The naked mole-rat (Heterocephalus glaber) is an interesting animal model for nociceptive mechanisms, as its nociceptive system has been shown to differ from that of other mammals. It has been reported that the animal completely lacks cutaneous C-fibre immunoreactive to substance P and calcitonin gene-related peptide (GRRP) (2, 3). In the hot-plate test, the animals were found to display aggressive behavior and hyperalgesia, instead of analgesia, when treated with morphine or pethidine (8, 9). This appears to be related to different properties of μ-, δ- and κ-receptors in mole-rats compared to other rodents (10). In the formalin test, however, stimulation with μ-, κ- and δ-opioid receptor agonists resulted in antinociceptive effects (11). The antinociceptive effects of non-steroidal and steroidal anti-inflammatory drugs have also been investigated in this animal (12, 13).

Other receptor systems, including the cholinergic receptor system, are known to be involved in nociceptive regulation and have been extensively investigated in other animal...
species [reviewed in (14)]. The role of the cholinergic receptor system in antinociception has been evaluated in several species, including rats (15-24), mice [reviewed in (25)], sheep (26,27), cats (28) and humans (29,30). To our knowledge, the cholinergic receptor system has not yet been investigated in the naked mole-rat. To our belief, studies of the nociceptive system in animal species of different phylogeny may provide important information about the underlying mechanisms of pain, we consider it important to investigate the effect of cholinergic receptor agonists and antagonists in the naked mole-rat.

The aim of this study was to characterize the potential use of the mole rat as a model for studying cholinergic system in pain transmission. For this purpose, the three commonly used analgesiometric tests – the tail-flick, hot-plate and formalin tests – were applied in order to evaluate effects of cholinergic agonists and antagonists in antinociception, and their possible interactions with the opioid system in the naked mole-rat. It was hypothesized that the cholinergic receptor system is involved in antinociceptive mechanisms in the naked mole-rat. This hypothesis was tested by demonstrating that intraperitoneal injection of the muscarinic receptor (mACHR) agonist oxotremorine, and the nicotinic receptor (nACHR) agonist epibatidine, would reduce pain-related behavior in the three applied tests, and that this reduction would be reversed by co-administration with the mAChR antagonist atropine, and with the nAChR antagonist mecamylamine, respectively. Furthermore, it has been hypothesized that the opioid receptor system is involved in the cholinergic regulation of nociception. This hypothesis was tested by demonstrating that the effect on pain-related behavior caused by oxotremorine and epibatidine in the three analgesiometric tests would be reversed by co-administration of the opioid receptor antagonist naloxone.

Materials and Methods

Animals. Naked mole-rats (Heterocephalus glaber) were captured from Kathekani and Kambu areas in the Kilwezi district, Kenya. The animals were captured under a permit issued by the Kenya Wildlife Services (KWS). The experimental procedures were performed after ethical approval of KWS and of the Institutional Animal Care and Use Committee (the Faculty of Veterinary Medicine Research Ethics Committee of the University of Nairobi). The experimental procedures were conducted in accordance with the guidelines set forth by the American Physiological Society (31). The animals were under health surveillance by veterinary staff, and found to be in good health throughout the duration of the experimental period. A total of 317 male and female adult animals weighing 30-35 g were captured and used in the study. Each animal was used only once, and euthanized with an intraperitoneal injection of pentobarbitone, 200 mg/ml, after testing was completed.

Housing and husbandry. Animals were housed in colonies of 50-70 in designated cages covered by non-transparent lids. The cages, made of plastic glass and painted with black super gloss on the outside surface, measured 70×50×20 cm. Each cage comprised of two compartments, with an interconnecting tunnel that measured 30×10×10 cm between the sub-dividing walls. Wood shavings mixed with sand were used as beddings and were changed once a week to ensure the cages were fresh and damp-free. To avoid the drying and scaling of the skin of the animals, the humidity in the room was maintained at 45-50%. The animals were fed on fresh carrots and sweet potatoes (4 g per animal and day). All naked mole-rats were acclimatized to the new environment for one month before experimentation. During this period and during the experiments, the naked mole-rats were handled and weighed daily.

