
Continued

[Contents](#) - [Previous](#) - [Next](#)

8. SILVANIDAE

The Silvanidae are a small family closely related to the Cucujidae. The adults are narrow and distinctly flattened, possess 11 -segmented antennae with a compact club. The elytra completely cover the abdomen, and there are five visible abdominal sternites. The tarsi are all 5-segmented. Most species are probably predacious, but two species of *Oryzaephilus* are of great importance as secondary pests attacking broken or milled cereals and oilseeds, while *Ahasverus advena* Waltl, the "foreign grain beetle", probably feeds on moulds and refuse, and is rarely found in grain which is in good dry condition. Another closely related species, *Cathartus quadricollis* (Guerin-Meneville) which resembles the saw-toothed grain beetle, but differs by the square prothorax which lacks the six large teeth on either side, is one of the most common beetles in stored corn in the Southern USA, and on damaged and exposed ears in the field. Its form and habit are also similar, while the larvae have the annoying habit of devouring the germs of the seeds in which they breed.

(i) *Oryzaephilus surinamensis* (L.), The saw toothed grain beetle.

Small (2.5-3 mm) brown beetle; distinguished by serrated lateral margins of thorax.

The eggs are laid loose amongst the substrate or tucked into creases in the grain. The pale yellow, elongate larva passes through four instars feeding and moving freely and eventually pupates within a cocoonlike structure of small grains or food particles. After a preoviposition period of 54 days the female lays some 375 eggs (at 30C) over a life-span of 6-8 months but may be prolonged at low temperatures. The adult may overwinter in temperate areas in the fabric of the building or granary.

The life-cycle can be completed over 20C to 37.5C, the optimum range being 31-34C. Development is relatively rapid and is completed in 22 days at 30C and 68 days at 20C (at 70% R.H.).

The species is very tolerant of low humidities and even at 30C and 10% R.H., larval mortality is less than 15%.

The adults rarely fly and during the day they tend to hide in cracks and crevices which enables them to escape the harmful effects of unfavourable temperatures and insecticide treatments. Aggregations occur at the edges of and in the dampest and warmest parts of a grain bulk and may contribute to grain heating.

O. surinamensis is a cosmopolitan secondary pest of cereals, mainly milled products such as flour, meals, breakfast foods, stock and poultry feeds, copra, confectionery and dried fruits.

(ii) *O. mercator* (Fauvel), The merchant grain beetle.

Small (2.5-3 mm) brown beetle: may be distinguished from *O. surinamensis* which it closely

resembles, by the eye diameter being larger than the temple region behind the eye, the more triangular head and generally darker and slightly larger.

Life-history and biology similar to that of *O. surinamensis* but *O. mercator* has a lower rate of increase because of its lower fecundity and slower development. Thus, the female produces an average of only 200 eggs and development takes 23 days at 30C and about 100 days at 20C (both at 70% R.H.). Additionally, the species is less tolerant of low humidities and extreme temperatures than *O. surinamensis* and is therefore less important in temperate regions.

O. mercator is more abundant in oilseeds, cakes and dried fruit than cereals and milled products although it will thrive in the latter.

(iii) *Ahasverus advena* (Walt!), The foreign grain beetle.

Small (2-3 mm) brown beetle: prothorax squarer than *O. surinamensis* and has only one prominent tooth at each apex.

Generally regarded as a mould feeder attacking a wide range of commodities such as damp cereals or cereal products, copra, groundnuts, oilseeds and cakes, dried fruits, etc., but can breed in clean cereals provided that yeast or germ is present. At 25C and 70 R.H. development is completed in 35-40 days but the species fails to breed below 60% R.H.

A. advena, although cosmopolitan, is most abundant in tropical and warm temperate regions.

9. TENEBRIONIDAE

This is one of the largest and most important families of small to moderate sized beetles nearly always unicolorous black or dark brown with 11 segmented antennae (rarely 10-segmented) and are generally thickened or clubbed towards their apices. The elytra completely cover the abdomen, and there are five visible ventral abdominal segments. The front and middle tarsi are 5-segmented and the hind tarsi are 4-segmented. It is useful since it distinguishes members of this family from nearly all other stored products Coleoptera.

They are particularly abundant in the subtropics, where most species are saprophagous, feeding on organic debris, and are also well adapted to dry conditions. Many are carnivorous; cannibalism being common, some are predators and others require their diet to be supplemented with insect remains.

Almost 100 species have been recorded as stored product pests. Their size varies considerably from about 21 mm in *Blaps mucronata* to 2.8 mm in *Alphitophagus bifasciatus*. The larvae are cylindrical, generally sclerotized, yellow to brown, and compared with pests such as *Sitophilus* and *Rhyzopertha*, the adults are longer lived, producing eggs throughout most of their life and the larvae pass through many more instars.

(i) *Tribolium castaneum* (Herbs"), The rust-red flour beetle.

Small (2.3-4.4 mm) reddish-brown beetle. Antennae with distinctive 3-segmented club. Width of eyes (seen from below) about equal to the distance between the eyes. The lateral margins of the

head are nearly continuous at the eyes.

The eggs, which are usually covered with particles and are laid singly. The larva, which is white to yellow and has rather obvious protruberances or horns on the terminal segment, passes through 6 or more instars. Newly emerged adults mate and start to lay eggs within 3 or 4 days. The adult may live for 14 months (average 9 months) and produces as many as 18 eggs per day during the first month of life when temperature, moisture and food supply are suitable. Although this high rate of egg production declines with increasing age the potential fecundity of any female may well exceed 1000 eggs. Both larvae and adults are cannibals and may consume eggs, young larvae and pupae.

The life-cycle can be completed over 20-42 C but the optimum lies between 32-37.5C. Development from egg to adult takes 22 days at 34C (72% R.H.) and 50 days at 24C (76% R.H.).

The adults fly readily at temperatures found 25C and are generally more numerous on the grain surface. Although cosmopolitan, *T. castaneum*, with its relatively high temperature optima is a pest of tropical or warm areas while in temperate or cold regions, it may be replaced by *T. confusum*.

T. castaneum is a secondary pest of a wide range of cereals, cereal products, legumes, oilseeds and cakes, nuts, spices and animal products and is regarded as an extremely successful omnivore particularly abundant in warm and often dry conditions. Quinones secreted by the adults produce an unpleasant musty taint under high population densities.

(ii) *T. confusum* Jacquelin du Val. The confused flour beetle.

Small (2.6-4.4 mm) flattened, oval, reddish brown beetle. Antennae without a distinct club, but expand gradually. The width of eyes (seen from below) about one third of distance between the eyes. The head margins are expanded and notched at the eyes. The head and upper parts of the prothorax are densely covered with minute punctures. Elytra ridged lengthways and sparsely punctate between the ridges.

Life-history and biology essentially as for *T. castaneum*. However, *T. confusum* is less fecund than *T. castaneum* and under favourable conditions lays only 14 eggs per day during the first month of life. The life-span is about 7 months but may exceed one year.

Development from egg to adult may be completed from about 19C to 37.5C, the optimum lies between 30C and 34C. The life-cycle is completed in 26 days at 34C (72% R.H.) and in 54 days at 24C (76% R.H.). Under identical conditions of temperature and humidity *T. confusum* develops more slowly than *T. castaneum*. Development may be prolonged at low humidities but mortality is little different from that at optimal conditions.

The adult seldom flies and the dispersal of the species is passive.

T. confusum is less susceptible to cold than *T. castaneum* and although cosmopolitan more abundant in temperate regions. A wide range of cereals cereal products, nuts, oilseeds and their cakes are attacked.

Several other members of the genus are also troublesome on cereals and cereal products. *T. audax* (Halstead), the American black flour beetle, is commonly found both outdoors and in warehouses

and is often confused with the European *T. madens* (Charpentier) which it resembles in appearance, biology and habits. Both are essentially similar to *T. confusum*, but are larger (3.55-5.2 mm) and black in colour. Another European import is *T. destructor* (Uyttenboogaart), a larger (4.5-5.5 mm) brown beetle which is not cold tolerant, and *T. anaphe* (Hinton) which is 3.6-5.1 mm long, brown, and is similar in biology to *T. castaneum*, but more susceptible to low humidities.

Because of the arrested development of some larvae at low temperatures *T. madens* appears better able to withstand cold conditions than the *Tribolium* species discussed. It is a secondary pest of some importance in granaries and flour mills in USA, Canada and eastern Europe.

(iii) *Tenebrio molitor* L. The yellow mealworm. Large (12-18 mm) polished dark brown to black beetle. Thorax is finely punctured, and the wing covers are longitudinally striated or grooved.

The bean-shaped, white eggs, which become covered in particles of flour and dust are laid singly or in clusters. The active larvae are yellowish in colour except for the brown head, prothorax and terminal segment and may moult as many as 23 times- the number being influenced by temperature. The adults live for up to 3 months which is considerably less than most *Tenebrionids*, and lay some 280 eggs.

The optimum temperature for development appears to be 25°C and at 75% R.H. The life-cycle is completed in about 250 days and there is normally one generation per year in temperate regions. The adults, which are quite strong fliers, may be seen in light traps at night.

T. molitor is a cosmopolitan but minor secondary pest that is more abundant in temperate and

subtropical areas than in the tropics. Found mostly in undisturbed residues in corners, under floors and under sacks, etc., and its presence is often indicative of poor hygiene.

(iv) *Tenebrio obscurus* F. The dark mealworm.

Large (12-18 mm) dull, pitchblack beetle.

Biology and life-history essentially the same as *T. molitor*. Adults live for two to three months and produce about 460 eggs over three months. The overwintering larvae usually begin to pupate before those of the yellow mealworm.

(v) *Alphitobius diaperinus* (Panzer). The lesser mealworm.

Medium sized (5.5-7 mm) blackish or very dark reddish-brown, beetle, resembling dark and yellow mealworms but much smaller. The eggs are generally stuck, in groups, onto a firm surface. The active cylindrical larvae, which vary in colour with increasing age from white to brown, pass through 8-11 instars and appear to require the presence of fungus for their proper development. The pupa, which is brown when mature, is found in a cell constructed by the larvae.

In a saturated atmosphere, the optimum temperature for development is approximately 32°C when development from egg to adult is completed in 46 days with 60% survival. At 16°C, development is extended to 97 days and survival is reduced to about 27%. Adult mortality is high and immature stages cannot survive in grain of 9% moisture content regardless of temperature and survive better in grain of 15% moisture.

A. diaperinus is a cosmopolitan secondary pest and is recorded from oilseed cakes and meals, cereal brans and meals, whole cereals and oilseeds. It is particularly abundant in damp out-of-condition grain and spillage and mostly occurs in poultry houses where it may be found in great numbers amongst the litter. It appears to act as a reservoir for avian leucosis.

(vi) *Alphotobius laevigatus* (F.), The black fungus beetle.

Medium sized (4.5-6 mm) black beetle. Differentiated from the lesser mealworm by the eyes being completely or nearly completely divided by a backwardly produced wide margin of the head. Life history and biology similar to that of *A. diaperinus*. Requires damp and mouldy conditions for survival. It infests oilseeds, oilseed cakes and meals, cereals and cereal products and other commodities but is not found in poultry houses. It is less tolerant of cold than *A. diaperinus* and has a more tropical distribution.

(vii) *Palorus subdepressus* (Wollaston), The depressed flour beetle.

Small (2-2.5 mm) reddish-brown beetle. Head with sides strongly explanate and flexed upwards. A distinct ridge above the eyes covering anterior portion. Eyes large with vertical diameter exceeding horizontal diameter.

The egg, which is sticky when laid and usually covered in food is laid loose amongst the substrate. The cylindrical larva, which are transparent when young and brown when mature, passes through 7-9 instars. The pupa is found within a cell amongst the substrate.

At 30C and 80% R.H., the female produces some 2.5 eggs per day with a maximum of 650 eggs throughout most of her life span of about 6 months. The development from egg adult can be completed over the range 20-35C; the optimum being 30-32.5C. Development takes 36 days at 32.5C and 124 days at 20C and relative humidities of 70-80% appear to be the most favourable.

P. subdepressus is a cosmopolitan, but not abundant, secondary pest in the residues of damp and mouldy grain.

(viii) *Palorus ratzeburgii* (Wissman). "The small-eyed flour beetle"

Small (2-3.0 mm) somewhat oblong, flattened reddish-brown beetle. Head with sides not strongly flexed upwards while the ridge above the eyes is feeble and indistinct. The eyes are small and round. Biology and life-history generally similar to *P. subdepressus*. The female is more fecund and, at 30C and 80% R.H., lays about 3.4 eggs per day.

The optimum temperature for development lies between 30C and 32.5C can be completed up to 37C. The species develops quicker than *P. subdepressus* i.e. 22 and 69 days respectively at 32.5C and 20C (70% R.H.) and can tolerate drier conditions.

The small-eyed flour beetle is cosmopolitan but generally is more common and has a wider distribution than the depressed flour beetle. It breeds in grain and milled products and is frequently found in basements. However, both species appear to be of relatively minor economic importance.

(ix) *Latheticus oryzae* Waterhouse, Long headed flour beetle.

Small (2.5-3 mm) rather slender flattened, yellowish-brown beetle. Head extended in front of eyes. Antennae distinctive: short and thickened with a five segmented club.

The eggs, which are sticky when laid; are usually covered in floury particles. The free living larva is white and passes through 6-7 instars. The pupa is white and is found naked amongst the substrate. The slow moving adult lives for up to 112 days and lays about 300 eggs.

The optimum temperature range for development is 33-37C with a minimum threshold of about 25C. Development from egg to adult is completed in 29-37 days at 31 C and 70% R.H. on sieved wheat.

L. oryzae is a cosmopolitan secondary pest of some importance in the tropics and sub-tropics but is of little significance in cold climates except in heated premises, because of its unusually high temperature preferences. Both damaged whole grains and cereal products are attacked and causes the same type of damage as the confused flour beetle. It has been recorded infesting wheat, rice, corn, barley, rye, flour and similar products.

(x) *Gnathocerus cornutus* (F.), The broad horned flour beetle.

Small (3-4 mm) reddish-brown beetle. Mandibles of male armed with a pair of broad, stout horns; edges of head expanded and strongly flexed upwards.

The egg, which is sticky when laid, is covered in floury particles. The free living larvae, which is yellowish-brown with a dark brown head and terminal segment, passes through 7-8 instars. The exarate pupa is yellow when young and brown when mature, and found loose amongst the substrate.

Females may produce as many as 1200 eggs over a period of 7-14 months. Development is possible over the range of 15-32C while the optimum range appears to be 24-30C and takes about 8 weeks. Low humidities extend the larval period without markedly increasing mortality.

G. cornutus is a cosmopolitan, but minor, secondary pest in flour mills and a scavenger in grain debris. It cannot tolerate low temperatures.

(xi) *Gnathocerus maxillosus* (F.). The slender horned flour beetle.

Small (3-4 mm) reddish-brown beetle, more slender than *G. cornutus*, with the life-history and biology being essentially similar. Development is possible over the range 20-35C and takes 35 days at 30C (91% R.H.) and 64 days at 23C (93% R.H.).

This pest is of little economic significance and is mostly recorded on maize or groundnuts in the tropics and sub-tropics.

10. TROGOSITIDAE

The members of this family vary considerably in size, form and habits. Adults are obovate (egg

shaped) to parallel without hairs or scales or at most with a few very short, microscopic hairs. The antennae are 11-segmented and terminate in a 3-segmented club. Elytra completely cover the abdomen. Tarsi are 5-segmented but the basal segment is sometimes so small that they appear 4-segmented (viewed with a hand lens).

Mostly tropical species often associated with decaying wood and preying on the larvae of other insects. Some species are mycophagous (fungivorous). Two species, the cadelle and the Siamese grain beetle (family Lophocateridae of Hinton and Corbett, 1972) are of economic importance in stored products. (Trogositidae previously known as family Ostomatidae in Munro's classification "Pests of stored Products").

(i) *Tenebroides mauritanicus* (L.). The Cadelle.

Large (5-11 mm) shining-black oblong beetle. Prothorax with base distinctly separated from the base of the elytra. Elytra possessing longitudinal ridges.

The eggs are laid loosely in batches in the substrate or are tucked into crevices and hatch in 7-10 days. The larvae, which are whitish and cylindrical becoming large and fleshy when matured are approximately 18 mm long and easily recognized by the two black horny terminal points and black head and thoracic shield. Feeds on damaged grain when young and later almost exclusively on the germ of the grains while cannibalism and predation are also common. There are four larval instars the last of which searches for a pupation site between the timbers of the store or burrows into the timber hollowing out a chamber within. In temperate regions the cadelle overwinters as an adult or larva leaving the grain to shelter in the fabric of the store. The female may produce more than

1000 eggs in a lifespan from 6 to 12 months, but some adults survive for nearly 2 years, making the cadelle one of the longest lived insects that attack stored grain.

The life-cycle may be completed in from 70 days to more than one year. Warm damp grain is preferred.

T. mauritanicus is a cosmopolitan pest whose habit of burrowing into timber structures, sacks and flour-mill silks causes much structural damage as well as providing hiding places for other pests. It is a difficult pest to control by application of insecticides. The lowering of germination caused by both adults and mature larvae feeding on the germ is probably the most appreciable form of grain loss. Both the adults and larvae can survive for long periods without food and often remain hidden in wooden bins for long periods after the grain has been removed. Newly harvested grain becomes infested within a relatively short period.

(ii) *Lophocateres pusillus* (Klug). The Siamese grain beetle.

Small (2.7-3.2 mm) flattened reddish-brown beetle. Prothorax with base closely attached to the base of the elytra which also possess longitudinal ridges. Both the margins of the thorax and wingcovers are more distinctly flattened.

The eggs are laid in clusters, generally in crevices, and there are four larval instars. The optimum temperature for development is approximately 30°C and can be completed down to 10% R.H. At 70% R.H. development from egg to adult is completed in 49 days at 35°C and 180 days at 20°C.

The Siamese grain beetle is now well established in the southern states of the USA, being commonly found in rice mills in Texas. Its economic significance has not been adequately demonstrated, but it often exists in large numbers on both paddy and milled rice in some Asian countries. Evidence seems to suggest that it is capable of feeding on more than just detritus in bulk grain, especially since the immature larvae are certainly capable of penetrating or utilizing any husk defects that can support populations of *S. oryzae* and *R. dominica*.

11. PTINIDAE

Because of their elongate legs and antennae, and stout rounded hairy bodies, they have generally been referred to as spider beetles. The antennae are situated in front of the head between the eyes and are generally close together. They are 11-segmented and never thickened epically. The base of the prothorax has a short and narrow constriction or neck. The elytra completely cover the abdomen and there are either four or five visible abdominal sternites. All tarsi are 5-segmented, with segments 1-4 decreasing in length. It is a small family of 500 described species, of which 24 have been found associated with stored food products in various locales throughout the world. Closely related to the Anobiid beetles previously described.

They are a cosmopolitan family, which is well represented in the tropics, but only of economic significance in temperate regions. Generally associated with stored foods as scavengers feeding on dead insects, excrement or dry vegetable matter.

The "hairy spider beetle" *Ptinus villiger* (Reitter) is a reddish brown beetle marked with four irregular white patches on the elytra. Approximately 3.5 mm long it is a common pest in the Prairie

Provinces of Canada and Northern USA. It attacks stored grain and all types of milled cereal products. The female lays about 40 eggs and under favourable conditions development takes 3-112 months. *P. raptor* Sturm is an allied species which is as common as the hairy spider beetle in Canada.

The "white-marked spider beetle" *Ptinus fur* (Linneaus) closely resembles the hairy spider beetle in appearance and habits. It is 2.0-4.3 mm long and feeds on both animal and plant matter and attacks flour, feed, grain and miscellaneous foodstuffs but is rarely abundant enough to inflict serious damage. The brown spider beetle *Ptinus clavipes* Panzer (= *P. hirtellus* Sturm) closely resembles *P. fur*, is 2.3-3.2 mm long, but can be distinguished from that beetle by the lack of white hairs or scales on the elytra. *Ptinus tectus* Boieldieu (= *P. ocellus* (Brown)) is also close in appearance, is 2.5-4.0 mm long, but the elytra are so densely clothed with brown or golden brown hairs that striae punctures and intervals are not distinct.

The female may lay about 1000 eggs, but 270 would be more typical during a life-span of 5-6 months. Access to water is essential for maximum fecundity.

Optimum temperature for development is between 23-27C and at 70% R.H., the time from egg to adult is 62 days (or 78 days at 20C).

The adults cannot fly, and are most active in dark damp places. The species is often associated with birds nests and bat roosts which may act as reservoirs for reinfestation. It is a hardy beetle and has a higher rate of increase compared to other *Ptinids*.

P. tectus is cosmopolitan but of particular importance in temperate regions and is the most destructive of the spider-beetles. All types of cereals, cereal products, oilseed meals, but not oilseeds themselves, are attacked as are many spices and animal products. Adults and larvae readily bore through paper or cardboard containers and sacks particularly through adjacent surfaces, causing the containers to split.

Mezium americanum (Laporte), "the American spider beetle is a small spider beetle with a golden head and thorax and shiny subglobular body that is characteristic of this genus. It is a scavenger generally found in cereals and oilseeds but of relative little importance. It is cosmopolitan compared to *M. affine* Boieldieu which is restricted to more temperate regions, even though it is best suited to the tropics (optimum humidity and temperature is 70-80% R.H. and 30-33C respectively, when development is completed in 62 days; females lay 460 eggs and have a life-span of 17 to 21 months).

A closely related species, the bag beetle, *Gibbium psylloides* (Czenpinsky) is a small (1.73.2 mm) dark brown shining beetle which is distinguished from *M. americanum* by the head and thorax, which are densely covered with small scales or scale like hairs.

A cosmopolitan species associated with a variety of produce such as grain residues, cotton seed, linseed, animal fertilizers, dried egg etc. It is most frequent in the tropics and sub-tropics, particularly on the outsides to bags, but it is of little economic consequence. It also is reported as occurring in the extreme southern portion of the USA, whereas *M. affine* is the common form in the Northern States and Canada. Females survive 10 months during which time she lays some 300 eggs. Optimum conditions are 33C when development takes 45 days (116 days at 20C) at 70% R.H.

(still possible down to 40% R.H.) and is regarded as cold tolerant.

Other Ptinids that maybe encountered are *P. pusillus* Sturm, *Tipnus unicolor* Piller and Mitherpacher, *Trigonogenius globulus* Solier and *Niptus hololeucus* Faldermann among others (identification of spider beetles from Hinton, H.E. (1941) "The Ptinidae of economic importance". Bull. Ent. Res. 31:331-381).

MISCELLANEOUS STORED PRODUCT BEETLES

CLERIDAE

The Cleridae or "chequered beetles" comprise rather a large family of 2000 species which are mostly of tropical distribution. They are brightly coloured pubescent insects of moderate size with 11-segmented antennae with the apical segments enlarged to form a distinct club. Elytra generally completely cover the abdomen, and there are five or six visible abdominal sternites. The tarsi are all 5-segmented, with the first and fourth often small and indistinct. They are mostly predaceous beetles feeding on wood borers.

1) *Necrobia rufipes* (Degeer). The copra or redlegged ham beetle.

Medium sized (4-5 mm) dark blue beetle with red legs.

The eggs are laid in batches of up to 30 in cracks and crevices. There are four larval instars, while the presence of a high protein substrate such as fishmeal greatly enhances development and

reduces larval mortality. The larvae are cannibalistic and predatory particularly attacking the larvae of fungivorous beetles. In the tropics, the females live for up to six months and may produce as many as 3000 eggs. However, if sufficient protein is lacking, both longevity and fecundity are greatly reduced. The adults fly readily.

The speed of development may be completed in as little as 42 days at 30C and 65-70% R.H. but can be completed at humidities down to 50% R.H.

N. rufipes is widespread in the tropics and will attack oilseeds, copra, dried and salted fish and cured meats such as ham and bacon.

2) *Paratillus carus* (Newman)

Medium sized (5-7 mm) beetle with red head and thorax and blue elytra with a transverse white stripe.

