

Original article:

Skeletal muscle relaxant property of baclofen by using rotarod on albino mice

Dr. N. Sundara Veena¹, Dr. K.Sivaji¹, Dr. G.V. Benerji¹, M.Farid Babu¹, D.Rekha Kumari¹

¹Associate Professor of Biochemistry RIMS, Srikakulam, AP

¹Associate Professor of Pharmacology, GSL Medical College Rajahmundry, AP

¹Professor of Biochemistry, KIMS&RF Amalapuram, AP

¹Assistant Professor of Biochemistry, KIMS&RF Amalapuram, AP

¹Assistant Professor of Biochemistry, KIMS&RF Amalapuram, AP

Corresponding author: Dr. G.V. Benerji

Abstract:

The comparison of skeletal muscle relaxant property of Baclofen, in experimental animal models. To evaluate skeletal muscle relaxant effect of Baclofen to evaluate the agents having good skeletal muscle relaxant properties. The earliest known use of muscle relaxant drugs dates back to the 16th Century. When European explorers encountered natives of the Amazon Basin in South America using poison-tipped arrows that produced death by skeletal muscle paralysis. This poison known today as curare, led to some of the earliest scientific studies in pharmacology. Its active ingredient, tubocurarine, an alkaloid as well as many synthetic derivatives, played a significant role in scientific experiments to determine the function of Acetylcholine in neuromuscular transmission. By 1943 neuromuscular blocking drugs became established as muscle relaxants in the practice of anesthesia and surgery. A muscle relaxant is a drug which effects skeletal muscle function and decreases the muscle tone. It may be used to alleviate symptoms such as muscle spasms, pain and hyperreflexia. The term "Muscle relaxant" is used to refer to two major therapeutic groups: neuromuscular blockers & spasmolytics.

This study was designed to evaluate the skeletal muscle relaxant properties of Baclofen, It is found that Baclofen appeared to be more effective and safe. Previous studies suggested that the CNS depression and the non specific muscle relaxation effect can reduce the response of the motor coordination.

Skeletal muscle relaxants are used to treat two different types of conditions.

1. Spasticity from upper motor neuron syndromes
2. Muscular pains or spasms from peripheral musculoskeletal conditions.

In this study centrally acting skeletal muscle relaxants Baclofen and used and muscle relaxants activity. The study was carried out in albino mice weighing 40gms.

Thus it was found that Baclofen Demonstrated muscle relaxant property but produced less muscle relaxant property as compared to Baclofen. Hence Baclofen is considered to have maximum muscle relaxant property. This may be due to its high lipid solubility.

Keywords: Muscle relaxant, diazepam, rotarod, albino mice

Introduction:

The earliest known use of muscle relaxant drugs dates back to the 16th Century. When European explorers encountered natives of the Amazon Basin in South America using poison-tipped arrows that produced death by skeletal muscle paralysis. This poison known today as curare, led to some of the earliest scientific studies in pharmacology. Its active ingredient, tubocurarine, an alkaloid as well as many synthetic derivatives, played a

significant role in scientific experiments to determine the function of Acetylcholine in neuromuscular transmission. By 1943 neuromuscular blocking drugs became established as muscle relaxants in the practice of anesthesia and surgery. A muscle relaxant is a drug which effects skeletal muscle function and decreases the muscle tone. It may be used to alleviate symptoms such as muscle spasms, pain and hyperreflexia. The term "Muscle relaxant" is used to refer to two major therapeutic groups: neuromuscular blockers & spasmolytics.

Neuromuscular blockers – Act by interfering with transmission at the neuromuscular end plate and have no central nervous system (CNS) activity. They are often used during surgical procedures and in intensive care and emergency medicine to cause paralysis.

Spasmolytics – Also known as "Centrally acting "muscle relaxants, are used to alleviate musculoskeletal pain and spasms and to reduce spasticity in a variety of neurological conditions.

While both neuromuscular blockers and spasmolytics are often grouped together as muscle relaxants.

Skeletal muscle relaxants are classified by their pharmacologic property as either Anti-spasticity and Anti-spasmodic agents.

