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Actellic 2% Dust as Pesticide in Feed Ingredients: Post-Weaning Tolerance, Growth Performance and Organ Weights of Growing Rabbits

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Abstract: The study was designed to evaluate the tolerance, survivability and performance of weaner rabbits fed varying levels of actellic dust. The pathological effects of the pesticide on the animals were also appraised. A uniform diet was compounded from ingredients certified to be apparently pesticide residue-free. This diet was partitioned into 5 sub-diets. The first partition A had no residue, serving as control while B, C, D and E had 0.01, 0.02, 0.03 and 0.04% inclusion of the actellic dust, respectively. A total of 20 rabbits of mixed breeds and sexes were used in the experiment. The animals were balanced for breed, sex and weight in a completely randomized design experiment. Four animals each were placed on each treatment diets for 45 days which were preceded by 3 weeks of acclimatization. Data on feed intake and weight gain were collected on weekly basis. Data on blood volume and organ weight were also obtained at the end of the study. Feed intake reduced with progressive inclusion of actellic dust. The average daily feed intakes were 71.40g (A), 66.04g (B) 64.82g (C), 67.56g (D) and 63.79g (E) though not significantly different ($P > 0.05$) Weight gain however increased with increase dosage of the animals with treatment E having the highest gain and significantly higher ($p < 0.05$) than treatment B but not higher ($p > 0.05$) than the control. Feed utilisation (feed efficiency ratio) was comparable in all treatments. Organ weights and blood volume (average values of 4.67-7.01% of body weight) were comparable among groups except for heart, spleen and liver that were significantly different ($P < 0.05$) indicating incidence of gross pathological changes in the group with the highest dose of actellic dust inclusion. Percentage mortality of 25 was also recorded from treatment with the highest dose. The results therefore show that weaner rabbits can tolerate the presence of actellic dust in feed up to 0.03%. At this dose, the actellic dust does not have adverse effects on growth performance. Higher concentration of 0.04% can result in manifestation of toxicity symptoms and sometimes death, such values are rarely naturally present in feeds. However, animals which are able to tolerate high values hardly manifest poor performances.

Key words: Actellic dust, tolerance, performance, organs rabbits

INTRODUCTION

Rabbit is becoming a very popular source of meat in Nigeria due primarily to their versatility in the feeding habits, limited space requirement, ease of handling and short gestation and generation interval (Onifade and Tewe, 1993). Adegbola *et al.* (1985) and Bamikole and Ezenwa (1999) emphasized the importance of concentrate in feeding rabbits.

Stored grains form the bulk of the ingredients in concentrate used as animal rations including rabbits (Olomu, 1995). One of the greatest losses of these ingredients is due to ravage of insects and other pests after harvesting when grains and other crops are in store. One of the most effective ways of controlling these pests is the use of chemicals called pesticides.

Since the widespread of strains of stored product pests resistant to malathion (Pieterse *et al.*, 1972) there has been considerable interest in Actellic 2% dust (a.i. pirimiphos-methyl) which has proved effective against all known strains of malathion-resistant stored product insects. The most effective method of protecting grains is by the admixture of actellic dust. Bullock (1974)

undertook numerous trials where pirimiphos-methyl was admixed with wheat, barley, maize and rice in husk. Mean initial residues of pirimiphos-methyl itself in the whole grains of wheat, barley and maize rarely exceeded the nominal treatment rate and were frequently in the range of 40-60% of the nominal dose. However, the stability of the deposit of pirimiphos-methyl on grains is quite remarkable and the half-life has been estimated to be of the order of 9 months (Bullock, 1974).

Domestic animals and man are of course, the unfortunate victims of these residues. The toxic effects may not often be attributed to the pesticide residues since the farmers and commercial dealers are either not inclined to expose themselves nor the end-users do not have the scientific knowledge or facilities to detect the deleterious effects on the animals. Usually these victims suffer grossly from the immediate impacts of pesticide residues and insidiously therefore, from the cumulative build up of the chronic presence of the toxicity. The effect of this to health and economy could be significant.

