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Abstract: Two long-acting neuroleptics were used to tranquilize nine captive cheetahs (*Acinonyx jubatus*). Perphenazine enanthate (3.0 mg/kg) and zuclopenthixol acetate (0.6 mg/kg) were each administered to separate groups of three cheetahs in a double blind trial. Both products were administered together to a third group of three animals at the same dosages. Behavioral effect, duration of effect, and possible side effects were observed by a predefined protocol. Under standardized holding conditions, the cheetahs were observed 5 days before drug administration and 14 days after administration. Daily activity was defined and statistically evaluated by a *U*-test. A significant reduction of activity was observed after administration in all three trials. Zuclopenthixol acetate at 0.6 mg/kg alone and in combination with perphenazine enanthate caused inappetence, ataxia, extra pyramidal reactions, akathisia, and prolapse of the third eyelid. Zuclopenthixol acetate should not be used in cheetahs. Perphenazine enanthate did not cause inappetence, reduced appetite, or any of the previously mentioned side effects when used alone. It produced satisfactory tranquilization and is suitable and safe for cheetahs at 3.0 mg/kg. This dosage should be varied depending on health, age, and temperament of the individual cheetah.

EVALUATION OF LONG-TERM SEDATION IN CHEETAH (*ACINONYX JUBATUS*) WITH PERPHENAZINE ENANTHATE AND ZUCLOPENTHIXOL ACETATE

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Abstract: Two long-acting neuroleptics were used to tranquilize nine captive cheetahs (*Acinonyx jubatus*). Perphenazine enanthate (3.0 mg/kg) and zuclopenthixol acetate (0.6 mg/kg) were each administered to separate groups of three cheetahs in a double blind trial. Both products were administered together to a third group of three animals at the same dosages. Behavioral effect, duration of effect, and possible side effects were observed by a predefined protocol. Under standardized holding conditions, the cheetahs were observed 5 days before drug administration and 14 days after administration. Daily activity was defined and statistically evaluated by a *U*-test. A significant reduction of activity was observed after administration in all three trials. Zuclopenthixol acetate at 0.6 mg/kg alone and in combination with perphenazine enanthate caused inappetence, ataxia, extra pyramidal reactions, akathisia, and prolapse of the third eyelid. Zuclopenthixol acetate should not be used in cheetahs. Perphenazine enanthate did not cause inappetence, reduced appetite, or any of the previously mentioned side effects when used alone. It produced satisfactory tranquilization and is suitable and safe for cheetahs at 3.0 mg/kg. This dosage should be varied depending on health, age, and temperament of the individual cheetah.

Key words: Cheetah, *Acinonyx jubatus*, zuclopenthixol acetate, perphenazine acetate, long-acting neuroleptics.

INTRODUCTION

Neuroleptic drugs suppress behavioral responses without affecting spinal or other reflexes. In humans, they provide antipsychotic effects and suppress alarm situations, anxiety, and psychomotor agitation.¹⁶ In animals, they relieve anxiety, decrease motor activity, and moderate excitement and agitation.¹³ Long-acting neuroleptics (LANs), or depot-tranquillizers, are neuroleptics that can be administered such that a single dose gives a therapeutic effect for at least 7 days.³ They are formulated by combining the active drug with a long-chained fatty acid, and the combination is hydrolyzed slowly in body tissues, releasing the neuroleptic drug into the vascular system over a prolonged period of time.^{3,9} Zuclopenthixol acetate is a thioxanthene, and perphenazine enanthate is a phenothiazine derivative. Both are used routinely for capture and translocation of ungulates in southern Africa.^{2,3,13} There is, however, little information on their use in felids. Experience in free-ranging ungulates shows that zuclopenthixol acetate is effective within 1 hr, and its sedative effect wears off at about 72 hr postinjection.^{2,3} Because of this relatively short duration of action, it does not fully qualify as a LAN according

to the previous definition. Its duration of effect is intermediate between such short-acting tranquilizers as acepromazine and LANs such as perphenazine enanthate and haloperidol decanoate. In ungulates, perphenazine enanthate begins to be effective 12 hr postinjection and reaches peak effect at 72 hr. The effect decreases steadily thereafter over approximately 14 days.^{2,3} Combination use of zuclopenthixol acetate and perphenazine enanthate has been recommended to keep animals in a steady state of tranquilization for 1–14 days.^{3,13}