Occasionally found in stored products but is really a predator of timber borers infesting the structure of the store.

3) *Thanoclerus buquetii* (Lefebvre)

Medium sized (5-7 mm) dark brown beetle.

A predator of *Lasioderma serricorne*, *Sitophilus zeamais*, *Tribolium castaneum* and other stored product pests.

NITIDULIDAE

A large family of more than 2,000 species which are extremely variable in form, structure and habits. Many inhabit flowers, others are found in decaying animal and vegetable remains, in fungi and in exuding sap while some larvae are predatory. Those important to stored products are attracted to mouldy grain, cured meats and dried or decaying fruit.

Adults are obovate or oblong beetles with 11segmented antennae terminating epically in a distinct 3segmented club. The elytra are shortened so that two or three abdominal tergites are visible, while there is always five visible abdominal sternites. All tarsi are 5segmented the fourth segment shorter than the rest, while the first three segments are dilated and hairy beneath.

1) *Caropophilus dimidiatus* (F.), The corn-sap beetle.

Small (2-3.5 mm), oblong shining beetle with short truncate elytra; colour is variable from black to brownish-yellow.

The eggs are laid on or in decaying plant materials such as mouldy grain. The larvae pass through four instars in the soft and mouldy parts of the fruit or residue, while the pupae are generally found in the soil. The females each lay about 1000 eggs over 34 months. The adults fly readily congregating at suitable breeding and oviposition sites.

The life-cycle may be completed in as little as 15 days at 32C and 49 days at 18C. Corn sap beetles, which are cosmopolitan, may be found in damp and mouldy grain, peanuts, copra, cotton-seed,

maize-cobs in both field and in storage. Often found in ricemills breeding in accumulations of broken rice or swarming and crawling over bags. It is not attracted to clean, dry grain in good condition.

2) *Carpophilus hemipterus* (L.). Dried-fruit beetle.

Small (2-4 mm) oblong shining black beetle with buff spots on the elytra.

Life-history and biology essentially as for *C. dimidiatus*.

C. hemipterus is a serious cosmopolitan pest of dried fruits, figs, dates and maize-cobs in the field.

LISTERIDAE

A large family of compact, hard, shining beetles with elbowed and listinctly clubbed antennae. The elytra are truncated leaving the terminal abdominal tergites exposed. They are mostly black or brown beetles but the elytra are sometimes marked with red and a few species ire metallic. Some are carrion feeders while others live beneath bark or in the burrows of wood borers. The larvae are generally carnivorous but of little significance in the control of pest species. One member, *Carcinops pumilio* (Erichson) a small to medium (1.5-9 mm), broadly oval, shining black beetle is cosmopolitan and common in grain residues and groundnuts.

STAPHYLINIDAE

A very large family of small to moderate sized beetles, usually slender and almost parallel sided

and somewhat flattened. The elytra are short covering only the first few segments of the abdomen, but the wings are functional and most species fly readily. The beetles are variable in colour and although many are entirely brown or black red, yellow and blue are common. Staphylinids are common in decaying organic matter, dead animals and fungi, many are predators, a few parasites and others herbivores. The larvae are active and pass through three instars.

Most staphylinids are predators but of little practical value in grain storage and their presence is more indicative of damp, unhygienic conditions favourable to infestations of other insects and mites.

CRYPTOPHAGIDAE

A small family of small (2-3 mm) pale or darkbrown beetles found in mouldy environments and in the nests of ants, bees, birds and small mammals. They frequently occur in small numbers in warehouses and live principally on moulds. Some 25 species of the genus *Cryptophagus* may be encountered in the storage environment such as *Cryptophagus affinis* Sturm., a small (1.7-2.3 mm) brown beetle, of wide distribution, often found in grain refuse and dried fruit.

LANGURIIDAE

A large family of small to medium sized oval shaped beetles which are mostly black, with orange or red spots, or metallic blue and scarlet. The adults and larvae of most species are fungus feeders.

1) *Pharaxonotha kirschi* Reitter. The Mexican grain beetle.

Medium sized (4-4.5 mm), shining, deep brown beetle with rather long antennae with a loose three segmented club. It somewhat resembles the confused flour beetle but is distinguished by its more polished surface and longer antennae. This species is found in maize, wheat, beans and flour, common in Mexico and Guatemala, but it is not known to be established in the U.S. Another Langurid, *Cryptophilus integer* (Heer) is a small (2-2.3 mm), shining, darkbrown beetle found in warehouses in Europe and North America.

LATHRIDIIDAE

A small family of very variably shaped beetles 1-3 mm long, usually obovate pale brown to nearly black beetles which are found in mouldy plant and animal materials, in vegetable refuse and under bark and stones. Some 35 species of general such as *Corticaria*, *Coninomus*, *Enicmus* and *Holoparamesus* are recorded from granaries, mainly as fungus feeders but none are of real economic importance although, at times attaining quite large populations.

The antennae of most common species are 11segmented with a rather compact 2 or 3-segmented club. The sides of the prothorax are explanate or dilated and is depressed along the middle and transversely near the base. The elytra completely cover the abdomen and there are five visible abdominal sternites. All tarsi are 3-segmented. Specific differentiation however, is difficult amongst members of this family (a full account is given by Hinton, H.E. (1941), "The Lathridiidae of Economic Importance", Bull. Ent. Res., 32:191-247).

1) *Corticaria pubescens* (Gyllenhai)

Small (2.3-3 mm), stout, convex reddish-brown beetle, the largest member of this genus which is cosmopolitan in distribution and often found in grain residues in buildings and the field. *Corticaria adelaide* Blackburn and *C. australis* Blackburn have been found in standing wheat crops in New South Wales in 1966 (Greening, 1978) but do not pose any threat after harvest where the adults are usually found dead.

MYCETOPHAGIDAE

A small family of small (1.5-5.0 mm), oblong to oval, flattened, densely pubescent beetles which are generally brown to black in colour with yellow or reddish spots on the elytra. The larvae and adults are fungus feeders occurring under bark in haystacks, on mouldy grain and in vegetable refuse. The family is of little economic importance. The antennae are 11segmented with a 2 to 5-segmented club. The elytra completely cover the abdomen and all tarsi are 4segmented except the front tarsi of the males which are 3-segmented.

1) *Litargus balteatus* LeConte

Small (1.7-1.9 mm), convex, oval shaped, dark brown, hairy beetle with several lighter patches on the elytra. A cosmopolitan species frequently infesting damp spillage or in harvester residues.

2) *Typhaea stercorea* (L.). The hairy fungus beetle.

Small (2.2-3.0 mm), convex, oval shaped, hairy brown beetle which is common in a wide range of mouldy cereals, groundnuts, tobacco, cocoa, haystacks and in mills, rail cars and domestic

premises.

It is also frequently found in cornfields where it is apparently attracted to the decaying kernels on exposed ears. After the corn is harvested and shelled, it is often heavily infested, but there is little feeding on undamaged grain. It rather closely resembles the drugstore beetle, *Stegobium panecium* (L.) but can be distinguished by the shape of the antennae, which are clavate instead of irregularly serrate.

Another funivorous Tenebrionid, *Alphitophagus bifasciatus* (Say), is cosmopolitan and frequently found around mills and warehouses where waste materials are allowed to accumulate, and in wet and damaged grain in the holds of ships. It is a small (2.5-3 mm) handsome elongate-oval beetle, which is reddishbrown with two transverse black bands near the apex and base of the elytra. Its biology is relatively unknown, but development is completed in round 30 days at 25C and 70% R.H., the larvae being reared in moist cornmeal and in spoiled cereals. It is of little economic significance, except like most fungus feeders, it is an indicator of damp and mould grain that is deteriorating and likely to be heavily infested by the more important grain feeders.

[Contents](#) - [◀ Previous](#) - [Next ▶](#)

[Home](#) [ar](#) [cn](#) [de](#) [en](#) [es](#) [fr](#) [id](#) [it](#) [ph](#) [po](#) [ru](#) [sw](#)

Moths of economic importance infesting stored products: Selected notes on

bionomics and identification

[Contents](#) - [◀ Previous](#) - [Next ▶](#)

INTRODUCTION

Moths, skippers and butterflies are some of the most common and familiar insects and make up the insect order Lepidoptera. The name of the order comes from two Latin derivatives meaning "scaly wings" which refers to the tiny flat scales that cover the two pairs of membranous wings like shingles and gives them their characteristic colours and patterns. It is the second largest order of insects exceeded only by the Coleoptera or beetles, and contains approximately 150,000 described species. They range in size from tiny leaf-miners with wingspans of only 3 mm, to the huge cossids, saturniids and birdwing butterflies (*Ornithoptera* spp.) which have wingspans exceeding some 25 cm. The colouring and elegance have given them considerable popular appeal, while the destructive qualities of the larvae of many species establish the economic significance of the order. There are Lepidoptera in various diverse habitats such as arctic tundras to tropical forests to arid deserts and in every environment they play a major ecological part in their relationship with plants and other animals.

Despite their numbers and complexity, the members of this order are surprisingly alike with respect to their food source, nearly all of which are green-plant feeders concentrating primarily on the foliage. The great majority of larvae chew up leaves, many tunnel inside leaves feeding on the soft juicy tissue, while some bore into fruits, seeds, plant galls or wood, or are scavengers on dead and decaying plant or animal matter. Some adults do not feed at all, while others usually visit

flowers for nectar and many are attracted to sap, juicy carrion, excrement or rotting fruit.

Moths and butterflies are therefore primary consumers feeding directly on plant matter and transforming this into animal matter, a process in which they are not rivalled by any other group of land animals, either numerically or quantitatively.

Unlike many other insect orders, only a small minority of moths feed on other animals as parasites or predators (i.e., members of the subfamily Gerydinae, the harvesters, while most other Lycaenid caterpillars are cannibalistic).

The evolution of the endopterygotes based on fossil history begins in the Permian period 250 million years ago, in which the remains of Neuroptera (lacewings, antlions and snakeflies) and Mecoptera (scorpionflies) are found. It is not difficult to imagine that an ability to fly might have evolved several times but it is unlikely that the same pattern of wing venation would have evolved more than once, which strongly suggests a monophyletic origin of the Pterygota. The Neuroptera remain as rather an isolated case, but evidence suggests from studies on wing venation that the Mecoptera played a more crucial role in endopterygote evolution. Loss of some veins and modification of others has led to the Trichoptera (caddie-flies) and Lepidoptera in one direction and to the Diptera and Syphonaptera (fleas) in another, while the Coleoptera and Hymenoptera were probably derived from a neuropteran ancestor. The first beetles appeared in the Permian period and there are traces of Hymenoptera from the Jurassic, but the explosion of diversity in the main orders did not begin until the end of the Cretaceous period 70 million years ago, when flowering plants began to appear (see Table 1). The adaptation for feeding on the nectar of flowers by a modification of the mouthparts, has allowed the Lepidoptera to make the most of the rich

food resources made available by the flowering plants that are insect dependent for cross-pollination, a symbiotic process called entomophily (see Fig. 1).

General life history of the order

Like other endopterygotes such as beetles, flies and wasps, moths, skippers and butterflies go through a complete metamorphosis (holometabolous) involving a decided morphological change from the immature forms to the reproductively mature adult or imago forms. This "complete" or "abrupt" metamorphosis involves four distinct stages: egg, larva (or caterpillar), pupa (or chrysalis) and finally, the adult.

Two types of eggs maybe distinguished in the Lepidoptera. A flat type with the micropyle at one end, and an upright type with the long axis vertical and the micropyle positioned at the top. They are small, usually hard-shelled while the chorion is sometimes smooth or may exhibit a regular or irregular sculptured pattern with prominent ribs. In some species, the female may produce as many as one or two thousand, laid individually or in groups.

The larva or caterpillar, exhibits a clear division into head, 3-segmented thorax, and 10-segmented abdomen, whose chief function is to eat, digest and grow. The head capsule is heavily sclerotized while the prothorax carries a large dorsal sclerotized area, the prothoracic shield. They are peripneustic, bearing one pair of lateral mesothoracic spiracles and one pair of spiracles on segments 1-8 of the abdomen. The antennae are usually short, 3-segmented, and bear several sensilla. Posterior to the antennae, the eyes are merely represented by two groups of 6 simple ocelli which maybe reduced in number or even absent. The mandibles are normally well developed and

dentate, adapted for biting and chewing, but are modified in the sap feeding instars of leaf-miners. The maxillae are well differentiated with 3-segmented palpi, while the labium or second maxillae bears a median spinneret connected to very large silk glands and a pair of minute, lateral 2-segmented palpi. There is nearly always three pairs of 5-segmented thoracic legs, which terminate in a single claw. Those found in stored foods may be immediately distinguished from the larvae of all other insects occurring in similar situations by having paired pseudopods or prolegs on abdominal segments 3-6 and 10. Each of these prolegs is armed with a fine, curved apical "teeth" (crochets) which maybe of different lengths to form a definite series and are usually arranged in a circle. The head and body of the larva bear setae and punctures which follow distinct patterns (chaetotaxy) and these setal maps are widely used for the identification of larvae.

The larva is the nutritive stage of the life cycle and consequently possesses an enormous and efficient digestive tract. As the larva grows, it must shed its entire outer covering or cuticle to accommodate the increase in size. This occurs a number of times, and at each moult forms a new and larger head capsule and integument, discarding the remains of the old cuticle. Sometimes the larval life may last a couple of weeks although is usually longer, or may extend as long as three years in some species under extreme or unfavourable conditions. Many larvae are relatively plain in shape and colour, with only sparse hairs such as the moth larvae that infest stored products, but others especially those that feed out in the open, show a startling variety of colours and patterns and may bear dense hairs or spines or strong projections which perform a protective function.

When fully grown the larvae moults for the last time and transforms into a pupa. During this seemingly inactive quiescent stage, incredible histological, physiological and morphological

changes are taking place internally as the adult structures develop. The pupa maybe formed inside a silk cocoon spun by the larva while a great many are formed in a cell in the ground. Most moth and skipper pupae are quite plain, smooth and brown while many butterfly pupae (or chrysalids) are irregularly shaped, often metallic and brightly coloured.

The pupae and pharate adult stages of all primitive endopterygotes are passed in a cell or cocoon of some kind, and it is only in some specialized Coleoptera, Lepidoptera, Hymenoptera and Diptera that do not possess one. In the higher Diptera (sub-order Cyclorrhapha) and some others the last larval cuticle forms the puparium which somewhat resembles a barrel (coarctate pupa). An exarate pupa is one in which the appendages are free and not cemented to the body wall conversely, an obtect pupa has the appendages more or less strongly cemented to the body, presumably by tanning of a particular protein in the moulting fluid, and the cuticle is much more strongly sclerotized than in exarate pupae (see Fig. 2).

Various modifications have evolved to allow the adult to escape from its cell or cocoon. In primitive forms, the pupal cuticle is not generally shed by the adult, until it has escaped by using its pupal mandibles (decticous pupae). A pupa without articulated mandibles, and therefore one which cannot be utilized by the pharate adult, are called adecticous pupae.

[Fig. 1. Modified mouthparts of a butterfly, adapted for sucking. \(Source: "Pests of stored products" by J. W. Munro\)](#)

[Fig. 2. The three kinds of pupae: \(a\) exarate; \(b\) obtect; \(c\) coarctate. \(Source: "Pests of stored products" by J. W. Munro\)](#)

The methods of emergence of pharate adults of this kind are very varied. When the pupa is exarate, ecdysis normally occurs within the cocoon and the adult simply bites its way out, as in most Coleoptera and Hymenoptera. When obtect as in most Lepidoptera, the mobile abdomen of the more primitive forms are often armed with backwardly directed dorsal spines which force the abdomen forward when the body is wriggled, and the pupae partially emerge from the cocoon before adult ecdysis takes place. Such pupae often possess cocoon cutters on the head as in both Lepidoptera and Diptera

In the more advanced forms, ecdysis occurs within the cocoon or shelter which often has a weak area to allow easy escape for the newly emerged adult. Some regurgitate a fluid which softens the silk while others cut a hole means by means of a sharp hook on the forewing.

On emergence, the adult is very much different from the workaday larva from which it developed. Its whole life is oriented towards sensitivity and mobility, enabling it to carry out its primary functions of mating, multiplying and dispersing its species. Feeding, though important is secondary. Its two large compound eyes are excellent for detecting motion, patterns and colours of preferred flowers, and for potential mates. If simple eyes are present, they are normally paired one above each eye. The antennae are many segmented, with or without scales, the flagellum of the male usually more specialized than the females and vary greatly in structure.

Mandibles are usually absent and rarely functional, while the maxillae (galeae) are greatly elongated, usually grooved internally and fastened together by interlocking spines to form the tubular haustellum (proboscis) which is coiled beneath the head. In some cases, the proboscis is vestigial or absent, therefore adults cannot feed and subsist on food reserves stored when they

were larvae. Labium is small, usually with well developed 3-segmented palpi.

The prothorax is small compared to the meso- and metathorax which house the powerful flight muscles. Wings sometimes reduced or absent in females of some families such as Cossidae, Oecophoridae, Lymantriidae and Arctiidae. Similarly, the three pairs of well developed thoracic legs may be reduced as in the Numphalidae and the hind legs in some Geometridae.

They are usually 5-segmented with 5-segmented tarsi which often bear various tibial spurs or bristles and in some species, sensitive taste structures on the soles of the feet capable of detecting minute concentrations of sugars which result in a reflex coiling of the proboscis. The abdomen is usually 10-segmented with functional spiracles on segments 1-7. A pair of auditory tympanal organs are situated laterally near the base in the Pyraloidea and Geometroidea, while they may be present on the metathorax of most Noctuoidea.

Many species are weak fliers, and then for only comparatively short distances. Others however are capable of sustained flight for hours or days, or a fast darting flight and some are even capable of flying backwards. The North American monarch, *Danaus plexippus*, is deservedly famous through its regular southward migrations and has crossed the Pacific oceans and colonized Australia and New Zealand. Members of Pieridae, Numphalidae and Hesperidae undertake regular migratory flights, while the noctuid, *Agrotis infusa* (Boisd) a serious pasture and field crop pest, is known to undertake a remarkable annual two-way migration.

The Lepidoptera cannot physically resist attacks with strong jaws, hard shells, or poison stings as many other insect groups employ, and have consequently evolved other means of protection and

adaptations that are unparalleled in any other group of animals. Many make cases in which the live hidden during critical periods, others have adaptations in form and colour that enable them to escape unnoticed or deceive predators, some are clothed in poisonous hairs and spines or loose scales which enable some moths to escape from spider webs, while others have developed chemical defenses from the plants they consume, and have also developed aposematic or warning colourations to advertise these defenses. Many unprotected species have taken advantage by mimicing these colourations and therefore gain similar protection, while others may resemble distasteful or poisonous wasps and lycid beetles.

Classification

The classification of the Lepidoptera is exceedingly complex, and is based on many technical features. Opinions vary considerably, but the order is divided into four suborders, twenty-three superfamilies, and in excess of one hundred or more families. Despite their prominence and familiarity, butterflies are grouped in one super family - the Papilionoidea - and skippers which are often confused with the butterflies, another-the Hesperoidea, which together comprise no more than 10% of the order, the rest being moths. There is no definite demarcation between butterflies, skippers and moths since many overlapping characteristics exist. Butterflies are thought to be diurnal while moths are nocturnal, but many moths are day-fliers. Butterflies are generally brightly coloured while moths are dull and drab but in many instances the reverse occurs. Butterflies have clubbed antennae while most moths are hairlike tapering or plummy, but some moths also possess clavate antennae. The majority of moths have a special wing-coupling apparatus (the frenulum) while butterflies rely largely on the friction of overlapping parts of the fore and hindwings (the humeral and jugal lobes), the amplexiform method of coupling. But a great many moths do not

possess this mechanism. At rest, the wings may be folded in various ways, the ancestral method of folding roofwise over the abdomen is characteristic of most moths. In the Papilionoidea, the wings are generally held back to back above the body. In the true skippers, flight is generally very fast, without the characteristic fluttering as in the butterfly flight. The antennae has a thinner end portion beyond the club and is hooked at the tip.

Lepidoptera associated with stored products are all moths and cause damage only in larval stages. One species the Angoumois grain moth, *Sitotroga cerealella* (Oliv.) is capable of destroying sound, unbroken grain kernels, but most infest broken damaged kernels or milled products. This distinction provides a grouping of species into major and minor or primary and secondary accordary to their pest potential. For example, species such as *Ephestia Keuhniella* (Zell.), *Ephestia cautella* (Walk.), *Plodia interpunctella* (Hbn.), and *Hofmannophila pseudospretella* (Staint.) the primary pests of processed cereals but secondary pests of whole grain. However, while grain usually contains a proportion of broken grain, thereby allowing the establishment of secondary species as serious pests, especially if the moisture content is high. A common characteristic associated with larval development is the production of webbing. Adult moths rarely live longer than 14 days.

Some seventy five species, all of cosmopolitan distribution, are known to infest a range of stored products. The major species are small and exhibit a remarkable consistency in structure and appearance, which has led to difficulties in identification.

Identification of Larval Specimens

Moth larvae are best prepared for identification by placing them in a test tube containing either water or, preferably ten percent aqueous alcohol and heating the liquid until they are killed. This technique causes the larvae to become fixed in an elongate position which is most suitable for easy handling and identification. The fine hairs or setae on the larvae and their position, relative to each other and to the spiracles, are used as a diagnostic feature. The presence or absence of small brown sclerotized areas around the base of some of these setae is also a distinguishing feature; such as in the genus *Ephestia*, the presence of these dark areas gives the larvae a characteristically speckled appearance.

Identification of Adult Specimens

The coloured patterns formed by the minute scales which cover the top side of the forewings of adult moths provide a useful means of direct visual identification. When the moth is sitting at rest these forewing patterns are readily visible and the hindwings are hidden. The scales, however, are easily removed by rubbing, and adult specimens of the stored products species then appear remarkably similar; for example, moths which are held in a collecting jar will rapidly become de-scaled by their fluttering which renders direct identification difficult. Other more accurate methods of identification which rely on alternative diagnostic features must be employed when this occurs.

To identify moths by means of the pattern on the wings, it becomes necessary to be familiar with the more important parts of the wings. The margin of the wing nearest the head is the costa, that nearest the body, the dorsum and that farthest from the body the termen: the costa and the termen meet at the apex, while the angle formed by the termen and dorsum is the tornus. In some

moths, the apices of the wings are acuminate produced almost to a point. Almost all of the moths infesting stored products have similar wing patterns in both sexes, the exception being *Paralipsa gularis*, the stored nut moth.

Two structural characters are employed in identification of adults, namely the form of the labial palpi and the position of vein 8 on the hindwing. The labial palpi are paired 3-segmented appendages visible in front of the head and arising between the eyes from beneath. These structures should not be confused with the much longer thread-like antennae situated above the eyes, or with the legs attached to the underside of the thorax. The basal segment is small and inconspicuous, but the second and third segments display considerable diversity in form and scaling. Some may project horizontally in front of the head and resemble an open or closed beak, or they may be curved upwards or downwards.

[Fig. 3. Larva of *Ephestia kuehniella*.](#)

[Fig. 4. Diagram showing parts of a moth. \(After Hintor and Corbet, 1972\)](#)

Wing venation

Moth wings are thin membranous expansions of the body which are strengthened by hollow veins which carry blood. The cell pattern formed by the vein structure can be utilized as a diagnostic feature. To examine the veins the wings should be removed by cutting the base with a fine scalpel and placing them in xylene. The scales can then be removed by scrubbing the immersed wings very carefully with a fine brush to expose the veins; the structure of veins can then be examined using a

X 20 hand lens.

From the venation of fossil insects and living primitive insects it is possible to construct a hypothetical primitive venational pattern from which the venation of living insects can be derived. This pattern is shown in Fig. 5 and the name and convex or concave nature of each vein is indicated by positive and negative signs).