Tail-flick test. The tail-flick test was performed using an IITC model 33D analgesimeter (IITC Inc., Woodland Hills, CA, USA) with a sensitivity setting of 10 and beam at 8. In order to protect the tail from tissue damage, heat cut-off was set at 10 seconds in the absence of a tail-flick. Before the start of the experiments, the naked mole-rats were acclimated to the restrainer for 30 min per day for a period of 30 days. Thirty minutes after drug or saline injection, the animal was placed gently in the restrainer and a radiant beam focused on the dorsal surface of the tail, at mid-point to the tip, and until occurrence of a flick or cut-off. The latency from the application of heat until the animal flicked its tail away from the heat source was recorded. Any animal that failed to respond before the cut-off at 10 sec would be assigned a response latency of 10 sec. However, no animal reached the cut-off time on any occasion.

Hot-plate test. The hot-plate test was performed using an IITC model 35D analgesimeter, which consisted of a copper plate (7×29 cm) enclosed in a 30×30×30 cm lidded perspex box. During experimentation, the plate was heated to a fixed temperature of 60°C. Before the start of the experiments, the naked mole-rats were acclimated to the plate at room temperature, 30 min per day for a period of 30 days. Prior to tests, the animal was examined to ascertain that it had normal sensorimotor function by observing its ability to explore and negotiate the corners of the perspex box. Thirty minutes after drug or saline injection, the animal was placed gently on the hot-plate and the latency (in seconds) to escaping or jumping (hot plate response latency) was recorded. To guard against damage to the paws, any animals that failed to respond by 60 sec was removed from the plate and assigned a response latency of 60 sec. However, no animal reached the cut-off time on any occasion.

Formalin test. The naked mole-rats were acclimatized to the observation chamber (measuring 30×30×30 cm) for 60 min daily during the 30-day acclimation period. Prior to the formalin test, each animal was acclimatized for 30 min. A 30-gauge needle (U-100 insulin syringe) was used to inject 20 μl of 10% formalin in 0.9% NaCl intradermally into the dorsal side of the rear right hind paw. The volume and concentration of formalin used was based on previous experiments (11, 12). The animal was returned to the observation chamber immediately after the formalin injection, and the observation period of one hour started. The time in seconds the animal spent licking the injected hind paw was recorded in blocks of five minutes.

Drugs and chemicals. Oxotremorine sesquisulfate salt, (+/-)-epibatidine dihydrochloride, atropine sulfate salt, and naloxone hydrochloride dehydrate were purchased from Sigma-Aldrich, Stockholm, Sweden. Mecamylamine hydrochloride was purchased from Tocris Bioscience, Copenhagen, Denmark. Formaldehyde 40%
was purchased from Merck, agents F & S, Nairobi, Kenya. All drugs were weighed and dissolved in physiological saline (0.9% NaCl) and stored as a stock solution at a temperature of 2-4˚C.

**Drug administration.** All the drugs except formalin were injected intraperitoneally 30 min prior to testing. The injections were given in a volume of 50 μl, using a U-100 insulin syringe with a 30-gauge needle. The doses were based on data from preliminary studies, as well as from doses used in similar studies (15-17, 21). The mAChR agonist oxotremorine was administered in doses of 10, 20, 50 and 100 μg/kg body weight (bw). Oxotremorine at 20 μg/kg bw was also tested in co-administration with the mAChR antagonist atropine (2.5 mg/kg bw), or the opioid receptor antagonist naloxone (2.5 mg/kg bw). The mAChR agonist epibatidine was administered in doses of 0.5, 1, 2 and 3 μg/kg bw. Epibatidine at 2 μg/kg bw was also tested in co-administration with the mAChR antagonist mecamylamine (50 μg/kg bw) or naloxone (2.5 mg/kg bw). Control animals were injected with 0.9% physiological saline. All treatments were administered in a randomized order. Co-administration of substances was performed in accordance with previous studies, where the same chemicals were tested (32). However, it should be pointed-out that the pharmacokinetics for these substances is unknown in the naked mole-rat. The experiments were performed in a sound-proof room, at a room temperature of 26-28˚C, between 8:00 and 14:00.

**Statistical analysis.** Data are presented in graphs as mean values±standard error of the mean (SEM). Data from tail-flick and hot-plate tests were analyzed with a one-way analysis of variance (ANOVA), with Tukey’s post-hoc test to compare latency between groups. Data from formalin tests were analyzed with a two-way ANOVA, with time and treatment as factors, and with Bonferroni’s post-hoc test to compare pain-related behavior between treatment groups. Statistics were calculated using the GraphPad Prism 5.01 (GraphPad Inc., La Jolla, CA, USA). p-values <0.05 were considered significant.

