must be administered orally at a dose of 500 mg. The *anupana* (fluid vehicle taken with or after medicine) is with juice of pomagranate, goat's milk, or water.

1.4 Rationale from Economic Perspective

The progressive increase of the social cost for treating urolithiasis could be related to an increased incidence of the disease and/or to an increase of costs for diagnosing and treating renal stones. In the course of the last century, the incidence of renal stones has progressively increased in Europe, North America, and other industrialised countries. This has been explained in terms of changing social conditions and the consequent changes in eating habits. In contrast, renal stones were less frequent than in developing countries of the world but in the last 20 years investigators began to report high incidences of upper urinary stone disease also from some areas of the Third World concurring with the changing of economic and social conditions. Each stone episode involves the costs for emergency visits, diagnostic work up, and medical or surgical treatment. Furthermore, we have to consider the costs of follow-up visits and the costs of testing and drugs for stone prevention. In adjunct of direct costs for diagnosis and treatment, we should also take into account the indirect individual and social cost of workdays lost. Finally, we should estimate the costs of complications and outcomes of treatment with particular attention to the costs of chronic renal failure secondary to stone disease. The strategy of treatment of each stone centre involves different costs for the treatment of each single stone episode. On the other hand the choice of treatment can be driven by National Health Systems and insurance companies by their policy of reimbursement for different procedures. The trends of renal stone incidence will have different impact on health care systems in different countries. In Europe and North America, the peak of incidence has been probably reached but the increase of costs for diagnosing and treating each single stone episode will still increase the social cost for managing stone disease. For this reason the actual objective should be to optimise protocols avoiding redundant or expensive diagnostic procedures or inappropriate treatments. In developing countries, the incidence of stone disease is still increasing and it could reach peaks even

higher as a consequence of hot climate in some geographical areas. In those countries the demand for treatment of symptomatic stones could dramatically increase involving a huge financial outlay (Trinchieri, A. 2006).

1.5 Laboratory Experiments

This research project was conducted using previously established pharmacological models based on animal studies. These proposed methodologies of various scientists are used to analyse the following aspects of SMB:-

1.5.1 Analgesic study

To investigate whether any drug has any analgesic effect the following experiment was carried out:

- *Acetic acid induced writhing test* – This experiment was carried out to find the existence of non-narcotic analgesic property of drugs. The pain sensation was initiated by using acetic acid. The acetic acid induced writhing is inversely proportional to the non-narcotic analgesic property, if present, of the drug.(Tang *et al* in 1984)

1.5.2 Neuropharmacological effects / side effects

To study the neuropharmacological effect/side effect of a drug using open field and hole board tests were carried out.

- *Hole board test* - The hole board test is carried out to investigate the effect of the test drug on the exploratory behavior of the laboratory animal model characterized by exploration, locomotion and emotional defecation. It has been considered that the exploration evoked under an unfamiliar environment is modified with physiological

factors such as curiosity, fear and anxiety and the modulation of these factors after the administration of a drug (Nakama *et al* in 1972).

- *Open field test* - It is a widely used procedure for examining the effect of the drug under consideration on the pattern of behaviour – locomotion, exploration and anxiety. A different and more complex environment is presented for exploration to the animal in this experiment (Choleris, E. *et al* 2001)

1.5.3 Gastrointestinal effects / side effects

In order to investigate any effect of drug on the gastrointestinal (GI) tract the following two experiments were carried out:

- *Gastrointestinal (GI) motility test* – The GI tract is innervated by both the parasympathetic and the sympathetic fibers of the autonomic nervous system. Its peristaltic movement is myogenic in character which is mainly initiated by the local reflexes and can occur without any neural connections to the brain or the spinal cord. Extrinsic nerves to the intestine appear to have only a minor role in modulating the peristaltic activity of the organ. GI motility test was carried out to find the effect of the drug on the peristaltic movement of the GI tract. This experiment was carried out by the method previously described by Chatterjee (1993).
- *Colon transit time test* – The colon transit time test is carried out to assess the effect of the experimental drug on colon. Distal colonic transit time is determined by monitoring the time required for expulsion of the glass bead (bead latency). Martinez *et al.* followed this model in 2002.

1.6 Objective of the Study

By using Ayurvedic medicine expensive and extensive procedures of clinical investigations can be avoided in many cases and people in these selected areas have the choice to get treatment at a cheaper price depending on their choice.

Considering the widespread use of Ayurveda as the popular form of treatment in Bangladesh, one cannot emphasize enough the need for establishing the safety profiles of Ayurvedic drugs. Keeping in mind, the present scenario this research work on Ayurvedic formulation, Sukramatrka Vati (SMB) explores a spectrum of its pharmacological aspects utilizing experimental animals. The objective is to have a better understanding of the possible pharmacological profile of the drug under study and, to some degree, to decide how justifiable the use of this drug is under the stated circumstances. The project will eventually result in supplementing and complementing the existing health care facilities and, in the long run, will ensure total coverage of the population in terms of public health.

This study was performed in an effort to evaluate the safety of this drug according to modern pharmacological parameters so as to fully fathom the safety of this Ayurvedic drug for Alternative Medical Care.

Chapter Two

Materials and Methods

2.1 Collection of Ayurvedic formulation

The entire research project was conducted by using five pharmacological models in order to study the analgesic, neuropharmacological and gastrointestinal effects of SMB, an Ayurvedic preparation. Sukramatrika Vati (Batch# 002) was collected from "Sree Kundeswari Aushadhalaya Ltd.", Chittagong, Bangladesh. The formulation was available in the form of tablets packed in suitable containers. A detailed discussion is given on the materials required and the methods followed for each of the experiments.

2.2 Dose and Route of Administration

The tablets were powdered, by grinding with the aid of a mortar and pestle, and made into a solution with distilled water. The solution was then administered at a volume such that it would permit optimal dosage accuracy without contributing much to the total increase in the body fluid. For all the pharmacological studies the drugs were administered by the oral route [per oral, (p.o.)] by means of a long feeding needle with a ball-shaped end at the doses of 100mg/kg, 200mg/kg and 400mg/kg body weight.

2.3 Experimental Animals

Male and female mice of the Swiss-Webster strain having a weight range between 20-40 gm were experimented on for pharmacological effects. The mice were bred in the animal house of the Department of Pharmacy, Jahangirnagar University, where they were kept in cages having dimensions of 30 x 20 x 13 cm and soft wood shavings employed as bedding in the cages.

The animals were provided with standard laboratory food and tap water '*ad libitum*' and maintained at the natural day and night cycle. They were fed with "mouse chow" (prepared according to the formula developed at BCSIR, Dhaka). Before starting an experiment, the

animals were weighed using an electronic balance (Shimadzu, Japan) and carefully marked at the base of their tails, which was later used as identification mark for a particular animal, so that the response of a particular mouse prior to and after the administration could be noted separately.

For each experiment there were two groups of mice – control group and drug treated group. Each group consisted of equal number of mice of the same sex. Six to ten mice were taken for both the control and the drug group and the experiments were simultaneously employed for both.

The mice in drug group were administered SMB solution orally of a specified dose and those in the control group were administered with distilled water as par the same volume as the drug treated group.

2.4 Experiments conducted and Doses employed

- Acetic acid induced writhing test: 100, 200 and 400mg/kg body weight.
- Hole board test: 100, 200 and 400mg/kg body weight
- Open field test: 100, 200 and 400mg/kg body weight
- GI motility test: 100mg/kg body weight.
- Colon transit time test: 100mg/kg body weight.

2.5 Pharmacological studies with Animal models

2.5.1 Acetic Acid (AA) Induced Writhing Test

Materials required:

20 female mice	Methanol (VWR, England)
10 separate mice cases for observation	Feeding needle
Counter	Sterile syringe
0.6% aqueous acetic acid solution (COO, Germany)	Injection needle

Method employed:

The 20 mice female mice were taken (body weight ranging between 0 to 5 units). Out of 20, 10 were randomly chosen for the drug (SMB) group and the remaining 10 for the control group. In case of each mouse muscular contraction was induced by the intraperitonial injection of Acetic Acid (0.6%, 0.25ml/animal). Then SMB was administered orally 45 minutes before the intraperitonial injection of 0.6% AA. It was placed in an observation box and number of writhes (painful abdominal muscular contractions and stretches) produced was counted for five minutes after fifteen minutes of acetic acid injection.

The average number of writhes was calculated and the percent protection was calculated as –

$$\% \text{ Protection} = 100 - [(\text{treated mean}/\text{control mean}) \times 100]$$

The readings were then compared to that of the distilled water treated control group.

2.5.2 Hole Board Test

Materials required:

12 female mice	Methanol (VWR, England)
Hole board apparatus	Feeding needle
Stopwatch	Counter
Cotton	

Method employed:

The 12 mice were divided into two groups (body weight ranging from 0 to 5 units); 6 for the drug group and 6 for the control group. Each mouse was placed on one corner of the hole board apparatus (a board containing a total of 16 holes, each 3 cm in diameter, was presented to the mouse in a flat space of 25 square centimeters) and the number of ambulation (expressed as the number of squares traveled) head dipping and number of fecal boluses expelled by the mouse were recorded for a period of 2 minutes at pre 30 minutes and post 30, 60, 120 and 240 minutes intervals and were compared with the control group.

2.5.3 Open Field Test

Materials required:

12 male mice	Methanol (VWR, England)
Open field apparatus	Feeding needle
Stopwatch	Counter
Cotton	

Method employed:

The 12 mice were divided into two groups (body weight ranging from 0 to 5 units); 6 for the drug group and 6 for the control group. Each mouse was placed on one corner of the open field apparatus (a box consisting of a half square meter floor divided into a series of squares alternatively coloured black and white and enclosed by walls 40 cm in height) and the number of ambulation (expressed as the number of squares traveled), the number of times it entered the center region, the number of stand ups and number of fecal boluses expelled by the mouse were recorded for a period of 2 minutes at pre 30 minutes and post 30, 60, 120 and 240 minutes intervals and were compared with the control group.

2.5.4 Gastrointestinal (GI) Motility Test

Materials required:

128 male mice	Dissection tools
$BaSO_4$ (Merck, India)	Feeding needle
Sodium CMC (Merck, India)	Meter ruler

Method employed:

The experiment was carried out by the method previously described by Chatterjee (1993).128 male mice were weighed (body weight ranging from 30 to 40 g) and divided into four sets to represent the action of the drug at four different hours (1^{st}, 2^{nd}, 3^{rd}, and 4^{th} hour) with respect to drug administration.

$BaSO_4$ milk was prepared by adding $BaSO_4$ at 15% w/v in 0.5% sodium CMC suspension. In this experiment, $BaSO_4$ acts as a non-absorbable marker to measure GI motility.

$BaSO_4$ milk was given to a group of 12 mice 15 minutes after administration of the test drug (SMB). The drug treated mice were subdivided into two groups and sacrificed by cervical

dislocation after 15 and 30 minutes after the administration of the milk. The distance traversed by $BaSO_4$ milk through the small intestine was measured and expressed as a percentage of the total length of small intestine (from pylorus to the ileocaeccal junction). The test drug was compared with the control group that was administered with distilled water. The experiment was performed in the same manner for all the sets of mice to study the effect of SMB on gastric peristalsis at four different hours.

2.5.5 Colon Transit Time Test

Materials required:

40 female mice	Marker
Glass beads	Feeding needle
Stopwatch	Feeding needle for rats
20 plastic cases	

Method employed:

The 40 mice (body weight ranging from 30 to 40 g) were divided into two groups.One hour after drug administration to the drug group, a single glass bead, 2 mm in diameter was inserted into the distal colon of each mouse at 2 cm from the anus, after which the mice were returned to their respective cages and observed closely. Distal colonic transit time was determined by monitoring the time required for expulsion of the glass bead (bead latency). Similarly, the experiment was repeated in the same manner for the control group (Martinez *et al* in 2002).

2.6 Statistical Analysis

The various degrees of significance on activity were determined by analyzing all the data obtained using SPSS (Statistical Package for Social Science) for WINDOWS™ (Version 12). Data were presented as Mean ± SEM (standard error of the mean) and unpaired "t" tests were done for statistical significance tests.

Upon analysis, a "p" value was obtained that determines the appropriateness of rejecting the null hypothesis. The "p" values range between 0 to 1; and the smaller it is the smaller the probability of rejecting the null hypothesis.

For all the data analyzed, $p = 0.05$ was assigned as the level of significance; $p = 0.01$ was assigned to represent a high level of significance; and $p = 0.001$ was assigned to represent a very high level of significance.

Chapter Three

Results and Discussions

3.1 ACETIC ACID INDUCED WRITHING TEST

Statistical findings

Writhing response

SMB treated female mice at dose 100 mg/kg showed an overall decrease in writhing compared to the control group throughout the five minutes study period. The decreased writhing response of the 2nd minute was statistically significant ($p = 0.047^*$). In addition, the overall reduction in pain perception from minutes 1 to 5 was found to be statistically significant ($p = 0.043^*$).

At a dose of 200mg/kg the SMB treated female mice showed an increase in their response in the 1st and 2nd minute but showed a decrease in writhing response in the 3rd, 4th and 5th minutes compared to the corresponding control group. None of the result was statistically significant ($p > 0.05$). The mean writhing response of the drug group was lower than the control group.

At a dose of 400mg/kg SMB treated female mice, a reduction in response in the 1st, 2nd and 4th minutes were observed compared to the control group present and in the 3rd and 5th minutes the response increased in comparison to the control group. The mean writhing response was also reduced compared to the control group. None of the result was statistically significant ($p > 0.05$).