The largest remaining free-ranging population of cheetahs (*Acinonyx jubatus*) lives predominantly on farmland in Namibia.^{11,20} Their livestock predation, with subsequent retaliatory action by farmers, limits their population size. Local livestock farmers often capture cheetahs in box traps and, in accordance with Namibian law, may kill them. As an alternative to killing, two local nongovernmental organizations collect cheetahs from farms and attempt to rehabilitate them. Because cheetahs are extremely susceptible to stress, LANs might reduce anxiety and motor activity after capture.^{12,18} Wild-caught animals, particularly cheetahs, may have difficulty adapting to captivity, with attendant novel environments, proximity to humans, and unfamiliar food. Escape attempts end traumatically, with excoriations and superficial lacerations caused by box traps and fencing material.^{4,7} Significant liver pathology is incurred by cheetahs during the first week of captivity, and significant renal pathology may also occur.¹² The secretion of adrenal glucocorticosteroids may also be affected.¹⁷ Severe gastritis in captive

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Table 1. Cheetahs used in the study, Salzburg Zoo Hellbrunn.

Cheetah no.	Sex	Age	Body Weight	Zucloperthixol (mg/kg)	Perphenazine (mg/kg)
1	F	2	31	0.6	—
2	M	2	35	0.6	3.0
3	M	2	44	—	3.0
4	F	2	25	0.6	3.0
5	M	2	40	0.6	—
6	F	7	25	0.6	—
7	F	1	20	0.6	3.0
8	F	4	30	—	3.0
9	M	2	40	—	3.0

cheetahs, probably caused by *Helicobacter* sp., may also be associated with captive housing-induced stress.⁶

In order to assess the effect of the LANs in cheetahs, a study of behavioral activity in medicated cheetahs was initiated with a predefined protocol and the greatest possible objectivity. Reduced activity and a subsequently increased resting behavior were equated with sedative effect. "Stress" is best assessed by examination of the systems used to cope with homeostatis disruption: the autonomic nervous and the neuro-endocrine systems, plus the animal's behavior.¹⁰ Recently captured Namibian cheetahs are often extremely agitated in the presence of humans during captivity.^{4,12} Long-acting neuroleptics relieve anxiety and moderate excitement and agitation in addition to decreasing motor activity, so we also discuss the effectiveness of perphenazine enanthate and zucloperthixol in possibly reducing stress over a period of several days.

MATERIALS AND METHODS

The study was conducted at the Salzburg Zoo Hellbrunn, Austria, in September 1998 in accordance with Austrian animal welfare legislation (permit no. 9/01-40.034/81-1998). Nine adult (1–7 yr old) cheetahs, four males and five females, were housed individually, except for one mother (animal 4)—juvenile daughter pairing, in 100–120-m² off-exhibit enclosures (Table 1). The animals had access to 4-m² heated shelters in which water was offered ad lib. Drinking behavior could not be monitored. The animals were fed once a day at about 1530 hours with approximately 1 kg of beef supplemented with a commercial mineral and vitamin powder supplement (Vitavet[®], Richter Pharma, Wels, Austria).