Anti-spasticity - Agents are used to reduce spasticity that interferes with function or daily living activities, such as in cerebral palsy, multiple sclerosis and spinal cord injuries.

Neuromuscular blockers:

Detailed view of a neuromuscular junction:

1. Presynaptic terminal
2. Sarcolemma
3. Synaptic vesicle
4. Nicotinic acetylcholine receptor
5. Mitochondrion

Antispasmodic- agents are primarily indicated as adjuncts to rest, physical therapy and relief of discomfort associated with acute ; painful musculoskeletal disorders.

Muscle relaxants are used to treat two different types of underlying conditions.

Spasticity from upper motor neuron syndromes.

Muscular pain or spasms from peripheral musculoskeletal conditions.

Although muscle relaxants have by convention been classified into one group, the Food and Drug Administration (FDA) has approved only a few medications in this class for treatment of spasticity. The remainder are approved for treatment of musculoskeletal condition.

Muscle relaxants for treatment of spasticity:

Spasticity is a state of increased muscular tone with exaggeration of the tendon reflexes. Some of the more common conditions associated with spasticity and requiring treatment include multiple sclerosis, spinal cord injury, traumatic brain injury, cerebral palsy and post-stroke syndrome. In many patients with these conditions; spasticity can be disabling and painful with a marked effect on functional ability and quality of life.

The upper motor neuron syndrome is a complex of signs and symptoms that can be associated with exaggerated cutaneous reflexes, autonomic hyperreflexia, dystonia, contractures, paresis, lack of dexterity, and fatigability.

Spasticity from the upper motor neuron syndrome can result from a variety of conditions affecting the cortex or spinal cord.

Muscle relaxants for treatment of musculoskeletal conditions:

Muscle spasm is defined as a sudden involuntary contraction of one or more muscle groups and is usually an acute condition associated with muscle strain (partial tear of a muscle) or sprain (partial or complete reupture of a ligament). Common musculoskeletal conditions causing tenderness and muscle spasms include fibromyalgia, tension headaches, myofascial pain syndrome, and mechanical low back pain or neck pain. If muscle spasm is present in these conditions. It is related to local factors involving affected muscle groups.

Efficacy:

Most studies have shown the skeletal muscle relaxants to be more effective than placebo in the treatment of acute painful musculoskeletal disorders and muscle spasm, while efficacy was less consistent when treating chronic disorders. When muscle relaxants were used alone, they were not consistently superior to simple analgesics in relieving pain. When the skeletal muscle relaxants were used in combination with analgesics, pain relief is superior to either agent used alone,. Studies have suggested that these drugs are effective, have tolerable side effects and can be an adjunct in the treatment of painful musculoskeletal conditions with associated muscle spasm.

Centrally acting skeletal muscle relaxants are generally prescribed either as single agents or as components of combination products. The Food and drug Administration has approved indications for these medications as adjuncts to rest and physical therapy for relief of acute, painful musculoskeletal problems. Clinically the mild pain associated with the majority of cases of minor muscle strains and minor injuries is self limiting. Most patients usually respond rapidly to rest. An anti-inflammatory drug may be useful when there is considerable tissue damage and edema. On the other hand, severe musculoskeletal strains and sprains, trauma and cervical or lumbar radiculopathy as a consequence of degenerative osteoarthritis, herniated disk, spondylitis or laminectomy, often cause moderate or severe and more chronic painful skeletal muscle relaxants alone or in combination with an analgesic are frequently prescribed. Results of some studies have suggested that a formulation of a muscle relaxant and an analgesic. Provides greater benefit in patients with acute musculoskeletal problems than similar doses of an analgesic alone.

Review of literature:

Skeletal muscle relaxants are a heterogeneous group of medications. As a class, they are structurally and pharmacologically diverse. Muscle relaxants are used to treat two different types of underlying conditions.