The percentage (%) protections provided by SMB at different doses are given below:

At a dose of 100mg/kg - 40.50%

At a dose of 200mg/kg - 7.69%

At a dose of 400mg/kg - 15.39

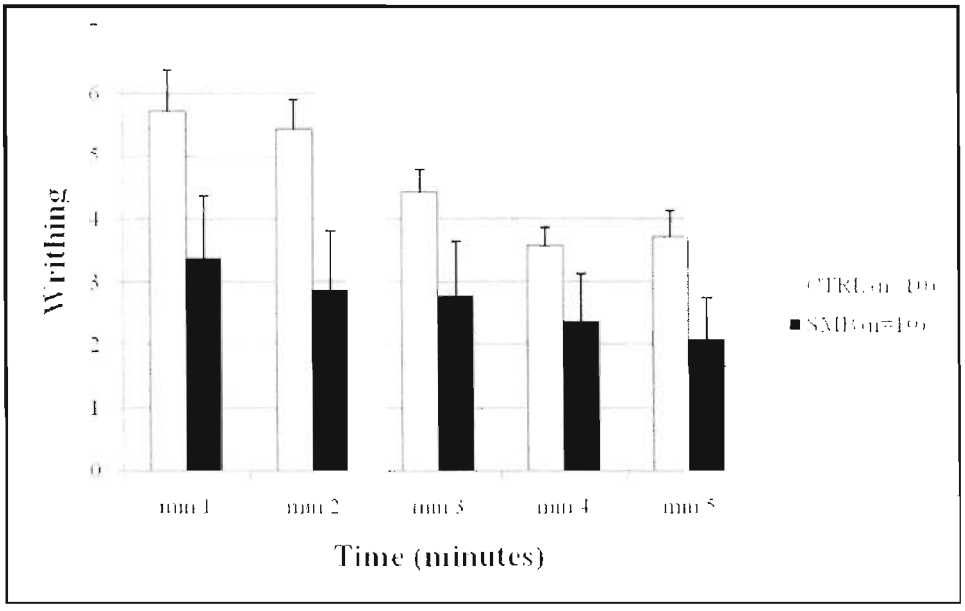
Tabular and Graphical presentation of the effect of SMB (100mg/kg, 200mg/kg and 400mg/kg) on the Acetic Acid Induced Writhing Test utilizing female mice.

3.1.1 Tabular presentation of the effect of SMB (100 mg/kg) in AA Induced Writhing Test using female mice

Group		1st Min	2nd Min	3rd t Min	4th Min	5th Min
Ctrl (n=10)		5.714±0.680	5.429±0.481	4.429±0.369	3.571±0.297	3.714±0.421
SMB (n=10)		3.400±0.968	2.900±0.912	2.800±0.854	2.400±0.748	2.100±0.674
t/p		1.956/0.070	2.160/**0.047***	1.751/0.105	1.455/0.172	2.032/0.061
95% confidence interval	Lower	-0.212	0.034	-0.397	-0.589	-0.088
	Upper	4.841	5.023	3.654	2.932	3.317

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.1 Graphical presentation of the effect of SMB (100 mg/kg) in AA Induced Writhing Test using female mice

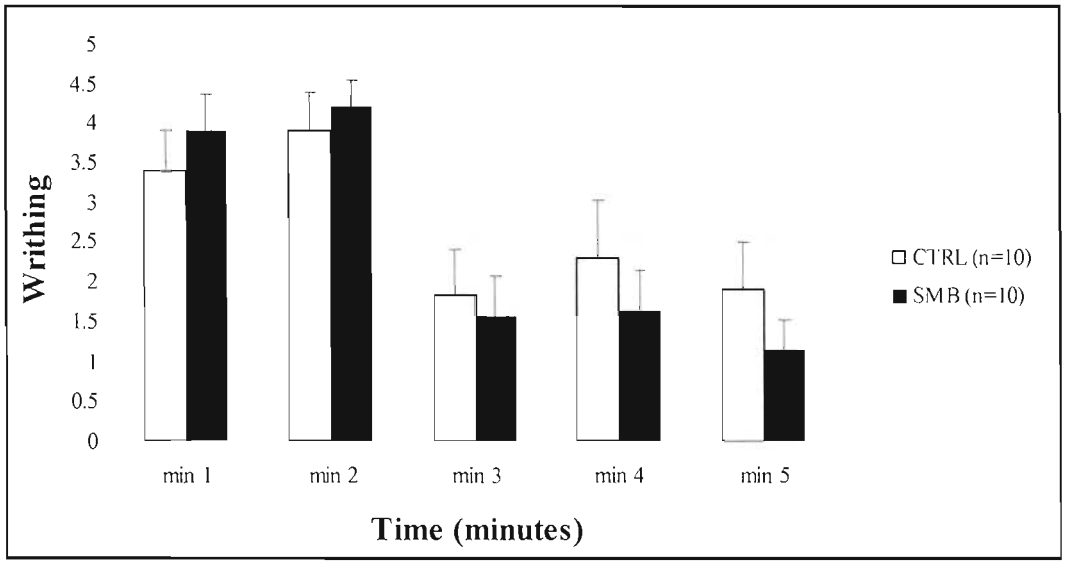


3.1.2 Tabular presentation of the effect of SMB (200 mg/kg) in AA Induced Writhing Test using female mice

Group		1st Min	2nd Min	3rd t Min	4th Min	5th Min
Ctrl (n=10)		3.400±0.499	3.900±0.482	1.829±0.578	2.300±0.727	1.889±0.597
SMB (n=10)		3.900±0.458	4.200±0.327	1.567±0.496	1.619±0.512	1.155±0.365
t/p		-0.738/0.470	-0.515/0.613	1.313/0.206	1.124/0.276	0.429/0.674
95% confidence interval	Lower	-1.923	-1.523	-0.600	-0.869	-1.193
	Upper	0.923	0.923	2.600	2.869	1.793

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.2 Graphical presentation of the effect of SMB (200 mg/kg) in AA Induced Writhing Test using female mice

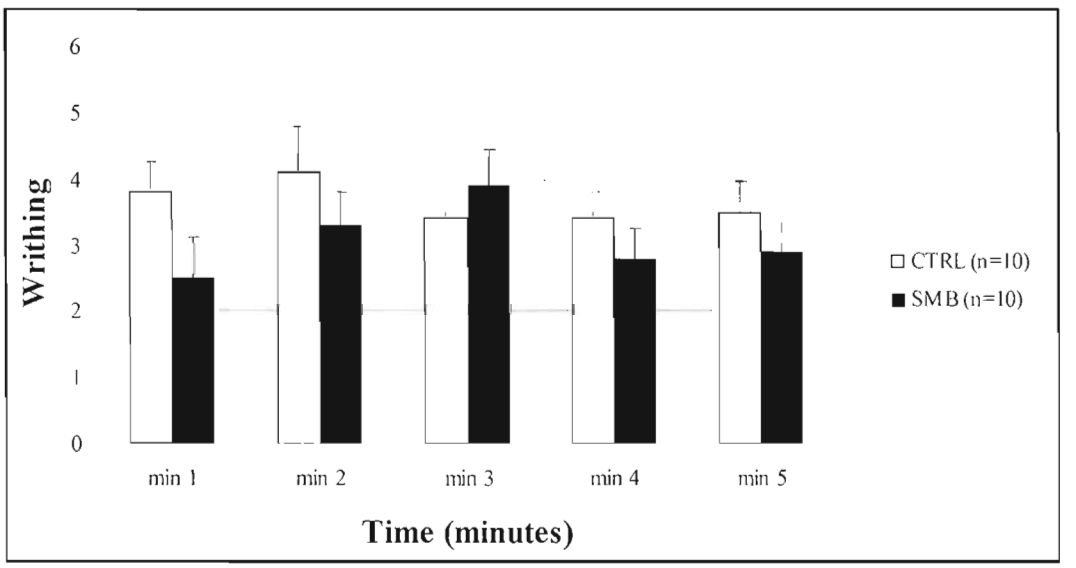


3.1.3 Tabular presentation of the effect of SMB (400 mg/kg) in AA Induced Writhing Test using female mice

Group		1st Min	2nd Min	3rd t Min	4th Min	5th Min
Ctrl (n=10)		3.800±0.467	4.1000±0.674	3.400±0.499	3.400±0.400	3.500±0.453
SMB (n=10)		2.500±0.619	3.3000±0.496	3.900±0.547	2.800±0.442	2.900±0.605
t/p		1.677/0.111	0.956/0.352	-0.676/0.508	1.006/0.328	0.794/0.438
95% confidence interval	Lower	-0.329	-0.958	-2.055	-0.653	-0.988
	Upper	2.929	2.558	1.055	1.853	2.188

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.3 Graphical presentation of the effect of SMB (400 mg/kg) in AA Induced Writhing Test using female mice

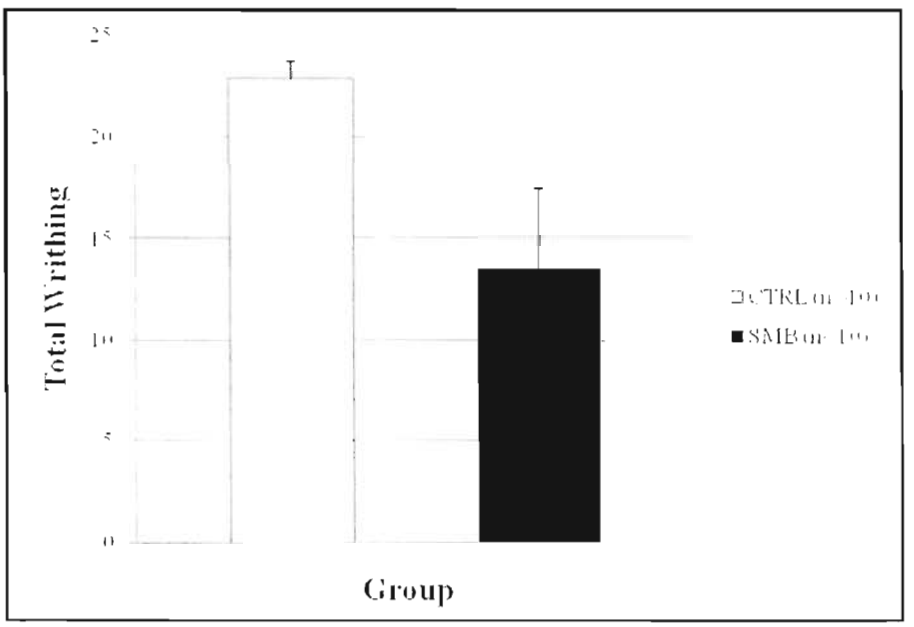


3.1.4 The effect of SMB (100mg/kg) in AA Induced Writhing Test during the five minutes study period

Group	PARAMETER	
Male mice	Min 0-5	% Protection
Control(n=10)	22.857±0.857	40.50 %
SMB (n=10)	13.600±3.899	

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.4 Graphical representation of the effect of SMB (100mg/kg) in AA Induced Writhing Test during the five minutes study period

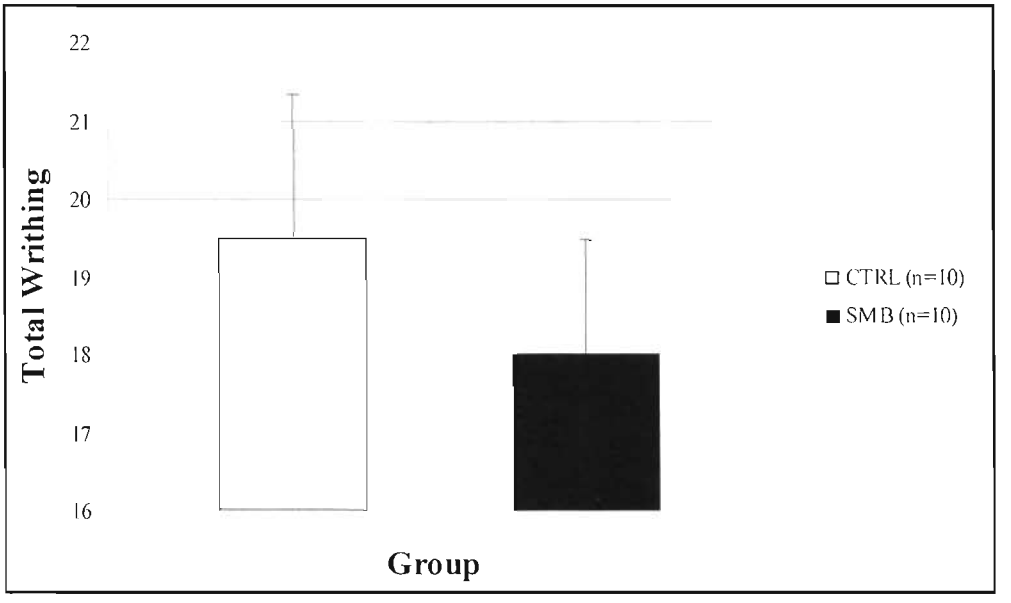


3.1.5 The effect of SMB (200mg/kg) in AA Induced Writhing Test during the five minutes study period

Group	PARAMETER	
Male mice	Min 0-5	% Protection
Control(n=10)	19.500±1.821	7.692
SMB (n=10)	18.000±1.483	

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.5 Graphical representation of the effect of SMB (200mg/kg) in AA Induced Writhing Test during the five minutes study period