Several chemicals are available for use as pesticides. However, preliminary surveys carried out through

unstructured questionnaire among agro chemical dealers have shown that Actellic 2% dust is one of the most popular pesticides used in the storage of grains in Edo State of Nigeria and its Environs. This study was therefore design to determine the tolerance level of Actellic 2% dust in feed of weaner rabbits, its effects on growth performance and signs associated with its toxicity.

MATERIALS AND METHODS

The experiment was carried out in the University of Benin Teaching and Research Farm, Benin City in Southern Nigeria. Benin City is located between latitude 6° and 6°30'N of the equator and longitude 5°40' and 6°E of the Greenwich Meridian. It also has an annual rainfall of about 2162mm and a mean relative humidity of 72.5%. The mean temperature of the rabbit house for the period the experiment lasted was 27.6°C.

The experiment was carried out using 20 mixed breed rabbits obtained from farms in Benin and environs. The animals aged between 6 and 7 weeks. The average initial weight of the rabbits (12 males and 8 females) was 670g (range = 470-720g).

On arrival, the rabbits were given neo-terramycin in drinking water, a combination of vitamins and antibiotics. This served as anti-stress agents and prophylaxis. The animals were housed individually in raised hutches in a house which had a concrete floor with metallic sheet roof. The house was designed to ensure cross ventilation and to exclude rodents and other pests. Raised metallic feeders were hung at a reasonable height to prevent wastage resulting from spillage while weighted clay bowls served as drinkers. On daily basis throughout the experiment, the rabbits were physically examined to ensure absence of external disease complications that are not products of the experimental treatments.

The experiment which lasted for 45 days was preceded by a 3-week adaptation period. Within this period, the rabbits were fed generally on the control diet without pesticide. The experimental diet was formulated based on the nutrient requirement of weaner rabbits recommended by NRC (1977). Ingredients used for the formulation of the diet were carefully sourced locally and were ensured that they were not previously stored with any pesticide to obtain pesticide-free diet. After compounding the diet, the quantity of the feed that would last only for 3 days for each treatment was weighed and the pesticide (Actellic 2% dust) was admixed to obtain pesticide contamination of 0g/kg feed (0%), 0.1g/kg feed (0.01%), 0.2g/kg feed (0.02%), 0.3g/kg feed (0.03%) and 0.4g/kg feed (0.04) forming treatment A, B, C, D and E, respectively. This was based on the recommended dose (0.5g/kg ingredient) by the manufacturer for normal storage. Each of these treatment diets was partitioned into 4 replicates of 250g and neatly tied in nylon bags

and stored in air-tight plastic containers. These were only opened during feeding to minimize volatilization.

After balancing for weight, sex and breeds differences, the rabbits were randomly allocated to the five experimental diets at four animals per diet in randomized complete block design with each rabbit serving as a replicate. The animals were fed twice daily *ad libitum*. Before feeding the following morning, the remnants from the previous day meal were collected into individual containers. Feed left after 3 days of admixture with pesticide was discarded as part of the remnant. Fresh clean water was also supplied *ad libitum* on daily basis. The animals were individually weighed weekly.

Physical examinations such as posture, activity of the animals, fur texture, urination etc. were made during the experiment. Mortality was also recorded. Three rabbits each from a treatment were sacrificed for autopsy. Vital organs such as the heart, kidney, liver, spleen and lung were carefully removed and weighed. All data were subjected to the analysis of variance procedure (SAS, 1995) as completely randomized design. Mean separation was done where there were indications of significance using Duncan's Multiple Range test (Alika, 2006).

RESULTS

The gross composition of the basal experimental diet is shown in Table 1. The chemical composition of the diet (Table 2) shows that it is able to adequately meet the nutrient requirement of the growing rabbits. The feed intake and growth performance of the rabbits are also shown in Table 3. The highest daily feed intake was recorded for the control treatment which was closely followed by treatment D (containing 0.03% pesticide). The least feed intake was observed in treatment E with the highest dose of the Actellic 2% dust. Though there was relative difference in absolute values of the feed intake, the difference was not significant ($P > 0.05$). Significant difference ($P < 0.05$) was observed in daily weight gain of the animals with animal receiving treatment E gaining more than others. Although the gain was progressive from the control, treatment B had the least weight gain.