Atrophy of certain vein stems in both fore and hindwings of the Lepidoptera, have resulted in an approximately triangular area called the discal cell and the veins closing it are known as the discoidals. Normally, the forewing has 10 veins arising from the discal cell or originate from veins arising from the cell, while 6 veins originate in this manner on the hindwing, but there are many variations with some veins absent or fused with one another.

In the Tineidae, vein 8 of the hindwing is free throughout its length and is situated near the costar margin of the cell, while in the Pyralidae, it often is united with vein 7 beyond the discal cell but separates again before reaching the costa (see figure).

Genitalia

Adult moths which are very similar in appearance or which have had their wings or wing patterns damaged may be positively identified by examination of their genitalia. This may be done by removing the abdomen soaking it in a ten percent aqueous solution of potassium hydroxide for twenty-four hours and then carefully dissecting the genitalia from it. However, it is emphasized that an examination of this sort is almost impossible with a hand lens and genitalia obtained by

this means are best examined by mounting them on a glass slide in a suitable medium for microscopic study (Winks, 1975).

BIOLOGY OF STORED PRODUCTS LEPIDOPTERA

GRAIN MOTHS

This term includes only those moths capable of destroying sound, unbroken grain kernels when stored at or below the safe moisture level. They are not as abundant in terms of the number of species involved, as compared to the flour moths or secondary pests, which are principally pests in broken or damaged kernels or in milled products.

FAMILY GELECHIIDAE

Sitotroga cerealella (oliv.) - The "Angoumois Grain Moth"

This moth is the only species in this family which is associated with stored products.

Description

A small buff or yellow-brown moth with a wing span of approximately 12 mm; the wings are narrow with wide fringes and the hindwings narrow sharply to points at the apices. When the moth is at rest, the wings are folded in a sloping manner over the abdomen, the antennae point backwards and are slightly raised above the wings, while the labial palpi are raised upwards like two horns (Fig. 7)

Biology

This moth is capable of attacking grains in the field and in store. The eggs are laid on the surface of the grain and after hatching the larvae bore into the grain and complete their development within the endosperm. The larvae are always inside the grain kernel except in the first instar. Under optimum conditions the life cycle is completed in approximately four to five weeks. Each female lays an average of 40 eggs deposited either singly or in clusters, but individual moths have been known to lay as many as 400 eggs. The eggs which are white when first laid, soon change to a reddish colour and are laid on wheat heads, exposed tips of corn ears in the field. or in stored grain. After hatching, each larva crawls to a kernel of grain and often spins a small entrance cocoon to assist in boring into the hard kernel. After entering the grain, it feeds on the endosperm or the germ until fully developed. It then channels out a groove to the outside of the seed and makes a weakly fastened flap over the exit hole. The last larval instar then spins a silken cocoon in preparation for the transformation into a reddish brown pupa. The adult moth emerges after pushing open the flap prepared by the larva, since the adult is incapable of chewing its way out. The larval period lasts from 2 to 3 weeks, and pupates in 5 to 6 days.

Fig. 5. Hypothetical primitive pattern of wing venation. (Based on various authors). C, costa; SC, subcosta; R, radius; Rs, radial sector; M, media; MA, anterior media; MP, posterior media; Cu, cubitus; A, anal.

Fig. 6. *Ephestia elutella*: wings de-scaled to show venation. (Numerical system of notation after Hinton and Corbett, 1972)

[Fig. 7. Neuration of *S. cerealella* \(Oliv.\), ♀:](#)

[Fig. 8. *Sitotroga cerealella* \(Olivier\), the "Angoumois grain moth".](#)

[Fig. 9. *Sitotroga cerealella* \(Oliv.\), the "Angoumois grain moth". Dorsal view of resting adult.](#)

Economic Importance

The advent of combine-harvesting has restricted the distribution of this pest; infestation often occurred in the field when harvested wheat was stacked to dry, but combining now reduces the possibility of this type of infestation occurring. Field infestation is now restricted to sorghum and maize (corn) when these grains are harvested without threshing or shelling. In bulk grain, infestations are restricted to the surface layers only.

COSMOPTERYGIDAE

This family contains 2 species associated with stored products, *Sathrobrotia rileyi* (Walsingham) the "pink corn worm" or "pink scavenger caterpillar", which is a pest of cotton and corn in the southern United States causing considerable damage in both the field and storage, and *S. badia* Hodges, which is of little economic importance.

Sathrobrotia rileyi (Wals.) "pink scavenger caterpillar" slightly smaller than the Angoumois grain moth, the forewings banded and mottled with yellow, reddish brown and black. The pale grey hindwings are very narrow and edged with long fringes.

A reliable indication of the presence of this pest is a large amount of loosely webbed frass that fills the interstices between the kernels or the cavities of partially consumed kernels. The white eggs are normally laid singly but sometimes in small groups of two or three eggs. The pinkish larva feeds on the seed, husk or cob with equal voracity and is capable of inflicting serious damage to corn if allowed to mature in the field.

TINEIDAE "clothes moths"

Many of the species of this family feed on animal products such as wool, fur, skins and rarely infest stored cereals or seeds. However, they are sometimes encountered in established infestation where the larvae are scavenging on dead bodies of other insects. The major species likely to be identified in these circumstances are:

Trichophaga tapetzella (Linnaeus). The white tip clothes moth.

Tineola bisselliella (Hummel). The common clothes moth.

Tinea pallescentella Stainton. The large pale clothes moth.

Tinea pellionella (Linnaeus). The case-bearing clothes moth.

Niditinea fuscipunctella (Haworth). The brown-dotted clothes moth.

Some species, however, will feed on dried material of plant origin.

i) *Nemapogon granella* (Linnaeus) "European Grain moth"

A small moth (wingspan 10-14 mm) approximately the same size as *S. cerealella* but is creamy white and heavily mottled with brown. It infest all kinds of grain, both in the field and in storage. The larva feeds on the grain and webs the kernels together but is generally considered a minor pest of grain and spillage in temperate climates, and is found through the northern states of the US, but is less abundant than *S. cerealella*. It is more important on rye, less important on wheat and of no economic importance on barley and oats.

ii) *Setomorpha rutella* Zeller. The tropical tobacco moth

This species infests seeds, tobacco, stored cereals, flour, and dried vegetable materials in tropical and subtropical regions. It is of little economic significance except in stored tobacco.

GALLERIIDAE

There are 9 species of the family associated with stored products, while only *Corcyra cephalonica* (Staint.) is of economic significance on stored grain and seeds. *Galleriinae* is considered to be a subfamily of *Pyralidae* by Hinton (1943), Corbett and Tams (1943) and Hinton and Corbett (1972), which also includes the *Phycitinae* as well. Wing venation of the forewing separates the two subfamilies according to Corbett and Tams (1943).

Corcyra cephalonica (Stainton). The rice moth.

A medium-sized moth, with a wing span of approximately 25 mm, the wings of which are uniformly pale buff brown or grey with the veins slightly darkened. It has a tuft or crest of scales on the head, the wings are folded in a sloping manner over the abdomen with the antennae lying straight on the wings. Labial palpi are close together and straight giving the appearance of a closed beak. It is a cosmopolitan species more important in tropical areas and attacks a wide range of commodities including whole grain particularly rice, milled and processed cereal products, oilseeds, and nuts, and occasionally dried fruits. Larvae produce dense webbings, and when feeding on grain form silken tubes, which bind the grain kernels together into lumps.

The moths live for 1 to 2 weeks, in which time the female lays between 90 and 200 eggs, laid indiscriminately and loosely in the grain mass. The incubation period lasts 4 to 6 days, the larval period 3 to 4 weeks and the pupal stage, approximately 10 days; while the total life cycle requires 97 days at 24C, the optimum being between 28C-32C.

Although it is frequently assumed to be predominantly a pest of rice, it is found that most other cereal grains and occasionally oilseeds and pulses are attacked. It is essential that the food source contains either linoleic acid or biotin, and the species flourishes on a low protein intake diet, but is limited by low levels of carbohydrates. Thus cereals are considered the best diet and at 30C and 70% RH, larval development takes 35.5, 39, 50 and 78 days on pearl millet, broken wheat, chick peas and black gram respectively. The number of larval instars is variable, generally seven to eight, with the males having one fewer than the females, but both sexes taking about the same time to develop.

The females mate only once during a 1-2 day period after emergence, and if copulation has not

taken place, the females lay unfertilized eggs and are disinclined to mate. Males mate several times (up to four times in a period of nine days) thus at a normal 1:1 sex ratio, many males may be deprived of the opportunity to copulate.

C. cephalonica is a cosmopolitan pest of mills in tropical climates where it fills the niche occupied by *E. kuehniella* in more temperate climates. The contamination of the food caused by webbing activity of the larvae, larval galleries, cocoons and frass may prove more economically significant in terms of reconditioning of the grain to make it saleable than the actual weight loss incurred through larval feeding.

Paralipsa gularis (Zell.). The "stored nut moth"

The forewings are greyish buff marked in the female with a distinct black patch or spot, while in the males, the spot is smaller and has a zig-zag, reddishyellow streak across the hindportion of the forewing. The wingspan is approximately 15-20 mm and is particularly attracted to walnuts, almonds, hazelnuts and ground nuts, but attacks a wide range of other commodities.

FLOUR MOTHS

Among the flour moths are some of the commonest and most serious of grain pests. They are designated as secondary grain pests because they seldom attack sound undamaged kernels and prefer broken grains, grains previously damaged by the primary grain feeders, and more especially milled and processed cereal products. The Indian meal moth, *Plodia interpunctella* (Hubner) and the meal snout moth *Pyralis farinalis* Linnaeus may under favourable conditions become

established in whole grain causing damage by consuming the germ. This is especially true if the grain is stored at an unacceptably high moisture content.

PYRALIDAE

The 9 species of this family associated with stored products are widely distributed and are general feeders in the larval stages upon cereals and cereal products. Infestations are indicative of poor storage hygiene associated with damp or "wet" storage conditions. *Pyralis* and *Aglossa* are the most important genera.

i) *Pyralis farinalis* (Linnaeus). The moth meal

A cosmopolitan species, more plentiful in temperate than tropical areas. The wing span is approximately 25 mm, and the forewings are characteristically patterned - overall light brown with basal and apical purple brown areas demarked by wavy transverse white lines. It breeds mainly in grain spillage, moist flour, bran, peanuts and hay that have been held for some time in damp conditions. *Pyralis manihotalis* (Guenee) is a cosmopolitan species that appears to take the place of *P. farinalis* throughout tropical regions.

The female lives for about one week and lays 200400 eggs (average 250). The mealmoth larvae is white and when fully grown some 25 mm long. The larva shows a contrast between the black of the head capsule and thoracic shield and the white of the remainder of the body, which is often tinged with orange towards each end. The larvae spin peculiar tubes of silk that contain mixed particles of food material. These- tubes are extremely resilient, and the larvae rest feed from the

openings at the ends. When fully developed, the larvae migrate from the tubes, spin silken cocoons which are also camouflaged by adhering food particles.

Economic Importance

Although common, this moth is not a serious pest of stored grain as it prefers damp conditions and commonly breeds on out-of-condition grain or grain spillage. The presence of this pest is usually indicative of poor hygiene and damp local conditions. It has, however, been recorded as attacking sound grains which have a relatively high moisture content.

Mealmoth larvae often attract much attention due to their capacity to "web-up" and bind together seeds of various kinds, as well as their capacity to cut through hessian sacks causing considerable damage and spillage, especially when present in large numbers.

ii) *Aglossa caprealis* (Hubner). The murky meal caterpillar.

A medium-sized moth, with a wing span of approximately 25 mm. The basal quarter of the forewing is deep ferruginous brown, outwardly defined by a narrow highly zigsagged pale pinkish buff antemedial line, sloping outwards from costa to dorsum; the pale, narrow, sigsagged postmedial line touching, or almost touching the termen between veins 2 and 6. Hindwing is greyish white and unmarked. It is cosmopolitan, and found associated with damp grain spillage, clover, and lucerne hay, but is of minor importance.

PHYCITIDAE

This family contains several notable species. These are *Ephestia cautella* (Walk.), *Ephestia elutella* (Hbn.), *Ephestia kuehniella* (Zell.), and *Plodia interpunctella* (Hbn.). They infest a wide range of commodities from cereals and tobacco, to dried fruits and nuts. Diapause has been recorded from species of the family. It is emphasised that it is usually necessary to examine genitalia to confirm identifications of species, particularly within the genus *Ephestia*.

i) *Plodia interpunctella* (Hubner). The Indian meal moth.

A small cosmopolitan species with a wing span of approximately 20 mm, the forewings of which are characteristically red-brown with a copper lustre, the inner third being creamy white. When the adult moths are at rest, the wings are folded touching each other closely along the posterior margin and lie flat on the abdomen. The antennae lay crossed over the wings, and the labial palpi are slightly separated and project straight forward giving an appearance of an open beak (see Fig. 13). It is a serious pest of stored grains, oilseeds, and of processed and milled products, nuts, dried fruits, and chocolate. Females lay approximately 200 eggs either singly or in clusters and the larvae burrow into the food mass where they remain till completion of larval growth.

When fully grown, the larva is approximately 12-14 mm long, and is sometimes dirty white, varying to greenish and pinkish hues. The larva spin a silken cocoon and transform into a light brown pupa, as well as creating webbed sheets in the grain mass. The larval period is about 3 weeks, but is known to live under adverse conditions for up to 2 years. The pupal stage lasts approximately 2 weeks. Pupation occurs in crevices in the fabric of the surrounding store, similar to *Ephestia* spp. Larvae are moderately resistant to low temperatures and overwinter in this stage; the life cycle in summer is 4 to 6 weeks. This species does not reach the high population levels in grain often seen

in other members of the family and infestations often tend to be localised.

P. interpunctella gets its common name from the United States where it was recorded as a pest of "Indian corn" or maize. It is perhaps of S. American origin, but is now cosmopolitan; and attracts more attention as a pest of dried fruit particularly raisins, currants and sultanas.

ii) *Ephestia elutella* (Hubner). The tobacco moth.

The adult is a small, grey or brownish-gray moth with a span of approximately 16 mm. The moth is similar in appearance to *E. cautella*, with markings on the top side of the forewings very similar, while *E. elutella* the outer transverse band is pale, well defined, wavy and bordered on each side by a narrow dark line. It is a cosmopolitan species which is most prevalent in temperate regions, attacking almost all grains, ground cereal products, tobacco and dried fruit. It is common in tropical and subtropical areas where its occurrence is restricted usually to stored tobacco. Females lay approximately 200 eggs which are deposited singly or in small groups on the food material. Winter is passed in the larval stage, in loose cocoons in cracks or crevices. The life cycle maybe completed in 5 to 6 weeks, while the final instar larva is responsible for enveloping commodities and surrounding structures in dense webbing during their migration.

Control of this moth is often difficult because the mature larvae finds pupation sites in the surrounding storage structure which renders it inaccessible to normal applications of contact insecticides. When feeding on raw cereals, the larvae will preferentially attack the germ.

Ephestia cautella (Walker). "the tropical warehouse moth".

The adult moth is cosmopolitan, grey in colour with a wing span of approximately 20 mm. The upper sides of the forewings are marked with two transverse bands, the inner one being straight, dark and continuous with a pale band along its inner edge.

Fig. 10. Neuration of *S. rutella* Zell., (after Diakonoff). (Note: Vein 8 of hindwing completed separated from vein 7, a distinguishing feature between the Tencid and Pyralid venation)

Fig. 11. Neuration of *Corcyra cephalonica* (Staint.),

Fig. 12. Neuration of *Pyralis farinalis* (L.)

Fig. 13. *Plodia interpunctella*, "the Indian meal moth". Dorsal view of resting adult.

Biology

As with other insect pests, the time required for this moth to complete its life cycle from the egg to the emergence of the adult varies greatly according to environmental conditions. Under the optimum conditions of 30C-32C and 70%-80% relative humidity the cycle is completed in approximately thirty days and the egg, larval and pupal stages account for 10%, 70% and 20 % of this time respectively. Development does not take place above 36C or below about 14C and at 15.5C the life cycle is extended to approximately 145 days.

The female moth lays about 250 eggs loosely in the infested commodity. Pupation takes place in small aggregates of grain held together with webbing.

Economic Importance

This is the most important moth pest of stored grains in Australia, being particularly active in the warmer states. The larvae will feed on a wide range of commodities including grains, grain products, dried fruits, nuts, cacao beans and chocolate. They can attack whole sound grains but prefer the seed coat to be slightly damaged; grain which is combine-harvested is usually damaged to a sufficient degree to render it suitable for infestation by this pest.

As with some of the other species in this family, *E. cause/la* tends, by preference, to attack the grain germ, moving from grain to grain consuming these parts; damage to the commodity is therefore much greater than the actual weight of grain consumed due to this high spoilage factor. Contamination of the infested commodity also occurs from the large quantity of webbing which is spun over its surface; in heavy moth infestations of stored grain this webbing looks like a fine white film covering the whole surface of the bagged stack or bulk.

E. cautella is also a serious pest in tropical flour mills where it infests ground cereal products. Webbing produced by the moth larvae is often responsible for damage to milling and conveying equipment.

Ephestia calidella (Guenee), and *Ephestia figuliella* Gregson, are of minor significance infesting dried fruits and meal in temperate regions.

Ephestia kuehniella (Zeller)-The Mediterranean Flour Moth.

Description

E. kvehniella which is a cosmopolitan species native to Europe, has a wing span of approximately 25 mm and is slightly larger than the two members of this genus which have just been described. The top side of the forewing is slate-gray in colour and the inner transverse band is almost z-shaped and consists of dark streaks and spots; it does not have a pale band along its inner edge.

Biology

The complete life cycle may require as little as six to seven weeks under optimum conditions. The female moth lays between 50 and 300 eggs during a period of five to seven days and there is no egg laying below 12C or above 34C. In temperate climates there is only one generation per year in unheated stores and four to six generations in heated flour mills. The life cycle may be completed in 8 to 9 weeks under optimum conditions.

Economic Importance

The larvae of this moth are prolific spinners of silk and it is this characteristic which has made them a serious pest in flour mills.

The copious webbing which they produce blocks milling machinery, conveying ducts and equipment, and generally interferes with production; large clumps of webbed flour also provide harbourage for other insect pests. Under experimental conditions the moth will breed in ground cereal food with a moisture content as low as 1%-2%; it very rarely attacks stored whole grains.

This pest was once widespread in Australian flour mills but its incidence is now restricted due to the introduction of more efficient control methods particularly fumigation specifically aimed at curtailing its development.

In tropical climates *E. cautella* (Walk) usually replaces *E. kuchniella* as the major Phycitid pest of flour mills.

General Notes on the Biology of the Phycitids

The Phycitids are undoubtedly the most important members of the stored product Lepidoptera associated with milling premises and grain storages. Consequently, efforts have been directed towards a better understanding of the behaviour of these moths with a view to more efficient control either from new techniques or more effective usage of existing control measures. The most vulnerable stage is the adult, but little is gained by controlling adults unless they are prevented from producing viable eggs, since the larvae is the most damaging trophic phase.

Although most females mate once only, up to 7 matings have been reported for some individuals. During mating activity, females of Phycitid moths emit sex pheromones to attract the males, and males emit pheromones to stimulate copulation. The nature of these pheromones is complex and is species specific although individual components may be produced by a number of species.

The fecundity of females of *E. cautella* has been found to be related with their weight there appears to be no correlation between longevity and weight. The fertility of males of *E. cautella* persists for up to 8 matings with subsequent matings resulting in decreased egg fertility and

fecundity of females. Females of *E. cautella* exposed to a lethal dose of insecticide discharge their eggs by involuntary abdominal peristalsis prior to death. Eggs have been found to be equally viable to those discharged normally, and this oviposition response to stress, has enormous implications in control using insecticides.

First instar larvae commence feeding immediately, forming small galleries of silk and frass. In large populations larvae may enter a migratory phase which aggravates the problems associated with silk production. In *E. keahniella*, larvae have been observed to secrete a droplet of fluid when encountering other larvae in dense populations causing increased wandering in the larvae, increased developmental period, reduced fecundity in the adult and influence the choice of egg-laying sites. These secretions are therefore density dependent and density regulating.

Mature larvae normally leave the food medium in which they develop and pupate within a cocoon of webbing in adjoining cracks or crevices. In bulk grain, the cocoon is constructed within several kernels of grain causing familiar grain aggregations.

Diapause has been observed to occur in a number of the Phycitids but is more a feature of populations in colder climates. In *E. elutella* and *P. interpunctella*, late instar larvae enter diapause in response to shortened photoperiod or to crowding. Again diapausing larvae increase the problems of obtaining sufficient control by insecticides due to their higher tolerance.

OECOPHORIDAE

The presence of species from this family, 3 of which are associated with stored products, is

indicative of poor hygiene and damp conditions. The hind wings are acutely narrowed at the tips, but less so than members of Gelechiidae, and generally have a larger wingspan (12-22 mm).

i) *Hofmannophila pseudospretella* (Stainton). The brown house moth.

A small to medium-sized moth (15-22 mm wingspan), the forewings of which are dark brown or bronze-brown with a number of blackish brown flecks. The larvae are omnivorous scavengers feeding on spillage, bagged flour, seeds, dried fruit, and clothes. Larvae prefer high humidities (80% R.H.) for development, and the life cycle in Summer is completed in 8-12 weeks, but varies between 152-266 days at 24C and 90% R.H. to 192-440 days at 20C and 90% R.H. Although the developing larvae require high humidities, the eggs and resting (diapause) larvae are tolerant to desiccation. Under most conditions, *H. pseudospretella* caterpillars enter diapause which may be prolonged and in temperate climates is univoltine (i.e., only one generation per year). According to studies by Woodroffe, the only biological check on the increase of *Hofmannophila* is the predacious prostigmatid mite, *Cheyletus cruditus*. Coombs and Woodroffe have also reported an interesting interaction under high population densities of *H. pseudospretella* in bulk wheat, where numerous mutilated beetles were found trapped in the copious amount of webbing on the surface. Presumably the caterpillars had been responsible for biting off the appendages.

ii) *Endrosis sarcitrella* (Linnaeus). The whiteshouldered house moth.

This moth has similar habits and is often found with *H. pseudospretella*. It is responsible for more damage to stored products particularly grain in bag stacks, and is less a scavenger than the previous species.

The head and "shoulders" (prothorax) of this moth are conspicuously white; the upper side of the forewing is shining buff or grayish-white and speckled with dark brown and blackish spots. The wing span is approximately 17 mm and the hindwings are slightly narrowed towards the tip but not nearly as noticeably as those of *S. cerealella*.

Biology

The adult female of this species lays between 50 to 230 eggs and lives between five to nine days. Complete development from eggs to adult requires 235 days approximately at 10C and 62 days at 24C and 90% R.H. High humidity levels are required for completion of the life cycle. It is multivoltine producing several generations in a year.

This moth is of sporadic economic importance, while both *H. pseudospretella* and *E. sarcitrella* are often associated with birds' nests which provide harbourage for these insects in otherwise clean warehouse.

[Contents](#) - [◀ Previous](#) - [Next ▶](#)

[Home](#) [''''''''''>](#) [ar.cn.de.en.es.fr.id.it.ph.po.ru.sw](#)

Section 6 - Inspection and detection methods for storage insect pests

Inspection procedures for grain handling facilities and methods for detecting stored grain insects

[Contents](#) - [◀ Previous](#) - [Next ▶](#)

By R. L Semple

I. INTRODUCTION

The inspection of storage facilities and the stored food commodities that they contain, is of paramount importance in preserving grain for human consumption. Stored grain is not an inert substance, but a respiring, living organic entity. It deteriorates during storage, either quantitatively by the amount of dry matter lost (DML), or qualitatively by discoloration, mould contamination, sprouting and caking, etc., promulgated by the activity of microorganisms, invertebrate (insects and arachnids) and vertebrate pests (rodents and birds). Further weight losses are incurred during the handling, transporting and processing of stored food commodities.