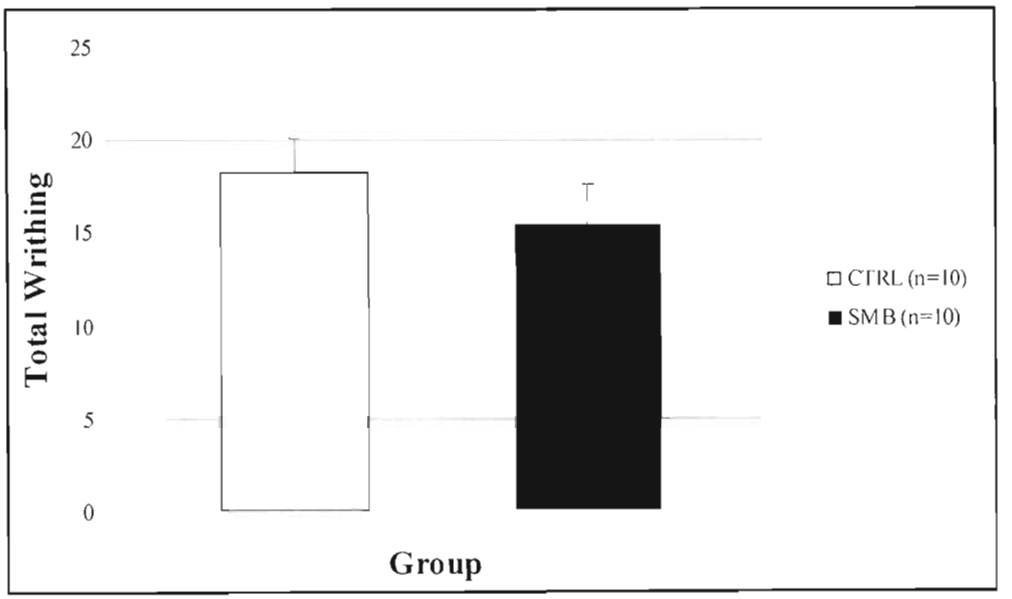


3.1.6 The effect of SMB (400mg/kg) in AA Induced Writhing Test during the five minutes study period

Group	PARAMETER	
Male mice	**Min 0-5**	**% Protection**
Control(n=10)	18.200±1.931	15.385
SMB (n=10)	15.400±2.177	

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.1.6 Graphical representation of the effect of SMB (400mg/kg) in AA Induced Writhing Test during the five minutes study period



3.2 Hole Board

Statistical findings

Emotional Defaecation

As evident from the graph, at a dose of 100mg/kg the SMB treated mice showed an increase in defaecation at minute 0, 60, 180 and 240 and showed a decrease in minute 30 and 120 compared to the control group. The reading at minute 0 was statistically highly significant ($p = 0.010^*$).

At the dose of 200mg/kg, SMB treated male mice showed a decrease in defaecation throughout all the time intervals compared to the control group. Of these, the reduction at minute 240 was observed to be statistically significant ($p = 0.044^*$).

With a 400mg/kg dose of SMB the defaecation increased from minute 0 – 60 and then there was no defaecation observed from minute120 – 240 when compared to the control group. No result with this dose was statistically significant ($p > 0.05$).

Ambulation

SMB treated male mice at a dose of 100mg/kg showed an overall decrease in ambulation throughout the experimental study period when compared with the control group.

On the other hand, at a dose of 200mg/kg the SMB treated male mice exhibited a decrease in ambulation at all time intervals throughout the experiment.

Again, the SMB treated male mice at a dose of 400mg/kg, demonstrated an increase in ambulation over the entire experimental study period on comparison with the control group.

None of the result was statistically significant ($p > 0.05$).

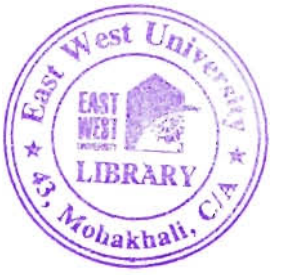
Head Dipping

SMB treated male mice at a dose of 100mg/kg exhibited an increase in the head dipping activity throughout all the time intervals except at minute 0 when no head dipping activity was shown compared to the control group.

At minutes 0, 60, 240 the head dipping of SMB treated male mice at the dose of 200mg/kg increased whereas at minutes 30, 120, 180 the head dipping decreased compared to the control group.

The head dipping of the SMB treated male mice at the dose of 400mg/kg decreased from minute 0 – 120. At minute 180 there was no reading of the control or the drug group and at minute 240 there was no reading compared to the control group.

None of the result was statistically significant.



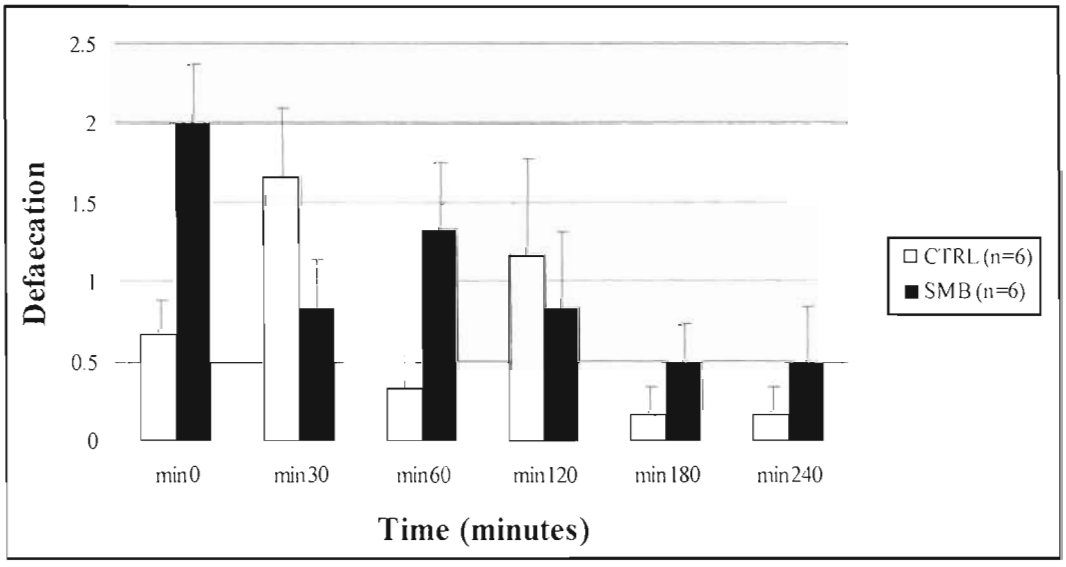
Tabular and Graphical presentation of the effect of SMB (100 mg/kg, 200 mg/kg and 400 mg/kg) on the Hole Board Test utilizing male mice.

3.2.1.1 Tabular representation of the effect of SMB (100 mg/kg) in defaecation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		0.667±0.211	1.667±0.422	0.333±0.211	1.167±0.601	0.167±0.167	0.167±0.167
SMB(n=6)		2.000±0.365	0.833±0.307	1.333±0.422	0.833±0.477	0.500±0.224	0.500±0.342
t/p		-3.162/**0.010***	1.597/0.141	-2.121/0.060	0.434/0.673	-1.195/0.260	-0.877/0.401
95% confidence interval	lower	-2.273	-0.329	-2.050	-1.377	-0.955	-1.180
	Upper	-0.393	1.996	.050	2.043	0.288	0.514

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.1.1 Graphical representation of the effect of SMB (100 mg/kg) in defaecation in Hole Board Test

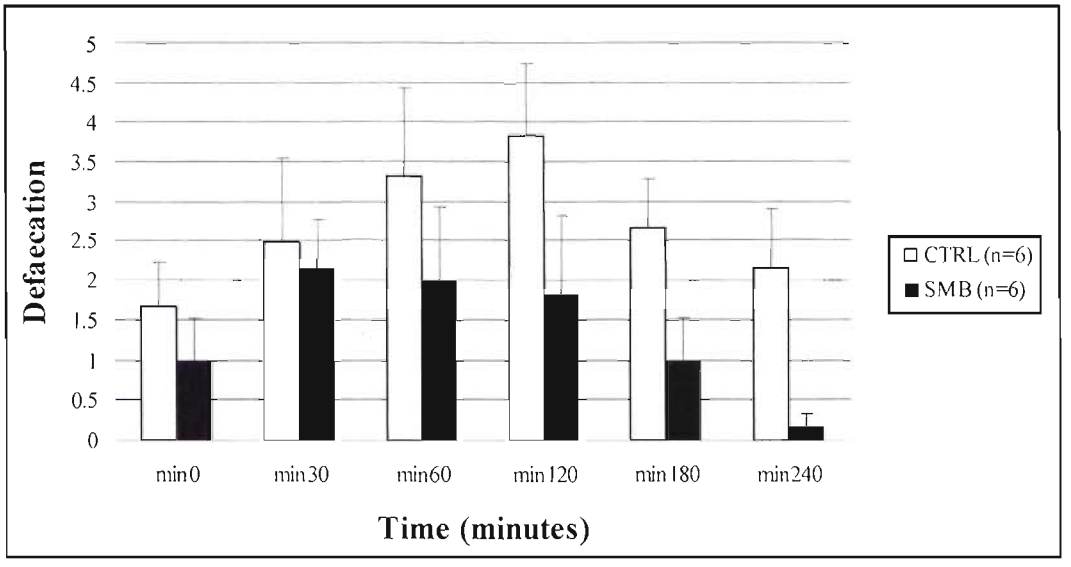


3.2.1.2 Tabular representation of the effect of SMB (200 mg/kg) in defaecation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		1.667±0.558	2.500±1.057	3.333±1.085	3.833±0.910	2.667±0.615	2.167±0.749
SMB(n=6)		1.000±0.516	2.167±0.601	2.000±0.931	1.833±0.980	1.000±0.516	0.167±0.167
t/p		0.877/0.401	0.274/0.790	0.933/0.373	1.495/0.166	2.076/0.065	2.606/**0.044***
95% confidence interval	lower	-1.027	-2.375	-1.853	-0.980	-0.122	0.080
	Upper	2.360	3.042	4.519	4.980	3.455	3.921

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.1.2 Graphical representation of the effect of SMB (200 mg/kg) in defaecation in Hole Board Test

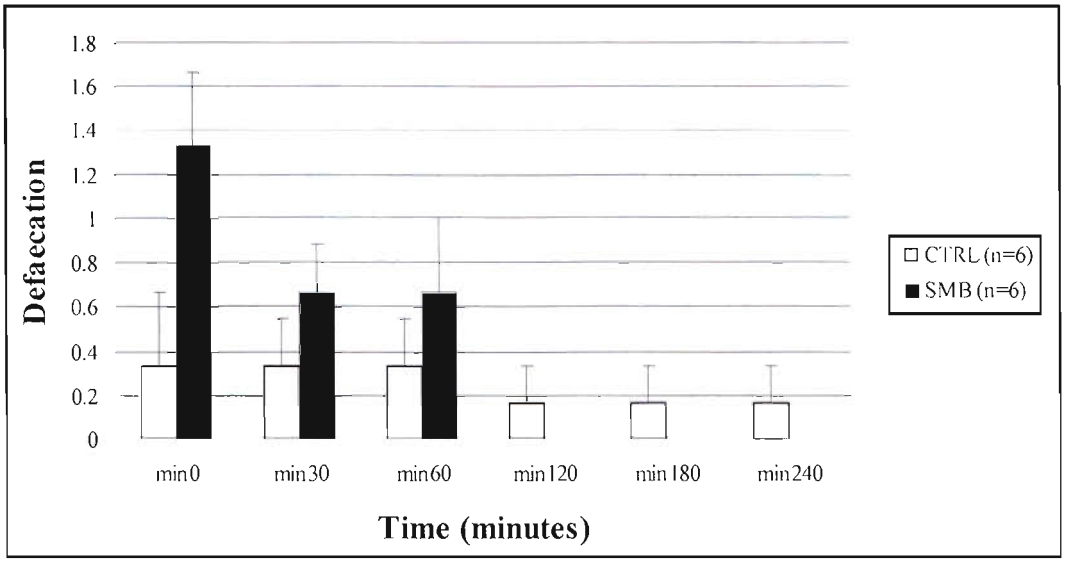


3.2.1.3 Tabular representation of the effect of ICB (400 mg/kg) in defaecation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		0.333±0.333	0.333±0.211	0.333±0.211	0.167±0.167	0.167±0.167	0.167±0.167
SMB(n=6)		1.333±0.333	0.667±0.211	0.667±0.333	0.000±0.000	0.000±0.000	0.000±0.000
t/p		-2.121/0.060	-1.118/0.290	-0.845/0.418	1.000/0.363	1.000/0.363	1.000/0.363
95% confidence interval	lower	-2.050	-0.998	-1.212	-0.262	-0.262	-0.262
	Upper	0.050	0.331	0.546	0.595	0.595	0.595

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.1.3 Graphical representation of the effect of SMB (400 mg/kg) in defaecation in Hole Board Test

