

## EFFICIENCY OF LIBRAX<sup>R</sup> AS AN ANTISTOMACHEC TO OVERCOME BAIT SHYNESS OF ZINC PHOSPHIDE TO BLACK RAT, *RATTUS RATTUS*

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### **Abstract**

Some laboratory trials were conducted to increase the bait acceptance of zinc phosphide against black rat, *Rattus rattus*, using Librax<sup>R</sup> drug (chlordiazepoxid 5mg + clidinium bromide 2.5mg) as antistomachec. Librax<sup>R</sup> 0.001% was tested alone and combined with different concentrations of zinc phosphide bait using non and free choice feeding methods. Results showed that in both methods, Librax<sup>R</sup> alone and mixed with zinc phosphid bait significantly increased the average daily consumption and the acceptability of rats. Also it enhanced the percent of mortality comparing with zinc phosphide bait lone.

### **INTRODUCTION**

Zinc phosphide still is an important rodenticide for its effectiveness against different rodent species and rapid reduction of large population of rodents. Also, zinc phosphide is probably the acute poison of choice when some rodent species show level of resistance to anticoagulant rodenticides. Bait shyness is the major drawback in using zinc phosphide against different rodent species (Meehan, 1984). Small quantity of zinc phosphide (1/4 LD50) consumed at the first time is likely to be sufficient to elicit unpleasant symptoms but not to be cause death. The fast onset to toxic enables rodents the associate cause and effect. Affected animals will usually refuse to consume the poisoned food on subsequent associate or may reduction to feed again from bait receptacles (Prakash, 1988). Many techniques have been used to maked poison-shyness behavior in rodent control process.

The resent study aims to overcome bait shyness phenomenon using antigestrointestinal spasm pain of zinc phosphide treatment.

## MATERIALS AND METHODS

### 1. Tested Compounds:

- 1.1. **Zinc phosphide 94 % (Zn<sub>3</sub> P<sub>2</sub>)** is an acute rodenticide was obtained from KZ Company, Egypt. It was mixed with crushed maize at 0.025, 0.25, 0.5, and 1% concentrations in this study.
- 1.2. **Librax<sup>R</sup> drug** (chlordiazepoxid 5mg + clidinium bromide 2.5mg) tablets for the treatment of organic manufactured EIPICO., Egypt. It was mixed with crushed maize at the 0.001% (calculated according to standard weight of man 75 kg) on the basis of the tablet weight.

### 2. Tested animals:

Black rats, *Rattus rattus*, were trapped from infested areas of Kafr Hakim village, Giza governorate. Animals were transported to laboratory and caged individually for two weeks for acclimatization. They were fed *ad libitum* on crushed maize and water. The immature, unhealthy and pregnant rats were excluded. Animals were weighed and given a reference number for each one.

### 3. Experiments:

#### 3.1. Non-choice feeding method:

##### 3.1.1. Effect of Librax on rat consumption from bait treated with different concentrations of zinc phosphide

Forty rats of, *Rattus rattus*, were divided into treatment groups (each of 5 animals) seven groups for the treatment and another one as check control. Forty grams of 1% zinc phosphide bait were offered for each rat in the 1<sup>st</sup> group while animals of the 2<sup>nd</sup> group were fed on 1% zinc phosphide mixed with Librax<sup>R</sup>. The 3<sup>rd</sup> group were fed on 0.5% zinc phosphide and the 4<sup>th</sup> on zinc phosphide + Librax<sup>R</sup>. The 5<sup>th</sup> group fed on 0.25% zinc phosphide and the 6<sup>th</sup> group on 0.25% zinc phosphide + Librax<sup>R</sup>. The last group fed on Librax bait only. Water was provided *ad libitum* to each animal. The consumed bait was estimated for each rat for 4 days pre-treatment and 24 h treatment. They were observed for 7 days and mortality was recorded.