- Spasticity from upper motor neuron syndromes.
- Muscular pain or spasms from peripheral musculoskeletal conditions

Although muscle relaxants have by convention been classified into one group, the Food and Drug Administration (FDA) has approved only a few medications in this class for treatment of spasticity. The remainder are approved for treatment of musculoskeletal conditions

History:

The earliest known use of muscle relaxant drugs dates back to the 16th century, when European explorers encountered natives of the Amazon Basin in South America using poison-tipped arrows that produced death by skeletal muscle paralysis. This poison known today as curare, led to some of the earliest scientific studies in

pharmacology. It is active ingredient, Tubocurarine the alkaloid curarine is transported in bamboo tubes so it is known as tubocurarine the active form of alkaloid dextroisomer so it is called as d-tubocurarine. As well as many synthetic derivatives, played a significant role in scientific experiments to determine the functions of Acetylcholine in neuromuscular transmission. By 1943 neuromuscular blocking drugs became established as muscle relaxants in the practice of anesthesia and surgery.

Materials and methods:

Materials:

➤ **Chemicals and solutions:**

- BACLOFEN
- Double distilled
Water
Normal saline

➤ **Animals:-**

- Albino mice weighing 40 gms

➤ **Equipment:-**

- Rota Rod
- Insulin syringes
- Measuring jar
- Glass beakers
- Animal weighing balance
- Animal cages
- Cotton
- Spirit

Stop Watch

➤ **Statistical Method :-**

- ONE WAY ANOVA

➤ **Rota rod:-**

- Instrument description

Operation

Techno Rota Rod:

The rotarod assembly is immensely useful for screening drugs effecting motor co-ordination. It consists of four experimental compartments with a rotating rod of about 25mm diameter and having speeds of approx – 5,10,15,20 & 25 revolution / min. Time interval counters are provided in each compartment. The apparatus works off 220/30 volts single phase 50 HZ Ac.

On the floor of each compartment there is a plat form balanced on a pivot and held n position by device. When the from of the plat form is raised a little, it sticks up and completes electrical circuit of its counter which records in seconds. When the rat falls off the rotating shaft this plat form is tripped.

Operation:

1. Raise the front end of the platforms a little if necessary.
2. Connect the instrument of 220 V AC supply and switch on
3. The pilot camp will light up.
4. Reset the counters to zero by pushing the reset switch.
5. Turn the (Change RPM) select or clock wise for required speed. The shaft will rotate.
6. The digits of the couner will begin to second time in seconds.
7. The counter of all platforms will stop when plate is pressed.
8. To restart the counter raise the plat form a little by one finger.
9. Shaft speed may be changed by selector switch to obtain fine RPM
Use fine mode control o adjust the required RPM (5-25 or what ever).
10. In practice all the plat forms are in on position and apparatus is switched on. So that the shaft begins to rotate and the counter also works. Now place a nice on shaft and press
reset switch simultaneously. Thus each counter will record will the endurance time of its corresponding animal.
11. To change shaft for rat and nice open the large hexanut anti clock wise at left side of the experimental rod and instrument. Now loose the screw of the experimental rod also anti clock wise and pull out the shaft. Insert another required ex. Rod & light it clock wise with their screw then retight large hexsawnut at their proper place.

Method:

Object:

To study the muscle relaxant property of drug (Muscle of drug (Muscle grip strength) in mice using Rota rod apparatus.

Principle:

One of the important pharmacological actions of anti anxiety agents of Benzodiazepin and centrally acting d₂ against class of drugs are muscle relaxing property.

The skeletal muscle relaxations together with taming or calming effect these agents reduce anxiety and tension. The loss of muscle grip is an indication of musclerelaxation. This effect can be easily studied in animals using inclined plane or rotating rods.

The difference in the fall off time from the rotating rod between the control and drug treated animal is taken on an index of muscle relaxations. The angle of the slope of the inclined plane or the rate of rotation of the rod should be adjusted such that a normal mouse can stay on the plane or on the rod for an appreciable period (3-5 min) of time.

Requirements:-

Animals -	Mice (35-40 gms)
Drug	- Baclofen
Equipment	- Rota Rod apparatus (Techno)

- Procedure:-**
1. The mice were weighed and numbered.
 2. The animals were placed one by one on the rotarod
 3. Turn on the Rota Rod. Select an appropriate speed (20-25 rpm is ideal)
 4. The animals were trained for 3 hrs in a day with a gap of 1hr in between and was trained 10 mins on rotarod.
 5. All the drugs were administered by intra peritoneal route by using 26G needle.
 6. Injected Baclofen (3,4,5 mg/kg) to all the animals. After 30- minutes repeat the experiment.
 7. Note down the fall of time.