There were no significant differences ($P > 0.05$) among the treatments in their effects on the average feed efficiency ratio (FER) of the rabbits. The lowest FER was however recorded with treatment B. Treatment E had the highest FER but without significant difference ($P < 0.05$) from the other treatments. Notably, all the treatments gave positive FER.

The highest crude protein intake (g/day) was observed in treatment A (control). Inclusion of actellic 2% dust to the diet had no significant influence on crude protein intake. The highest protein efficiency ratio (PER) of 0.94 was observed with treatment E while the least value of

Table 1: Ingredients and calculated Nutrient composition (%) of Experimental Diets

Ingredients	Composition (%)
Maize	35.00
Maize cob	7.00
Palm kernel cake	40.75
Soybean meal	14.00
Bone meal	1.00
Oyster shell	1.50
Salt	0.50
*Premix	0.25

*Composition of the mineral and vitamins mix per kg of diet: vitamin A, 7,000IU; vitamin D₃, 1,400IU; vitamin E, 5IU; vitamin K, 2.0mg; vitamin B₁, 1.5mg; vitamin B₂, 4.0mg; vitamin B₆, 1.5mg; vitamin B₁₂, 0.1mg; Niacin, 15mg; pantothenic acid, 5.0mg; folic acid 0.5mg; Mn, 75mg; Zinc, 45mg; Fe, 20mg; Cu, 5mg; I, 1mg; Se, 0.1mg; Co, 0.2mg; Choline chloride, 100mg.

Table 2: Proximate composition (%) of the experimental diet before the admixture of actellic2% dust

Nutrient	%
Dry matter	91.10
Crude protein	18.12
Crude fibre	10.32
Ash	5.64
Ether extract	5.70
Nitrogen free extract	60.22
DE (kcal/kg)	2848.73
ME (kcal/kg)	2591.65

0.70 was recorded in treatment B. The PER was not significantly different ($P > 0.05$) among the treatments of the animals. Highest feed intake as percent body weight was observed in treatment B. This was not significantly higher ($P > 0.05$) than the values for other treatments. Result also indicated that the digestible energy (DE-kcal) intake/day decreases with increase concentration of pesticide in the feed. Highest intake of DE was noticed in the control treatment which was not however significantly different from the least value obtained from treatment E.

The results of the weight of organs as percent body weight are presented in Table 4. Liver weight of animals on treatment E was significantly heavier ($P < 0.05$) than for other treatments. Variations among the weights of kidney and spleen were minor and not significantly different. The heart was enlarged in animals in treatment E and differed significantly ($P < 0.05$) from the rest of the groups. There was also relative decrease in the mean weight of the lung with increasing dosage except for treatment D. Though there was relative marked difference in absolute values between the control (7.01g) and treatment E (5.50g) but there was however no significant difference between the extreme mean weights. Blood volume as percent body weight was comparable in all the group treatment. Generally, 25% mortality was recorded among animals in treatment E.

DISCUSSION

The nutrient composition of the diet could ensure adequate availability of nutrients for growing rabbits according to the standard of NRC (1977).

Results from the study showed that feed intake was progressively depressed by the corresponding increase in the level of Actellic 2% dust. The depression was not significantly different ($P < 0.05$) from the control. This agrees with the report of Rajini *et al.* (1987) that the feed intakes were comparable in all groups when 20 rats were administered 0, 50, 100 and 200mg of 90.5% purity of pirimiphos-methyl/kg bw/day. Berry and Gore (1975) however recorded a reduced feed consumption for male rats dose at 5ppm (statistically significant) and 8ppm (not statistically significant). Possible reasons for the depressed feed intake could be ascribed to the repulsive odour and astringent characteristics of the Actellic 2% dust. There was an increase weight gain with increase level of pesticide. This was contrary to reports of Rajini *et al.* (1987); Rajini and Krishnakumari (1988) who reported that weight gains were not comparably significant among rats. The only interpretation to the observation in the increase weight gain in this study could be drawn from the findings where Gage (1971) indicated that the metabolites of pirimiphos-methyl are natural products of body metabolism. Their roles were however not clear, hence the explanation to this is still difficult and subject to further investigations.