**Results**

Atropine, mecamylamine, and naloxone had no effect in any of the three tests described below, when administered alone.

**Tail-flick test.** The effect of oxotremorine at 10-100 μg/kg bw on tail-flick latency is shown in Figure 1A. The tail-flick latency in animals treated with 20, 50 and 100 μg/kg bw oxotremorine was significantly different from that of the saline-treated animals. The difference was found to be dose-dependent. Co-administration of 2.5 mg/kg bw atropine was found to reverse the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 1B. Co-administration of naloxone 2.5 mg/kg bw was found to enhance the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 1C.

The effect of epibatidine at 0.5-3 μg/kg bw on tail-flick latency is shown in Figure 2A. The tail-flick latency in animals treated with epibatidine at 1, 2 and 3 μg/kg bw was significantly different from that of the saline-treated animals. The difference was found to be dose-dependent. Co-administration of 50 μg/kg bw mecamylamine was found to reverse the effect of epibatidine at 2 μg/kg bw, as shown in Figure 2B. Co-administration of naloxone 2.5 at mg/kg bw was found to enhance the effect of epibatidine at 2 μg/kg bw, as shown in Figure 2C.

**Hot-plate test.** The effect of oxotremorine at 10-100 μg/kg bw on hot-plate response latency is shown in Figure 3A. The hot-plate response latency in animals treated with 20, 50 and 100 μg/kg bw oxotremorine was significantly different from that of the saline-treated animals. The difference was found to be dose-dependent. Co-administration of atropine at 2.5 mg/kg bw was found to reverse the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 3B. Co-administration of 2.5 mg/kg bw naloxone was found to enhance the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 3C.

The effect of epibatidine at 0.5-3 μg/kg bw on hot-plate response latency is shown in Figure 4A. The hot-plate response latency in animals treated with epibatidine at 1, 2 and 3 μg/kg bw was significantly different from that of the saline-treated animals. The difference was found to be dose-dependent. Co-administration of 50 μg/kg bw mecamylamine was found to reverse the effect of epibatidine at 2 μg/kg bw, as shown in Figure 4B. Co-administration of 2.5 mg/kg bw naloxone was found to enhance the effect of epibatidine at 2 μg/kg bw, as shown in Figure 4C.

**Formalin test.** The time the animals spent in licking the injected paw (referred to as pain-related behavior) during the one-hour observation period is shown in Figure 5. It was found that pain-related behavior occurred in two phases: The first phase during the first five minute block, and the second phase from 35 to 60 min after injection, in comparison to animals injected with saline.

The effect of oxotremorine at 10-100 μg/kg bw on pain-related behavior is shown in Figure 6A. The pain-related behavior in animals treated with oxotremorine at 20, 50 and 100 was significantly different from control animals, both in the first and second phase. Co-administration of 2.5 mg/kg bw atropine was found to reverse the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 6B. Co-administration of naloxone at 2.5 mg/kg bw was found to enhance the effect of oxotremorine at 20 μg/kg bw, as shown in Figure 6C.

The effect of epibatidine at 0.5-3 μg/kg bw on pain-related behavior is shown in Figure 7A. The pain-related behavior in animals treated with 0.5 μg/kg bw epibatidine was significantly different from that of the saline-treated animals in the second phase only (45-55 min). The pain-related behavior in animals treated with epibatidine at 1, 2 and 3 μg/kg bw was significantly different from that of the saline-treated animals in both the first and second phase. The difference was found to be dose-dependent. Co-administration of 50 μg/kg bw mecamylamine was found to reverse the effect of epibatidine at 2 μg/kg bw, as shown in...
Figure 1. Tail-flick latency [mean±standard error of the mean (SEM)] in the African naked mole-rat. A: Latency after treatment with oxotremorine at 10 μg/kg bw (OXO 10), 20 μg/kg bw (OXO 20), 50 μg/kg bw (OXO 50), and 100 μg/kg bw (OXO 100), (all n=6), in comparison with control animals (saline) (n=8). Significantly different latency a, from saline; b, from OXO 10; and c, from OXO 20, as determined by one-way ANOVA with Tukey’s post-hoc test. B: Latency after treatment with OXO 20 in co-administration with atropine at 2.5 mg/kg (OXO 10 + ATR 2.5) (n=6), in comparison with treatment with saline, OXO 20, and atropine at 2.5 mg/kg alone (ATR 2.5), (n=6). a, Significantly different latency from saline. C: Latency after treatment with OXO 20 in co-administration with naloxone at 2.5 mg/kg (OXO 10 + NAL 2.5), (n=6), in comparison with treatment with saline, OXO 20, and naloxone 2.5 mg/kg alone (NAL 2.5), (n=6). Significantly different latency a, from saline; b, from NAL 2.5; and c, from OXO 20.