Accurate information on all these forms of loss can only be obtained by thorough and regular inspection and sampling procedures. This is imperative in formulating sound management decisions involving the adoption of any remedial action against these biodeteriorating agents or the disposal of grain with due cognizance to the condition of both the commodity or the storage facility. Regular inspection helps maintain a storage environment which is conducive to good grain quality by monitoring any significant buildup in pest populations, grain temperature, moisture migration, spillage and grain residues.

It further promotes or encourages general maintenance of the storage structure and surroundings in a sound condition thereby denying access to pests as far as practicable. It also removes potential sites for infestation developing in cracks and crevices within the storage fabric and the removal of disused or obsolete machinery, used and often infested bags and dunnage.

Connell (1975) stated that, "Inspection and sampling work involves the exercises of judgement based principally on experience and an intimate knowledge of the nature, properties and physical behavior of grain. Inspection, although considered of major importance, is more often than not underestimated or in fact neglected, because it is time consuming and precise methods cannot be adequately specified. General principles however are the same whether the object of the inspection for pests is for quarantine, grading or quality determination, or a scientific study; or whether it is performed in a grain warehouse, mill or processing plant, farm or ship. Information on the following is required:

- the presence of any pests
- if so, what species, population densities, location
- damage inflicted prior to the inspection
- potential for further development of these populations (their growth potential) or,
- whether the populations will eventually die out if conditions persist or they remain uncontrolled (seriousness)
- duration at safe storage of commodities or,
- the need for treatment or disposal. (urgency)

Subjective estimates must be critically avoided. While a numerical estimate of population numbers

is seldom a practical approach, some quantitative estimate based on an arbitrary, internationally accepted and comparative scale must be pursued.

It will be expanded in a later section, but it is important to realize that after adopting a thorough inspection programme, the inability to find any insect infestation does not automatically preclude their absence, and especially in the humid tropics of Southeast Asia, the presence of low level populations now (i.e. 1 insect per 100 kg), will result in damaging population densities developing within four months. Continuous monitoring is therefore the essence of preventing serious deterioration of grain in storage.

II. THE OBJECTIVES

The objective of any sampling and inspection programme is to formulate the basis for future planning and action. Armed with precise and complete records of the history of stocks on hand, their present condition and presence of pests, the initiation of a feasible and economic control programme can be undertaken. Secondly, it forms an important base for determining its monetary value and whether the condition and quality of grain will satisfy the requirements of any potential buyer. Decisions based on a sample of grain that does not statistically reflect the overall condition of the bulk will result in erroneous conclusions being drawn concerning its fitness for human consumption. This problem is further compounded by the multiplicity of acceptance in quality standards; the nil tolerance for live insects in international export wheat as well as white flour in Western countries where it is condemned both in the eyes of the consumer and legally. Conversely, quite heavily infested grain or grain products maybe considered the norm in some developing countries depending on the economic status of the community.

2.1 Problems in systems assessment

It is considered a prerequisite that the inspector has a working knowledge of the insect species that are commonly occurring in the produce with which he is dealing. Recognition of the taxonomic and morphological differences becomes increasingly important in establishing the status on endemic pests as well as identifying the introduction of any exotic species. Any species that is subject to quarantine regulations must be accurately identified by an appropriate authority. It is also advantageous for the inspector to have some knowledge on the biology, ecology and behaviour of various species which he may encounter, the differences between primary, secondary, field and associated pests, as well as beneficial parasites and predators.

Other practical difficulties have been elucidated by Howe (1966), and can be summarized as:

- inaccessibility due to bad stacking or to inadequately designed buildings and machinery
- sheer bulk of produce and size of premises
- variety of ecological niches and habitats
- insect behaviour: detection of latent or hidden infestation remaining inside the stack or bulk
- non-random or uneven distribution or insect populations

Problems associated with inspection techniques all focus on the desirability of investigating locales where insects are likely to abound, or to search for signs of the presence of insects rather than the insects themselves, particularly by recording rises in temperature (hotshots) with the use of thermocouples or thermistors inserted into bulk or built-in to bagged stacks.

III. INSPECTION AND SAMPLING TECHNIQUES

Normal visual inspection of storage facilities and stored grain is subjective in nature, and therefore any results can only be recorded in a descriptive way. So long as the methodology remains uniform, direct comparison and therefore useful appraisals of different situations can be used. For more accurate and quantitative measurements it becomes necessary to sample the grain whereby the results can be interpreted by physical or chemical methods.

3.1 Qualitative inspection methods

Inspection is carried out with the objective of assessing the degree of insect infestation, although all forms of deterioration should be subsequently identified. Freeman (1948) first developed the need for standardization of estimates of infestation into defined categories for all species and all products, and in some cases his estimate have been modified for the tropics where the degrees of infestation are somewhat higher than in Britain (Hall, 1953).

Recording the results of a visual inspection have further been simplified by a useful shorthand notation as described by Ashman (1966; 1970) and has been widely adopted by inspectors. (Table 1).

For example,

Living adults (A) Living larvae (L) Living pupae(P) Dead adults (a) Dead larvae (l) Dead pupae (p)

General inspection may involve checking grain held at storage facilities for any obvious infestation without drawing grain samples or it may involve looking for sources of residual infestation within the fabric of the storage structure or the immediate surrounds.

3.1.1 General inspection of grain in store.

The following developed by the Ministry of Agriculture, Fisheries and Food Inspectorate, Britain, have international sanction and are therefore recommended for any general commodity inspection.

3.1.2 Inspection of storage facilities and handling equipment.

The following categories apply to general inspections of the structural condition of the warehouse, silo or mill, or any handling and conveyance equipment that may offer suitable localities for endemic or residual insect infestations.

Table 1: Notation used for recording the results of a visual inspection or general inspection without taking samples and commodity inspection after drawing samples (After Ashnan. 1970)

<i>1. Stack inspection</i>	
C = Clear or none	No insects discovered in the course of a prolonged search.
F = Few or light	Small numbers of insects occurring infrequently or irregularly.

MN = Moderate numbers	Insects obvious, encountered regularly, sometimes forming small populations or aggregations.
LN - Large numbers	Insects immediately obvious where large numbers are actively crawling over the entire surface of the commodity, i.e. stack or bulk.
VLN = Very large numbers	Insects extremely active and numerous that they are audibly present within the confines of the bulk or stack. Live insects or exuviae (cast skins) forming a continuous carpet around the perimeter of the stack or bulk.
<i>2. Storage inspector</i>	
C = Clear or none	No obvious insects or populations signs.
VF = Moderate numbers	Insects occurring regularly and frequently, often forming populations but not obvious enough to be immediately noticeable.
LN = Large numbers	Insects immediately obvious on commencement of inspection crawling actively on walls and in other situations.
VLN = Very large numbers	Insects present in very large numbers, often forming dense populations on numerous surfaces as well as in any grain residues present on the floor, in mill augers, used sacks, dis-used machinery, bins, etc.
<i>3. Sampling inspector</i>	

C = Clear or none	No insects obvious on stacks or sacks or any of the samples. (Require protection from cross-infestation and regular inspection).
VL = Very light	Insects not obvious on sacks, or in sample of produce before sieving. < 20 insects per 90 kg sieved sample (Requires disinfestation in near future)
L = Light numbers	Between 20-300 insects per 90 kg sieve sample.
M = Moderate numbers	Between 50-300 insects per 90 kg sieved sample.
H = Heavy numbers	Between 300-1500 insects per 80 kg sieved sample.
VH = Very heavy numbers	> 1500 insects per 90 kg sieved sample

In describing the species of insects present, it becomes necessary to employ the scientific generic and specific names, since the common names associated with insects sometimes vary between different countries. If the inspector is not absolutely positive of completely identifying species that are found, the uncertainty should be specified in the report, with every effort made to have the specimens in question identified by a specialist taxonomist.

The location where the species of insects were collected during the inspection should also be established in the report. They may not necessarily have complete distribution within the warehouse unless very heavy infestations are involved and this information will become an

important consideration when embarking on any follow-up control procedures. Individual inspectors may develop their own shorthand notations or use floor plan sketches to supplement their report.

For example, the findings of an inspection maybe recorded as follows:

Sitophilus oryzae	VLN (a)
Latheticus oryzae	NM (A,L)
Trogoderma granarium	F (a), MN (L)
Tribolium spp	VF (1), VLN(A,a)

The main activities associated with a worth while inspection programme are making the effort for a prolonged search, observation and accurate recording of results. There exists no satisfactory alternative to be becoming actively involved either by getting into the grain, on to the stock, into the premises around handling equipment and physically searching for live insects, or the obvious signs for their presence. Because of the time-consuming nature of an inspection, the initial phase should

concentrate on observing places where insects if present are likely to congregate. However, there are certain practices that maybe employed to make the job a little easier, most of which aim at stimulating the insects to come to you rather than the reverse.

3.1.3 Additional aids for visual inspection. During an inspection, a few bags should be opened at

random and the folds of sacking and bag corners examined; *Trogoderma granarium* larvae and *Tribolium castaneum* adults are frequently found this way. Some bags should be at least lifted and set aside and the exposed surfaces of neighbouring bags quickly examined for adults and more carefully for larvae and pupae; such as for *Ephestia cautella*.

For the detection of light infestation, a more detailed examination is required. The following techniques may prove rewarding.

(i) agitation of bags:

This is effective for low population densities of sitophilus granaries, *S. orgzae* and *S. zeamais* which will often walk out of sacks after they have been sufficiently disturbed. A long stick maybe drawn over surfaces of vertical stacks or they can be hit to activate small numbers of adult moths which are therefore more readily observed.

(ii) the feel of grain in bulk:

Walking across the surface of bulk grain with bare feet may prove an excellent guide to its general condition. If it feels cool and free flowing chances are there is no cause for immediate concern. However, if a hotspot exists, this can be exemplified by solid caked patches indicative of high dust content and moisture migration with subsequent rises in temperature.

(iii) traps:

Various traps have been designed to exploit the activities of many species of insects. Tube traps, consisting of a smooth inner surface and rough external surface (approximately 7.5 cm long x 2.5 cm diameter) can be inserted into bags to catch species such as *Tribolium castaneum* which are unable to escape by climbing the smooth inner surfaces. It is a useful cumulative trap, but is not effective in trapping *Sitophilus* sp. or *Oryzaepilus surinamensis* (plus others) and may become ineffective where the webbing activity of moth larvae is apparent.

An extension of this simple design for trapping insects at varying depths in bulk grain has been developed in Canada (Loschiavo and Atkinson, 1967).

Various forms of home-made, fly paper-type, sticky traps are commonly used to give an indication of the presence of flying insect pests at an early stage of infestation. Attractant traps, such as light taps (incandescent, fluorescent or black light) as well as suction traps, can be helpful in large warehouses where suitable power is available to give reliable early indications of the presence of moths and beetles that fly readily. Traps that employ a sex attractant or pheromone as a lure offers a potential approach not only for estimating the degree of infestation but ultimately as control measure. Various traps have been adequately described by Bailey (1975).

(iv) artificial crevices:

Sections of corrugated cardboard (4 cm wide x 20 cm long) can be placed between bags to attract pupating moth larvae. It can be examined after 24 hours for insects, but it is more applicable if the bulk is only lightly infested in which case examination may be done after 4 to 5 weeks.

Plank traps, consisting of two strips of wood 15 cm wide and 60 cm long, hinged together but held 3 to 4 mm apart, is use ful for Trogoderma granarium larvae and Tribolium castaneum adults. These are inserted in bags and left for several days before withdrawal and examination.

(v) dead insects:

When residual protectants have been applied as a surface treatment and dead insects continue to accumulate, the conclusion might be reached that the treatment has been fully effective. Usually it indicates a source of live insects in the area, or from infested bags, some of which bemaay obscured within the stack. If these insects are removed and the location marked for future reference, any further accumulation of dead insects indicates the need for further action.

(vi) repellents:

Insecticidal formulations that possess a strong repellency action such as pyrethrin or synergised pyrethroids, can be particularly useful in exposing hidden insects in cracks and crevices. A light application will often stimulate insects to crawl onto exposed surfaces before they finally succumb to the insecticide.

(vii) grain temperature and moisture content:

As mentioned earlier, it is offer more rewarding to investigate for the signs of insect presence rather than looking for live insects. Localized rises in grain temperature or moisture within a bagged stack or grain bulk are most important indicators of insect activity.

Dr. R. W. Howe has further discussed how the knowledge of insect pests and the physical behaviour and properties of stored grain should be used to help the inspector. He states, "Most storage insects are inconspicuous and secretive, and as a consequence are difficult to find. Nearly all storage insects are more easily found in dark premises because they are more active in the dark than in the light. They also lay eggs more readily in the dark.

The proportion of insects at or near the surface of produce varies with the insect species and the produce concerned. This is related to the size of the insect and its developmental instars and to the grain size of the product. Packing of stacks, diurnal rhythms, a tendency to stay near boundaries when brought to them by random movement, upward movement stimulated by disturbance, and outward movement stimulated by heating-all tend to bring insects near the surface of bagged stacks where the inspector has some chance of finding them.

Some of the reactions of insects to stimuli also help the inspector. Most prefer the dark; some are thigmotactic and collect in cracks between bags or under rubbish; most seek out wetter spots and many drink; yet others react to temperature gradients. Therefore, the inspector should examine dark places, the conical tufts of sprouting grain under leaks in the roof, the wet surfaces of bags and areas of produce known or thought to be wetter than the rest, and the tops of stacks especially those under metals roofs if *Trogoderma* is likely to be present."

3.2 Quantitative sampling inspections

The aim of drawing random samples of the commodity is to determine the mean value and the variability of the level of infestation or contamination (% discolored kernels) in any given situation.

Ashman (1970) devised a tentative "sequel sampling" procedure, involving spear samples of the commodity taken at random and then examined by sieving (NB: does not account for latent infestation of the immature stages hidden within individual kernels). The procedure involves collecting a number of spear samples from several bags (dependent on the total number of bags in the consignment, the number of which should not be less than the square root of the total number) until a 1 kg sample is obtained, and examined for insects by sieving. Resampling occurs if low numbers are found and may involve a further three consecutive sampling occasions consisting of 3, 9 or 22 kilograms.

Degrees of infestation and need for appropriate action was as follows:

N.B. Population of *Trogoderma granarium* Everts requires control measures at any degree of infestation.

3.2.1 Spear sampling. There are many inherent problems with such a classification and could therefore be open to a variety of interpretations.

- **Different insects preferentially attack different commodities and consequently inflict greater damage (i.o. pest status is variable)**
- **When insects are present in low numbers and are unevenly distributed, sampling spears are likely to give an inadequate assessment of the infestation, either grossly over or underestimated.**
- **The sampling size required to give an accurate assessment of the infestation using sampling spears is often laborious or time-consuming if the consignment is 10 bags then not less than**

10 bags should be sampled at random: (1000 bags require 32 bags to be sampled).

- **The sample taken is rarely a random sample, due to the difficulty of sampling from the central portion of the stack.**
- **Samples taken from individual bags is rarely a random sample or truly representative of the condition within the bag. At least six more samples should be removed from each bag to make up the primary sample, a practice that is rarely adhered to (approximately 1 kg sample).**
- **Sampling spears damage sacking and consequently create potential for more spillage.**

Problems related to spear sampling are further exemplified in Figures 1 and 2 as depicted by Golob (1977). Insects do not distribute themselves randomly or uniformly in any container of grain. They are most often found in pockets associated with dust, broken grain, and foreign materials towards the bottom or in areas of localized heating or wetting. It is very difficult to sample with a grain trieur or spear close to the peripheral margins of the bags, especially at the top and bottom. Therefore, a large population crawling on the bottom could be completely missed. Alternatively, small pockets of insects within the bulk maybe, by chance, included in a spear sample. For example, 6 sitophilus oryzae in a 500 g sample is not equivalent to 1200 individuals in a 100 kg. bag because of nonuniform distribution, and in fact the bag may contain less than 10 individuals altogether.

Typical spears for sampling bagged grains are represented in Figure 3. If used with due recognition of their limitations, they offer a cheap, simple and quick method in obtaining grain from bagged produce. To obtain more representative samples, a sectional probe should be utilized. These are available in sizes suitable for bags, or in larger sizes for probing deeper piles in bins, sheds, transport vehicles, etc. For larger samples in bulk grain, suction samplers consisting of a small

hollow spear, hollow extension rods and suitable end attachments for the connection of an industrial type vacuum cleaner are adequate for drawing samples from up to 10 m below the surface. A more sophisticated version consisting of double construction interlocking aluminum extension tubes connected by flexible hose to an electrically driven vacuum pump is available.

Samples are drawn into a collecting compartment, with the advantage that grain friction can be minimized by the vacuum and the probe inserted to any grain depth. The major disadvantage is its relative cost.

3.2.2 Other direct sampling methods involve either snaking of bags, coning and quartering and sieving.

(i) bag snaking:

A number of bags maybe emptied by pulling the open bag backwards over the floor surface, allowing a small stream of grain to flow out gradually. Most visible insects will be concentrated in the latter portion and will be readily observed at the sides of the band.

(ii) coning and quartering:

It is a simple and cheap method of obtaining highly representative samples (approximately only 10% sampling error and therefore more accurate than spear samples) but suffers from time and capacity constraints. The procedure involves tipping bags into the floor forming a cone, constantly mixing materials from the periphery to the apex of the cone, then spreading it evenly and dividing

into 1/4, 1/8, 1/16, etc. subsamples depending on the volume required.

(iii) sieving:

"Hand held sieves" are particularly useful in assessing the dust content and live insects from small samples. Different-sized mesh openings can be used for different particle size, or a combination of appropriate sizes can be used for mixed commodities varying in particle size.

"Sack sieves" have also been developed to sample an entire sack; the time taken can be between 5-15 minutes. The recovery of insects is dependent on insect species, time of sieving, slope of the oscillating sieve mesh and mesh size, but tests have shown better than 90% recovery of insects and is independent of population density.

Hugh and Simmonds (1978) have also developed a vibratory screen detector applicable to free ranging adult forms. Insects pass through the vibrating screen along with fine materials which accompanies them. Commercially available models with a throughput of 1 kg. min⁻¹ is available from Eriez Magnetics Pty. Ltd., Sydney, Australia, and this equipment has the ability for scaling up if larger throughputs (10 kgs. min⁻¹) are required. Similar flow rates have been developed by Sweco, Inc., Los Angeles, California.

[Fig. 1. Typical spears for sampling bagged grain. A= Closed spear for sampling large particles, such as maize or coffee; B= closed spear for sampling small particles, such as wheat or rice C= open spear.](#)

Fig. 1. Typical spears for sampling bagged grain. A= Closed spear for sampling large particles, such as maize or coffee; B= closed spear for sampling small particles, such as wheat or rice C= open spear. - continue

For greater accuracy and representation, it is advantageous to take larger primary samples and then take subsamples to form a suitable working sample. Various methods (such as coning and quartering mentioned earlier) can be employed, or by using specific apparatus designed for the purpose. These consist of the gravity mechanical types such as the Boerner conical divider, a simplified alternate channel box divider or the motorized centrifugal types such as the Gamet divider. Simplified dividing trays are also available.

TSPC developed the produce flow sampler (PFS) for taking samples from entire bags originally designed for sampling incoming loads from road transport. The time it takes is approximately 20 seconds for a 100 kg. bag, all flowable commodities can be sampled, and because samples are completely random, it is far more accurate than spear sampling.

IV. DETECTING HIDDEN INFESTATIONS

Most of the damage and weight loss caused by insects on grain are inflicted by the primary grain feeders. They are capable of penetrating sound whole kernels of grain and their life cycle is completed entirely within the kernel in which the egg is laid or entered by the first instar larva.

The absence of any live adults of storage insects in grain samples separated by the previous methods, does not necessarily mean the absence of an infestation and consequently many

methods have been devised to identify individual kernels that have become the home of the immature stages of the major insect pests.

4.1 Concentration of infested grains in a sample Any detection method is greatly enhanced if the infested material can be adequately separated from sound grain and hence reduce the number of grains and the amount of dust and broken kernels that have to be examined. It becomes desirable to concentrate the insect-damaged kernels as a preliminary step. For this purpose, flotation in an air stream or liquid maybe employed.

4.1.1 Flotation separation in liquids. The feeding activity of insect larvae progressively reduces the density of the grain, and by immersing the grain sample in a liquid of suitable specific gravity, the infested grains should float and the sound ones sink. At specific gravities between 1.050 and 1.190, the floating layer contains only infested kernels and approximately 50 to 70% of all infested kernels were separated (White, 1956). Absolute separation is therefore unlikely, but the presence of hidden infestations can be estimated quite accurately; while a general indication of the severity of infestation (the degree) will also be obtained.

4.1.2 Flotation separation in the air. A vertical column with a fan which produced a stream of air sufficient to float the grain sample was used by Milner (1953). By progressively increasing the intensity of blowing, it was noted that virtually all insect-damaged kernels were removed in the first two fractions, from which no emergence had occurred. The detection of insect-damaged grain (i.e. those containing exit holes) can then be a relatively quick and efficient operation, and may speed up the exit-hole inspection procedure in commercial samples by a factor of ten or better.

4.1.3 Projection separation in air. By projecting the grains of a sample through the air at an initially constant velocity, they should separate according to their relative density (Bailey, 1975). Air drag and gravity then further separate the grain according to kernel size, shape and the surface texture which also determine the bulk density or test weight of any particular grain example. Infested kernels tend to fall short of sound kernels of the same size and shape.

This separation has enormous practical implication. Wheat that has been designated as heavily infested can be separated into different bins reclaiming as much as 50% of the grain for food purposes rather than condemning the entire consignment for feed purposes.

4.2 Methods available for detection of insect infestation

4.2.1 Physical methods:

(i) **Acoustic detection** (Adams et al., 1953; Bailey and McCabe, 1965). This method provides an immediate answer if active immature forms are present, and has an advantage of not destroying the sample. A microphone, low-noise amplifier, and loud speaker or a cathode ray oscilloscope display tube are required, and to limit extraneous laboratory background interference, the sample and microphone needs to be insulated or alternatively, the grain can be directly linked to a piezo-electric crystal.

Vibrations are noted at several characteristic frequencies, e.g., 200 cycles.sec⁻¹ are associated with movement and dispersal, while frequencies round 1200-1500 cycles.sec⁻¹ are associated with feeding.

This method is potentially valuable for detection of insect activity within silos and other storage facilities, and possibly transport vehicles, provided extraneous noises can be successfully eliminated. The system can be modified for adequate sensitivity under field conditions.

Other limitations of acoustic detection are that quiescent stages (pupae) and eggs cannot be identified. However, a commercial unit has been developed by SASAD, Budapest, Hungary called the Insectofon (R).

Acoustic microscopy. involving the use of high frequency ultrasound (ca. 100 mHz) which had different velocities in tissues of different densities could be developed for insect detection in grain (Bruce and Street, 1978).

(ii) Grain Radiography (Milner et al., 1950 b). This method is appropriate for detecting hidden insects at most stages of development and has been developed and used extensively under commercial applications in the United States.

The use of X-rays, discovered in 1895 by Roentgen, was generally restricted to the examination of high density materials. Claussen and Shehan (1942) developed a process for making beryllium as a window material on X-ray tubes, and enabled the use of radiography with low density materials such as grain.

Several researchers at Kansas State University (Katz et al., 1950; Milner et al., 1950 b, 1952) pioneered radiography in its application to agriculture, and developed a method for detecting hidden infestation that was marketed by the General Electric company known as the "grain

inspection unit". It suffered from being time-consuming (approximately 15 minutes) and was not suitable for routine spection in of grain. Stermer (1972) developed a completely automated X-ray system to inspect grain on kernel-tokernel basis that was more reliable and practical for use. His study also resulted in the development of a procedure using fine-grain "mammography" film which obtained a contrast of about 75%, after only 2.5 minutes exposure. The film is examined by low-power binocular microscope (6x to 30x) with transmitted light, and obtained an efficiency of close to 100% with fully grown larvae and pupae and 80 to 90% with early instar and eggs.

The automated X-ray analyzer requires four major phases.