##### 3.1.2. Effect of Librax<sup>R</sup> on rat consumption of sub lethal dose zinc phosphide bait

Two groups of roof rats, *R. rattus*, (5 rats each) were used for this test and another one as control. The 1<sup>st</sup> group was afforded sub-lethal dose 0.025% of zinc phosphide mixed with 1% molasses. The same bait+ Librax<sup>R</sup> were offered to animals of the 2<sup>nd</sup> group.

#### 3.2. Free choice feeding method:

After one week, free choice test was conducted on the previous two groups for 2 successive days using 0.5% zinc phosphide bait mixed with 1% molasses for 1<sup>st</sup> group

and plus Librax<sup>R</sup> (0.001%) for the 2<sup>nd</sup> group. Forty grams of treated bait were offered for each animal and another untreated in separated dishes. Their position was daily alternated to avoid feeding preference for certain location. The consumed amount of treated and untreated was estimated daily for each animal. Rats were offered food and water *ad libitum* and were observed for 7 days. Mortality was recorded.

The acceptability effect was calculate using the following equation (Mason, *et al*/1989)

$$\text{Acceptability \%} = \frac{\text{Average daily consumption of treated food (g)}}{\text{Total average daily consumption of (treated + untreated) food (g)}} \times 100$$

## RESULTS AND DICUSSION

### 1-Non choice test:

#### Effect of 0.001% Librax drug with zinc phosphide bait

Librax<sup>R</sup> drug consists of chlordiaze epoxide (5mg) and clidium bromide (2.5mg). The first compound has effect in the symplomatic treatment of gastrointestinal spasm pain or discomfort of the gastrointestinal tract may be associated with spasm of the smooth muscle of the gut. The second compound is a quaternary ammonium antimuscarinic with peripheral effect similar to those of atropine (Sweetman, 2002).

Date in table(1) showed the effect of 0.001% Librax<sup>R</sup> drug with different concentrations of zinc phosphide bait against black rat, *Rattus rattus*, using non-choice test.

Results revealed that the average daily consumption of bait decreased from 18.58 to 2.64g for pre-treatment and treatment respectively, of 1% zinc phosphide. Also, the consumption of pre-treatment reduced from 21.76g to 4.59g in case of mixed Librax<sup>R</sup> drug with 1% zinc phosphide bait. The percent of acceptability was raised from 12.44% of zinc phosphide bait alone to 17.42% with Librax<sup>R</sup> addition. Both treatments achieved complete mortality 100% with 24h time of death.

Regarding the treatment of 0.5% zinc phosphide the average daily bait consumption reduced from 15.34 and 14.79g pre-treatment to 5.39 and 7.25g for zinc phosphide alone and mixed with Librax<sup>R</sup>, respectively. Librax<sup>R</sup> increased the percent of acceptability to 32.37% comparing with 26.0% in case of zinc phosphide alone. Mortality percent was 100% with time required to death 24 h for both treatments.

In case of 0.25% zinc phosphide treatment, the average daily bait consumption was 7.02 and 6.01 g for zinc phosphide alone and mixed with Librax<sup>R</sup>, while it was 10.96 and 10.48 g for pre-treatment consecutively. The addition of Librax<sup>R</sup> decreased the acceptance of bait from 39.04% for zinc phosphide alone to 36.47%.

From these results it was noticed that when Librax<sup>R</sup> was mixed with zinc phosphide bait, the acceptance increased with the decrease of zinc phosphide concentrations. For the concentration of zinc phosphide with Librax<sup>R</sup>, the acceptance was always high than for the concentration of zinc phosphide (0.25%). However higher mortality 80% resulted from using Librax<sup>R</sup>, it was 60% only of using zinc phosphide treated alone.

Further more time to death in the first case was lower (24h) than in the second case (48h). Further studies are needed to investigate this phenomenon. Hazards to none target species will substantially be reduce due to the decrease in the amount of poison bait offered to rats in the field.

**Asran (1994)** stated that, in free choice and non- choice feeding tests on, *Arvicanthis niloticus*, with 1% zinc phosphide bait gave 80% and 90%, mortality, respectively. These values were 80% and 80% for, *Rattus rattus*, and 70%, 70% for *R. norvegicus* while in case of, *Mus masculus*, mortality was 90% and 100% respectively.