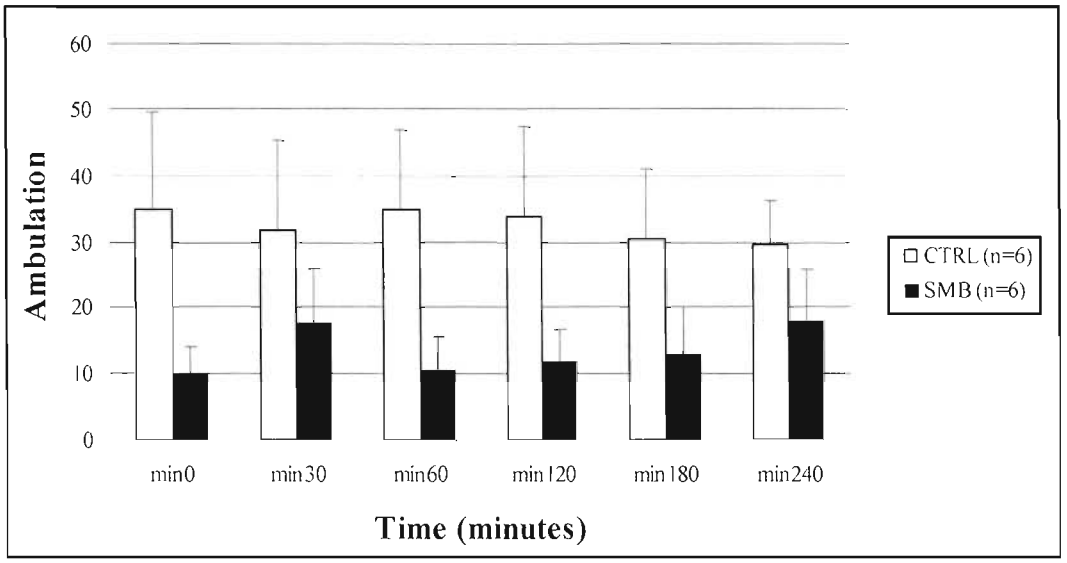


3.2.2.1 Tabular representation of the effect of SMB (100 mg/kg) in ambulation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		35.000±14.408	31.833±13.350	34.833±11.876	33.833±13.593	30.500±10.452	29.500±6.672
SMB(n=6)		10.167±3.953	17.667±8.110	10.667±5.018	12.000±4.683	13.000±7.188	18.000±7.590
t/p		1.662/0.127	0.907/0.386	1.875/0.090	1.519/0.178	1.380/0.198	1.138/0.282
95% confidence interval	lower	-8.457	-20.638	-4.559	-13.111	-10.765	-11.016
	Upper	58.123	48.972	52.892	56.778	45.765	34.016

N.B :*(< 0.05) =Significant. ** (< 0.01) = Highly Significant. *** (< 0.001) = Very Highly Significant

3.2.2.1 Graphical representation of the effect of SMB (100 mg/kg) in ambulation in Hole Board Test

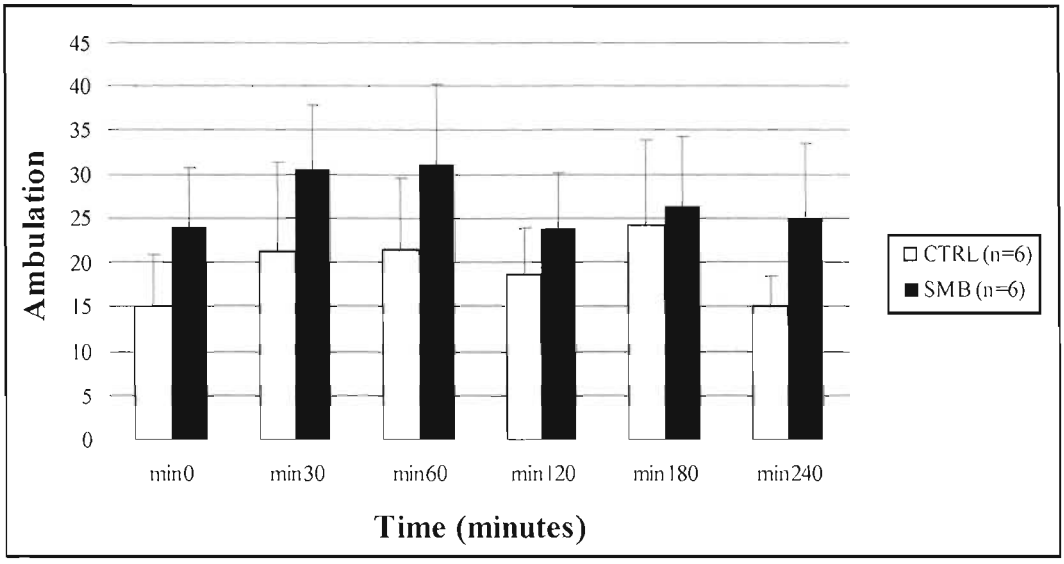


3.2.2.2 Tabular representation of the effect of SMB (200 mg/kg) in ambulation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		35.000±14.408	31.833±13.350	34.833±11.876	33.833±13.593	30.500±10.452	29.500±6.672
SMB(n=6)		10.167±3.953	17.667±8.110	10.667±5.018	12.000±4.683	13.000±7.188	18.000±7.590
t/p		1.662/0.127	0.907/0.386	1.875/0.090	1.519/0.178	1.380/0.198	1.138/0.282
95% confidence interval	lower	-8.457	-20.638	-4.559	-13.111	-10.765	-11.016
	Upper	58.123	48.972	52.892	56.778	45.765	34.016

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.2.2 Graphical representation of the effect of SMB (200 mg/kg) in ambulation in Hole Board Test

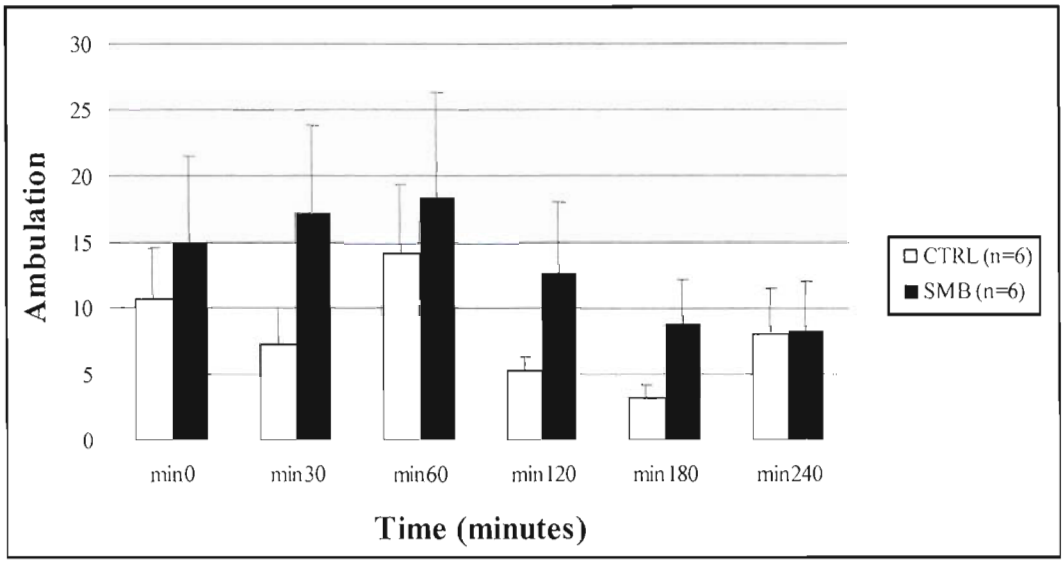


3.2.2.3 Tabular representation of the effect of SMB (400 mg/kg) in ambulation in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		10.667±3.844	7.333±2.777	14.167±5.076	5.333±0.989	3.167±1.078	8.000±3.435
SMB(n=6)		15.000±6.377	17.167±6.680	18.333±8.011	12.667±5.258	8.833±3.331	8.333±3.712
t/p		- 0.582/0.576	- 1.359/0.218	- 0.439/0.670	- 1.371/0.225	- 1.619/0.156	- 0.066/0.949
95% confidence interval	lower	-21.428	-27.109	-25.298	-20.817	-14.221	-11.602
	Upper	12.761	7.443	16.964	6.151	2.887	10.935

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.2.3 Graphical representation of the effect of SMB (400 mg/kg) in ambulation in Hole Board Test

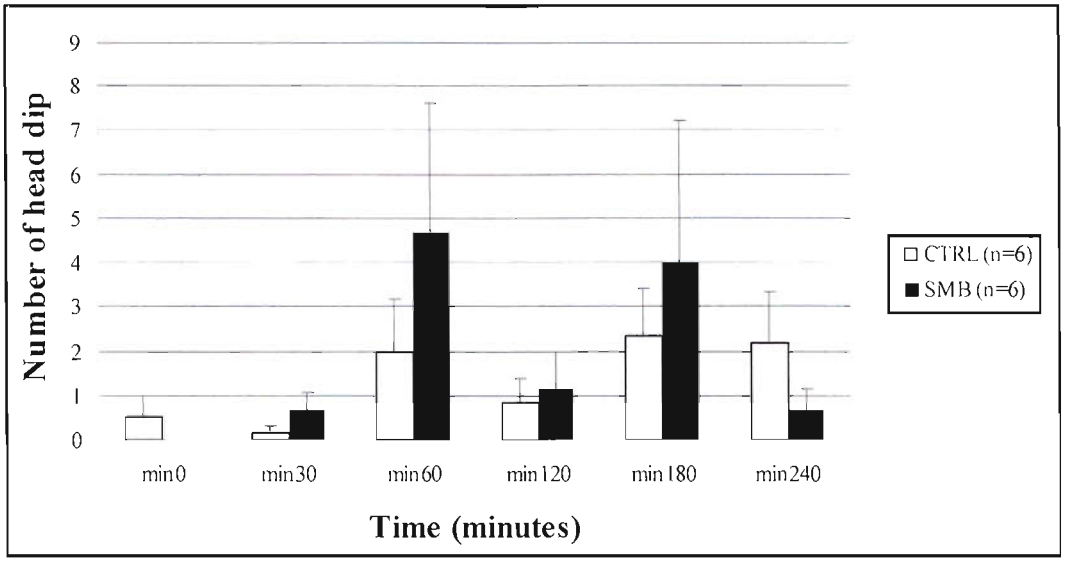


3.2.3.1 Tabular representation of the effect of SMB (100 mg/kg) in head dipping in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		0.500±0.500	0.167±0.167	2.000±1.183	0.833±0.543	2.333±1.085	2.167±1.167
SMB(n=6)		0.000±0.000	0.667±0.422	4.667±2.963	1.167±0.833	4.000±3.235	0.667±0.494
t/p		1.000/0.363	-1.103/0.309	-0.836/0.433	-0.335/0.744	-0.488/0.636	1.184/0.264
95% confidence interval	lower	-0.785	-1.588	-10.315	-2.549	-9.270	-1.323
	Upper	1.785	0.588	4.982	1.882	5.937	4.323

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.3.1 Graphical representation of the effect of SMB (100 mg/kg) in head dipping in Hole Board Test

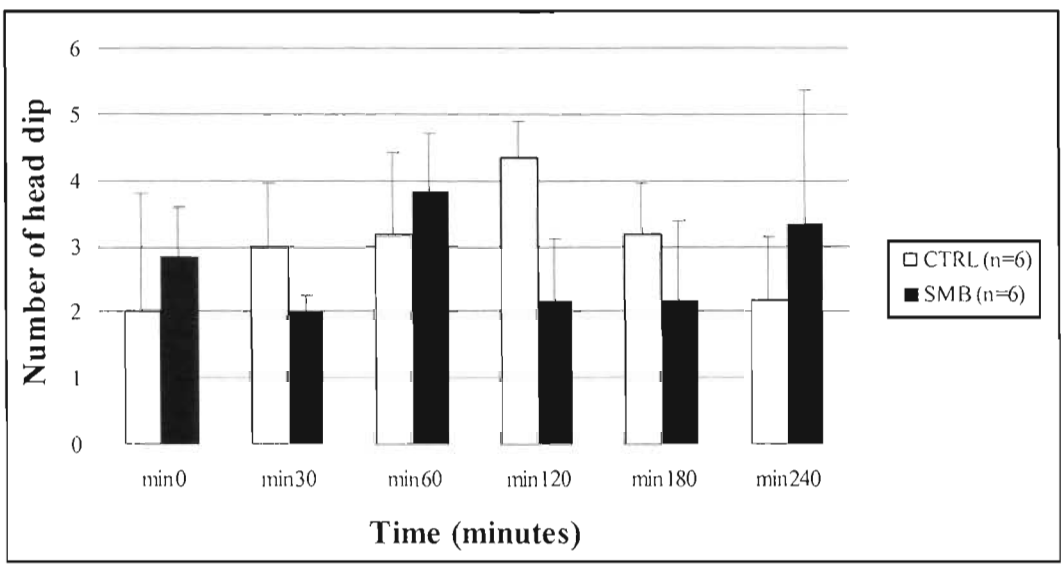


3.2.3.2 Tabular representation of the effect of SMB (200 mg/kg) in head dipping in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		2.000±1.807	3.000±0.966	3.167±1.249	4.333±0.558	3.167±0.792	2.167±0.980
SMB(n=6)		2.833±0.749	2.000±0.258	3.833±0.872	2.167±0.946	2.167±1.222	3.333±2.028
t/p		-0.426/0.679	1.000/0.358	-0.437/0.671	1.973/0.077	0.686/0.508	-0.518/0.616
95% confidence interval	lower	-5.193	-1.477	-4.062	-0.280	-2.246	-6.185
	Upper	3.526	3.477	2.729	4.613	4.246	3.851