The automated X-ray analyser suffers from being an expensive piece of laboratory equipment, does not distinguish between live and dead insects and treating with K₂CO₃ as a constrasting agent was a lengthy process.

Simplicity of operator has been enhanced in various films such as self processing (Polaroid (R)) and cassettes for the Hewlett-Packard Faxitron(R) X-ray unit. The problem of interpretation of the radiograph which makes the X-ray techniques so different has prompted the USDA Federal Grain Inspection Services to scrap it altogether as a quick inspection method (Bruce and Street, 1978).

The low-intensity X-ray imaging scope (Lixiscope) developed by NASA, is a fully portable lightweight unit powered by a penlight battery, and may find application for grain inspection.

(iii) Nuclear magnetic resonance (Street, 1971). The application is similar to the X-ray techique but suffers from the same limitations of time constraints, sampling efficiency and relative cost.

(iv) Carbon dioxide evolution. This method gives an accurate measurement of the total metabolic rate of the grain, and therefore cannot be specifically applied to insects. The method requires enclosing a quantity of grain in a gas tight bottle at 35C for 24 hours, then drawing a sample of intergranular air and analyzing it for percent CO₂ evolved. Dry uninfested grain is normally < 0.25%, between 0.3-0.5% suggests a light insect infestation (or a mc > 15%), and if the CO₂ evolution is > 0.5% in 24 hours, the grain is definitely unsuitable for storage without any further treatment (Howe and Oxley, 1944).

A further development requires the detection of respired CO₂ by infrared absorption spectroscopy which is extremely sensitive, and is applicable to all stages except possibly the egg. The procedure requires a chamber containing the grain sample to be purged with a carrier gas, and then sealed during which time the CO₂ evolved by insect activity reacts a concentration sufficient for detection. This takes approximately one minute and is followed by flushing with a carrier gas for 2 minutes, and moves into the infrared detector by a pulse-flow movement. It is capable of picking up slight increases in CO₂ above the natural atmospheric background concentration of 300 ppm and can detect one 4th instar larva of *Sitophilus oryzae* (L) in 350 ppm grams of wheat (Bruce and Street, 1974).

Two commercial prototypes based on their original concept were developed by Technico Instruments Inc., USA, which can process three samples simultaneously, and by Horiba Instrument Inc. of Japan which is a smaller portable unit. The Lawrence Livermore Laboratories of the USA developed a miniaturized atmospheric carbon dioxide detector system (MACDS) which is extremely sensitive and portable (Bruce and Street, 1978).

Other sophisticated detection systems that may find future application in the grains industry involve infrared radiation detection such as Far infrared (FIR) imaging, photoacoustic spectroscopy and thermal imaging with a pyroelectric vidicon.

All these detection systems are based on the principle that all physical bodies with a temperature greater than absolute zero have an infrared radiation spectrum which is a function of the body's absolute temperature and that living metabolizing tissues (such as insects) generally have a higher temperature than its surrounding environment, thus giving off different radiation characteristics.

(v) Breeding out. Grain suspected of being infested maybe incubated thus allowing insects to complete their life cycle. The biggest disadvantage is the time factor since even under the optimum conditions of temperature and moisture content (2630C; 14-16% m.c.), at least 4-6 weeks will be required to breed out the full population of grain weevils and even longer incubation periods will be required for many other storage species.

(vi) Visual examination of exit holes is the simplest method. It has been standardized by the FDA of the United States that > 3 holes per 100 grains was cause for rejection. An experienced observer can also detect the presence of mature, late instar larvae and pupae of weevils in grain by changes in colour transmitted through the seedcoat. This only indicates the presence of an advanced infestation, but neither the degree nor the severity of the infestation.

(vii) Grain dissection will reveal any internal infestation, and gives a valuable indication of the stage of development of an infestation relevant for any impending control method. It is best done under a binocular microscope, and dissected with a sharp scalpel after the grains have been

presoftened by soaking for 2 hours.

(viii) Cracking flotation has long been employed to determine the internal insect content of grains. Insect fragments are released after coarse grinding of the sample. Concentration of insect material is achieved from an alcoholic solution by flotation separation with the addition of mineral solvents. This enables microscopic examination and identification, and provides objective data on the number of insects present as well as an indication about the stages of development within the test sample.

The technique is complicated and laborious, requires the facilities of a grain test laboratory and sufficient skill and experience on the part of the technicians.

(ix) Electrical capacitance and resistance methods have been investigated by Wirtz and Shellenberger (1962) where changes in both these properties occur when insects are present within the grain.

Additionally, temperature testing as mentioned earlier is of extreme value in detecting the presence of insects, as well as the effectiveness of applied control measures (i.e. fumigation) as indicated by a gradual decline in grain temperature compared to the ambient.

4.2.2 Chemical Methods:

(i) Alkali treatment. This method is often referred to as the sodium hydroxide gelatinisation procedure which depends on rendering the seedcoat and endosperm translucent so that the more

advanced stages of immature insects become visible. Tests by Keppel and Harris (1953) have shown that it has too many constraints to be adaptable as a quick and efficient method, although its reliability is unquestionable.

(ii) Egg plug staining. (Milner et al., 1950 a). A soluble fluorescent dye (berberine sulphate) is used to stain the gelatinous plug secreted by female Sitophilus spp. to cover the egg cavity in the grain. Grains are soaked in a dilute solution of 20 ppm followed by rinsing and examining the kernels under ultra-violet light for the greenish-yellow plugs. The degree of internal infestation can be then estimated by the number of egg plugs observed. This method is not particularly accurate, it is time-consuming, gives no indication on the stage of insect development and is only useful for weevil infestation.

(iii) Uric acid test (Pixton, 1961). Measurement of the uric acid content of infested grain will give an indication of the past insect infestations which may have been concealed during processing. The method was found too insensitive to detect present infestations, and was only useful when population densities were high, in which case they were visibly obvious anyway.

(iv) Chemical detection of insect phenols (Pottets and Shellenberger, 1952). A test based on spectrophotometry analysis for the concentration of a hydroxyphenol occurring in insect cuticle, which produces phenolidophenol dyes when chemically treated with 2, 6- dichloroquinone chlorimide was proposed. Bailey (1975) stated that the method showed particular merit based on Food and Drug Administration evaluations, but at that stage required more work to perfect the method.

(v) Ultra-violet light illumination. Ashman (1986) stated that insects or insect fragments when present in finely ground commodities will appear red when stained in crystal violet and illuminated with ultra-violet light. Similar to the uric acid test, it provides useful information of any previous insect infestations in the grain before processing.

(vi) Ninhydrin system for insect detection. Dennis and Decker (1962) described a machine for detecting pre-adult stages of insects in wheat utilizing the chemical ninhydrin. In practice, the grain and any internal insects are crushed between filter paper which has been impregnated with a 0.7% solution of ninhydrin in acetone. If the grains are infested, the amino acids contained in the body fluids of live insects are absorbed by the paper and chemically react with ninhydrin to produce a strong purple colouration. The machine developed was automated, with a sample flow rate of 300 kernels per minute, spot paper was previously heated to approximately 120C. The test paper could be kept as a visual record and retained for more than two years. Unreacted treated paper loses some of its sensitivity with aging, resulting in a slight colour intensity reduction if the paper is used several months after treatment. This machine was limited because of its size (92 x 61 x 40 cm weighing 250 lbs.), and a more versatile smaller prototype was developed at the Tropical Stored Products Centre (TSPC and reported by Ashman et al., (1970).

Compared to the X-ray technique for insect detection in a series of laboratory trials, the AshmanSimon Insect Infestation Detector proved superior in many respects, with the following deserving mention:

1. Cost of the X-ray technique was substantially higher.

2. Both techniques provide permanent records (impregnated paper can be photocopied and therefore retained indefinitely).

3. Determination of infestation was simpler and quicker.

4. The detector will detect 5-14% of eggs and early instar larvae; 40-60% of moderate sized larvae, 80-90% of late instar larvae in small cereal grains such as wheat, while the X-ray technique is incapable of detecting eggs and small larvae and is not significantly better in estimating mature larvae or pupae in grains.

5. Skilled technicians and expensive laboratory equipment are not required.

6. The machine is adapted to both manually cranked and/or electrical operation and is capable of dealing with a wide range of food grain of various particle size by altering the gap between the rollers (interchange of rollers with different diameters).

Some problems that may interfere with the ninhydrin test have been enumerated by Bailey (1975). Moisture content above 16% m.c. will give a faint general reaction, especially if contaminated by storage molds. Other forms of interference listed were 1) kernels damaged by cracking or checking; 2) kernels damaged externally by insect feeding or mechanical injury; 3) previously infested kernels where adult emergence has been completed; 4) kernels containing dead insects; 5) sooty or heat-damaged kernels; 6) kernels with a high free fatty acidity (FFA) value. These interfering reactions tend to produce distinct, sharply outlined spots.

When estimation hidden infestation of *Sitophilus* spp. in maize, individual kernels maybe infested by more than one larva, which produce an overlapping stain recorded as a single spot. The ninhydrin estimate in this case generally underestimated even though the grains have to be initially kibbled, but appears to be predictable and consequently a correction factor could be applied. The Ashman-Simon Insect Infestation Detector was originally designed for field applications to give a more subjective assessment of the infestation rather than a numerical estimate.

V. RECORDING OF RESULTS

Accurate and complete records give essential background information especially when compiled over a period of time. A standardized inspection record sheet should be used so that information is recorded in a systematic and comprehensive manner for convenient analyses. (Appendix III).

Some of the points that need to be considered in compiling a report are:

weather conditions have an important influence on the development of a pest infestation and are less variable in tropical climates where temperature and relative humidity (which are functions of the EMC) approach the optimum requirement of many major insect pests.

- the structures used for storage of produce have an important effect on produce quality and subsequent build-up of populations. Accurate information of these structures, their condition, and potential sources for cross-infestation should be noted.**
- the type of produce, its variety and grade, and observations on insect damage will often**

prove valuable.

- **origin, destination and length in storage of commodities inspected have an important effect on future implementation of control and,**
- **previous applied control measures should be documented as an assessment to their efficiency.**
- **a plan of the storage layout (bagged stacks and their dimensions) will also prove useful.**

SUMMARY

The inspection of stored grain and storage facilities becomes necessary for the following reasons:

(i) To detect the presence or absence of insects and to determine stability of grain for export purposes, milling, etc.

(ii) To detect insects as they may affect grade and price.

(iii) To determine the suitability of grain for further storage.

(iv) Whether control measures will have to be implemented to secure its end usefulness for human consumption.

(v) If control measures have been applied previously, whether their implementation was successful or whether further controls are required.

(vi) In research work, survey and inspection for the detection insects becomes necessary in the

evaluation of biological, ecological and insecticide resistance studies of grain pests.

(vii) To continuously monitor for the possible introduction of pests subject to quarantine regulations (i.e. Khapra beetle).

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Annex

1.1 Grain Purification:

The table above records the results of an experiment where the grain was projected from a moving belt into 16 separate fractions. The original sample was heavily infested (85%) and each fraction was characterized radiographically for internal infestation. Considerable concentration of the original infestation occurred in bins 1-7, which were closest to the point of projection, while grain samples from hoppers 12-16 yielded approximately half (48.9%) of the original sample weight with an infestation level of only 15% ($7.4 \times 100/48.9 = 15.13$). This represents a 6x purification ratio which could then be successfully be treated and sold for human consumption.

1.2 Ninhydrin detector:

The detector, consisting of a hopper (H) leading to a pair of roughsurfaced steel rolls (D and C) through the 'nip' of which passes a continuous strip of specially treated paper (from A). The grain sample under test (G) is crushed by the action of the rolls on to the paper, and the body juices of any infestation present are expressed, forming an easily recognisable purple stain (B) on the paper. An arithmetical count of these marks then gives a clear indication of the level of infestation in the sample passed through the small machine. Two spring scrapers (E and F) keep the paper and exposed roll D free from crushed grain (J).

The detector measures 35.6 cm x 22.9 x 30.5 cm, weighs approximately 9.5 kg and is conveniently shaped with a built-in handle for local carrying by an operator.

Operating procedure is extremely simple and is as follows:

- 1. Select rolls appropriate to the size of the grain under test.**
- 2. Lift off side cover, fit rolls, thread paper and replace cover.**
- 3. Place a 50-9 sample of the grain in the top hopper.**
- 4. Turn operating handle until sample has completely run through the machine; the crushed grain drops out of the base, of the machine.**
- 5. Tear off the the extruded length of paper, and wait for marks to develop (the time lapse can be reduced to a few seconds by applying gentle heat to the paper).**
- 6. Examine paper visually and record number of marks.**

[Fig. 2. Line drawing of the operation \(after Ashman et al, 1970\).](#)

Cereals and pulses-Determination of hidden insect infestation- Part 2: Sampling

Cerales et legumineuses-Determination de l'infestation cache par les insectes-Partie 2: Echantillonnage

UDC 633.1 :635.65:632.7

Descriptors: agricultural products, cereal products, leguminous grains, determination, insects, contamination, sampling.

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Cereals and pulses-Determination of hidden insect infestation Part 2: Sampling

0 Introduction

This International Standard deals with methods for the determination of hidden insect infestation in cereals and pulses.

Part specifies methods of sampling. ISO 6639/3 and ISO 6639/4 describe the reference method and rapid methods for determining hidden insect infestation, respectively, while ISO 6639/1 describes the general principles of the methods.

1 Scope and field of application

This part of ISO 6639 specifies methods of sampling cereals and pulses, in bags or in bulk, for the determination of hidden insect infestation. The methods are applicable as a routine to grain) in

any form of store or vehicle at any level of trade from producer to consumer.

2 References

ISO 950, Cereals-Sampling (as grain).

ISO 951, Pulses in bags-Sampling.

ISO 6639, Cereals and pulses-Determination of hidden insect infestation

ISO 950, Cereals-Sampling (as grain).

ISO 951, Pulses in bags-Sampling.

ISO 6639, Cereals and pulses-Determination of hidden insect infestation

Part 1: General principles.)

Part 3: Reference method.)

Part 4: Rapid methods.)

ISO 6644, Cereals and milled cereal products Automatic sampling by mechanical means.

3 Definitions

For the purpose of this part of ISO 6639, the following definitions apply.

3.1 consignment: The quantity of grain delivered at one time and covered by one set of shipping documents. It may be composed of one or more lots (see the notes to 3.2).

3.2 lot: A stated quantity, to be sampled using a particular sampling plan.

NOTES

1 There is no need to restrict the size of the lot when sampling for hidden insect infestation. A consignment of the same origin and history may be regarded as one lot may be split into several lots for sampling, whichever is the more convenient. If the consignment is received in several barges, railway waggons, lorries, stacks, etc., it is usually more convenient to treat each part as a separate lot for sampling purposes. Any parts of a consignment known to be of different origin and/or history are sampled as separate lots.

2 It should be noted that the definition of lot for the purposes of sampling for determination of hidden insect infestation differs from the definition of lot in International Standards relating to sampling of grain and pulses for the determination of other characteristics.

3.3 increment: A small quantity of grain taken from a single position in the lot.

3.4 bulk sample: The quantity of grain obtained by combining and mixing the increments taken from a specific lot.

3.5 laboratory sample: The quantity of grain removed from the bulk sample, or an increment (see 10.1), intended for examination.

4 General principles

NOTE - Usually there is little or no prior information on the size or distribution of any insect

population that may be present in a lot to be sampled. In these circumstances, it is not possible to adopt a sampling scheme which is soundly based on statistical theory. Therefore, sampling schemes described in this part of ISO 6639 do not necessarily enable insect populations to be measured precisely, but have been designed to give a maximum of information in a practical manner.

4.1 Special care is necessary to ensure that all sampling apparatus is clean and dry before, during, and after the sampling of each lot. Sampling shall be carried out in such a manner as to prevent insects from elsewhere entering the samples, sampling apparatus and sample containers.

4.2 Laboratory samples shall be enclosed in sample bags (5.5) and shall be protected from direct exposure to sunlight, wetting or other extreme environmental conditions. Airtight containers shall not be used for samples as these may cause any insects present to be asphyxiated.

4.3 If related information about the grain, such as moisture content, is required, separate samples should be taken in accordance with ISO 9p>

Samples may be taken at any point from farmer to final destination.

NOTE - If samples are to be taken at different points in the distribution chain, it is useful to establish standardized sampling operations at all points and to collect the sampling data in order to form a more comprehensive picture.

Sampling is most easily carried out as the commodity is moved into and out of the storage

structure or transit vehicle (railway waggons, lorries, containers, ships, lighters, etc.). If a commodity is stored for a long period in bulk or in bags, sampling becomes more difficult but more important. However, owing to the life cycle of the common species of insects responsible for grain infestation and to the need for any infestation to migrate to the areas where samples will be taken, it is, in general, not useful to take samples from commodities that have been stored for less than 3 weeks.

7 Pre-sampling inspection and identification of lots

7.1 The parties concerned shall agree as to what constitutes the lot or lots to be inspected and shall specify the species of insects (live or dead) to be reported on.

[Continue](#)

[Contents](#) - [◀Previous](#) - [Next▶](#)

[Home](#)"" """"> [ar.cn.de.en.es.fr.id.it.ph.po.ru.sw](#)

Continued

[Contents](#) - [◀Previous](#) - [Next▶](#)

NOTES

1 In the case of grain for export, due regard should be paid to any regulations concerning scheduled pests, and tolerances for such pests, in the importing country. Internal trade may also be affected by such regulations.

2 It should be borne in mind that it is possible for a hidden infestation to mature and produce large numbers of free-living adults shortly after a lot has been reported to be free from infestation or only lightly infested. Rapid changes in insect population density or distribution can result from variations in ambient temperature, cross-infestation or some other reason.

7.2 Inspection of bags, buildings, structures and transport shall be carried out before sampling of the commodity. Information recorded during this inspection may help in the assessment of samples. Any free-living insects found in the samples being taken should be collected and forwarded in a separate sample bag to the laboratory for identification.

8 Sampling of bulk grain

8.1 Extracting samples from moving bulks

At flow rates of 100 t/h, or less, the lot to be sampled shall be not greater than 5 000 kg (5 t) or smaller than 1 000 kg (1 t) and the increments should be equivalent to a minimum of 1 kg per 1 000 kg. Higher rates of flow may require the designation of larger lot sizes, to allow the sampling equipment to cope, but the size of the increments should be proportionately the same. An automatic sampling device or Pelican scoop (see 5.1) shall be used for collecting samples from free-falling grain. If there is no point of free fall, alternative mechanical sampling equipment or hand

scoops may be used.

NOTE- It should be pointed out that samples obtained from conveyor belts are less representative than those extracted from a point of free fall.

8.2 Extracting samples from static bulks

In vertical bulk storage bins, sampling for hidden infestation shall first performed at the surface to a depth of 100 mm, or 250 mm if air temperature above the grain is below 15C. An increment weighing at least 1 kg shall be extracted from each 1 000 kg sample unit in this surface layer, using a hand scoop (see 5.2).

The number of increments to be taken, n, is given by the equation

a) if sampling to a depth of 100 mm

$$n = \frac{Am}{1000}$$

b) if sampling to a depth of 250 mm, by the equation

$$n = \frac{Am}{400}$$

where

A is the surface area, in square metres;

m is the mass of 1 hal of the grain.

Round the value of n to the next highest integer.

If possible, an equivalent number of similar sized increments shall be taken from the bottom of the bulk by running grain out of the outlet spout.

Samples from below the surface shall be obtained using a cylindrical sampler or suction sampler (see 5.3) inserted at selected points on the surface. Increments taken at regular intervals along vertical lines from these surface points shall weigh not less than 1 kg.

Sampling grain for insects in flat bulk stores shall be carried out as described in the preceding paragraph if the surface of the bulk is reasonably level. Pre-sampling information shall be used to select parts of the bulk for sampling.

9 Sampling of grain in bags

9.1 Selection of bags to be sampled

For a stack about to be dismantled, or a lot about to be unloaded from a railway waggon, lorry, ship or lighter, the number of bags to be sampled shall be as specified in the table.

Table Number of bags to be sampled

Number of bags in the lot	Number of bags to be sampled
Up to 10	Each bag
10 to 100	10, drawn at random
More than 100	Square root (approximately) of total number, taken at random

In a stack of bags which is to remain in position, it is only possible to sample the top layer. Since most insects are found in the outer bags, including the top layer, no serious disadvantage is incurred. The scheme for selecting sample units described above may be used, substituting the expression "(in the lot)" by "in the top layer)" The selected sample units shall always include the four corner bags since these are especially prone to infestation. Bags needed to make up the required number to be sampled shall be selected at random.

9.2 Extraction of increments from bags.

A device (see 5.4) capable of taking a representative sample of the contents of a bag shall be used, because of the non-random distribution of insects.

10 Preparation of laboratory samples

10.1 All samples to be submitted for laboratory examination shall be referred to as laboratory samples, whether they are original increments or samples obtained by the reduction of bulk samples.

If information on the distribution of insects within a lot is required, the increments shall not be combined, and each shall be considered as a laboratory sample.

10.2 If necessary, increments shall be combined and thoroughly mixed to form a bulk sample. The bulk sampled shall then be reduced, by the method described in ISO 950 or ISO 951 or any other relevant standard, to a laboratory sample that shall weigh not less than 1 kg.

11 Packaging and labelling of laboratory samples

11.1 Packaging

Laboratory samples shall be packed in sample bags (5.5) which have been cleaned and disinfected.

Sample bags containing laboratory samples shall be closed by knotting the tie ribbons tightly around the bag necks, and shall be secured by attaching metal seals (5.8) to the tie ribbons after closure. Seals shall be attached in such a way as to guarantee the inviolability of the samples.

11.2 Labelling

If paper labels are used for labelling the samples, they shall be of a suitably high quality for the purpose and, if they are to be attached to the outside of sample bags, the eyelet holes shall be reinforced.

External labels shall be attached by the tie ribbons at the time of closing the sample bags and shall be secured by the metal seals. Alternatively, labels may be placed inside sample bags before they

areclosed and sealed, and the bags marked indelibly with simple identification marks. Each label shall bear the information required by terms of the contract.

NOTES

1 It is important to indicate that the samples are intended for the determination of hidden insect infestation and not for the determination of other characteristics of the lot.

2 For examples of the type of information required for the label, see ISO 950 or ISO 951.

12 Despatch of laboratory samples

Laboratory samples shall be despatched as soon as possible, and only in exceptional circumstances more than 48 h after sampling has been completed. Samples shall be packed for transit in such a manner as to protect them from the hazards of the journey.

13 Sampling and inspection reports

A sampling report shall be prepared, giving the usual information and making reference to the condition of the grain sampled, including signs of insect infestation visible in the warehouse or silo, or during working the vessel or other carrier The report shall also refer to the technique applies, if this is other than that described in this part of ISO 6639, and to all the circumstances that may have influenced sampling.

Cereals and pulses Determination of hidden insect infestation Part 3: Reference method

***Crals et Igumineuses - Dterminatio de l'infestation cache par les insectes - Partie 3: Mthode do
reference***

UDC 633.1: 635.65: 632.7

Descriptors: agricultural products, cereal products, leguminous grains, determination, insects, contamination, analysis methods.

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Cereals and pulses-Determination of hidden insect infestation Part 3: Reference method

O Introduction

This International Standard deals with methods of determining hidden insect infestation in cereals and pulses.

This part specifies the reference method. The aim of this method is to count all the individuals, at every stage of life, of every insect species that normally feeds and develops within cereals and pulses.

ISO 6639/4 specifies rapid methods of determining hidden insect infestation, while ISO 6639/2 specifies methods of sampling for this purpose.

ISO 6639/1 describes the general principles of the methods.

1 Scope and field of application

This part of ISO 6639 specifies the reference method for determining the nature and number of hidden insects in a sample of cereals or pulses.

It is a slow method because it allows each insect to complete its developmental cycle and to emerge as an adult from the grain before it is removed. It can be used reliably for species that normally feed within within grains, but not for species that occasionally feed in holes or cracks in grains. These may be shaken from the grains or be induced to leave them by the disturbance of handling at any stage of the life cycle and some are likely to be killed in the process. The numbers of such species will therefore be underestimated.