On the other hand, in case of 0.001% Librax<sup>R</sup> bait the average daily bait consumption increased from 9.91g (pre-treatment) to 13.38g with 57.45% acceptance. It's evident that average of rat consumption using Librax<sup>R</sup> was higher 35% with increasing value of 1.35 fold.

The response of black rat to sub lethal dose of zinc phosphide bait 0.025% alone and combined with Librax<sup>R</sup> was shown in Table (2). Data indicated that, in the pre-treatment the average bait consumption was 11.48 ad 13.68g, respectively. For the treatment the values were 7.22 and 11.4g for 0.025% zinc phosphide only and mixed with Librax<sup>R</sup>, consecutively. Acceptability ratio for treated and untreated Librax<sup>R</sup> was 45.45% and 38.59%, respectively.

From these results, the average daily consumption of poison bait treated with Librax<sup>R</sup> increase by 4.18g and the acceptance increased 1.17 folds higher than zinc phosphide only with ratio of 6.84.

## **2. Free choice test:**

Response of black rat which previously treated with sub- lethal dose of zinc phosphide to 0.5% zinc phosphide alone and combined with Librax<sup>R</sup>. Data in Table (3) showed that in the first day the average bait consumption of rats treated with zinc phosphide only was 0.37g compared with 10.25g. In the second day, values were 0.61g and 10.61g respectively. The mean daily consumption for the two days was 0.49 and 10.44g, respectively for treated and untreated animals.

In case of using bait with Librax, the first day average consumption treated and untreated was 0.53 and 10.08g respectively while the second day these value were

1.44 and 13.03g, consecutively. The mean consumption was 0.99 and 11.55g, respectively. The average of poison bait consumption in the second day was higher than the first day 2.72 folds in case of using Librax<sup>R</sup>.

This means that the addition of Librax<sup>R</sup> increased the average consumption of bait treated with zinc phosphide with the increase of time exposure. Also Librax<sup>R</sup> enhance the acceptance of rats to zinc phosphide bait from 4.69% to 8.57% in case of zinc phosphide alone and mixed with Librax<sup>R</sup>, respectively. Results gave this new approach to improve applied pest management and control techniques.

Different other substances were added to zinc phosphide to increase its acceptability by various rodent species. EL-Deeb, *et al* (1991) and Rezk (2000) stated that zinc phosphide bait shyness could be solved with changing preferred bait and additives.

Discussing the previous results, it could be stated that the antigestrointestinal spasm pain of Librax drug prevent the pain of zinc phosphide treatment. This action leads rats to accept the bait of zinc phosphide and eat more. Richared and Pamela (2000) and Sweetman(2000) mentioned that the antispasmodic drug has effect of the symptomatic treatment of gastrointestinal spasm of discomfort of the gastrointestinal tract that may be associated with spasm of smooth muscles of gut. Gabr and Rizk (2010) found that the antispasmodic agent (octylonium bromide 0.05%) enhanced the efficiency of zinc phosphide bait against albino and wild Norway rat, *Rattus norvegicus*,. The compound lead rats to accept zinc phosphide bait causing high percent of mortality.

Also EL-Deeb, *et al* (2011) indicated the addition of spasmomen 0.1% or 0.05% to zinc phosphide bait, all animals of Norway rats, roof rats, house rats and shawi jerd accept the bait again after 10 days of treatment of sub- lethal dose of zinc phosphide bait alone. Also, the population reduction raised with treatment of spamomen mixed with zinc phosphide than zinc phosphide alone under field conditions. Saxina and Mathur, (1995) showed that the intensity of aversion depended on the period of exposure to the poisoned bait, whereas the aversion lasted for 15 days and 25 days exposure.