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.3.2 Graphical representation of the effect of SMB (200 mg/kg) in head dipping in Hole Board Test

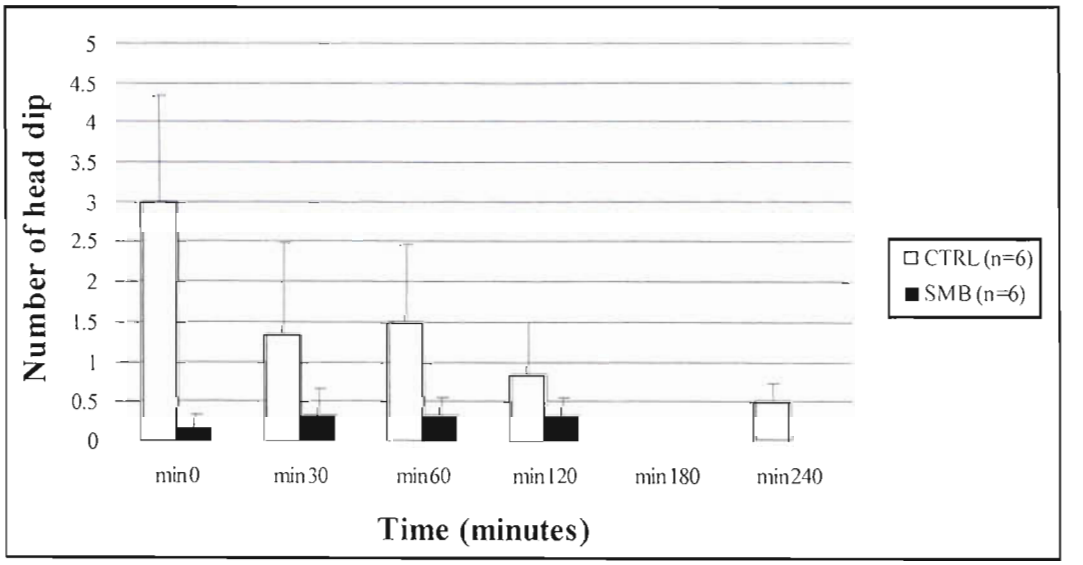


3.2.3.3 Tabular representation of the effect of SMB (400 mg/kg) in head dipping in Hole Board Test

Group		Min0	Min30	Min60	Min120	Min 180	Min240
Ctrl(n=6)		2.000±1.807	3.000±0.966	3.167±1.249	4.333±0.558	3.167±0.792	2.167±0.980
SMB(n=6)		2.833±0.749	2.000±0.258	3.833±0.872	2.167±0.946	2.167±1.222	3.333±2.028
t/p		-0.426/0.679	1.000/0.358	-0.437/0.671	1.973/0.077	0.686/0.508	-0.518/0.616
95% confidence interval	lower	-5.193	-1.477	-4.062	-0.280	-2.246	-6.185
	Upper	3.526	3.477	2.729	4.613	4.246	3.851

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.2.3.3 Graphical representation of the effect of SMB (400 mg/kg) in head dipping in Hole Board Test



3.3 OPEN FIELD TEST

Statistical findings

Total ambulation

At a dose of 100mg/kg, the SMB treated male mice exerted an overall increase in total ambulation compared to the control group. The increase at minute 0 was only slight. None of the result was statistically significant ($p > 0.05$).

The SMB treated male mice at a dose of 200mg/kg was found to exhibit a reduction in the total ambulation from minute 0 – 180. At minute 240 there was a slight increase when compared to the control group. None of the result was statistically significant ($p > 0.05$).

SMB treatcd male mice at the dose of 400mg/kg showed an overall decrease in the ambulation in comparison to the control group. The results obtained at minute 60 and 120 were statistically highly significant ($p = 0.004^{**}$) and ($p = 0.004^{**}$). The reading obtained at minute 180 was statistically significant ($p = 0.010^{*}$).

Total center ambulation

The SMB treated male mice at a dose of 100mg/kg showed an increase in ambulation at minute 0 compared to the control group. No other readings were observed and none was statistically significant ($p > 0.05$).

At a dose of 200mg/kg the mice did not give any ambulation in the center region.

There was a decrease in ambulation throughout the entire experimental study period in the SMB treated male mice at a dose of 400mg/kg in comparison to the control group. But the results obtained were not statistically significant ($p > 0.05$).

Total standing up behaviour

SMB treated male nice showed a decrease in standing up at a dose of 100mg/kg at minutes 0, 30, 60 and 180 and an increase in standing up at minutes120 and 240 when compared to the control group. But there was no statistically significant result.

With a dose of 200mg/kg the SMB treated male mice exhibited an increase in only minute 0 an in all the other minutes the standing up decreased compared to the control group. None of the result was statistically significant.

At a dose of 400mg/kg of SMB,the standing up behaviour decreased throughout the experiment and none of the result was statistically significant ($p > 0.05$).

Emotional defaecations

As seen from the graph the emotional defaecation decreased at minutes 0, 30, and 240 at a dose of 100mg/kg of SMB treated mice and at minutes 60, 120 and 180 the readings increased compared to the control group. t minute 0 the readings are the same for both the control group and the SMB treated mice at a dose of 200mg/kg. At minutes 30 and 240 the defeacation increased and at the other times the readings decreased.

SMB treated male mice at a dose of 400mg/kg showed a decrease in reading from minute 0 – 60 and an increase in emotional defaecation from minute 180 – 240 when compared to the control group.

But none of the result was statistically significant ($P > 0.05$).

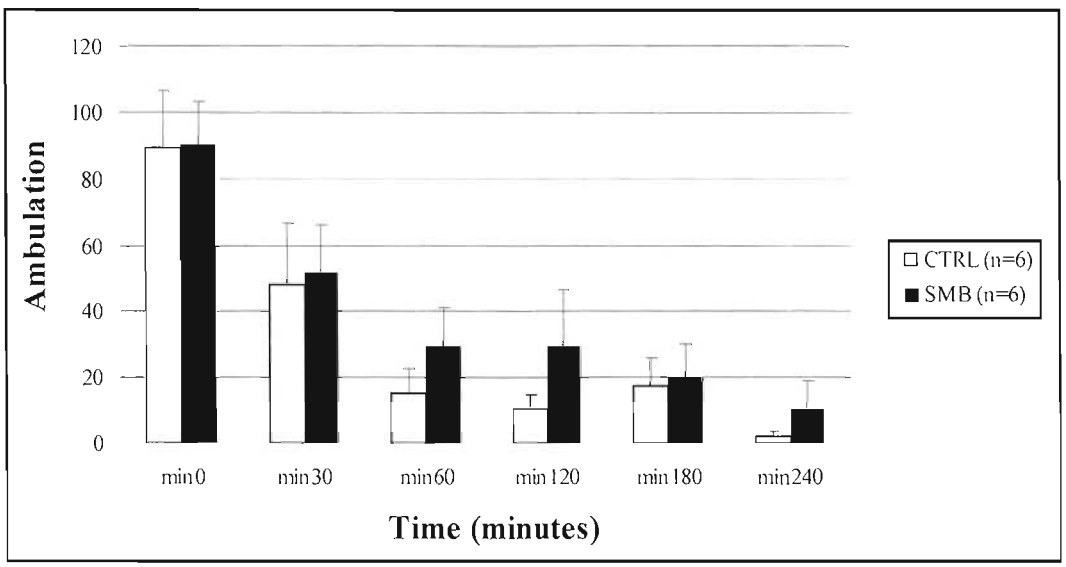
Tabular and Graphical presentation of the effect of SMB (100 mg/kg, 200 mg/kg and 400mg/kg) in Open Field Test using male mice

3.3.1.1 Tabular representation of the effect of SMB (100 mg/kg) in ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL (n=6)		89.333±16.891	48.333±18.500	15.500±7.060	10.667±3.947	17.667±8.024	2.167±1.515
SMB (n=6)		90.333±12.865	52.000±14.288	29.500±11.803	29.667±17.001	19.833±10.274	10.333±8.755
t/p		-0.047/0.963	-0.157/0.878	-1.018/0.333	-1.089/0.321	-0.166/0.871	-0.919/0.380
95% confidence interval	Lower	-48.309	-55.749	-44.645	-62.587	-31.213	-27.963
	Upper	46.309	48.415	16.645	24.587	26.879	11.630

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.1.1 Graphical representation of the effect of SMB (100 mg/kg) in ambulation in Open Field Test

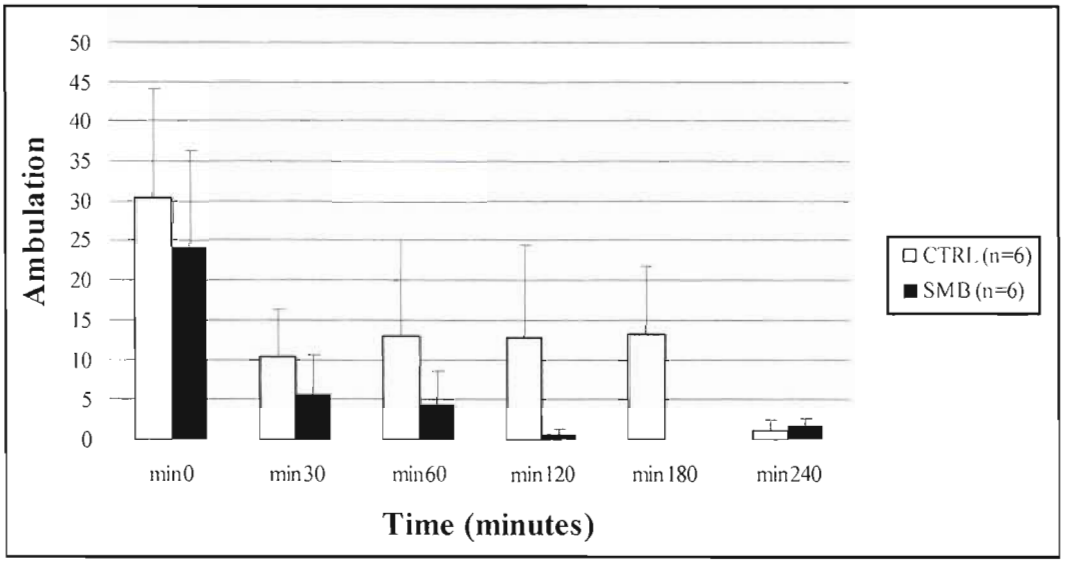


3.3.1.2 Tabular representation of the effect of SMB (200 mg/kg) in ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL (n=6)		30.333±13.747	10.333±5.903	13.000±12.217	12.667±11.885	13.167±8.696	1.167±1.167
SMB (n=6)		24.167±12.136	5.667±4.883	4.333±4.333	0.667±0.667	0.000±0.000	1.667±0.955
t/p		0.336/0.744	0.609/0.556	0.669/0.519	1.008/0.359	1.514/0.190	-0.332/0.747
95% confidence interval	Lower	-34.692	-12.403	-20.217	-18.541	-9.188	-3.859
	Upper	47.026	21.736	37.550	42.541	35.522	2.859

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.1.2 Graphical representation of the effect of SMB (200 mg/kg) in ambulation in Open Field Test

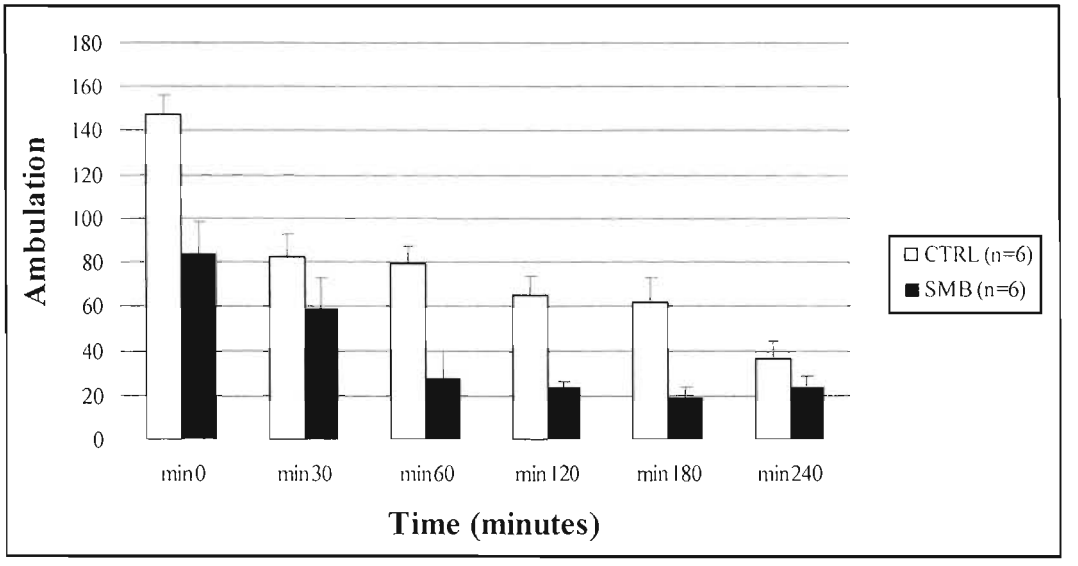


3.3.1.3 Tabular representation of the effect of SMB (400 mg/kg) in ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL(n=6)		147.500±8.872	82.833±9.840	79.667±7.526	65.167±8.654	62.000±11.334	36.167±8.336
SMB(n=6)		83.667±14.405	59.333±13.630	28.000±11.869	23.667±2.765	18.667± 5.512	24.000±4.810
t/p		3.773/0.004	1.398/0.192	3.676/**0.004****	4.568/**0.004****	3.438/**0.010***	1.264/0.235
95% confidence interval	Lower	26.137	-13.957	20.353	19.279	13.730	-9.278
	Upper	101.530	60.957	82.981	63.721	72.937	33.611