2 References

ISO 712, Cereals and cereal products - Determination of moisture content (routine method).

ISO 5223, Test sieves for cereals.

ISO 6639, Cereals and pulses - Determination of hidden insect infestation

Part 1: General principles.

Part 2: Sampling.

Part 3: Rapid methods.

3 Definitions

For the purpose of this part of ISO 6639, the following definitions apply.

3.1 initial observed infestation: Those free-living insects that are immediately apparent to the eye when the sample is first examined.

3.2 hidden infestation: Those insects which are present within individual grains either because they are at juvenile stages and have developed from eggs laid inside the grains, or because they have entered the interior of individual grains through cracks or other damage, usually to feed. (Hidden infestation is not normally apparent upon first examination of the sample.)

4 Principle

Maintaining test samples at a controlled temperature and relative humidity such that the greatest

possible proportion of the insects present in the sample when collected can develop to the adult stage. Removal of insects that emerge from the grains, identification and counting, at close intervals, to enable the number initially present to be identified.

5 Apparatus

Ordinary laboratory apparatus, and in particular

5.1 Airtight containers, for storage of samples prior to the determination of moisture content (see ISO 712).

5.2 Balance, accurate to about 1 9 and capable of weighing about 300 g.

5.3 Transparent containers, preferable made of glass or plastic, of a size capable of holding up to 300 9 of the sample to be tested at a depth not exceeding 50 mm.

5.4 Closures, to allow exchange of air, but to prevent insects and mites entering or leaving the containers (5.3).

NOTE - Filter papers sealed in place with wax have been found to be suitable.

5.5 Sieves, of suitable aperture sizes to retain the grain but to allow individual insects to pass.

NOTE - For cereals, a sieve of aperture size 2 to 2,5 mm should be suitable, but for pulses a larger aperture size would be necessary to remove some Bruchid beetles. It is desirable for the sieve to

have a deep bottom pan to collect the insects removed (see ISO 5223).

5.6 Shallow trays, preferably white enamel, of dimensions about 450 x 300 mm, with a rim about 10 to 20 mm deep, on which large samples can be spread or transparent Petri dishes, of diameter about 200 mm for smaller samples.

5.7 Flexible (entomological) forceps or small brush of soft hair about 10 mm long and not more than 2 mm in diameter, insecticide free.

5.8 Room or incubator, capable of being maintained to within 1 C of a temperature in the range 25 to 30C and at either 60 to 65 %, or 65 to 70% relative humidity.

NOTE - It is essential that all apparatus and rooms used in connection with this method be kept free of insecticides or other chemicals harmful to insects.

6 Sampling

Use samples obtained as described in ISO 6639/2. The samples shall be protected from extremes of temperature and humidity and from exposure to direct sunlight.

7 Procedure

7.1 Determination of the moisture content of the laboratory sample.

Determine the moisture content of a test portion taken directly from a laboratory sample

intended for the determination of insect infestation or a separate sample, according to ISO 712, or failing that, a rapid method.

7.2 Test portion

Weigh the laboratory sample to the nearest 1 g and divide it into test samples, each weighing not more than 300 g if the moisture content of the grain is less than 15% (m/m) or not more than 100 g if the moisture content of the grain is more than 15 % (m/m). Place each test sample in a container (5.3) with a suitable closure (5.4).

7.3 Determination

7.3.1 If Insects are abundant and active, use the test sieve and pan (5.5) to extract them from the sample, taking care not to load the sieve more than three grains deep (if necessary, divide the sample for this purpose).

After sieving, or if the insects are not abundant and active, spread the grain in a single layer on a tray or dish (5.6) and remove all the insects found, using the flexible (entomological) forceps or small brush (5.7).

Identify all insects found in the test sample and record separately for each species the number of adults, and where possible pupae and larvae. If required, the numbers of living and dead insects shall be recorded separately.

After removing all insects return the test sample to its container (5.3)

Replace the closure (5.4) on the container and place the sample in the room or incubator (5.8).

If the moisture content as determined in accordance with 7.1 was above 15% (m/m) ensure that the relative humidity of the room or incubator in which the sample is placed is between 60 and 65%. If the moisture content is at or below 15% (m/m) maintain the relative humidity between 65 and 70%.

Table - Incubation periods (in days) for the detection of the hidden stages of insects in cereal and pulses samples kept under the suggested conditions

Species	English common name	Incubation period (days)	
		at 25C	at 30C
Acanthoscelides obtectus (Say.)	Dried bean weevil	56	42
Araecerus fasciculatus Deg.	Coffee bean weevil	84	56
Callosobruchus maculatus(F.)	Cowpea beetle	49	35
Rhyzopertha dominica (F.)	Lesser grain borer	70	49
Sitophilus granarius (L.)	Grain weevil	56	42

Sitophilus oryzae (L.)	Rice weevil	56	42
Sitophilus zeamais Motsch.	Maize weevil	56	42
Sitotroga cerealella (Oliv.)	Angoumois grain moth	49	42
Zabrotes subfasciatus (Boh.)	Mexican bean weevil	56	42

7.3.2 Repeat the procedures specified in 7.3 at regular intervals of 3 or 4 days for a period of at least 36 days. The actual length of the incubation period will depend upon the temperature at which the samples are stored, the type of grain involved and the species of insect present.

The recommended lengths of incubation for some insect species are given in the table. If more than one insect species is present in the sample, the incubation period for the species with the longest development shall be adopted.

8 Expression of results

NOTE - An example of a suitable data record sheet is given in the annex.

8.1 Record the numbers of insects found in each test portion at the first examination, by species and by stage (i.e. adults, pupae, larvae and eggs) and whether dead or alive, as required. Calculate the totals for all the test portions and, using the mass of the laboratory sample recorded in 7.2, express the initial observed infestation as number per kilogram for each species and stage.

8.2 Record the numbers of insects found in all the test samples at each subsequent examination by species and stage and calculate the totals for all the test portions.

8.3 At the end of the final examination, calculate the totals for all the examinations and, using the mass of the laboratory sample recorded in 7.2, express the hidden infestation as number per kilogram for each species and stage.

If any adult insects emerge from the test samples during the first 7 days of the examination period, adults of the same species emerging after the period recommended in the table will be deducted from the total counted before the value for hidden infestation is calculated.

NOTE - It is assumed, in this case, that late emerging insects are progeny of adults emerging after the initial observed infestation has been removed, and, therefore, that they do not belong to the total infestation present at the time of sampling.

9 Interpretation of results

9.1 For each species, the pattern of emergence represents the age distribution at the time the sample was taken. The pattern, when plotted on a graph from right to left, will present a picture of the proportions of life stages from egg to adult in equal time periods.

A high proportion of young stages (late emergents) is a sign that the population in the zone sampled is increasing while a low proportion is a sign that the population is decreasing.

9.2 The significance of the number of insects found depends upon the temperature at which the product is stored. At temperatures below 15C, none of the species listed in the table can multiply quickly enough for small populations to be dangerous, but at temperatures above 25C, the presence of even a single individual per kilogram of any of the listed species is a serious hazard.

10 Test report.

The test report shall show the method used and the results obtained. It shall also mention all operating not specified in this part of ISO 6639, or regarded as optional, together with details of any incidents likely to have influenced the results. The test report shall include all the information necessary for the complete identification of the sample.

Creles and pulses Determination of hidden insect infestation Part 4: Rapid methods

Creles et Igumineuses - Dtermination de l'infestation cache par les insectes - Partis 3: Mthodes rapides

UDC 633.1: 635.65: 632.7

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Cereals and pulses Determination of hidden insect infestation

Part 4: Rapid methods

0 Introduction

This International Standard deals with methods of determining hidden insect infestation in cereals and pulses.

This part specifies rapid methods. ISO 6639/3 specifies the reference method against which the rapid methods can be checked, and ISO 6639/2 specifies methods of sampling for his purpose. ISO 6639/1 describes the general principles of the methods.

1 Scope and field of application

This part of ISO 6639 specifies five rapid methods for estimating the degree of, or detecting the presence of, hidden insect infestation in a sample of a cereal or pulse.

The method described in section one (determination of carbon dioxide production) is primarily intended for testing whole grains. It is not applicable for testing

a) finely ground grain products, as there is a risk that particles of material will be sucked up with air samples; or

b) grain products with moisture contents greater than 15% (m/m), because of the risk of carbon dioxide produced by the products themselves and by micro-organisms interfering with the results.

In addition, the method is not applicable to the rapid testing of grain products on to which carbon dioxide has already been adsorbed in large quantities, for example grain stored in a confined atmosphere or when there are clear external indications of heavy infestation.

The method can be used for coarsely milled or kibbled grain products, provided that they have been sieved before testing to remove fine particles and loose insects.

The method does not permit the presence of dead adults, pupae, larvae or eggs to be detected.

The method described in section two (ninhydrin method) is applicable to any dry grain prone to internal insect infestation, particularly wheat, rice and similar sized grains.

Large grains, such as maize, have to be partially broken (kibbled) before they can be tested. This

treatment of large grains can cause some insects to be lost or fragmented, thus rendering the interpretation of results unreliable. Numbers of eggs and early instar larvae may be underestimated, but, in this respect, the method is no less efficient than any other.

The method described in section three (whole grain flotation method) is suitable for detecting hidden infestation in most cereals and pulses but only on a qualitative basis.

The method described in section four (acoustic method) is suitable for detecting living insect adults and larvae feeding inside grains. It does not permit dead adults and larvae or living eggs and pupae (nonfeeding stages) to be detected.

The method described in section five (X-ray method) is suitable for detecting living and dead larvae and adults within grains. Insects which have been recently killed (for example by fumigation) may be difficult to distinguish from those still living.

2 References

ISO 520, Cereals and pulses-Determination of the mass of 1 000 grains.

ISO 565, Test sieves-Woven metal wire cloth, perforated plate and electroformed sheet-Nominal sizes of openings.

ISO 712, Cereals and cereal products-Determination of moisture content (Routine method).

ISO 950, Cereals-Sampling (as grain).

ISO 591, Pulses in bags-Sampling.

ISO 6639, Cereals and pulses-Determination of hidden insect infestation

Part 1: General principles.

Part 2: Sampling.

Part 3: Reference method.

Section one: Method by determination of carbon dioxide production

3 Principle

Incubation of a test portion of the material at a standard temperature and estimation, by a gasometric method or an infra-red method, of the amount of carbon dioxide generated during a standard period as a measure of the total metabolism of the material.

NOTE - This method is based on work in which it was shown that respiration could be detect insects in produce and that the volume of airspace is approximately constant in bulk grain packed tight. The metabolic rate of dry grain, or a grain product is very low. That of insects is so much higher that the generation of carbon dioxide in dry grain or grain product can be regarded as a sign of infestation, provided care has been taken to avoid contamination with this gas and to ensure that none is adsorbed on the grain.

4 Apparatus

4.1 Sieve, of suitable aperture size such that fine particles and insects can pass, but the material under test is retained (see ISO 565).

4.2 Balance, accurate to 0,1 g

4.3 Apparatus for gasometric analysis (see figure 1).

4.3.1 Airtight sample containers, of capacity not exceeding 750 ml. Each container shall be closed with a rubber septem.

4.3.2 Syringes and needles, for withdrawing samples of interstitial air. The syringes shall be completely airtight and shall be of sufficient capacity for the analysis. All-glass syringes of capacity 20 ml are suitable.

4.3.3 Incubator or climatic chamber, capable of being maintained at 25 1 C (see 4.4.1).

4.3.4 Gas analysis apparatus, suitable for measuring carbon dioxide concentrations to within 0,2 % (V/V)

4.4 Apparatus for infra-red gas analysis (see figure 2).

4.4.1 Controlled climate room.

The analytical apparatus should be housed in a controlled climate room, preferably maintained at 25 1 C and a relative humidity of 70 5%.

4.4.2 Infra-red gas analyser, with two interchangeable measurement ranges for carbon dioxide (0 to 50 ul/l and 0 to 500 ul/l), capable of operating with dry air as the carrier gas supplied by a

compressed air cylinder, and air pressure line or a leakproof diaphragm pump at a flow rate of 2 000 ml/mint

4.4.3 Airtight sample containers, of capacity not exceeding 750 ml. These containers comprise a cylinder made of gasproof material, approximately 100 mm in diameter, sealed at the bottom and accommodating a removable lid with an airtight closure at the top (see 4.3.1), having two orifices with nozzles permitting air to be introduced into the lower part of the cylinder after connection to the purified air line (see figure 2) and to be expelled at the top.

4.4.4 Supply of compressed dry air (air pressure line, compressed air cylinder or diaphragm pump) with a pressure reducing valve. A flow regulating valve and a flowmeter are necessary in the circuit.

4.4.5 Three-way valves, manually or electrically controlled.

4.4.6 Air washing and drying tubes, installed in the circuit before the sample container. The washer comprises a flask to allow the air to be bubbled through 10 % (m/m) sodium hydroxide solution. The desiccator contains desiccant, for example anhydrous calcium chloride.

4.4.7 Moisture indicator, placed between the sample container and the analyser (silica gel with saturation indicator).

5 Sampling

Use samples obtained as described in ISO 663912.

6 Procedure

6.1 Preparation of test sample

Use the sieve(4.1) to remove any fine particles and insects from the sample. If required, the insects may be identified and the number of adults, pupae and larvae recorded separately for each species.

In order to bring the sample to a suitable condition for testing, keep it for 24 h in the incubator (4.3.3), controlled at 25C, or in the controlled climate room (4.4.1) in a close-woven cloth bag, or a widemouthed jar, tray or open tin, suitably covered to prevent the entry or escape of free living insects, while allowing exchange of air (see ISO 6639/3, subclause 5.4).

Before preparing the airtight sample container (6.2), resieve the sample to remove any insects which may have emerged during the preparatory period.

Spread the sample thinly on a tray or other suitably flat surface, and leave to air for 15 to 30 min (to permit adsorbed carbon dioxide to escape). Airing is less important for infra-red analysis, but, if this is not done, the test report (clause 9) shall mention the fact.

Immediately before filling the airtight sample container, determine the moisture content of the sample by the method described in ISO 712, using test portions obtained in accordance with ISO

950 or ISO 951.

6.2 Preparation of the airtight container for test and test portion

Weigh the airtight sample container (4.3.1 or 4.4.3) to the nearest 0,1 g, having first ensured, by leaving it open, that it contains no trace of carbon dioxide.

Pour approximately 300 g of the test sample into the airtight container. Tap the container to shake the sample down, and add more of the test sample until container is completely full.

Weigh the container containing the test portion to the nearest 0,1 g and deduct the mass of the test portion.

NOTE - Constancy of filling and packing of the airtight sample container is not essential if the infra-red method is used.

Seal the container hermetically by means of its airtight device (see 4.3.1 and 4.4.3).

Return the prepared sample container to the incubator or climatic chamber (4.3.3) and leave for 24 h if the carbon dioxide is to be measured by the gasometric method. If the infra-red method is to be used, the prepared sample container may be connected to the gas analyser immediately.

6.3 Determination by the gasometric method

Expel all air from the syringe (4.3.2), insert the needle through the rubber septum on the sample

container and move the piston of the syringe backwards and forwards several times so as to mix the air in the needle thoroughly with the atmosphere in the container. Draw about 10 ml of the atmosphere in the container into the syringe and withdraw the needle from the septum.

Promptly transfer a suitable quantity of the gas sample from the syringe to the gas analysis apparatus (4.3.4). (If the gas sample cannot be transferred promptly, insert the needle into a rubber bung). Determine the concentration of carbon dioxide in the gas sample, expressing it as a percentage by volume. Repeat the analysis on the same test portion.

6.4 Determination by the infra-red method

Position the valves (4.4.5) so as to isolate the circuit near the container containing the test portion. After 5 min of scanning with purified air at a rate of 1 l/min, set the analyser to zero and to the most sensitive scale (measuring range 0 to 50 = l/l).

Connect the sample container nozzles to the air inlet pipe and to the analyser (see figure 2).

Direct the flow of air through the sample by operation the three-way valves, with the analyser now set on the least sensitive scale (measuring range 0 to 500 = l/l). Circulate the purified air at a rate of 1 l/min through the sample for 15 min. Then switch the analyser to the most sensitive scale (measuring range 0 to 50 = l/l). Take the reading, in microlitres per litre per minute, of the emission of carbon dioxide in the sample directly from the analyser screen or from the recorder.

NOTE - The automatic operation of the valves and sensitivity scales may be performed by and

electronic programmer and electric control valves. The measurement may also be carried out cyclically, but an integration system is required to measure the area of the successive peaks and for accurately determining the production of carbon dioxide in the sample.

[Figure 1-Apparatus for gasometric analysis](#)

[Figure 2-Diagram of apparatus for infra-red gas analysis with operating accessories](#)

With analysers with a non-linear scale, the value obtained should be converted into microlitres per litre using the analyser calibration curve.

6.5 Number of determinations

Carry out two determinations on the same test portion.

7 Expression of results

7.1 Gasometric method

7.1.1 Calculation and formula

The concentration, expressed as a percentage by volume, of carbon dioxide in the intergranular air of 1 kg of grain after 24 h incubation at 25C is given by the formula

$$\frac{C1 + C2}{2} \times \frac{1000}{m_0}$$

where

C1 and C2 and the results of the two determinations of the carbon dioxide concentration, as percentages by volume, measured on each test portion;
m₀ is the mass, in grams, of the test portion.

Take as the result the arithmetic mean of the values obtained in the two determinations, if the repeatability conditions are met.

7.1.2 Repeatability

The difference between the results of two determinations carried out one after the other by the same analyst should not exceed 0,2 % (V/V).

7.2 Infra-red method

7.2.1 Calculation and formula

The concentration, expressed in microlitres per litre, of carbon dioxide produced in 1 min in the intergranular air in 1 kg of grain is give by the formula

C 1 000 / m₀

where

C is the concentration, in microlitres per litre, of carbon dioxide produced in 1 min in the intergranular air of the test portion;

m₀ is the mass, in grams, of the test portion.

Take as the result the arithmetic mean of the Values obtained in the two determinations, if the repeatability conditions are met.

7.2.2 Repeatability

The difference between the results of two determinations, carried out one after the other by the same analyst, should not exceed 2 = 1/1 min.

8 Test report

The test report shall show the method used, the number of determinations carried out, and the results obtained. It shall also mention any operating details not specified in this part of ISO 6639, or regarded as optional, together with details of any incidents likely to have influenced the results.

The test report shall include all the information necessary for the complete identification of the sample.

9 Interpretation of results

9.1 Gasometric method

For wheat, peas, split, haricot beans, butter beans, polished rice, small yellow maize, and similar small huskless hard grains, tested by the gasometric method, the interpretation given in table 1 applies.

NOTE - For other grains, it is necessary to make a correction for the characteristic volume of interstitial air and the observed carbon dioxide concentration should be multiplied by the correction factor. Some correction factors are:

- **linseed 0,89**
- **large white maize 1,18**
- **barley 1,25**
- **oats 1,39**

Table 1 Interpretation of results obtained by the gasometric method

Production of Carbon dioxide, % CO₂(V/V) for 1 kg after 24 h incubation	Interpretation
---	-----------------------

	Probably no infestation
< 0,2	present, Repeat test on another sample to confirm.
	Possible light infestation.
0,2	Repeat test on another sample to confirm.
	Light to moderate infestation.
	Grain unsuitable for storage longer than 2 months without treatment.
0,3 to 0,5	Moderate to heavy infestation.
0,6 to 0,9	Grain should be fumigated immediately.
	Heavy infestation. Grain in dangerous condition and Highly unsuitable for storage.
1,0 and higher	

9.2 Infra-red method

The interpretation given in table 2 applies.

Table 2 Interpretation of results obtained by the infra-red method

Rate of carbon dioxide production, -l/l min. for 1 kg of grain	Interpretation
	Probably no infestation
	present, Persistent small
< 10	peaks could indicate a very
	light infestation. Repeat test
	on another sample to confirm.
	Possible light infestation.
1,0	Repeat test on another
	sample to confirm.
	Light to moderate infestation.
	Grain unsuitable for storage
2,0 to 3,0	longer than 2 months without

	treatment.
	Moderate to heavy infestation.
4,0 to 6,0	Grain should be fumigated
	immediately.
	Heavy infestation. Grain in
6,0 and higher	dangerous condition and
	highly unsuitable for storage.

Section two: Ninhydrin method

10 Principle

Crushing a test portion, from which any visible living insects have been removed, against white paper impregnated with ninhydrin.

When an infested dry grain is crushed, the amino acids in the body fluid of insects with the ninhydrin in the paper to give a purple spot, but the amino acids of the grain are not released and do not react.

Counting of the purple spots on the paper. The number of spots is taken to indicate the level of hidden infestation in the sample.

11 Apparatus

11.1 Sieve (see 4.1).

11.2 Kibbling device, if required, to partially break large grains.

11.3 Grain sample divider (see ISO 950).

11.4 Infestation detector, manually or electrically operated, which consists essentially of two rough surfaced steel rolls 0,75 mm apart between which passes a continuous strip of ninhydrin treated paper (see figure 3).

NOTE - The Ashman Simon apparatus is suitable.

11.5 Ninhydrin treated paper

Use a roll of white paper 57 mm wide and 50 m long, already impregnated with ninhydrin, or prepare as follows.

Pass the untreated paper through a 10 g/l solution of ninhydrin in industrial denaturated alcohol. Roll the paper up and leave it to dry at 20 to 25C and 40 to 60% relative humidity, in a dark place for at least 3 days. Wrap the dry treated roll in metal foil and store away from light, if possible at 20 to 25C and 40 to 60% relative humidity. Under these conditions, the ninhydrin treated paper will remain stable for 2 to 3 years.

11.6 Balance, accurate to 0,1 g.

12 Sampling

Use samples obtained as described in ISO 6639/2.

13 Procedure

13.1 Preparation of test sample and test portion

Use the sieve (11.1) to remove all foreign matter and free insects from the sample. If required, the free insects may be identified and counted according to species and stage.

Weigh the sifted sample and divide it, using the grain sample divider (11.3), to obtain the test portions required (see 13.3 and clause 15). Each test portion should contain at least 1 000 grains (see ISO 520). Test portions of large grains should be kibbled and resifted before testing.

Weigh a test portion and/or count the number of grains in it. Prepare the infestation detector (11.4) and pass the test portion through it in accordance with the manufacturer's instructions.

13.2 Determination

Remove the paper strip corresponding to the test from the detector, taking care only to handle the ends of the strip as amino acids on the skin of the fingers also react with ninhydrin to give purple stains (this may be obviated by wearing of surgical gloves or using tweezers), and allow time for

purple spots to develop. At 20C and at higher ambient temperatures, purple spots develop within 1 h, although they can take up to 24 h to reach maximum intensity. At lower temperatures, or if more rapid development is required, the paper can be heated in an oven maintained at 50C, or it can be passed cautiously (to prevent burning) over a spirit lamp flame or electric light bulb.

Figure 3 - Apparatus for ninhydrin detection of hidden insect infestation

When purple spots have developed, mark the boundary of each with a pencil line, taking care to distinguish spots which may be so close as almost to merge.

Ignore any spots on the paper which are not purple in colour.

Count the number of marked spots.

13.3 Number of determinations

Carry out two determinations on the same test sample. (See also clause 15.)

14 Expression of results

Express the infestation as the number of hidden insects per kilogram or per 100 grains, and take as the result the arithmetic mean of the two determinations.

15 Interpretation of results

If no insects are detected in the first pair of test portions, the test should be repeated with up to a total of 10 test portions before it can be reasonably concluded that the grain is free from infestation. Even then it should be remembered that eggs and small larvae can escape detection by the method. Therefore, if it is desired and practical, apparently infestation-free grain should be tested again after 2 to 4 weeks.

The efficiency of method also varies according to the species of insect and size and type of grain under test. It is doubtful whether a correction coefficient valid for different grain types and different insect species can, or should, be recommended or whether it is necessary in commercial practice.