Feed utilization was similar in all treatment groups. This agreed with the report of Berry and Gore (1975). The feed efficiency ratio (FER) was however -29, 4, 0 and 50%, respectively of control value for treatments B, C, D and E. The FER were lower than 0.18-0.26 reported by Biobaku and Dosumu (2003) and Bamikole *et al.* (2000). There was also a reduction in crude protein intake with increasing dosage of Actellic 2% dust. The utilization of the protein also followed the pattern of FER. The values obtained in the study are similar to those reported by Maigandi and Hadejia (2003).

Full post-mortem examinations were performed and the major organs and any gross abnormality of interest were observed. There was little or no deviation of colour variation and fat deposit of the organs from the control treatment. The result however showed atrophy of the heart and liver at the highest dose. The values obtained were significantly different ($P < 0.05$) from other treatment means. This finding agreed with the report of Rivett *et al.* (1973) where there were increase in liver weights and liver body ratios seen in the group of dogs given 10mg/kg bw/day. Wekhe *et al.* (2003) reported cardiac hypotrophy in an inverse trend to rising dosage of rabbit with crude oil. The liver and lung also responded in a similar inverse trend with a paler coloration.

In this study, no treatment- related pathological changes were observed in the lungs and kidneys. The weight of

Table 3: Feed intake and performance of rabbits exposed to different levels of actellic dust

Parameters	A (0.00%)	B (0.01%)	C (0.02%)	D (0.03%)	E (0.04%)	SEM
Feed intake (g/day)	71.40	66.04	64.82	67.56	63.79	5.87
Wt gain (g/day)	9.65 ^{ab}	7.46 ^b	9.69 ^{ab}	10.25 ^{ab}	11.84 ^a	1.01
Ave feed efficiency						
Ratio	0.14	0.10	0.16	0.14	0.17	0.03
Crude protein intake (g/day)	12.94	12.09	11.75	12.2	11.53	1.08
Protein efficiency						
Ratio	0.75	0.70	0.93	0.75	0.94	0.15
Feed intake as % body wt	6.62	6.97	6.65	6.60	6.97	0.60
DE (Kcal) intake per day	203.40	188.55	184.64	192.45	181.73	16.75
Mortality (%)	0	0	0	0	25	

Mean in a row with different superscripts are significantly different (P < 0.05).

Table 4: Percent Organ Weights and Blood Volume of Body Weight

Parameters	A (0.00%)	B (0.01%)	C (0.02%)	D (0.03%)	E (0.04%)	SEM
Liver (%)	2.88 ^{ab}	2.32 ^b	2.65 ^{ab}	2.81 ^{ab}	3.25 ^a	0.25
Kidney (%)	0.68	0.62	0.63	0.56	0.65	0.07
Spleen (%)	0.06 ^a	0.03 ^b	0.05 ^{ab}	0.06 ^a	0.04 ^{ab}	0.01
Lung (%)	0.53	0.45	0.44	0.50	0.41	0.05
Heart (%)	0.23 ^b	0.21 ^b	0.22 ^b	0.22 ^b	0.34 ^a	0.03
% blood volume of body wt	7.01	4.67	7.16	6.03	4.77	1.02
% viscera organs	29.93 ^{ab}	30.13 ^{ab}	31.19 ^a	29.99 ^{ab}	29.65 ^a	0.42

Means in a row with different superscripts are significantly different (P < 0.05).

the spleen was statistically different (P < 0.05) among the treatment groups. This agreed with the observation of gross or histopathology, which was not striking but slight to moderate haemopoiesis of the spleen (Clark, 1970). Dose-effect relationship was virtually non-existent despite statistically significant decrease in mean values. A wide variation in mean values between the control (0.06%) and the treatment B (0.03%) also renders the interpretation difficult.