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.1.3 Graphical representation of the effect of SMB (400 mg/kg) in ambulation in Open Field Test

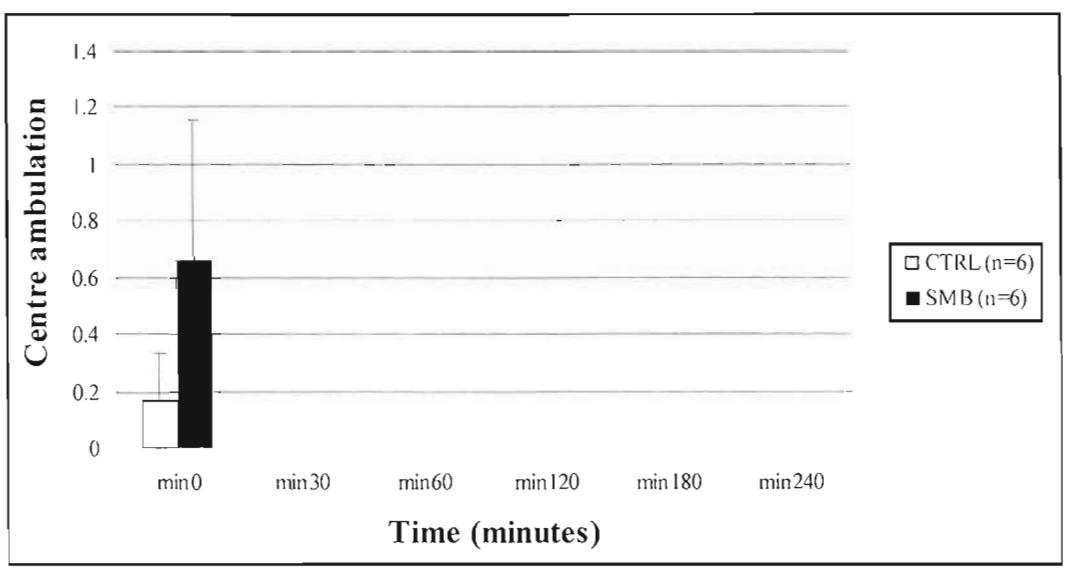


3.3.2.1 Tabular representation of the effect of SMB (100 mg/kg) in center ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL (n=6)		0.167±0.167	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000
SMB (n=6)		0.667±0.494	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000
t/p		-0.958/0.360	0.000/0.000	0.000/0.000	0.000/0.000	0.000/0.000	0.000/0.000
95% confidence interval	Lower	-1.663	0.000	0.000	0.000	0.000	0.000
	Upper	0.663	0.000	0.000	0.000	0.000	0.000

N.B :*(< 0.05) =Significant. ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.2.1 Graphical representation of the effect of SMB (100 mg/kg) in center ambulation in Open Field Test

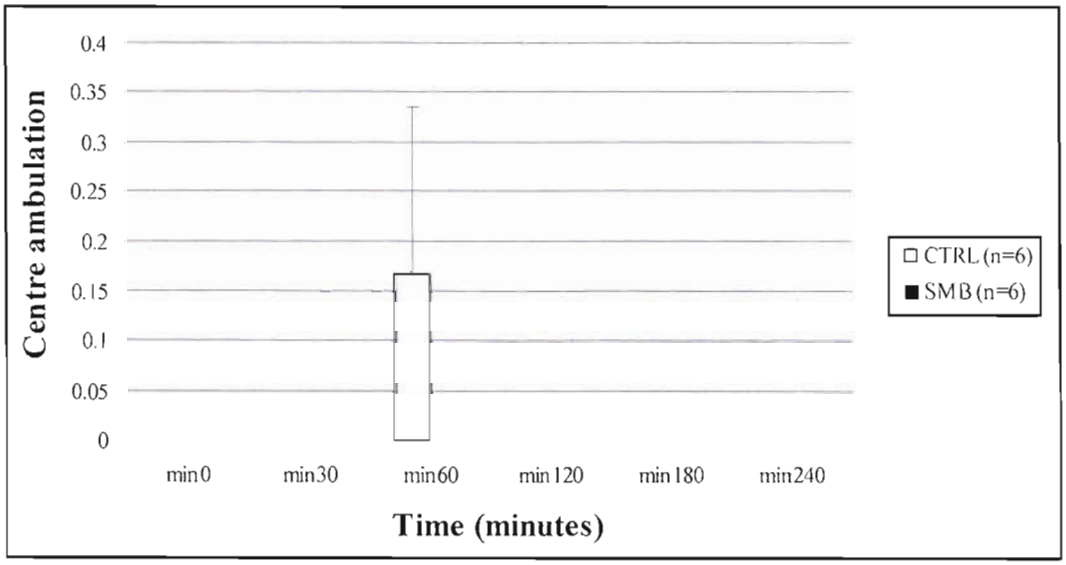


3.3.2.2 Tabular representation of the effect of SMB (200 mg/kg) in center ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL (n=6)		0.000±0.000	0.000±0.000	0.167±0.167	0.000±0.000	0.000±0.000	0.000±0.000
SMB (n=6)		0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000	0.000±0.000
t/p		0.000/0.000	0.000/0.000	1.000/0.363	0.000/0.000	0.000/0.000	0.000/0.000
95% confidence interval	Lower	0.000	0.000	-0.262	0.000	0.000	0.000
	Upper	0.000	0.000	0.595	0.000	0.000	0.000

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.2.2 Graphical representation of the effect of SMB (200 mg/kg) in center ambulation in Open Field Test

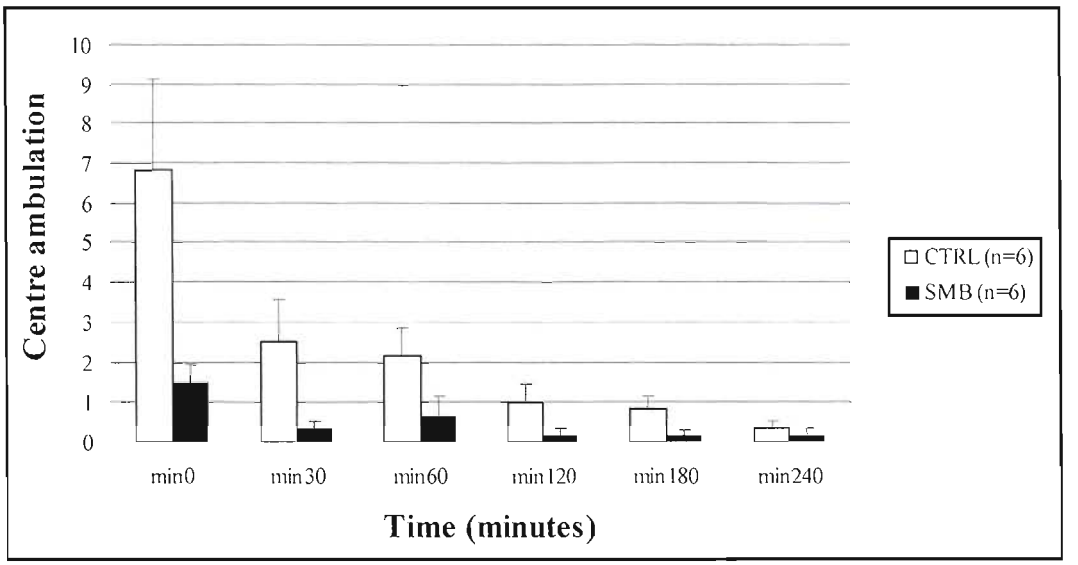


3.3.2.3 Tabular representation of the effect of SMB (400 mg/kg) in center ambulation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
CTRL(n=6)		6.833±2.301	2.500±1.057	2.167±0.703	1.000±0.447	0.833±0.307	0.333±0.211
SMB(n=6)		1.500±0.428	0.333±0.211	0.667±0.494	0.167±0.167	0.167±0.167	0.167±0.167
t/p		2.279/0.068	2.011/0.096	1.745/0.112	1.746/0.111	1.907/0.086	0.620/0.549
95% confidence interval	Lower	-0.568	-0.543	-0.415	-0.230	-0.112	-0.432
	Upper	11.234	4.876	3.415	1.897	1.446	0.765

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.2.3 Graphical representation of the effect of SMB (400 mg/kg) in center ambulation in Open Field Test

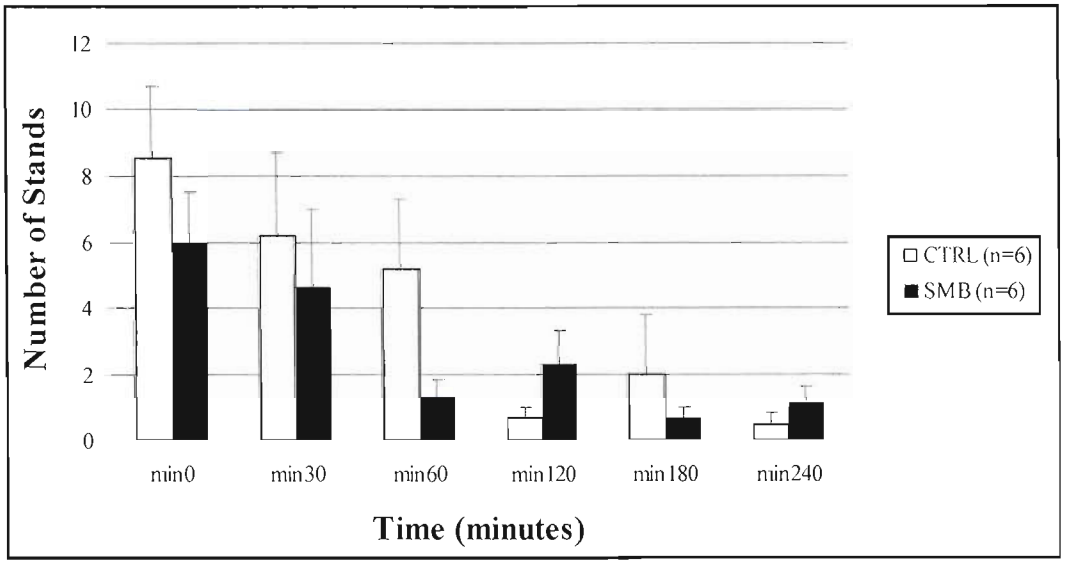


3.3.3.1 Tabular representation of the effect of SMB (100 mg/kg) in standing up behaviour in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		8.500± 2.187	6.167± 2.522	5.167± 2.120	0.667± 0.333	2.000± 1.807	0.500± 0.342
SMB(n=6)		6.000± 1.528	4.667± 2.290	1.333± 0.494	2.333± 0.989	0.667± 0.333	1.167± 0.477
t/p		0.937/ 0.371	0.440/ 0.669	1.761/ 0.133	-1.597/ 0.160	0.725/ 0.485	-1.136/ 0.282
95% confidence interval	Lower	-3.444	-6.091	-1.602	-4.208	-2.762	-1.974
	Upper	8.444	9.091	9.268	0.874	5.428	0.641

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.3.1 Graphical representation of the effect of SMB (100 mg/kg) in standing up behaviour in Open Field Test

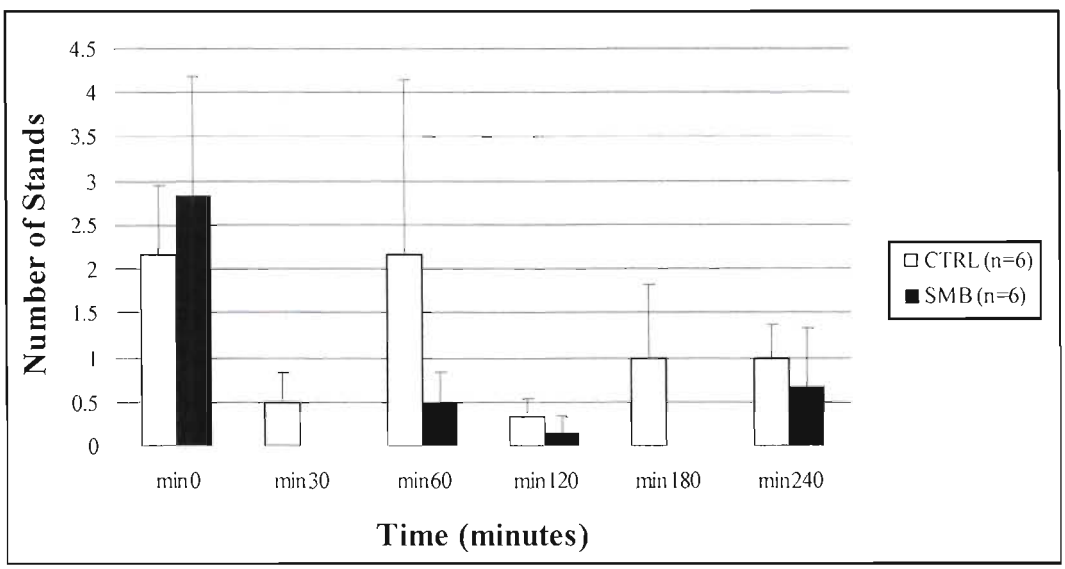


3.3.3.2 Tabular representation of the effect of SMB (200 mg/kg) in standing up behaviour in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		8.500± 2.187	6.167± 2.522	5.167± 2.120	0.667± 0.333	2.000± 1.807	0.500± 0.342
SMB(n=6)		6.000± 1.528	4.667± 2.290	1.333± 0.494	2.333± 0.989	0.667± 0.333	1.167± 0.477
t/p		0.937/ 0.371	0.440/ 0.669	1.761/ 0.133	-1.597/ 0.160	0.725/ 0.485	-1.136/ 0.282
95% confidence interval	Lower	-3.444	-6.091	-1.602	-4.208	-2.762	-1.974
	Upper	8.444	9.091	9.268	0.874	5.428	0.641