Figure 2. Tail-flick latency [mean±standard error of the mean (SEM)] in the African naked mole-rat. A: Latency after treatment with epibatidine at 0.5 μg/kg bw (EPI 0.5) 1 μg/kg bw (EPI 1), 2 μg/kg bw (EPI 2) and 3 μg/kg bw (EPI 3) (all n=6), in comparison with control animals (saline) (n=8). Significantly different latency a, from saline; b, from EPI 0.5; and c, from EPI 1, as determined by one-way ANOVA with Tukey’s post-hoc test. B: Latency after treatment with EPI 2 in co-administration with mecamylamine at 50 μg/kg (EPI 2 + MEC 50) (n=6), in comparison with treatment with saline, EPI 2, and mecamylamine at 50 μg/kg alone (MEC 50) (n=6). Significantly different latency a, from saline; and b, from EPI 2. C: Latency after treatment with EPI 2 in co-administration with naloxone at 2.5 mg/kg (EPI 2 + NAL 2.5), (n=6), in comparison to treatment with saline, EPI 2, and naloxone at 2.5 mg/kg alone (NAL 2.5), (n=6). Significantly different latency a, from saline; b, from NAL 2.5; and c, from EPI 2.
Figure 3. Hot-plate response latency [mean±standard error of the mean (SEM)] in the African naked mole-rat. A: Latency after treatment with oxotremorine at 10 μg/kg bw (OXO 10), 20 μg/kg bw (OXO 20), 50 μg/kg bw (OXO 50), and 100 μg/kg bw (OXO 100) (all n=6), in comparison to control animals (saline) (n=8). Significantly different latency a, from saline; b, from OXO 10; c, from OXO 20; and d, from OXO 50, as determined by one-way ANOVA with Tukey’s post-hoc test. B: Latency after treatment with OXO 20 in co-administration with atropine at 2.5 mg/kg (OXO 20 + ATR 2.5) (n=6), in comparison with treatment with saline, OXO 20, and atropine at 2.5 mg/kg alone (ATR 2.5) (n=6). Significantly different latency a, from saline; b, from ATR 2.5; and c, from OXO 20 + ATR 2.5. C: Latency after treatment with OXO 20 in co-administration with naloxone 2.5 mg/kg (OXO 20 + NAL 2.5), n=6, in comparison with treatment with saline, OXO 20, and naloxone 2.5 mg/kg alone (NAL 2.5), n=6. Significantly different latency a, from saline; b, from NAL 2.5; and c, from OXO 20.

Figure 4. Hot-plate response latency mean±standard error of the mean (SEM)] in the African naked mole-rat. A: Latency after treatment with epibatidine at 0.5 μg/kg bw (EPI 0.5), 1 μg/kg bw (EPI 1), 2 μg/kg bw (EPI 2), and 3 μg/kg bw (EPI 3) (all n=6), in comparison to control animals (saline) (n=8). Significantly different latency a, from saline; b, from EPI 0.5; c, from EPI 1; and d, from EPI 2, as determined by one-way ANOVA with Tukey’s post-hoc test. B: Latency after treatment with EPI 2 in co-administration with mecamylamine at 50 μg/kg (EPI 2 + MEC 50) (n=6), in comparison to treatment with saline, EPI 2, and mecamylamine 50 μg/kg alone (MEC 50) (n=6). Significantly different latency a, from saline; b, from MEC 50; and c, from EPI 2 + MEC 50. C: Latency after treatment with EPI 2 in co-administration with naloxone at 2.5 mg/kg (EPI 2 + NAL 2.5) (n=6), in comparison with treatment with saline, EPI 2, and naloxone at 2.5 mg/kg alone (NAL 2.5) (n=6). Significantly different latency a, from saline; b, from NAL 2.5; and c, from EPI 2.
Figure 7B. Co-administration of naloxone at 2.5 mg/kg bw resulted in pain-related behavior similar to the effect of epibatidine at 2 μg/kg bw, as shown in Figure 7C.