The percent blood volume of body weight also showed a comparable result among the treatment means. Blood volume was inconsistently depressed below control level but the effect was generally greatest at the mid-dose. The values compared closely to % blood volume of body weight of other apparently health animals; horse (9.7%), cow (7.7%), sheep and goat (8.0%) and dog (7.2%) reported by Frandson (1974). However, dose-effect relationship do not appear to be present due to the inconsistency and coupled with the fact that there was no significant difference (P > 0.05) among the treatments. The percent viscera organs of the live weight increased with increasing dosing except for the highest dose of pesticide which was statistically different (P < 0.05) from the mid-dose. This provides evidence of rapid elimination through faeces and urine noticed by Hawkins and More (1979); Hawkins *et al.* (1989); Lock and Johnson (1990) in several animals.

Pathologically, signs of poisoning appeared the second week of dosing and increased in severity as the experiment progressed. Severity was also dependent on quantity of actellic 2% dust inclusion. These included occasional episode of vomiting, watery stools, abnormal

gaits, loss of appetite, dullness, rough fur and loss of hair. Some of these clinical signs were acute and persistent throughout the experimental period. Several of these persistence were however intermittent. Many authors have reported from a complete absence of clinical signs (Roberts *et al.*, 1983; Chester and Hart, 1986), to mild (Lock and Johnson, 1990; WHO, 1992) and acute toxicity (Clark, 1970; Gage, 1971). Water consumption and urination could also be said to have increased due to the quantity of water normally left at the end of the day and the level of wetness of faeces on the floor since the animal tend to excrete the residue from the body. Bratt and Jones (1973) observed that rats and dogs had 73-83% of the dose of pirimphos-methyl excreted in the urine during the 24 hours after dosing. This may also account for the low toxicity.

Conclusion: The results from this study show that Actellic 2% dust with its well protective properties against pests, fear of possible acute or chronic toxicity is unfounded. The presence of Actellic 2% dust though depresses feed intake, this does not negatively or adversely affect performance and feed utilization. High concentration values of about 0.04% in feed can result in manifestation of toxicity symptoms and sometimes death, such values are rarely naturally present in feeds. Besides, animals which are able to tolerate high values hardly manifest poor performance. Therefore, it could be concluded that Actellic 2% dust has low dietary toxicity and is slightly hazardous. It could be recommended for storage of feed ingredients but must be within the limits of specified conditions.