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.3.2 Graphical representation of the effect of SMB (200 mg/kg) in standing up behaviour in Open Field Test

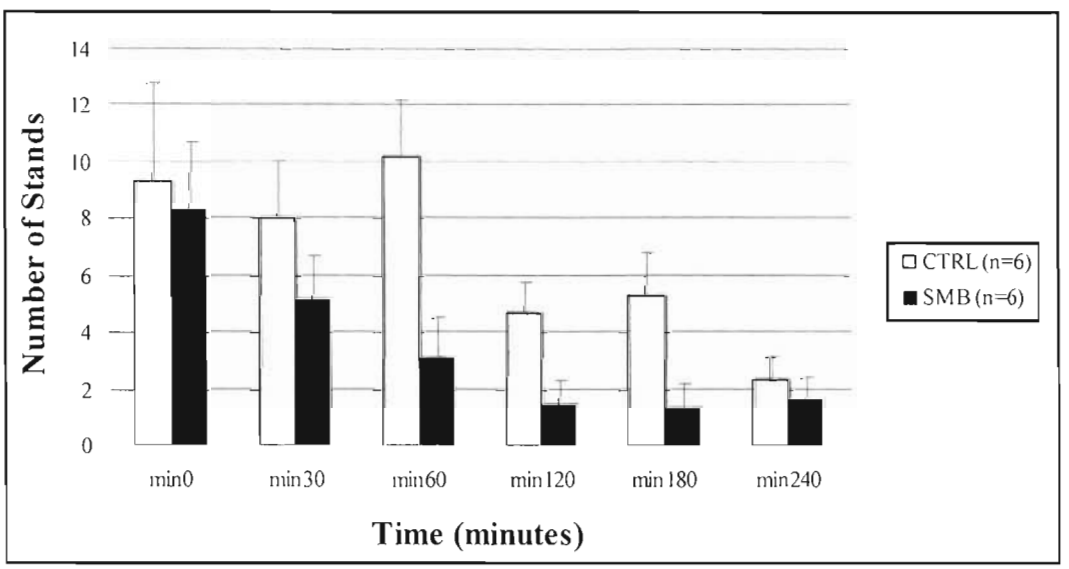


3.3.3.3 Tabular representation of the effect of SMB (400 mg/kg) in standing up behaviour in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		8.500± 2.187	6.167± 2.522	5.167± 2.120	0.667± 0.333	2.000± 1.807	0.500± 0.342
SMB(n=6)		6.000± 1.528	4.667± 2.290	1.333± 0.494	2.333± 0.989	0.667± 0.333	1.167± 0.477
t/p		0.937/ 0.371	0.440/ 0.669	1.761/ 0.133	-1.597/ 0.160	0.725/ 0.485	-1.136/ 0.282
95% confidence interval	Lower	-3.444	-6.091	-1.602	-4.208	-2.762	-1.974
	Upper	8.444	9.091	9.268	0.874	5.428	0.641

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.3.3 Graphical representation of the effect of SMB (400 mg/kg) in standing up behaviour in Open Field Test

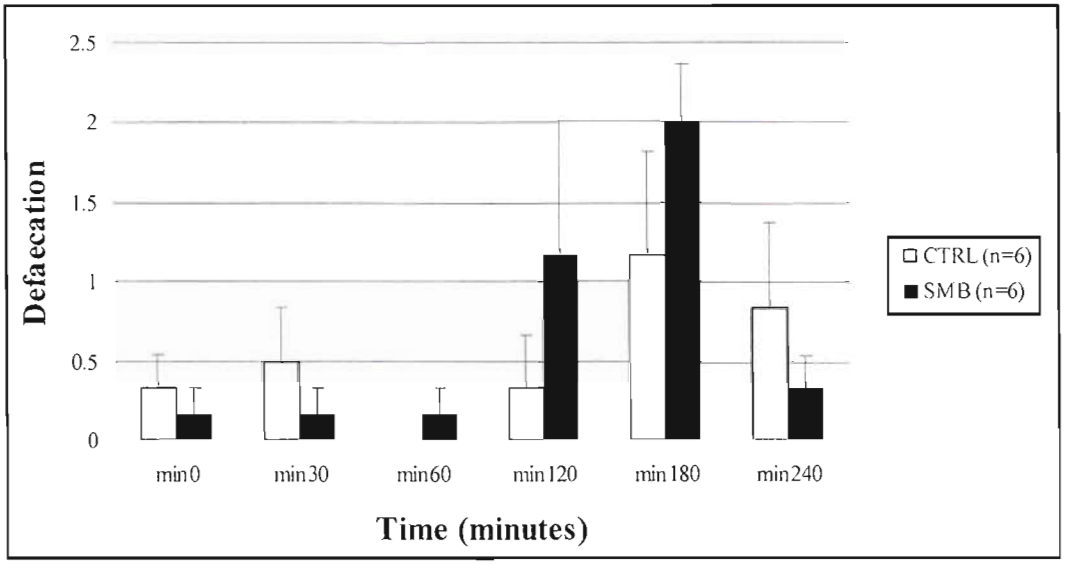


3.3.4.1 Tabular representation of the effect of SMB (100 mg/kg) in emotional defaecation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		0.333± 0.211	0.500± 0.342	0.000± 0.000	0.333± 0.333	1.167± 0.654	0.833± 0.543
SMB(n=6)		0.167± 0.167	0.167± 0.167	0.167± 0.167	1.167± 0.833	2.000± 0.365	0.333± 0.211
t/p		0.620/ 0.549	0.877/ 0.401	-1.000/ 0.363	-0.928/ 0.375	-1.112/ 0.292	0.859/ 0.421
95% confidence interval	Lower	-0.432	-0.513	-0.595	-2.833	-2.502	-0.899
	Upper	0.765	1.180	0.262	1.166	0.836	1.899

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.4.1 Graphical representation of the effect of SMB (100 mg/kg) in emotional defaecation in Open Field Test

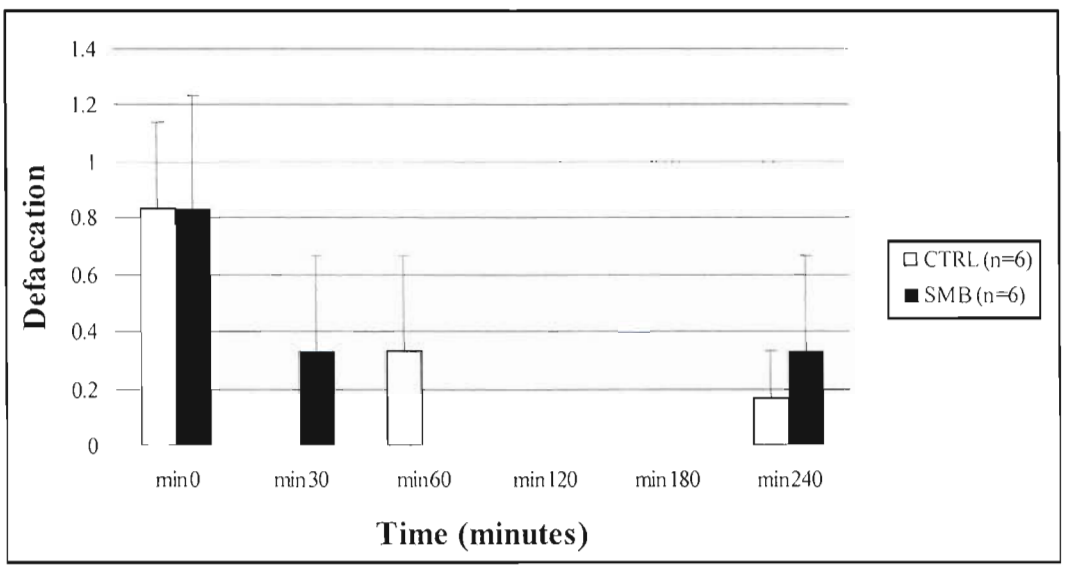


3.3.4.2 Tabular representation of the effect of SMB (200 mg/kg) in emotional defaecation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		0.833± 0.3073	0.000± 0.000	0.333± 0.333	0.000± 0.000	0.000± 0.000	0.167± 0.167
SMB(n=6)		0.833± 0.401	0.333± 0.333	0.000± 0.000	0.000± 0.000	0.000± 0.000	0.333± 0.333
t/p		0.000/ 1.000	-1.000/ 0.363	1.000/ 0.363	0.000/ 0.000	0.000/ 0.000	-0.447/ 0.664
95% confidence interval	Lower	-1.126	-1.190	-0.524	0.000	0.000	-0.997
	Upper	1.126	0.524	1.190	0.000	0.000	0.664

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.4.2 Graphical representation of the effect of SMB (200 mg/kg) in emotional defaecation in Open Field Test

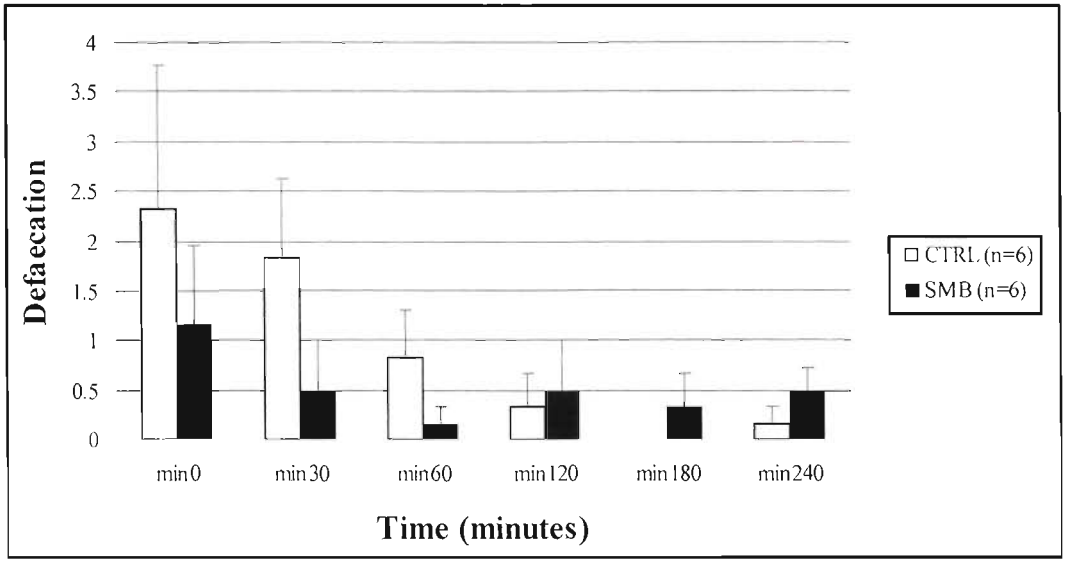


3.3.4.3 Tabular representation of the effect of SMB (400 mg/kg) in emotional defaecation in Open Field Test

Group		Min0	Min30	Min60	Min120	Min180	Min240
Ctrl(n=6)		2.333± 1.430	1.833± 0.792	0.833± 0.477	0.333± 0.333	0.000± 0.000	0.167± 0.167
SMB(n=6)		1.167± 0.792	0.500± 0.500	0.167± 0.167	0.500± 0.500	0.333± 0.333	0.500± 0.224
t/p		0.714/ 0.492	1.423/ 0.185	1.319/ 0.217	-0.277/ 0.787	-1.000/ 0.363	-1.195/ 0.260
95% confidence interval	Lower	-2.476	-0.754	-0.460	-1.506	-1.190	-0.955
	Upper	4.809	3.421	1.793	1.172	0.524	0.288

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant

3.3.4.3 Graphical representation of the effect of SMB (400 mg/kg) in emotional defaecation in Open Field Test



3.4 GASTROINTESTINAL (GI) MOTILITY TEST

Statistical Findings

The pattern in the change in GI motility of SMB treated female mice at dose 100 mg/kg is depicted below:

1st hour

GI motility of the drug (SMB) group was found to be similar to that of the control group in 15 minutes study. In 30 minutes study, SMB was found to increase GI motility slightly when compared to the respective control group.

2nd hour

GI motility of SMB treated mice was found to be less than the control group in both 15 and 30 minutes study.

3rd hour

After 15 minutes study it was observed that the gut motility of the mice in drug (SMB) group decreased slightly than those in control. In contrast, gut motility was found to increase in drug group than the corresponding control in 30 minutes study.

4th hour

The observations made in SMB treated mice were similar to that in the 3rd hour – a slight decrease in GI motility in the 15 minutes study followed by an increase in the 30 minutes study (when compared to the control group).

However, none of the results obtained in the study were statistically significant ($p > 0.05$).