EL-Deeb, *et al* (2007) determined the required period for different rodent species to accept zinc phosphide bait again after previous intake of sub- lethal dose (1/4LD<sub>50</sub>) through several laboratory experiments. Results cleared that the period of bait shyness differed considerably according to rodent species. This period was relatively long for rat species, i.e. Norway rats, *Rattus norvegicus*, (87 days) and roof rats, *R. rattus*, (61 days) followed by the gerbil, *Girbillus girbillus*, (59 days). It was relatively short in the house mouse, *Mus musculus*, (32 days) and spiny mouse, *Acomy*

*cahirinus*, (38 days), They concluded that the difference between these periods might be due to differences in feeding patterns among the rodent species.

Table 1. Effect of Librax<sup>R</sup> 0.001% on different concentrations of zinc phosphide bait against black rat, *Rattus rattus L*, using non-choice method.

Tretment	Average daily bait Consumption		Acceptability (%)	Mortality (%)	Time of dead(h)
	Pretreatment	Treatment			
Zinc phosphide (1%)	18.85	2.64	12.44	100	24
Zinc phosphide (1%) +Librax <sup>R</sup>	21.76	4.59	17.42	100	24
Zinc phosphide (0.5%)	15.34	5.39	26.0	100	24
Zinc phosphide (0.5%) +Librax <sup>R</sup>	14.79	7.25	32.37	100	24
Zinc phosphide (0.25%)	10.96	7.024	39.04	60	48
Zinc phosphide (0.25%) Librax <sup>R</sup>	10.48	6.014	36.47	80	24
Librax <sup>R</sup>	9.91	13.38	57.45	0	0

L.S.D 3.52

Table 2. Consumption of black rat, *R.rattus*, of zinc phosphide bait (sub-lethal dose) alone and mixed with Librax<sup>R</sup> in non-choice method.

Treatment	Average daily bait Consumption (g)		Acceptability (%)
	Pre-treatment	Treatment	
Zinc phosphide (0.025%)	11.48	7.22	38.59
Zincphosphide(0.025%)+ Librax <sup>R</sup> (0.001%)	13.68	11.4	45.45

Table 3. Effect of 0.001% Librax<sup>R</sup> mixed with 0.5% zinc phosphide bait against black rat, *Rattus rattus L.*, using free choice method after one week of sub-lethal dose treatment.

Treatment	Average daily bait consumption (g)						Acceptability (%)	Mortality (%)	Time of death (h)
	1 <sup>st</sup> day		2 <sup>nd</sup> day		Mean				
	Treated	untreated	Treated	untreated	Treated	untreated			
Zinc phosphide	0.37	10.25	0.61	10.61	0.49	10.44	4.69	20	24
Zinc phosphide +Librax <sup>R</sup>	0.35	10.08	1.44	13.03	0.99	11.55	8.57	20	48

L.S.D. 1.22

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## كفاءة عقار الليبراكس كمضاد للألم في التغلب علي ظاهرة النفور من طعام فوسفيد الزنك للجرذ الاسود

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أجريت بعض المحاولات المعملية بهدف إستساعة و قابلية الجرذ المتسلق لطعم فوسفيد الزنك بإستخدام عقار الليبراكس كمضاد للألم والتقلص المعوي الناتج من فوسفيد الزنك. تم إختبار عقار الليبراكس 0.001% منفردا و مضاف علي تركيزات مختلفة من طعام فوسفيد الزنك بإستخدام طريقة التغذية الإختيارية و الإختيارية. وقد أوضحت النتائج أنه في كلا الإختبارين أظهر عقار الليبراكس زياده واضحة في نسبة إستساعة الجرذان لطعم فوسفيد الزنك حينما عوملت بعقار الليبراكس منفردا أو عند خلطه بطعم فوسفيد الزنك، حيث إرتفع معدل إستهلاك الجرذان لطعم فوسفيد الزنك بدرجات ملحوظة معنويا عند المعاملة بعقار الليبراكس. كذلك إرتفعت النسبة المئوية للموت في الجرذان المعاملة بالعقار بالمقارنة بتلك المعاملة بفوسفيد الزنك فقط.