In general, a positive result should be taken to indicate that the grain is potentially unsafe for storage. Relatively few purple spots occurring irregularly on the paper indicate a light to moderate infestation, and that the grain cannot be stored for more than 2 months without treatment. Many purple spots indicate a heavy infestation requiring immediate fumigation. However, before taking any action, it should be determined whether the grain has already been effectively treated, and how recently. This is because dead insects continue to give positive results by the method until their body fluids have dried up. A large dead insect can take several weeks to dry out.

16 Test report

The test report shall show the method used, the number of determinations carried out, and the results obtained. It shall also mention any operating details not specified in this part of ISO 6639, or regarded as optional, together with details of any incidents likely to have influenced the results.

The test report shall include all the information necessary for the complete identification of the sample.

Section three: Whole grain flotation method

17 Principle

Hidden insect infestation reduces the mass of grain. When a mixture of sound and infested grains is immersed in a test solution in which sound grains just sink, infested grains float to the surface. The separation is usually imperfect because grains containing early instar larvae tend to sink, and uninfested grains with air pockets under the testa or some other defect may float. Floating grains are dissected, to confirm the presence or absence of insects.

18 Apparatus

18.1 Hydrometer floats, to measure relative densities in the range 1,100 to 1,300.

18.2 Measuring cylinder, of capacity 500 ml.

18.3 Sieve (see 4.1).

18.4 Balance, accurate to 0,01 g.

18.5 Grain sample divider (see ISO 950).

18.6 Beaker, of capacity 1 000 ml.

18.7 Skimmer, for removing floating grains.

19 Sampling

Use samples obtained as described in ISO 6639/2.

20 Test solution

A suitable test solution can be prepared by dissolving sodium silicate, ammonium nitrate or glycerol in water. The quantity of solute required to make 1 000 ml of test solution of approximately the correct relative density can be calculated by reference to figure 4. Check the relative density of the solution by using a suitable hydrometer float (18.1) and the measuring cylinder (18.2). If necessary, add small amounts of solute or water until the relative density is within 0,005 of that required.

NOTE - As grain densities vary according to type, variety and other factors, the required densities of test solutions also vary. Where practical, the required relative density should be determined by experiment. The following values for relative densities of test solutions are intended only as a guide:

- **wheat: 1,15**
- **maize and sorghum: 1,19**

- milled rice: 1,27
- peas: 1,27

[Figure 4 - Guide to the preparation of test solutions of given relative densities](#)

[Figure 5 - Limits of repeatability for the grain flotation method \(the curve represents the 95% confidence limits of repeatability for representative samples of about 500 grains\)](#)

21 Procedure

21.1 Preparation of test sample and test portion

Use the sieve (18.3) to remove all foreign matter from the sample. Weigh the sifted sample and divide it, using the grain sample divider (18.5), into test portions, each containing about 500 grains. Count the grains in a test portion.

21.2 Determination

Place the test portion in the beaker (18.6) containing the test solution. Mix thoroughly and allow to stand for 10 min. stirring briefly at 1 min intervals to release air bubbles by the grains. When the grains have settled for the last time, use the skimmer (18.7) to remove all floating grains. Sort out and count all grains bearing visible evidence of insect infestation ("windows" in the testa or tunnels visible through it). Cut open the remaining grains with a suitable instrument, and count those found to contain insect larvae, pupae or adults.

21.3 Number of determinations

Carry out two determinations on the same test sample.

22 Expression of results

22.1 Calculation

Express the infestation as a percentage of grains which are infested and take as the result the arithmetics mean of the two determinations.

22.2 Repeatability

The difference between the result of either determination and the mean shall not exceed the limit indicated in figure 5. If this limit of repeatability is exceeded, repeat the determination on other test portions until the requirement is satisfied.

23 Interpretation of results

In view of the limitations referred to in clause 17, the method is most likely to produce an underestimate of the level of infestation present. Therefore, results are of qualitative, rather than quantitative, value. One of the more accurate methods should be used if quantitative results are important.

24 Test report

The test report shall show the method used, the number of determinations carried out, and the results obtained. It shall also mention any operating details not specified in this part of any incidents likely to have influenced the results.

The test report shall include all the information necessary for the complete identification of the sample.

Section four: Acoustic method

25 Principle

Placing a test portion in a sample container inside a well soundproofed box. An acoustic vibration sensor, fitted inside the sample container and connected to an amplification system, transmits noise from the feeding activity of hidden insects for direct listening or for recording. Estimation of the approximate degree of hidden infestation from the noise level transmitted or recorded.

26 Apparatus

26.1 Acoustic detection equipment, comprising the following elements (see figure 6).

26.1.1 Box, sound-proofed by an internal lining of high performance sound-insulating material (for example rock wool or high density polyester foam with lead), the opening of which has a seal impervious to external noise, and within which is placed a removable sample container in the form of a thick plastic cylinder (for example high density PVC) fitted with a hermetic closure. A vibration

sensor is placed in the centre of the sample container and connected by a flexible or expanding cable to a socket outlet placed on the outer face of the box.

The box is mounted on an elastic suspension system (for example rubber pads).

NOTE - The arrangement of the various components of the soundproofed box may vary while achieving the same insulation result for airborne and mechanical vibrations.

26.1.2 Electronic amplification system, composed of a pre-amplifier giving an amplification of 50 to 100 dB, according to the equipment, compatible with the characteristics of the vibration sensor, with a pass band of 600 to 4 000 Hz and a signal to back ground noise ratio, on average, of-120 dB/V.

NOTE-In order to restrict the background noise, a filter may be added to reduce the width of the pass band (the central tuning frequency for the rice weevil is approximately 2 kHz). A filter is recommended when using a recording system.

26.1.3 Headphones or recording system:

a) headphones or loudspeaker connected to the amplifier for direct listening to the noise produced by the hidden insects;

b) voltage threshold detector and recording system for electrical impulses exceeding the adjustable threshold.

26.1.4 Mat of high density insulating material, placed between the box and the horizontal support to limit the transmission of mechanical vibrations. (This precaution is optional with wellinsulated boxes.)

26.2 Sieve (see 4.1)

26.3 Grain sample divider (see ISO 950).

27 Sampling

Use samples obtained as described in ISO 6639/2.

28 Procedure

NOTES

1 It is essential to carry out the test on samples having a temperature greater than or equal to 20C.

2 Some equipment incorporates a system for heating the sample in order to increase insect activity; this preliminary heating requires about 20 min before each determination.

28.1 Preparation of the test sample and test portion

Use the sieve (26.2) to remove all free insects. If required, the insects may be identified and counted according to species and stage. Divide the sample, using the grain sample divider (26.3),

into the required number of test portions (see 28.3). Each test portion shall be equal to or slightly in excess of the quantity required to fill the cylinder of the box (26.1.1).

28.2 Preparation of the apparatus

Place the box on the insulating mat (26.1.4) if necessary. Close the empty sample container, place it in the box and close the box. Connect the amplifier and listening or recording system and adjust the gain until a low continuous background noise is obtained. Switch off the now pre-set apparatus.

NOTE - With a recording system, the threshold setting should be calibrated periodically (for example with a test recording on magnetic tape).

28.3 Determination

Fill the sample container with the test portion. Settle the grain down gently by vibration, seal the container, place it in the box and seal the box. Wait for 5 min for the grain to stabilize, then switch on the detection system. Listen for the characteristic noises of insect activity in five listening periods of 1 min or by recording for a period of 5 min.

NOTE - When recording, the direct listening device can be used for checking any defects in recording and adjusting the setting of the detection threshold.

Once the apparatus is switched off, remove the test portion from the cylinder and weigh it to the

nearest 0,1 9.

28.4 Number of determinations

Carry out two determinations on the same test sample.

[Figure 6 Acoustic detection equipment](#)

29 Expression of results

29.1 Direct listening

The result of each 1 min listening period shall be noted separately, with an indication of the existence or absence of activity of hidden insects. The relative intensity of the insect may be assessed in order to classify the extent of infestation.

29.2 Recording

The number of impulses recorded over 5 min shall be converted to the mean number per minute.

30 Interpretation of the results

With less than one period of insect activity activity recorded per minute, the sample shall be free from infestation.

With one period of activity per minute, the sample is probably infested but the presence of hidden insects has to be confirmed.

31 Sample

Use a sample as described in ISO 6639/2. 32 Procedure

35.1 Sieving

Remove all free living insects from the sample using the sieve (33.2)

35.2 Test portion

35.2.1 Standard test portion (recommended in cases of dispute)

Take, and weigh to the nearest 0,1 g, a test portion that is sufficiently large to cover completely a minimum film area of 750 cm² when placed in a layer one grain thick.

NOTE - This quantity corresponds to approximately 10 000 grains of wheat or 3 000 grains of maize.

35.2.2 Reduced test portion

It may be possible to detect infestation to an acceptable degree of accuracy by using a smaller test portion (for example 1 000 to 1 200 grains of wheat). This reduced test portion, which is

particularly applicable for rapid checking, may be substituted for that specified in 35.2.1 on agreement between the interested parties.

35.3 Spreading the test portion

Place the wire grid on the envelope containing the film. Spread the test portion in a layer one grain thick. In this way, it is ensured that all the grains will lie on one side or other of the grid lines when the radiograph is examined.

35.4 Identification of the film

At the side of the grains, prains, place figures or letters made of X-ray opaque material which appear on the fillm after exposure and will allow the film to be identified.

35.5 Exposure

During exposure, the film remains inside its lighttight envelope. Position it in accordance with the instructions for the apparatus being used.

Ensure that all safety conditions have been fulfilled.

Choose a duration of exposure to suit the nature of the sample and the fillm being used, so as to reach a satisfactory film density (see 38.1).

If an apparatus for measuring the density is available, a density of 1,0 should be sought.

35.6 Development

After exposure, develop the film in accordance with the menu facturer's instructions (see 38.1).

35.7 Examination and interpretation of the radiograph (see 38.1).

Examine the radiograph using the negatoscope or viewing screen (33.5) and count the infested grains.

In general, cereals or pulaes appear white or grey on the negative. Any cavity within a grain is represented by a dark region and an insect within the cavity appears light in colour.

35.8 Number of determinations

Carry out three determinations on the same test sample.

33 Expression of results

36.1 Count the number of infested grains found in the three determinations and calculate the number of infested grains per kilogram.

36.2 The result may also be expressed as the number percentage of infested grains, provided that the number of grains in the test portions has been counted.

34 Test report

The test report shall show the method used, the number of determinations carried out and the results obtained, indicating clearly the method of calculation used. As far as possible, the stages of development of the insects present should be recorded. It shall also mention any operating conditions not specified in this part of ISO 6639, or regarded as optional, together with details of any incidents likely to have influenced the results.

The test report shall include all the information necessary for the complete identification of the sample.

35 Notes on exposure and development of the film and interpretation of radiographs

38.1 Exposure and development

The exposure and voltage required vary according to the product being examined and the degree of penetration and contrast required. Low voltages give less penetration of grain than high voltages. For small grains, it may be preferable to use a low voltage in order to achieve the image resolution required to detect eggs, for example.

The moisture content of the grain is also important: a grain with a high moisture content will require a high voltage for satisfactory penetration by the X-rays.

It is essential to develop the film in accordance with the manufacturer's instructions, for example for the developer concentration and the temperature. Time of film development will be variable and, until experience has been gained, the middle of the manufacturer's range should be

chosen.

The most satisfactory exposure time may be determined in the following way:

- a) Expose the entire area of the film covered by the grains for 15 at 20 kV and 5 mA, for example;**
- b) Cover one-third of the area of the film by placing a sheet of tin plate, steel or copper (of thickness 1,25 mm) over the grains and expose for a further period of 5 s;**
- c) Cover a further third of the surface and expose again for 5 s;**
- d) the film now has areas which have been exposed for 15, 20 and 25 s.**

If, after determination of the most satisfactory exposure time, the penetration of X-rays at 20 kV seems too great or too little, repeat the procedure

as described above, adjusting the voltage in steps of 5 kV between 15 and 30 kV in order to find the most satisfactory voltage.

After development and fixing of the film or films under the above conditions, the most satisfactory voltage and period of exposure may be selected, and used on future occasions for similar grains.

38.2 Interpretation of the radiograph

Eggs and small larvae can occasionally be recognized in a general test exposure. However, the

proportion found will depend on the orientation of the grain at the time of exposure, the voltage of the apparatus, the insect species, the grain type and the operational conditions. The X-ray technique cannot be relied on for the detection of every egg or early larval instar. If this point is important, the test portion should be kept at 25C after the test and re-examined at appropriate intervals.

Living larvae may sometimes be distinguished from recently dead larvae by a blurring of the image which is caused by the movement of live individuals during long exposure of the film. This requires considerable skill to detect, and furthermore, a living individual may not move for some minutes.

The X-ray technique gives an accurate assessment of late larval instars pupae and adults.

If it is desired to check the infested grains, they may be cut open and examined for the presence of larvae.

36 Disposal of test portion

When the test portion used in the test is being disposed of, it should be borne in mind that materials sold for food after irradiation may be required to comply with national legislative requirements.

[Contents](#) - [◀ Previous](#) - [Next ▶](#)

[Home](#) "" "" "" "" "" > [ar.cn.de.en.es.fr.id.it.ph.po.ru.sw](#)

Section 7 - Biology and control of other storage pests

[Contents](#) - [◀Previous](#) - [Next▶](#)

[Rodents in storage and their control](#)

[Rodent control research and future needs in the philippine grain storage system](#)

[Birds as pests of grain stores](#)

[Post-harvest microbial infection of cereal grain](#)

Rodents in storage and their control

Melinda M. Boque

I. Economic Importance

With the sole exception of man, the most successful and abundant mammals on earth today are rats and mice. They would not have enjoyed this without man's inadvertent help. Rats and mice are considered commensal for the fact that these animals live at man's expenses, invading his home, eating his food and destroying his commodities. They are also capable of transmitting diseases to man, who thus derives no benefits from the relationship.

Stored foods are particularly prone to rodent attack, with the items concerned varying in different regions. The most common and therefore most vulnerable, are maize, rice, sorghum, millet, barley, oats wheat and cereal products. Much food loss occurs as the result of contamination, supplies being rendered unfit for human consumption by rodents hairs faecal droppings and urine, which are shed liberally as the animals forage nightly for their food.

While rodent attack on stored foods is widespread, estimates of damage are poorly documented. The best information available has been summarized by Holf and co-workers (1976) and is presented in Table I. The amount of food lost through direct consumption by rodents is considerable. An average-sized Norway rat eats about 259 of food a day, or the equivalent of 9kg in a year. It was found that small colonies of Norway rats (10 to 26 animals), each with access to sack wheat for 12 to 18 weeks, contaminated 70% of the grain and caused a 4.4% loss in weight. The main monetary loss however, resulted from damage to the sacks. Total losses amounted to 18.2% of the value of the wheat and the sacks.

Three species of worldwide distributions are the most important commensals: the Norway or brown rat (*Rattus norvegicus*) the roof rat (*Rattus rattus*) sometimes called the black rat or shiprat and the house mouse (*Mus musculus*). Their characteristics are considered briefly in here. This paper therefore will discuss the reproduction, ecology and control of commensal rodents which are essentially pests in storage.

II. Taxonomy and Distribution

The identifying characteristic of commensal rats and mice are summarized in Table II.

The Norway rat (*R. norvegicus*) is essentially a temperate climate species. It is more abundant and widely distributed across central Asia, Europe and Northern America. The range of this species continues to expand due to changes in urban environments favorable to its habits and to occasional introductions resulting from the traffic. This species frequently live in and around residences, in cellars stores, warehouses, slaughter houses, docks and sewers. On farms they infest silos, granaries, piggeries, poultry houses, stables, warehoused and dockside structures in port areas.

The roof rat (*R. rattus*) is at home indoor or out, depending on the climate. It is a semi arboreal species, climbing shrubs, vines and trees in habitats ranging from river banks to tropical rain forest. This species inhabits a wide range of buildings in temperate areas, including houses, shops and large foodstore, warehouses, poultry houses, barns, market, restaurants and grain elevators. It also lives in close association with man in many cities and villages in the tropics. This species is more extensively distributed worldwide than is *B. norvegicus*. The ancestral home of the roof rat is the southern Asian mainland, Southern China, parts of India, Indonesia and the Philippines but it is also distributed in several countries of the Southern and Northern hemispheres.

The house mouse (*M. musculus*) is distributed in temperate, tropical and semi-desert regions.

TABLE 1. ESTIMATED DAMAGE AND LOSSES OF STORED CROPS AND OTHER FOODSTUFFS DUE TO COMMENSAL RODENTS IN TROPICAL AND SUBTROPICAL AREAS (Hopf et al 1976)

Area	Type of storage	Commodities attacked	% Damage or loss
Brazil	Stacks, sacks, cribs	Rice, maize, beans	4-8
Bangladesh	-	Rice, pulses, grains	2-5
Egypt	Open and closed stores	Cereal grains	0.5-1
Ghana	-	Maize, rice, grain.	2-3
India	Warehouses, sacked	Cereal grains	5-15
Korea, Republic of	Sacks in houses and stores	Rice, barley	20
Laos	Stores	Rice, maize	5-10
Malawi	-	Maize, rice	1-7
Mexico	Granaries, sacks, cribs	Maize, rice, groundnuts	5-10
Malaysia (Sarawak)	Cribs	Rice	5-10
Nepal	Sacks	Maize	3-5
New Hebrides (Vanuatu)	Covered platform	Yams	10
Nigeria (Kano State)	Temporary or closed stores	Pulses and groundnuts	3-5
Philippines	Warehouses, sacks	Rice, maize, legumes	2-5
Sierra Leone	Temporary cribs or sacks	Rice, maize, groundnuts	2-3
Solomon Islands	-	Yams	5

Thailand	Sacks, cribs	Maize, rice, copre	5
Turkey	Warehouses, sacks	Wheat, rice, maize, legumes	5-15
Tunisia	Warehouses	Cereal grains, legumes	6-8

TABLE 2. FIELD CHARACTERS AND MEASUREMENTS OF COMMENSAL RODENTS

Character	Norway rat Rattus norvegicus 150-600 gm	Roof rat Rattus rattus 80-300 gm	House mouse Mus musculus 10-21 gm
Head and body	nose blunt, heavy, stocky	nose pointed, slender body,	nose pointed, slender body,
	body, 18-25 cm	16-21 cm	6-10 cm
Tail	shorter than head plus body,	longer than head plus body,	equal to or a little longer
	darker above and lighter	uniformly dark colored,	than head plus body,
	below, with short, stiff	marked, 19-25 cm	uniformly dark coloured,
	hairs, 16-21 cm		naked, 7-11 cm
Ears	relatively small, close-set,	large, prominent, thin and	prominent, large for size of

	appear half-buried in fur,	hairless, stand well out from	animal, 15 mm or less
	rarely over 20-23 mm	fur, 25-28 mm	
Fur	brownish-gray on back,	brownish-gray to blackish on	one subspecies brownish-gray
	greyish on belly	back, belly may be white, grey	on back, greyish on belly,
		or grayish-black	another greyish on back and
			grayish-white on belly
Habits	burrows, swims and dives	agile climber, gnaws, often	climbs, sometimes burrows,
	easily, gnaws, lives indoors	lives off the ground in trees	gnaws, lives indoors and
	and outdoors, in sewers and	vines, etc., lives indoors and	outdoors
	drains	outdoors	

It habitually infest food storage and other premises in both urban and rural surroundings and it is also found occupying such varied habitats as cold stores, rice, sugarcane and cereal grain fields, garbage dumps, salt marshes, and coal mines.

Other rodents develop into commensals where their habits bring them into close contact with man. Most notable of these species are the lesser bandicoot rat, *Bandicota bengalensis*, and multimammate rat, *Mastomys natalensis*.

The bandicoot rat is distributed in South and Southeast Asia. While it is mainly a pest of agriculture and the predominant field rat in many parts of India, Bangladesh, Burma and Thailand, it has invaded cities, towns and villages and become the main urban commensal in Bombay, Calcutta, Madras, Dhaka, Rangoon and Bangkok. This species feeds extensively on field crops but also infests food stores where it feeds on paddy or wheat, both in warehouses and farmers' houses.

The multimammate rat (*M. natalensis*) is regarded as a peridomestic rat in most of Africa where it is found in close association with man. It nests in underground burrows or in dark protected areas when living in human habitations. It is abundant in farm houses as it is in the fields.

III. Biology

Reproductive activity in commensal rats and mice is characterized by early sexual maturity, short gestation period, post-partum oestrus, breeding throughout much of the year and large litter size. These traits give commensal rats and mice the potential for very rapid population growth and for

quick recovery when their numbers are reduced by poisons, traps or other means.

The results of reproductive studies of female commensal rodents are summarized in Table 3 and 4. Under conditions of optimum climate, surplus food and abundant shelter, commensal rodents population tend to breed throughout the year. These conditions most commonly occur indoors in food stores, warehouses, farm buildings and on ships.

Male animals generally remain in breeding condition throughout the year although the testes may be retracted into the abdominal cavity during periods of cold weather when they might appear to be infertile. Norway rats construct a nest of grass, waste paper, twine or other suitable materials in a separate chamber within a burrow system in the natural spaces within buildings. Likewise, house mice build nest in walls and roof cavities, in stacks of food or cabinets or drawers. Roof rats living outdoors often build in shrubs or trees, constructing them from twigs, leaves, grass and other plant materials. The young for all three species need constant maternal care for at least three weeks after birth.

IV. Ecology

The important physical elements necessary to sustain commensal rodent populations are food, water and shelter. Their abundance and distribution has a direct bearing upon how many rodents can be supported in a given environment. Commensal rodent populations thrive when all three resources are abundant and close together.

The main food sources for commensal rodents are stored food and garbage in urban areas, and in

rural areas, field crops, natural vegetation and seeds. Stored food are available in mills, warehouses and godowns, port facilities, food processing plants. Feed bins and corn cribs, waste food and spilled grain are other important food sources. Warehouses containing food stored in bags or in bulk are particularly vulnerable to rodent attack unless they are protected. Improperly stored and handled garbage increase rat problems in the urban environment and it is a major cause of the persistence of rat populations in many cities and towns.

Water , generally available to rodents in urban and rural localities but its supply can be a problem for Norway rats, particularly those living in well designed warehouses or in areas with dry seasons. The Norway rat needs food with a high moisture content or a supply of free water, the lack of which restricts its distribution and spread into new areas. House mice, which utilize metabolic water more effectively, tolerate dry habitats without difficulty and the roof rat can also withstand water deprivation better than the Norway rat.

A rodent population that has been reduced in size recovers slowly at first and then at an increasingly faster rate. As it approaches the capacity of the environment, the primary limitations being food, water and shelter, growth slows down and the population tends to level off. In an uncharged environment, the population would be expected to remain at essentially the same level but in practice it tends to fluctuate in size, around the carrying capacity of the environment.

Evidently, a confined colony of rodents competes more fiercely for food, shelter and living space as the population density increases. At very high densities, rats and mice spend abnormal time and energy in aggressive attacks and in the defense of territories. Reproductive success of females can also be seriously lowered as the result of aggressive behavior.

IV Methods of Control

The control of commensal rodents is important to safeguard human health and to prevent economic and other losses. Most control work is directed towards preventing rats and mice from living in and around buildings in both urban and rural areas or eradicating populations that have already become established in them. Thus, varied control methods have been devised to come up with an appropriate strategy for certain types of condition. These include environmental sanitation, physical, chemical, and biological control and other methods.

A. Environmental Sanitation

Environmental sanitation concerns the orderly management of the stored product. Basically it means good housekeeping, the proper storage and handling of food stuffs and organic waste and elimination of harborage for rodents and other pests. Commensal rodents are opportunistic and readily take advantage of man's misuse of the environment. Poor sanitation generally results in food stuffs and harborage being abundant and easily available to rodents and it enables their rapid establishment. Almost any pile of debris indoors or out, can provide rodents with suitable cover for nesting and breeding. In general, the maintenance of a cleared area, as extensive as possible, can do much to deter rats and mice infesting food stores.