Trials in domestic cats aided determination of dosages of perphenazine enanthate and zucloperthixol acetate.⁴ All cheetahs were immobilized on days 0, 7, and 14 with tiletamine—zolazepam (Zo-

letil[®] 100 Virbac SA, Louvain la Neuve, Belgium; 4.0 mg/kg i.m.) by blow dart into the rear leg muscles. On day 0, they were weighed, given LAN by deep i.m. injection, and had blood samples collected. Blood samples were also collected on days 7 and 14. The nine cheetahs were allocated into three groups of three cheetahs. On day 0, the members of one group were given zucloperthixol (Ciatyl-Z Acuphase[®], 50 mg/ml, Bayer AG, Leverkusen, Germany; 0.6 mg/kg i.m.) deep into the semimembranosus. A second group was given perphenazine enanthate (Decentan[®] Depot, 100 mg/ml, Merck, Darmstadt, Germany; 3.0 mg/kg i.m.). The third group was given both drugs in the above mentioned dosages and routes. Both drugs were diluted in sterile sesame seed oil and well mixed by passing the mixture back and forth 100× between two syringes connected by a three-way valve. To guarantee correct dosing, zucloperthixol acetate was diluted to 5 mg/ml and perphenazine enanthate to 10 mg/ml. To avoid subjective influences from the observer, this was a double blind study.

The effects of the LANs were evaluated by a behavioral observation protocol during previously determined main activity time periods (0800–1000 hours; 1400–1600 hours). Observation was started 5 days before administration and continued for 14 days after day 0. Behavior observed before LAN application served as a control for each individual according to previously described methods of small *N* research design (A₁-B-A₂-study)¹⁵ (Fig. 1). Scan sampling was used to record the behavioral data. The observation periods were divided into 60-sec sample intervals.^{1,8} At the beginning of each interval, the observed behavior was noted in coded form. One observer was used throughout the trial period to eliminate interobserver reliability problems. Time was measured with a digital timer (PC 80A, Conrad Electronics, Hirschau, Germany). The code system was based on a predefined ethogram (Table 2). Activity behavior was defined (Table 2,

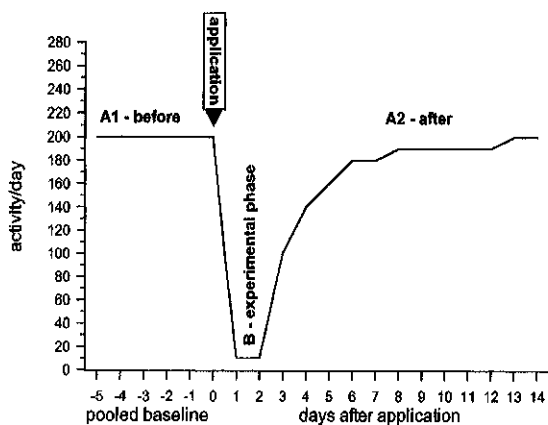


Figure 1. Schematic description of the study (A_1 - B - A_2 -study). A_1 represents the pre-LAN application phase, B is the phase after LAN application and significant reduction in activity, and A_2 represents the return to initial baseline activity.

bold print) and compared with rest behavior. In developing the ethogram and the subsequent definition of activity and resting behavior, we took the reduced behavioral potential of the holding facilities into consideration. Daily activity was statistically evaluated by a *U*-test. Observational data from days 0 and 7 were excluded from the statistical evaluation of this study. Because of animal welfare permit reasons, the number of trials was limited to one trial per cheetah; therefore, significance was established at $P < 0.05$ where appropriate and discussed individually. The daily activity before LAN administration was determined and pooled to provide a behavioral baseline (BL) for each individual. This BL was used as an individual control and compared with the daily activity after the LAN application.

RESULTS

For each animal, 240 individual behavioral scans per day were documented, and 1,200 pre-LAN application scans served to determine the individual behavioral activity baseline. Post-LAN application, 3,360 scans were recorded to determine the effect and duration of the applied LANs. All animals treated with perphenazine enanthate and/or zuclopenthixol acetate demonstrated a significant reduction of their daily activity compared with their individual BL data. The three animals treated with zuclopenthixol acetate alone showed a significant (*U*-test, $P < 0.05$) reduction of activity on day 1 only corresponding to the time frame of 24–36 hr postapplication (Fig. 2). However, significant side effects that lasted for 24–48 hr were noted in all

Table 2. Behavioral protocol used in code form. Behavior evaluated as activity is listed in bold print; resting behavior is listed in lightface print.