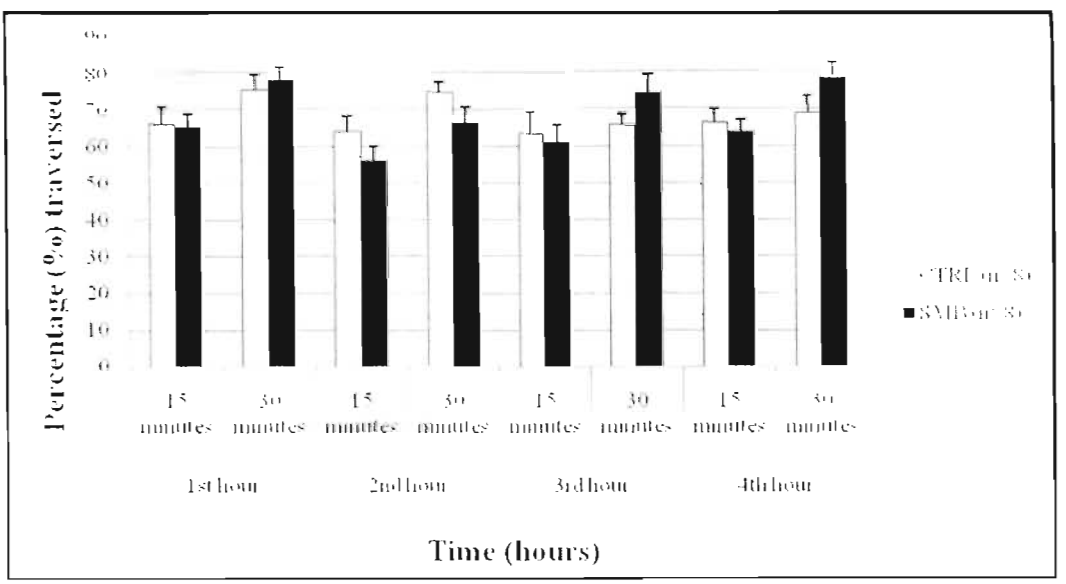
Tabular and Graphical presentation of the effect of SMB (100 mg/kg) on the Gastrointestinal Motility Test utilizing female mice.

3.4.1 Tabular presentation of the effect of SMB (100 mg/kg) in Gastrointestinal (GI) Motility Test using female mice

Group		Percentage (%) Traversed							
		1st hour		2nd hour		3rd hour		4th hour	
		15 min	30 min	15 min	30 min	15 min	30 min	15 min	30 min
CTRL (n = 8)		66.043± 4.831	75.398± 4.454	64.087± 4.124	74.863± 2.736	63.523± 6.032	65.691± 3.179	66.201± 4.161	68.826± 5.106
SMB (n = 8)		65.317± 3.593	78.323± 3.310	56.234± 3.908	66.498± 4.284	61.230± 4.773	74.790± 4.825	64.174± 3.135	78.989± 3.877
t/p		0.121/ 0.906	-0.527/ 0.606	1.382/ 0.189	1.689/ 0.115	0.292/ 0.775	-1.613/ 0.131	.389/ 0.703	-1.585/ 0.135
95% confidence interval	Lower	-12.186	-14.827	- 4.331	-2.332	-14.667	-21.282	-9.146	-23.914
	Upper	13.640	8.979	20.038	19.062	19.262	3.084	13.200	3.588

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant.

3.4.1 Graphical presentation of the effect of SMB (100 mg/kg) in Gastrointestinal (GI) Motility Test using female mice



3.5 COLON TRANSIT TIME TEST

Statistical findings

Bead latency time

SMB treated female mice at a dose of 100mg/kg showed a decrease in bead latency time compared to the control group. But the result obtained was not statistically significant.

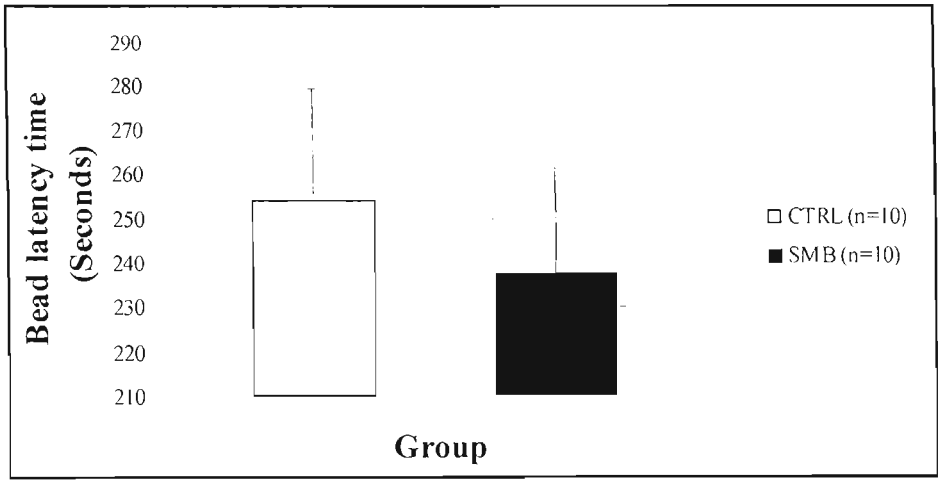
Tabular and Graphical presentation of the effect of SMB (100mg/kg) on the Colon Transit Time Test utilizing female mice.

3.5.1 Tabular representation of the effect of SMB (100mg/kg) on Colon Transit Time Test.

Group		Bead latency time (Sec)
Ctrl (n=20)		253.737±25.489
SMB (n=20)		237.632±23.495
t/p		0.465/0.645
95% confidence interval	Lower	-54.199
	Upper	86.410

N.B :*(< 0.05) =Significant, ** (< 0.01) = Highly Significant, *** (< 0.001) = Very Highly Significant.

3.5.1 Graphical representation of the effect of SMB (100mg/kg) on Colon Transit Time Test.



Chapter Four

Conclusion

4. Conclusion

The treatment and management of renal diseases have always been expensive but cost-effective Ayurvedic medicines have been used to help with urolithiasis through anti-inflammatory, diuretic, litholytic, antispasmodic, and antimicrobial actions, though many of these properties are speculative. Indeed, the most common proposed hypothesis is that herbal therapies alter urinary composition thereby changing supersaturation and reducing stone formation. Though used for thousands of years, reports of side effects and contraindications are poorly documented. Animal studies to support the role of Ayurvedic medications in the management of nephrolithiasis are scarce. In this paper the analgesic, neuropharmacological and gastrointestinal effects were studied using animal models. From the analgesic study it could be concluded that Sukramatrika Vati possibly could be utilized as a useful and potential analgesic for the palliative treatment of urolithiasis. In addition, the results from the neuropharmacological studies show that the preparation probably also possesses no anxiogenic effects. Therefore, it would be right to emphasize that the Ayurvedic preparation Sukramatrika Vati has minimum side effects. However, random data were obtained with the gastrointestinal studies. Hence, further studies using sophisticated models need to be performed to fully fathom the safety of Sukramatrika Vati for Alternative Medical Care.

REFERENCES

Alam, M.(2009). 20m kidney patients in country. *The Daily Independent*. p. A1.

Ayurveda (2009). Retrieved from the Web December 10, 2009, from Herbs2000. http://www.herbs2000.com/h_menu/t_ayurveda.htm

Ayurveda – History (2009). *History of Ayurveda*. Retrieved from the Web December 10, 2009, from Medindia. http://www.medindia.net/ayurveda/index.asp

Ayurveda with no side effect (2007). Retrieved from the Web December 10, 2009, from Medicines of India. http://mediciensofindia.blogspot.com/2007/09/disease-healthremedy-and-research.html

Ayurvedic medicine: India's science of life (2009). Retrieved from the Web December 10, 2009, from Life123. http://www.life123.com/health/alternative-medicine/ayurveda/ayurvedic-medicine-indias-science-of-life.shtml

Ayurvedic yog – SUKRAMATRKA VATI. (n.d.). Retrieved from the Web December 10, 2009, from easy Ayurveda™.

http://www.ayurvedaconsultants.com/formulation_display_name1.aspx?string=sukrama trka vati

Boissier, J.R., Simon, P. & Lwoff, J.M. (1964). *Use of a particular mouse reaction (Hole board method) for the study of psychotropic drugs*. Therapie, 19, 571-583.

Chatterjee, T.K. (1993). *Handbook on Laboratory Mice and Rats*. Department of Pharmaceutical Technology, Jadavpur University, India. 1st ed, 157.

Choleris, E., Thomas, A.W., Kavaliers, M.,& Prato, F.S. (2001). A detailed ethological analysis of the mouse open field test: effects of diazepam, chlordiazepoxide and an extremely low frequency pulsed magnetic field. *Neuroscience and Biobehavioural Reviews*. 25(3), 235-60.

Eddy, N.B.& Leimbach, D. (1953). *Synthetic analgesics: II Dithienylbutenyl and dithienylbutylamines*. J. Pharmac. exp. Ther., 107, 385-393.

Gokshura Compositions.(n.d.) Retrieved from the Web December 10, 2009, from TribulusTerrestris.net.

http://tribulusterrestris.net/Tribulus-product/Tribulus-composition.asp

History of Ayurveda (n.d.) Retrieved from the Web December 10, 2009, from AyurvedicCure.com. http://www.ayurvediccure.com/ayurveda_ayurvedic_herbs/ayurveda_history.htm

Islam. H.A. (1991). Traditional medicine in Bangladesh. *In Touch, 10* (99), 8-9.

Jadavpur University (1993). *Handbook on Laboratory Mice and Rats* (Edition 1, p. 157). Jadavpur, India: Chatterjee, T.K.

Kieley, S., Dwivedi,R.& Monga,M.(2008). Ayurvedic Medicine and Renal Calculi. *JOURNAL OF ENDOUROLOGY,*22, 613.

Korenchan, M. C.(n.d.) *History of Ayurvedic Medicine.* Retrieved from the Web December 10, 2009, from eHow. http://www.ehow.com/about_5382579_history-ayurvedic-medicine.html

Martinez, V., Wang, L., Rivier, J.E., Vale, W. & Tache, Y. (2002). *Differential actions of peripheral corticotropin-releasing factor (CRF), Urocortin II, and Urocortin III on Gastric Emptying and Colonic transit in mice: Role of CRF receptor subtypes I and II.* Journal of Pharmacology and Experimental Therapeutics, 301(2), 611-617.

Mishra, L. C. (2004). Introduction in *Scientific Basis for Ayurvedic Therapies*. CRC Press, USA.

Nakama, M., Ochiai, T., & Kowa, Y. (1972). *Effects of Psychotropic Drugs on Emotional Behavior; Exploratory Behavior of Naive Rats in Holed Open Field*, Japan J Pharmacol; 22, 767-775.

National Center for Complementary and Alternative Medicine. (2009). *Ayurvedic medicine: an introduction*. Retrieved 15 September 2009, from NCCAM. http://nccam.nih.gov/health/ayurveda/introduction.htm

Open field locomotor activity (2005). Retrieved 26 October 2009, from *The Behavior Core*. http://www.psychiatry.uc.edu/BehaviorCore/HTML/openfield.html

Open-field test (n.d.). Retrieved 26 October 2009, from *Panlab*. http://www.panlab.com/panlabWeb/Software/SMART%20JUNIOR/pdfProtocols/AS%20Open%20Field%20with%20smart%20JUNIOR.pdf

Parvez, S.(2009, July 12). Herbal medicines get new lease of life. *The Daily Star*. Retrieved December 5, 2009 http://www.thedailystar.net/newDesign/index.php

Pierce, N.F., Carpenter, C.C.J., Elliot, H.I., Greenough, W.B. (1971). Effects of prostaglandins, theophylline and cholera exotoxin upon transmucosal water and electrolyte movement in canine jejunum. *Gastroenterol.* 60, 22–32.

Santos, A.R.S., Filho, V.C., Niero, R., Viana, A.M., Moreno, F.N., Campos, M.M., Yunes, R.A., Calixto, J.B., (1994). *Analgesic effects of callus culture extracts from selected species of Phyllantus in mice.* Journal of Pharmacy and Pharmacology 46, 755–759.

Shahriar, M., Bhuiya, M.S., Khan, M.T.H., Gafur, M.A., Choudhri, M.S.K. (2000). Pharmacological study of *Symplocos racemosa* Roxb. *Ham Med.*, 43, 8-18.

Sridevi,V., Lakshmi,I.R. & Rao, I.S.(2004). *Urolithiasis (Mutrashmari), Scientific Basis for Ayurvedic Therapies*. CRC Press, USA.

Tang, Xican, Lin, Zhigong, Cai, Wen, Chen, Nian, Shen and Li (1984). Anti-inflammatory effect of 3-acetylaconitine. *Acta Pharmacologica Sinica*. 5, pp. 85–89.

Trinchieri, A. (2006). Epidemiological trends in urolithiasis: impact on our health care systems. *Urological Research*,34, 151-156.

Woolfe, G. & MacDonald, A.D. (1944). *The Evaluation of the Analgesic Action of Pethidine Hydrochloride (Demerol)*. J Pharmacol Exp Ther; 80, 300-307.

ABBREVIATIONS

The following abbreviations were used throughout this paper:

Acetic acid	AA
Control	CTRL
Female	F
Male	M
Gastric Emptying	GE
Gastrointestinal	GI
Gram	g
Hour	h
Sukramatrika Vati	SMB
Intraperitoneal	i.p.
Per oral	p.o.
Kilogram	kg
Minute	min
Milligram	mg
Millilitre	ml
Second	s
Weight	wt