Compared the fall of time of animals before and after treatment.

Observations and results:

Table-1: Comparison of Baclofen at the dose of 3mg/kg & 4mg/kg & 5 mg/kg body Weight

SI. No	Treatment	3mg/kg	4mg/kg	5mg/kg
1	Baclofen	33.33	79.50	89.28
2	Baclofen	42.85	77.78	91.26
3	Baclofen	46.15	81.28	90.62
4	Baclofen	34.78	72.73	94.73
5	Baclofen	36.27	80.26	90.24
6	Baclofen	39.51	86.67	90.32
7	Baclofen	25	64.29	90.74
8	Baclofen	32.72	64.29	87.75

Table-2: Comparison of Baclofen a the dose of 3mg/kg & 4mg/kg & 5 mg/kg body Weight

Treatment	Mean	S.D	S.E
Baclofen(3mg/kg)	36.326	1.015	+ 2.322
Baclofen(4mg/kg)	75.850	8.107	+ 2.866
Baclofen(5mg/kg)	90.618	1.985	+ 0.702

Discussion:

This study was designed to evaluate the skeletal muscle relaxant properties of Baclofen, It is found that Baclofen appeared to be more effective and safe Previous studies suggested that the CNS depression and the non specific muscle relaxation effect can reduce the response of the motor coordination.

Skeletal muscle relaxants are used to treat two different types of conditions.

1. Spasticity from upper motor neuron syndromes
2. Muscular pains or spasms from peripheral musculoskeletal conditions.

In this study centrally acting skeletal muscle relaxants Baclofen and used and muscle relaxants activity The study was carried out in albino mice weighing 40 gms. Eight mice were included in each group to evaluate the muscle relaxants property in different concentrations such as 3,4, and 5 mg/kg Baclofen, There are various screening techniques to assess muscle relaxants property of a drugs Baclofen, baclofen and quinine. For initial screening of a drug mouse is one of the best animals as it is easy to handle can be used repeatedly since the animal is not sacrificed by rotarod method. Second choice of animal is rat.

There are various screening techniques to assess muscle relaxants property of a drug Baclofen, For initial screening of a drug mouse is one of the best animals as it is easy to handle can be used repeatedly since the animals is not sacrificed by rotarod method. Second choice of animal is rat.

In the present study it was found that the Mean % increases in activity for Baclofen 81.201 with 3 mg/kg,85.159 with 4 mg/kg and 92.419 with 5mg/kg. When rotarod method was used, whereas it is 36.326 with 3 mg/kg. 75.850 with 4mg/kg, when assessed by rotarod method.

At 3 mg/kg does the Mean % of increases in activity for Baclofen is 81.201

At 4 mg/kg dose the mean % of increase in activity for Baclofen is 85.159.

At the 5 mg/kg dose the mean % of increase in activity for Baclofen is 92.419. The mean difference is low but it is marked difference.

This study shows at this concentration the Baclofen and it increases the muscle strength property is less than the Baclofen This study shows at all the concentrations muscle relaxant property when compared with baclofen and quinine, baclofen is more effective when compared to quinine.

Summary

This study carried out to comparison of muscle relaxant property of Baclofen by rotarod test. It was found that the drug Demonstrated muscle relaxant action.

Conclusion:

The present study was carried out to compare the muscle relaxant property of Baclofen given in different concentrations is experimental model. The experimental model is rotarod test in Albino mice. The Baclofen and were given in concentration of drug increases, the muscle relaxant property is also increase when assesses by rotarod. Thus it was found that Baclofen Demonstrated muscle relaxant property but produced less muscle relaxant

property as compared to Baclofen. Hence Baclofen is considered to have maximum muscle relaxant property. This may be due to its high lipid solubility.