-
1. Resting
 - 1.1. Lying on the side
 - 1.1.1. Lying on the side, head up
 - 1.1.2. Lying on the side, head down
 - 1.2. Lying in sternal position
 - 1.2.1. Lying in sternal position, head up
 - 1.2.2. Lying in sternal position, head down
 - 1.3. Sitting
 2. Activity on the spot and locomotion
 - 2.1. Standing
 - 2.2. Walking
 - 2.3. Trotting
 - 2.4. Stalking
 - 2.5. Sprinting
 - 2.6. Climbing in tree
 - 2.6.1. Lying in tree
 - 2.6.2. Sitting in tree
 - 2.6.3. Standing in tree
 3. Social behavior including playing
 - 3.1. Grooming
 - 3.2. Yawning
 - 3.3. Stretching
 - 3.4. Rolling
 - 3.5. Sharpening claws
 - 3.6. Rubbing
 - 3.7. Playing
 4. Metabolism—excretion
 - 4.1. Urinating/urine marking
 - 4.2. Defecating/defecation marking
 5. Olfactory examination
 - 5.1. Object sniffing
 6. Metabolism—intake
 - 6.1. Eating—piece with bone
 - 6.2. Eating—piece without bone
 - 6.3. Chewing on an old bone
 7. Not visible
-

three cheetahs. The side effects consisted of decreased food intake, ataxia, extrapyramidal reactions (head swaying), akathisia (motor restlessness and urge to move constantly), and prolapsed third eyelid. The sedative effect (as defined in a decrease of activity) of perphenazine enanthate was significant (*U*-test, $P < 0.1$) from day 1 until day 6 except on day 4 (*U*-test, $P = 0.17$). The effect maximum was reached on day 2. No side effects were noted in any of the three animals treated only with perphenazine enanthate. An apparent overall tranquilizing effect, though not statistically significant, could be observed additionally from day 8 until day 11 (Fig. 3). The cheetahs injected with a combi-

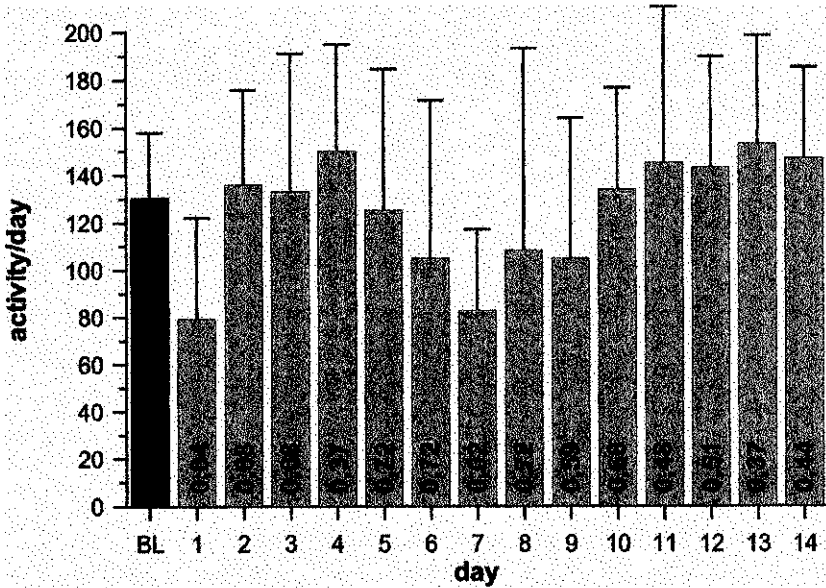


Figure 2. Daily activity of the three cheetahs injected with 0.6 mg/kg zuclopenthixol acetate on day 0 in comparison with the activity baseline (BL). Significance levels are given in the individual bars.

nation of both drugs were statistically less active on day 1 but apparently less active until day 6. Side effects similar to those noted in the animals injected with zuclopenthixol acetate only occurred on day 1 in these three animals also. As with the perphenazine enanthate-only group, an apparent prolonged sedative effect that was not statistically significant could be observed until day 11.