Discussion

Acetylcholine and cholinergic receptors appear to be an essential neurotransmitter system in antinociception, particularly at the spinal cord level. It has been suggested that an increased release of spinal acetylcholine after treatment with an mAChR agonist is associated with an increased pain threshold in rats, while mAChR antagonist treatment reduces the acetylcholine release and is associated with hyperalgesia (15). Further studies in rats have shown that acetylcholine release is increased after systemic or intraspinal treatment with several substances associated with antinociception, such as lidocaine (16), α2-adrenoceptor agonists (17), epibatidine (22), and serotonergic receptor agonists (23). It has also been suggested that spinal acetylcholine release is under tonic regulation by the GABA-receptor activity (24). In addition, an indication of acetylcholinergic involvement of the central antinociceptive actions of morphine (20), ketamine (19) and peripheral analgesics (18) has been shown.

The data presented in the present investigation suggest that the cholinergic receptor system is involved in antinociceptive mechanisms in the naked mole-rat, in accordance with the hypothesis. Oxotremorine is a well-characterized mAChR agonist, and has been applied in numerous studies to investigate the involvement of mAChRs in various species and experimental settings (15, 25, 28, 33, 34). Involvement of mAChRs in antinociception has been demonstrated e.g. in rats, using similar dose ranges as in the present study (15). The same is valid for the mAChR antagonist atropine (15, 16, 34, 35). Similarly, the nAChR agonist epibatidine and the nAChR antagonist mecamylamine have been extensively used to test for the involvement of nAChRs in antinociception (16, 17, 21, 22, 33, 36). Thus, it is proposed that both mAChRs and nAChRs are involved in antinociceptive mechanisms in the naked mole-rat. Furthermore, since the antinociceptive effect of both muscarinic and nicotinic agonists is similar in all three applied nociceptive tests, it could be speculated that both mAChRs and nAChRs activate a common mechanism. However, to what extent mAChRs and nAChRs are present in the naked mole-rat and their distribution in this animal remains to be discovered.

The finding that naloxone enhanced the antinociceptive effect of both oxotremorine and epibatidine was contrary to our hypothesis. Previous studies have shown antinociceptive effects of μ-, κ- and δ-opioid receptor agonists when using the formalin test in the naked mole-rat (11). This suggests that all opioid receptor subtypes are involved in antinociception in this species. Since opioid receptors are known to play a key role in antinociception in most species [reviewed in (34, 37)], it was believed that the effects of cholinergic receptor agonists in the analgesiometric tests would be mediated via a pathway involving opioid receptors. It was, therefore, expected that co-administration with naloxone would reverse the antinociceptive effects of oxotremorine and epibatidine. Furthermore, it was believed that if the cholinergic effect was not mediated via such an opioid pathway, naloxone would have no effect on cholinergic agonists. The fact that the opposite effect was observed indicates a complex organization of interaction between the cholinergic and opioid receptor systems in the regulation of antinociception in the naked mole-rat, possibly much different from that of other mammals. This is supported by a previous finding based on different properties of opioid receptor subtypes in the naked mole-rat compared to that of other rodents, as stimulation of κ- and δ-receptors produced antinociception, while μ-receptor stimulation produced hyperalgesia in the hot-plate test (10). How this interaction is mediated is not possible to explain from the present experimental setup. It is possible that opioid blockage removes a possible tonic opioid inhibition of ACh release, which leads to an enhanced agonistic effect on cholinergic receptors. Alternatively, opioid blockage may facilitate cholinergic antinociception by opening an antinociceptive gateway not involving opioid receptors. This could be of great importance for future pain therapies, since opioids have...
Figure 6. The effect of injection of 20 μl of 10% formalin on licking the injected paw (pain-related behavior) in the African naked mole-rat, 30 min after pre-treatment with drugs. Data are presented as mean ± standard error of the mean (SEM). A: Pain-related behavior 30 min after pre-treatment with saline (n=8) and oxotremorine at 10 μg/kg bw (OXO 10), 20 μg/kg bw (OXO 20), 50 μg/kg bw (OXO 50), and 100 μg/kg bw (OXO 100) (all n=6). Two-way ANOVA with Bonferroni’s post-test was used to demonstrate differences in pain-related behavior between groups. Significant difference a, between saline and OXO 20; b, between saline and OXO 50; c, between saline and OXO 100; d, between OXO 10 and OXO 50; and e, between OXO 10 and OXO 100. B: Pain-related behavior 30 min after pre-treatment with OXO 20 in co-administration with atropine at 2.5 mg/kg (OXO 20 + ATR 2.5) (n=7), in comparison to treatment with saline (n=8) OXO 20, and atropine at 2.5 mg/kg alone (ATR 2.5) (n=6). Significant difference a, between saline and OXO 20; b, between ATR 2.5 and OXO 20; and c, between OXO 20 and OXO 20 + ATR 2.5. C: Pain-related behavior 30 min after pre-treatment with OXO 20 in co-administration with naloxone at 2.5 mg/kg (OXO 20 + NAL 2.5) (n=6), in comparison with treatment with saline, OXO 20, and naloxone at 2.5 mg/kg alone (NAL 2.5) (n=6). Significant difference a, between saline and OXO 20; b, between saline and OXO 20 + NAL 2.5; c, between NAL 2.5 and OXO 20; d, between NAL 2.5 and OXO 20 + NAL 2.5; and e, between OXO 20 and OXO 20 + NAL 2.5.
several shortcomings, such as tolerance development and opioid-induced hyperalgesia (38). Further histological, molecular and pharmacological characterization of this issue is thus necessary.