REFERENCES

- Adegbola, T.A., E.U. Tibi and D.C. Asugwa, 1985. Feed intake and digestibility of rabbits on all-forage, forage plus concentrate and all-concentrate diets. *J. Anim. Prod. Res.*, 5: 185-191.
- Alika, J.E., 2006. *Statistics and Research Methods*. Ambik Press, Benin., pp: 269.
- Bamikole, M.A. and I. Ezenwa, 1999. Performance of rabbits on guinea grass and Verano stylo hays in dry season and effect of concentrate supplementation. *Animal Feed Sci. Technol.*, 80: 67-74.
- Bamikole, M.A., I. Ezenwa, M.K. Adewumi, A.B. Omojola, V.O. Adetimirin, O.M. Arigbede and S.A. Orisadeyi, 2000. Alternative feed resources for formulating concentrate diets of rabbits: unthreshed grain Amaranth seedhead. *World Rabbit Science*, 8: 125-129.
- Berry, D. and C.W. Gore, 1975. Pirimiphos-methyl: determination of noeffect level during a 28-day rat feeding study. Report received from ICJ Central Toxicology Laboratory. Submitted to the WHO by ICI.
- Biobaku, W.O. and E.O. Dosumu, 2003. Growth response of rabbits fed graded levels of processed and unhulled sunflower seeds. *Nig. J. Amin. Prod.*, 30: 179-184.
- Bratt, H. and L.A. Jones, 1973. Pirimiphos-methyl: metabolism in rats and dogs. Report from ICI Industrial Hygiene Research Laboratories.
- Bullock, D.J.W., 1974. Pirimiphos-methyl: residues in stored grains, bread, flour and milled products. ICI Plant Protection Limited. Report no. Ar 2537a.
- Chester, G. and T.B. Hart, 1986. Pirimiphos-a study for monitoring the health of spray operators engaged in knapsack application of "actellic" 50ec and 40wp formulations. Report from ICI Plant Protection Division. Submitted to WHO by Imperial Chemical Industries.
- Clark, D.G., 1970. The toxicity of pp: 511 (0-c2-deathylamino-6-methylpyrimidin-4-yl) 0, 0-dimethylphosphorothoate). Report from ICI Industrial Hygiene Research Laboratories.
- Franson, R.D., 1974. *Anatomy and physiology of farm animals*. 2nd Ed., Lea and Febiger, Philadelphia, pp: 174-225.
- Gage, J.C., 1971. Pirimiphos-methyl (pp: 211) and pirimiphos-methyl (pp: 511): acute and subacute oral toxicity in the rat of the plant metabolite, 2-diethylamino-4-roxy-6-methylpyrimidine. ICI Industrial Hygiene Research Laboratory.
- Hawkins, B., C. Gopinath and J.C. Hadley, 1989. Acute delay neurotoxicity study with Pirimiphos-methyl in the domestic hen. Report from Huntingdon Research Centre, submitted to WHO by Imperial Chemical Industries, Ltd.
- Hawkins, D.R. and D.H. Moore, 1979. Tissue levels of radioactivity after repeated oral doses of ¹⁴c-pirimiphos methyl to rats. Report from Huntingdon Research Centre, submitted to WHO by ICI, Ltd.
- Lock, E.A. and M.K. Johnson, 1990. Delayed neuropathy and acute toxicity studies with pirimiphos-methyl in the hen. *J. Appl. Toxicol.*, 10: 17-21.
- Maigandi, S.A. and A.A. Hadejia, 2003. Performance of rabbits fed varying levels of *Faidherbia albida* pods. Proceedings: 28th Annual Conf. of NSAP, Ibadan, Nigeria, pp: 336-339.
- NRC, 1977. *Nutrient requirement of domestic animal*. No. 9. Nutrition requirement of rabbit. Second revised edition. NRC, Washington, D.C.
- Olomu, J.M., 1995. *Monogastric Animal Nutrition: Principles and Practice*. A Jachem publication, Benin City, Nigeria, pp: 320.
- Onifade, A.A. and O.O. Tewe, 1993. Alternative tropical energy feed resources in rabbit diets: growth performance, diet digestibility and blood composition. *World Rabbit Sci.*, 1: 17-24.
- Pieterse, A.H., G.G.H. Schulten and W. Kuyken, 1972. A study of insecticide resistance in *trobium castaneum* in Malawi. *J. Stored prod. Res.*, 8: 183.
- Rajini, P.S. and M.K. Krishnakumari, 1988. Toxicity of pirimiphos-methyl: i. The acute and subacute oral toxicity in albino rats. *J. Environ. Sci. Health*, 23: 127-144.
- Rajini, P.S., S. Viswanatha and M.K. Krishnakumari, 1987. Effects of pirimiphos-Methyl, an organophosphorus insecticide on haematological parameters in albino rats. *Ind. J. Exp. Biol.*, 190-193.
- Rivett, K.F., B. Edwards, E. Street and A.J. Newman, 1973. pp: 511, oral toxicity study in beagle dogs. Repeated daily dosage for two years. Report from Huntingdon Research Centre.
- Roberts, N.L., C. Fairley, R.H. Almond and D.E. Prentice, 1983. The subchronic delayed neurotoxicity of pirimiphos-methyl to the domestic hen. Report from Huntingdon Research Centre, submitted by Imperial Chemical Industries, Ltd.
- SAS (Statistical Analytical System), 1995. *User Guide: Statistics* SAS Institute, Cary, N.C.
- Wekhe, S.N., M.A. Yahaya and O.O. Mgbere, 2003. Crude oil pollution impact on the tolerance and survivability of domestic rabbits. Proceedings: 28th Annual conf. Nsap, Ibadan, Nigeria, 58-60.
- WHO (World Health Organization), 1992. *The who recommended classification of pesticides by hazard and guidelines to classification, 1992-1993*. (who/pcs/92.14). Available from the international programme on chemical Safety, world health organization, geneva, switzerland.