DISCUSSION

The sedative effect (defined as reduction of activity) of 0.6 mg/kg zuclopenthixol acetate was statistically significant on day 1 only (U -test, $P < 0.05$). The duration of effect in the cheetahs from the moment of injection (day 0) until the effect of zuclopenthixol acetate ended was approximately

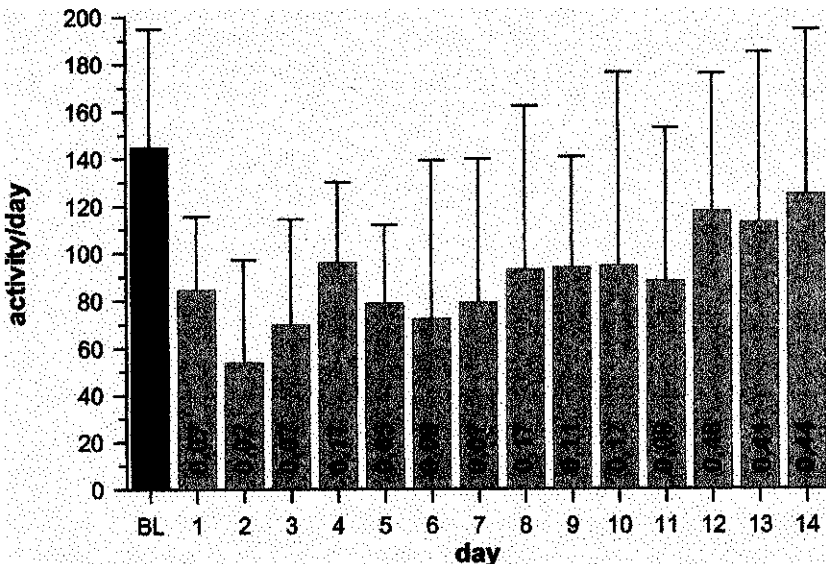


Figure 3. Daily activity of the three cheetahs injected with 3.0 mg/kg perphenazine enanthate in comparison with the activity baseline (BL). Significance levels are given in the individual bars.

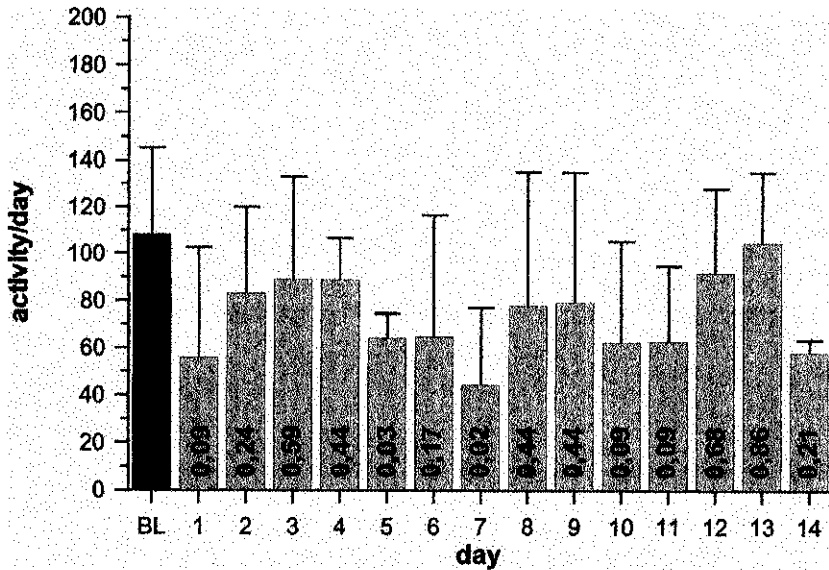


Figure 4. Daily activity of the three cheetahs injected with 0.6 mg/kg zuclopenthixol acetate and 3.0 mg/kg perphenazine enanthate in comparison with the activity baseline (BL). Significance levels are given in the individual bars.