Oxotremorine and epibatidine produced antinociception in all of the three applied tests, which could be blocked with atropine or mecamylamine, respectively. The tests were selected because of their involvement of different nociceptive mechanisms. The tail-flick test measures a spinal reflex response to an acute noxious thermal stimulus (39, 40), and is thus not considered to involve any supraspinal mechanism. The hot-plate test also measures a
response to an acute noxious thermal stimulus, but this test is considered to involve more complex mechanisms at supra-spinal levels (7, 41). The formalin test measures the response to two different stimuli (42). In the first phase, the animals’ paw-licking behavior is most likely a response to the acute pain from the direct nociceptor activation by formalin, while in the second phase, it has been suggested to relate to an inflammatory reaction in the tissue (43, 44). The results, therefore, indicate that the cholinergic receptor system is an essential mediator of antinociception in the naked mole-rat, involved in mechanisms activated by thermal, chemical and inflammatory mechanisms. As the tail-flick test is considered to mainly reflect a spinal reflex, part of this mediation is suggested to occur, directly or indirectly, at the spinal cord level. However, it should be pointed out that the anatomical structure of descending mechanisms in the naked mole-rat is unknown; hence the spinal portion of the antinociception in this species needs further investigation.

In conclusion, the naked mole-rat appears to be a relevant animal for studying cholinergic involvement in antinociceptive mechanisms. The present study has demonstrated that intraperitoneal injection of the mAChR agonist oxotremorine, and the nAChR agonist epibatidine, reduced pain-related behavior in the tail-flick, hot-plate, and formalin tests, and that the reduction was reversed by co-administration with the mAChR antagonist atropine, and with the nAChR antagonist mecamylamine, respectively, suggesting involvement of muscarinic and nicotinic receptors. Furthermore, it was shown that the effect on pain-related behavior caused by oxotremorine and epibatidine was enhanced by co-administration of the opioid receptor antagonists naloxone in all three tests. This suggests an interaction between the cholinergic and opioid receptor systems in the regulation of nociceptive transmission in the naked mole-rat, which could prove to be important for the understanding of the nociceptive regulation in this species. However, in order to fully-understand the involvement and interaction, more research is needed to determine the presence and distribution of muscarinic, nicotinic and opioid receptors, as well as the pharmacological binding properties of various ligands to these receptors.

Competing Interests

The Authors declare that they have no competing interests.

Acknowledgements

The Authors are very grateful to the Government of Kenya, Ministry of Livestock Development, which granted some financial assistance; to Mr. Marete and Kamonde, who took care of the experimental animals; and to the Director, Kenya Wildlife Services for granting a permit for capturing animals which enabled us to perform this study.

References


Received August 2, 2013
Revised November 19, 2013
Accepted November 20, 2013