Bibliography:

1. Definition of Muscle relaxant.” MedicineNet.com. © 1996-2007. Retrieved on September 19, 2007.
2. “Muscle relaxant.” Medilexicon. © 2007. Retrieved on September 19, 2007.
3. “Muscle relaxants.” WebMD. Last Updated: February 15, 2006. Retrieved on September 19, 2007.
4. “Skeletal Muscle Relaxant (Oral Route, Parenteral Route).” Mayo Clinical. Last updated: April 1, 2007. Retrieved on September 19, 2007.
5. Miller, R.D. “skeletal Muscle Relaxants,” in, “Basic & Clinical Pharmacology: Seventh Edition,” by Bertram G. Katzung. Published by Appleton & Lange, s
6. Bowman, W.C. “Neuromuscular block.” Br. J. Pharmacol. January 2006.
7. C.R. Craig, R.E. Stizel (2003) Modern Pharmacology with clinical applications Lippincott Williams & Wilkins ISBN 0781737621 p339
8. Rang, H.P. & Dale, M.M. “Drugs Used in Treating Motor Disorders” in, “Pharmacology 2nd Edition” published by Churchill Livingstone London, 1991, p684-705.
9. Standaert, D.G. & Young, A. B. “Treatment Of Central Nervous System Degenerative Disorders” in, “Goodman & Gilman’s The pharmacological Basis of Therapeutics 10th Edition” by Hardman, J.G. & Limbird, L.E. published by McGraw Hill, 2001, p.550-568.
10. Charney, D.S., Mihic, J & Harris, R.A. “Hypnotics and Sedatives” in, “Goodman & Gilman’s the pharmacological Basis of Therapeutics 10th Edition” by Hardman, J.G. & Limbird, L.E. published by McGraw Hill, 2001, p.399-427.
11. Cazalets JR, Bertrand S, Sqalli-Houssaini Y, Clarac F (1998). “GABAergic control of spinal locomotor networks in the neonatal rat”. Ann. N. Y. Acad.Sci. 860: 168-80. PMID 9928310.
12. Young R.R. (editor). “Symposium: Role of tizanidine in the treatment of spasticity.” Neurology. 1994, Vol. 44 (Suppl. 9)p.1.
13. Bras H, Jankowska E, Noga B, Skoog B (1990). “Comparison of Effects of Various Types of NA and 5-HT Agonists on Transmission from Group II Muscle Afferents in the Cat” (12): 1029-1039. PMID 12106064.
14. Jankowska E, Hammar I, Chojnicka B, Heden CH (2000). “Effects of monoamines on interneurons in four spinal reflex pathways from group I and/or group II muscle afferents”. Eur.J. Neurosci. 12 (2): 701-14. PMID 10712650.
15. Young, R.R; Weigner, A.W. “Spasticity.” Clin. Orthop. 1987, Vol.219, p. 50
16. “Motor neurons” <http://www.wikipedia.com>.
17. Baclofen. Pubchem. National Institute of Health: National Library of Medicine (2006). Retrieved on 2006-3-11.
18. Baclofen. Medical Subject Headings (MeSH). National Library of Medicine (2006), Retrieved on 2006-03-10
14. Jankowska E, Hammar I, Chojnicka B, Heden CH (2000). “Effects of monoamines on interneurons in four spinal reflex pathways from group I and/or group II muscle afferents”, Eur.J. Neurosci. 12 (2): 701-14. PMID 10712650.

15. Young, R.R; Weigner, A.W. "Spasticity." Clin. Orthop. 1987, Vol.219, p. 50
16. "Motor neurons" <http://www.wikipedia.com>.
17. Baclofen. Pubchem. National Institute of Health: National Library of Meicine (2006), Retrived on 2006-3-11.
18. Baclofen. Medical Subject Headings (MeSH). National Library of Medicine (2006) , Retrieved on 2006-03-10.
21. Baclofen: indications. Rxlist.com (January 24, 2005). Retrieved on 2006-03-11.
22. Baclofen: description. Rxlist.com RxList Inc. (January 24, 2005).Retrieved on 2006-03-10.
23. Baclofen: Side Effects. RxList.com Retrieved on September 26, 2006.
24. Baclofen: abuse and dependence. Rxlist.com. RxList Inc. (January 24, 2005). Retrieved on 2006-03-10.