24–48 hr. The duration of action of zuclopenthixol acetate in ungulates has been estimated to last from 1 hr until 72 hr postapplication, which agrees with our results.^{3,14} In the six cheetahs that received zuclopenthixol acetate, side effects such as inappetence, ataxia, extrapyramidal reactions (head swaying), akathisia, and prolapsed third eyelid occurred for the first 24–48 hr. These side effects disappeared completely by 72 hr post-LAN administration. Similar side effects have been described in ungulates when high dosages of haloperidol are used.³ Side effects were not noted when 1.0 mg/kg zuclopenthixol acetate was used for the transport of a single leopard (*Panthera pardus*) (Walzer, unpubl. data). The same dosage was used in a single wild-caught cheetah in Namibia with similar side effects.⁴ Lower dosages of 0.1–0.3 mg/kg zuclopenthixol acetate in four wild-caught cheetahs in Namibia did not elicit a sedative effect, but inappetence was noted.⁴ The zuclopenthixol acetate dose of 0.6 mg/kg used in this trial was established in a previous study with 20 domestic cats (*Felis catus*). In that study, reduced activity without side effects was recorded.⁴ Side effects such as restlessness, salivation, ataxia, and even an inability to walk have been described in humans. Recommended dosages in humans are 0.6–1.9 mg/kg every 2–3 days, depending on the severity of the condition and the obtained response. In ungulates treated with zuclopenthixol acetate, inappetence or other side effects were not noted.^{2,3} Generally, animals caught in the

wild and brought into a stressful and new captive environment may have decreased appetite. In contrast to our results with cheetahs in this study, several authors have reported that zuclopenthixol acetate-treated ungulates began to eat and drink earlier than nontreated animals.^{2,3,13} A marked difference in the effect of zuclopenthixol acetate exists in different species, as well as between different members of one family (e.g., Felidae, domestic cat and cheetah). Furthermore, anecdotal evidence suggests different effects of zuclopenthixol acetate in cheetahs and leopards. Because of the side effects noted in our study and elsewhere, zuclopenthixol acetate is not recommended for use at any dose in cheetahs.

In those six cheetahs that received 3.0 mg/kg body weight perphenazine enanthate, a statistically significant decrease in activity was noted on days 1–6 except for day 4. Maximum sedative effect occurred on day 2 (24–48 hr postadministration). The sedative effect decreased subsequently until day 14 when the observation period ended. The duration of the overall effect seemed to be even longer than 14 days because the pre-LAN activity BL was not reached until after the end of the observation period. The slight decrease in significance on day 4 is probably due to the small number of subjects in this trial. The noticeable sedative effect of perphenazine enanthate in ungulates begins 16–20 hr after injection, reaches its maximum at 72 hr, and shows effects until 7–10 days after injec-

tion.^{2,3,13} The effect of perphenazine enanthate on three cheetahs during their introduction into new enclosures at the Munich Zoo has been described anecdotally.¹⁹ The duration of effect observed in the Munich cheetahs was similar to the duration described in ungulates and the cheetahs in our study. This observation leads to the conclusion that the onset and the duration of perphenazine enanthate induced sedation are comparable among various ungulates and cheetahs. Side effects were not seen in the three cheetahs that were medicated with perphenazine enanthate alone. The time devoted to food intake appeared shorter after LAN application, but the difference was not statistically significant. Reduced food intake from day 7 until day 11 after the application of 1.15 mg/kg body weight perphenazine enanthate has been described in a lynx (*Lynx lynx*).⁵ In that species, the inappetence may have been caused by an overdose of perphenazine enanthate, although the occurrence of the decreased appetite during the period of decreasing effect makes this unlikely. Inappetence was not seen in domestic cats treated with perphenazine enanthate.⁴ At the Munich Zoo, perphenazine enanthate at a dosage of 0.5–0.6 mg/kg was used in three cheetahs for introduction into a new enclosure.¹⁹ A 10% reduction of activity occurred in the cats, and no side effects were mentioned. We used a similar low dosage in domestic cats without seeing a reduction of activity under standardized conditions, but satisfactory tranquilization without side effects was seen at 3.0 mg/kg.⁴ A possible reason for the large discrepancy in the dosages could be that the cheetahs in the Munich Zoo were moved into a new enclosure during the trial and thus were not under standardized conditions. The low-dose LAN application time was not disclosed in the study, and the change in behavior could have been a normal part of the animals' habituation to a new enclosure. After the present study, perphenazine enanthate (3.0 mg/kg) was used with satisfactory tranquilization at the Salzburg Zoo during the transport of two cheetahs. Perphenazine enanthate at approximately 2.5 mg/kg was administered to five cheetahs in Safari Beekse Bergen, Hivarenbeek, The Netherlands, 2 days prior to transport with good success (Huber, unpubl. data). A dose of 2 mg/kg was administered to two Siberian tigers (*Panthera tigris altaica*) and five African lions (*Panthera leo*) for translocation into new enclosures, and satisfactory tranquilization was observed (Kaandorp, pers. com., 2000). In humans, 0.6–2.5 mg/kg every 2–4 wk is recommended. The dosage should be individualized and adjusted according to the severity of the condition and the response desired.

A combination of a short-acting, rapid-onset neuroleptic drug such as zuclopenthixol acetate with a longer acting, slower onset neuroleptic has been recommended for tranquilization of ungulates.³ In our study, the combination of zuclopenthixol acetate with perphenazine enanthate did not significantly reduce the daily activity more than when the individual drugs were used alone, although there was a subjective shortening of the activity. These two drugs may not only complement each other in their time of onset but may also have a significantly synergistic sedative effect. The combination group and the group treated with only zuclopenthixol acetate showed similar side effects on days 1 and 2. When used alone, perphenazine enanthate led to reduced activity on day 1 and reached its maximum effect on day 2. This rapid onset eliminates the need for a second neuroleptic drug in this species.

The effects of neuroleptics in humans are evaluated by questioning people who are treated about their subjective feelings and experiences, but this cannot be done as easily with animals. Our double blind methodology was developed to measure the effect of neuroleptic drugs with the greatest possible objectivity on the basis of behavioral criteria. Ethologic studies are subjective because often only one observer makes the observations. We sought to avoid subjectivity with a predefined protocol under standardized holding conditions. However, reduced activity may not equal reduced stress. Except for the anesthetic events, none of the animals was stressed by activities beyond its normal daily routines, and effects under actual stress could be different. Examination of additional variables could help judge the effect of LANs in cheetahs.¹⁴ Whereas statistical evaluation of data gives objective results, it may underestimate effects when the number of subject animals is low. This was clearly illustrated in a study of the same LANS in a larger number of domestic cats.⁴ In our study, a marked subjective decrease in activity was observed for all LANs beyond the point at which the decrease could be defined statistically. Such a reduction could be clinically relevant.

CONCLUSIONS

All LANs used in this study, alone or in combination, significantly reduced activity, and LANs are useful adjunct drugs in tranquilization of non-domestic felids. Zuclopenthixol acetate alone and in combination with perphenazine enanthate should not be used in cheetahs because of significant side effects. Perphenazine enanthate at 3.0 mg/kg reduces activity in captive cheetahs from days 1–11 postadministration, but this dosage should be ad-

justed to the individual animal and its temperament. Lower dosages should be used in old and very young animals.

Additional studies on the use of LANs in different species are needed because dosages and pharmacodynamic data cannot necessarily be extrapolated between species. Standardized techniques should be used to determine the effectiveness of psychoactive drugs and to facilitate their clinical use.

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