B. Physical Control Methods

These have been appropriate measures since they do not basically contaminate the stored products. Some of the approaches are ratproofing buildings where rodents are denied shelter and

food. It is the most effective and long-term remedy against rats. Preventing entry of rats and mice is important for successful rodent control. This includes keeping openings to buildings and bins tightly closed when not in use. Heavy wire screening or sheet metal barriers on lower portions of openings will help keep rodents out. However, this is a difficult task since commensal rodents like mice can pass through 12 mm apertures and very young rats can enter 14 mm openings.

The method is only ideal where storage activity is less. Trapping is the preferred method of killing or capturing rodents in situations where the use of rodenticides is considered undesirable. This technique has to be properly done. Because of the cautious behavior of rats, they tend to be wary of traps and abundance of alternative foods, makes them difficult to capture or control by this approach. In campings against them, traps should be placed near runs and at other locations where there are clear signs of rat activity. Trapping success can be improved by leaving the traps baited but unset for a few days and using baits of proven acceptability to rodents which include bacon, peanut butter, fresh, smoked or dried fish, and ground meats or bread for Norway rats. Baits that dry out or spoil should be replaced immediately by fresh ones. Traps should be examined daily to remove dead rats, which should be buried or incinerated, and to reset those traps that have sprung.

For economic or other reasons, traps are of little value in controlling large infestations of rats and mice but these are useful in helping to catch the survivors of any poison treatment.

The effectiveness of snap-traps against rodents especially on house mice is largely determined by the sensitivity of the traps, their placement and number of traps used. Traps should be set close to walls and in other areas where active runs are evident.

Electric fences which have been used on occasion, both to exclude and enclose rats in the field, can be effective in protecting stored product but obvious caution should be emphasized for possible shocks or electrocution.

The use of very high frequency sound, ultra sound, has been proposed as a means of preventing rats or mice from freely moving into a building or from one area to another. Field and laboratory studies have generally failed to support these contentions. Reports on the repellency of four commercially available ultrasound generating machines tested against a population of wild Norway rats in a large outdoor enclosure using an uninterrupted beam of sound 0.5 m from where they were accustomed to feed shows initial and partial repellency for a day or so but thereafter the feeding behavior of the rats was unaffected.

C. Chemical Control

Most measures to control commensal rodents depend on the application of poison incorporated in either bait, dust or water formulation. Rodenticides are usually classified as either chronic (multiple dose, slowacting) or acute (single dose; quick acting) compounds. Of most widespread use and particular importance are the anti-coagulant poison, since these slow-acting compounds are now regarded as first choice rodenticides against commensal rodents in most control operations. Acute rodenticides still have a part to play, but they are principally and most effectively employed in situations demanding a rapid reduction of high-density population.

1. Anticoagulant rodenticides have a physiological action in that they disrupt the mechanism that controls blood-clotting and cause fatal internal hemorrhage to develop. Examples of this

rodenticides are warfarin, coumachlor, coumatetralyl, diphacinone, and chlorophacinone. The new anticoagulants which are so called "second-generations" are difenacoum, brodifrocoum bromadiolone and flocoumafen.

2. Acute rodenticides. The physiological action of this type is either on the stomach or nervous system. This is categorized based on hazardinuse such as:

a. Compounds that are highly toxic and extremely hazardous to man and animals, e.g. arsenic trioxide, Flouroacetamide, sodium furoacetate (1080)

b. compounds that are both moderately toxic and hazardous to man and animals, requiring considerable care in use, e.g. ANTU (alphanaphthyl-thiourea), calciferol-zinc phoaphide.

c. compounds or relatively lower toxicity that are least hazardous to man and animals. Some of these compounds are norbormide and red squill.

Attention has been given to the possible use of reproductive inhibitors or chemosterilants for the control of rodents. Field trials in England show some indication of reduction of rat populations due to ceased breeding. However, 6-14 months after treatment, a resumption in breeding was observed. This was attributed to a return to normal fertility of long surviving resident rats ang to immigration from nearby populations.

The future of suitable chemosterilants in the field of rodent control is uncertain, though this has and advantage over rodenticides in that they pose a less immediate hazard to human beings, pers

or domestic animals. The major disadvantage is that rodent populations treated in this manner decline rather slowly. Meanwhile, damage, contamination and disease problems have to be accepted.

Fumigants are also used to kill rodents and their ectoparasites living in accessible areas in buildings, ships or in burrows in the soil.

Fumigants most commonly used against rodents are calcium cyanide, methyl bromide, chloropicrin and hydrogen phosphide. These are used mainly indoors for insect control and can also be used against rodents in storage. Experience and skill are required therefore in their application.

Repellants are chemicals that are distasteful to rodents; their use in preventive measures is based on the extreme sensitivity of rodents to certain compounds and odors. A number of compounds have been found to possess repellents. The majority are also odorous or toxic to man however and, besides being difficult to work with, few of them have been found to have a long lasting effect.

D. Biological Control

This includes predation, disease and parasitism and genetic manipulation. There are dangers in the introduction of predators which may become pests themselves. The use of diseases and parasites has temporary effects on the population and rats can easily adopt and become resistant. Most of the potential infectious agents like Salmonella also cause disease in man and are therefore unsafe for use.

Genetic mutations through in radiation that induces mutations have been tried on *R.r. mindanesis* and *R. argentiventer* (Medina et al, 1973). More rsearc on inducing lethal genes and sterile male strains need to be done before this approach can become practical.

E. Other Methods

1. Rats as food

These was a time when people were encouraged to eat rats ("STAR meat") to help control infestation and to augment people's protein requirements. Rats are now socially acceptable as food in many rural areas of the Philippines.

2. Bounty System

Paying cash reward for dead rats as evidenced by tails or head as one control/ method in the field may be useful in rat control in storage areas.

Based on experience, the use of any combinat ion of different methods provide a better result than just making use of one control method

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*** Most of the contents of this paper were taken from this reference.**

Rodent control research and future needs in the Philippine grain storage system

by ENWIN A. BENIGNO

INTRODUCTION

Rodents are serious pests in both agricultural field and storage. Although the same problem species may be involved in both cases, their presence is less tolerated in storage than in the field. This is understandable since food in storage is nearer to human consumption and we can ill afford to lose at this stage. In the field, for instance, early crop damage by rodents can still be partly

recovered by the plants' ability to produce more than normal to compensate for the injury or damage. On the other hand, whatever rodents consume in storage is total loss; only the spilled or contaminated grains can be partly recovered but at an extra cost of re-processing.

In the Philippines, Rubio (1972) placed annual rodent damage at 0.80 to 4.12 cavans (40-206 kg) per rice mill-warehouse in Laguna. Aganon (1982) estimated annual grain loss per warehouse in Nueva Ecija due to rodent consumption, contamination and spillage at 1.92 to 2.93 cavans (96-14 Kg). Sayaboc et al. (1984) observed an average loss of 3.6 kg/day in commercial grain storage.

Added to these, there is permanent damage to the storage structure itself by the rodent's gnawing habits hence, food losses are potential. This is also one main reason why these pests must be controlled.

Rodent Research

The need for rodent research was deeply felt with the rodent outbreaks that devastated the farmlands in the island of Mindanao in the 1950's. With assistance from US-AID, the Philippine Government established a Rodent Research Center (RRC) on the campus of the University of the Philippines at Los Banos in 1968. Among the concerns of the center was the training of local expertise at the University and the Denver Wildlife Research Center at Colorado, USA. Quite naturally, the first research focused on rodent control in ricedields where most of the rodent problems were encountered. Research later branched out to include other crops like corn, coconut and sugarcane; and also research was initiated on birds. The activities of the RRC were absorbed by the National Crop Protection Center in 1976.

Storage problems came as extension of the rice production system. Surveys of private mill warehouses were conducted as part of student theses (Rubio, 1972; Aganon, 1982). RRC also studied rodent problems in farm storage in upland multiple cropping. More in-depth research in storage is now the concern of the National Post Harvest institute for Research and Extension (NAPHIRE).

Research Studies

Population dynamics mainly focused on reproductive potential and movements inside and outside the warehouse. One of the significant products of these studies was the inclusion of rodent losses in a warehouse stock inventory system. Bird pests were also studied although to a lesser extent than rodents. Some of the specific findings are presented here to help us in our discussions.

Rodent Species

The major species affecting food in storage in the Philippines are *Rattus norvegicus*, the Norway rat, *Rattus rattus mindanensis*, the common ricefield rat, and *Mus musculus*, the house mouse.

These species differ in their habits, *B. norvegicus* is expected to be dominant over the other species because of its size. Conversely the mice will be confined to a smaller area in the warehouse.

Damage patterns in the warehouse. In a survey of 20 ricemill-warehouses in Nueva Ecija, Aganon (1982) discovered that the amount of rat damaged grain is a function of the number of sacks

gnawed ($r = 0.50$). Further regression analysis based on the summarized annual data showed this relationship (disregarding partial recovery of spilled or contaminated palay) to be:

$$y = 92.2 + .369X$$

where

X = number of gnawed sacks

Y=total grains loss in kg. (consumed + contaminated + spilled)

In warehouse adjacent to ricefields, damage is not related to rainfall and air temperature but to the crop stage. Damage is highest at land preparation and lowest during the rice reproductive stage. Ricefield rats also migrate to the warehouse during land preparation (Sayaboc, et al., 1984).

Larger capacity warehouses had lower incidence of gnawed sacks ($r = 0.81$) and warehouse near to garbage dumps, slum dwellings, and commercial establishment showed more damage. The different types of warehouses had the same amount of damage (Table 1 and 2).

Some previous studies showed that rat proofing is Types I and 11 warehouses did not significantly reduce infestation and subsequent physical losses, maybe because they were not properly maintained.

The stomach contents of trapped rodents from modern government warehouses (Type I and II) were observed to have a 99.5 percent grain component, while those collected from private warehouses (Type III and IV) contained 90 percent grains. This indicates that rats in government

warehouses depend only on stored grains for food. Rats in private warehouses have alternate foods available such as feeds, grasses, fruits, etc.

It was also observed that while feeding, rodents spill 7.5 times as much as they consume indicating potentially more serious losses. Spilled grains were infected with *Aspergillus flavus* and *A. ochraceus*, the storage fungi associated with the production of carcinogenic compounds (mycotoxins) capable of causing liver and kidney damage in man. Samples were also infected with bacteria.

Poor sanitation, irregular baiting programs and improper warehouse design, i.e. unfit doors, drainage canals and gutters, are some of the factors that contribute to the rodent problem in both private and government warehouses (Sayaboc et al., 1984).

Rodent Control with Anticoagulants

Baiting was found effective and economical in controlling the rodent population in one warehouse. Prior to control, losses for 6 months were estimated at 976.65 kg paddy valued at P 1,659.09. During the first and second months of baiting, losses were reduced to 161.03 (94.72 kg) and 58.87 (34.63 kg) respectively.

No signs of rodent infestation such as damaged grains, gnawed sacks, feces, were observed during the third and fourth months of implementing the control program. The cost of control which includes: amount of poison, rice brewers, labor and cost of bait station was P 485.05 for six months. A benefit cost ratio of 1.36 was achieved using this control program on a sustained basis

(Table 3).**Control Threshold**

A control threshold (ETL) of 62 rats or 8.65 kg of spilled grains was computed based on consumption and baiting experiments. This figure still has to be validated.

Modeling

A rat population model was constructed using a leslie matrix. With consumption and control functions, rodent control corresponding losses can also be simulated. Results of our simulations are summarized in Table 4.

The model indicated that if an initial population were low (a pair of rats) rat problems occur starting at the 10th month of storage. Without any control measure the simulated loss is 5669.36.

The model also showed that it pays to have a high population control success (80-90%). For 90% control a two-year cost/benefit ratio of 1:14 is simulated.

**P = Philippine peso
(14 P = 1 USD, 1986)**

Future Needs

At present, there is no formal research project on rodent control in storage, although the

importance of rodent pests in storage is still recognized and included in all training courses of NFA and NAPHIRE. Besides research, there is also a need for computerized data storage and retrieval to facilitate information exchanges among researchers.

The future needs of rodent research in storage will depend largely on the type of storage system in the future. The trend of storage system seems to be slanted towards bulk storage and storage with controlled atmospheres. It has been observed that insects bore into plastic covers enclosing CO₂ treated stacks stored for 8-10 months. Going into the second year of storage, rodent gnawing of the sheets would be a greater potential problem, than insect boring. Likewise, rodents may damage structures in warehouses sealed for controlled atmosphere storage (CAST).

Presently we do not have information on how rodents would react physiologically and behaviorally to high concentrations of CO₂. We have observed rodent footprints and droppings on flatbed dryers. Rodent population can possibly become established in grain silos as they have established in cold storage. These are the unknown areas for rodent research.

Table 1. Classification of warehouse site included in the field survey (Sayaboc et al., 1984)

TYPE	OWNERSHIP	PERCENTAGE CONCRETE	PERCENTAGE GI SHEET	OTHER FEATURES
I	government	80-100	—	Elevated floors, hanging stairs,
				equipped with center weights,

				screened windows, gutters, and drainage.
II	-do-	60	40	Floor at the ground level, screened windows, gutters and drainages
III A	government- leased	100	—	Conventional design, no provision for rodent exclusion
III B	private	100	—	-do
IV	private	—	100	-do

Table 2. Average rodent population per warehouse according to type/design and daily consumption of paddy (Sayaboc et al., 1984)

Type of Warehouse	Rodent Populations	Consumption1 (kg)
Type I (NFA-GID)	57	1.6
Type II NFA-GID	69	1.9
Type IIIA (NFA-LEASED)	89	2.5
Type III B (Private)	119	3.4

Type IV (private)	223	6.4
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1/Non-significant at 1% and 5% levels

Table 3. Cost-benefit analysis for six months (Sayacoc, et al., 1984)

ITEM	Amount of Losses	Value(P)	TOTAL(P)
Condition:			
a) Without control	976.65		1,659.99
b) Control program implemented	128.65	217.85	
Cost of control		485.90	702.90
			957.90
Benefit/Cost Ratio			1.36

Table 4. Simulated two-year rat control programs with varying effectiveness in a warehouse starting with a pair or rats.

Items	Percent population (at ETL)					
	- 0	10(540)a/	20(270)	50(108)	80(67)	90(60)

Number of months w/ control	0	8	10	10	6	5
Total Control Cost at fixed rate of P80/mo.	0	640	800	800	480	400
Benefits due to control (P)	(-5669.36)	1521.28	3295.49	5163.38	5390.26	5453.21
Cost/Benefit Ratio	—	1:2.36	1:4.12	1:6.45	1:11.23	1:13.63

a/ETL corresponding to percent control

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I. Agencies Conducting

Rodent and Bird Research

- **Rodent Research Center (1968-1976)**
- **National Crop Protection Center (1976 to present)**
- **National Post Harvest Institute for Research & Extension (NAPHIRE)**
- **National Food Authority**
- **Bureau of Plant Industry**
- **Ministry of Agriculture and Food**
- **International Rice Research Institute**
- **Philippine Coconut Authority**
- **Philippine Sugar Commission**
- **University of the Phil. at Los Baos**
- **Central Luzon State University**

II. Rodent Research in Storage

- **Surveys (species; damage)**

- **Feeding habits**
- **Population dynamics**
- **Control (Physical and chemical)**
- **Warehouse stock inventory**
- **Population modeling**

III. Other Vertebrate Pests:

- **Birds**
 1. **Survey (species; damage)**
 2. **feedig habits**
 3. **control (nets traps and repell ants)**

IV. Researchable areas:

- **Intra-and interspecies competition**
- **Simplified warehouse stock inventory**
- **Model validation**
- **Longterm IPM program in existing storage system**
- **Control in other storage systems (e.g. controlled atmosphere, bulk storage)**

Birds as pests of grain stores

[Contents](#) - [Previous](#) - [Next](#)

by Filipinas M. Caliboso*

INTRODUCTION

Birds belong to the Class Abes of Phylum Chordata. They are the only animals with feathers. Among the vertebrates they are the most highly specialized. They are also the most numerous of vertebrates, with roughly 28,500 species and subspecies as compared with 15,000 for mammals and 20,000 for fishes.

As a group they have become adapted to aerial life, although there are a number of exceptions. Adaptive features include light hollow bones, loss of right ovary and oviduct in females of most species; the exceptionally well-developed eyes; the highly specialized lung and air-sac system; the modifications of the forelimbs to form wings; the presence of feathers which although light in weight offer effective resistance to air when the bird is in flight. Like the mammals, birds have a constant body temperature, independent of the environment (homiothermous.).

The feet and beaks of birds also exhibit various adaptations to different modes of life and different kinds of food. There are birds whose feet are adapted for swimming, perching, and

running. Some feed on insects, others on fruits, seeds (grain). A number of grain-eating birds have become important agricultural pests in many countries.

In the Philippines, there are various species of bird pests which feed on such crops as rice and sorghum. Collectively, these birds are known locally as maya, and are also called Philippine weavers.

PROBLEM

Many species are attracted by ripening grain crops and to drying and threshing floors at harvest time. Some have developed a close association with the more permanent sources of cereals and cereal products and have become a nuisance in food stores and warehouses.

Grain spilled by careless handling in and around stores attracts birds and regular spillage may lead to establishment of a resident population. Accumulations of grain dust and flour residues around mills and processing plants will also attract birds as well as heavy insect infestation on the surface of stocks. Once attracted to a building, birds quickly learn how to enter. Bird populations inside a store pose some major problems such as:

- 1. They may settle on top of stacks and peck holes in wooven sacks in order to reach the food inside. This can cause spillage and in extreme cases, collapse of the stack.**
- 2. They will roost and nest inside large buildings unless access is completely restricted. In a warehouse, birds, may damage screens in order to gain entry.**

- 3. Nests in guttering and downpiping can cause blockages and lead to flooding and water damage to commodities.**
- 4. They become hosts for lice and mites which become occasional pests of man when birds nest in buildings.**
- 5. Nesting materials will provide harbourage and breeding sites for several of the stored insect pests.**
- 6. As a result of their activity, bird droppings, feathers and decaying bodies may contaminate the foodstuffs, packaging and handling facilities. Droppings are likely to be infected by food poisoning bacteria (Salmonella) and also constitute a major source of infection for the diseases known as Histoplasmosis, Cryptococcosis and Aspergillosis which are caused by fungal spores.**

CLASSIFICATION AND IDENTIFICATION

The Philippine weavers belong to the Order Passeriformes (Perching birds) whose common feature is the perching foot. The major bird pests belong to the genera Lonchura (Family Estrildidae) and Passer (Family Ploceidae). Another species, Padda oryzivora has also been observed but in limited number and limited distribution. Studies conducted by NAPHIRE-TRED showed that warehouse bird populations are predominantly *Passer montanus* or mayang simbahan. The weavers are relatively tiny birds (weighing 10-30 gm) which build covered nests, often beautifully woven. They are mainly nonmigratory. Species are easily distinguished by their plumage. However, there is no sexual dimorphism among these birds. A guide to the identification of the different species is

summarized in Table 1.

METHODS OF COLLECTION

1. Mist nets-Fine silk or nylon net with 3/4 inch mesh, 7 ft. wide and 18 to 38 ft. long. A taut frame of stout twine crossed by horizontal braces called "shelfstrings" is used in conjunction with the mist net, the net and the shelfstring being tight, while the net is loose. The excess netting is arranged in a loose bag or pocket 3 or 4 inches deep below each shelfstring except the topmost one. A bird striking the net from either side carries the net beyond the shelfstrings and hangs in the pocket of the net. A net properly hung, with 4 shelves, is about 6 ft. high (Giles, 1971).

Mist nets are more effective when set on a calm day against a dark background such as tall grasses in the weavers roosting area, trees and buildings. Wind greatly reduces catch. The mist nets should be placed where birds tend to fly back and forth fairly low but where there is a minimum chance of people, and animals crashing into a net. It is best to remove birds from mist nets within a few minutes before they become entangled and die from exposure or injury.

Mist nets require great care. They are easily tangled or torn by contact with vegetation. A person operating the nets should wear clothing with the fewest buttons, exposed pencils and other things to avoid being entangled to the net in the process of removing the birds. To expedite cleaning of nets, caught birds should be placed in a holding. Between uses, nets are best preserved by removing them from the poles and storing them in plastic or cloth bag. Wet nets should be dried out to avoid fungal attack.

2. "Korag" - This is manually operated net trap composed of two rectangular nets (1" mesh, light net) each measuring 1 1/2 m x 2 m. The widths are framed by light poles and the lengths by nylon stings. The nets are set flat on the ground, parallel to each other 3 meters apart such that when activated, they close in perfectly. The nets are hung at the ends and are flipped over the birds by a jerk on the pullcord by the operator.

The operator must be hidden from the birds by a blind. Decoy birds are needed to attract the birds. The "Korag" can be used where mist nets are ineffective such as in newly harvested rice fields and open grasslands. The trap area should be level and cleared of vegetation and other debris.

3. Modified Australian crow trap-this trap is a large cage made of mesh wire with wooden frames measuring 1 1/2 m wide x 2 m long x 2 m high. The midsection of the truncated V-shaped cage top is provided with holes or slots through which birds can enter. This trap is self-operating and so must contain food, water and some of the captured birds as a decoy.

4. "Kaliked" - this is a funnel-shaped bird trap made of bamboo sticks or coconut midribs used to catch brooding birds. The trap is fitted into the mouth of the nest and another entrance is opened for the opposite end of the nest. The bird is trapped as it seeks its way out after incubating the eggs. A trap could only catch one bird.

BIOLOGY AND BEHAVIOUR

Not much is known about the Philippine weavers. Field observations reveal that the main diet of

these birds consists of rice and Echinochloa seeds. They are also observed to feed on corn tassel, sorghum and certain algae. Feeding begins just after dawn until about 10:00 A.M. and at about 3:00 P.M. to dusk.

The sparrows are primarily seed-eaters. In cage and field experiments, the birds were observed to consume 30% of their body weight per day. (3 grams for Lonchura spp. and 6 grams for Passer montanus). The same study also revealed that birds infesting private stores are 15% heavier than those found at NFA (National Food Authority, National grain marketing agency in the Philippines) warehouses. Apparently, the "openness" or loose construction of private warehouses provide easy access by birds to feed inside. On the other hand, NFA-constructed warehouses partially excluded populations through its better design and provision for bird proofing such as screens. Another reason for the observed discrepancy in weight is the presence of hog and poultry pens around the warehouse. Feeds are also available in these pens and birds obviously feed on them. Results further show that grain composed 91-97% of the gizzard contents of birds collected from private and NFA stores. Other components are weed seeds, grasses and stones.

Birds are abundant in the ricefields especially around harvest, in towns and villages. They roost on pill, starapple, ipil-ipil and bamboo trees as well as houses including warehouses. Their populations follow a definite cropping pattern in an area, that is, it increases when the grains are formed and ripened. Aside from cereals, they also subsist on weed seeds, especially Echnochloa spp. and some algae. Philippine weavers are gregarious. Different species are often together is feeding, roosting and even in nesting activities.

The breeding season of these birds may start as early as February with peak of nest building and

egg laying in April and May. The breeding season may extend until October. The male and female birds work together building nests in stands of tall grasses (cogon, talahib, marker grass), palms and citrus. Mating was also observed to occur in the process. In a warehouse, they build nests on walls, between window slots, crevices of walls, beneath the roofs, along gutters and on the sliding mechanism of doors.

An average of 6 small eggs (5-8) are laid in the nest at one day interval. The eggs are oval measuring 16 mm (greatest length) by 11 mm (greatest diameter) and white or pale ochraceous-salmon in color. Eggs are hatched after a 10-day incubation period.

Newly hatched birds are naked with eyes closed. On the 7th day after hatching, chirping becomes audible while on the 8th day, eyes start to open and primary feathers appear. The body and feathers develop further and the birds are ready to fly when they are 17-19, days old.

Present information about the different species are summarized in Table 2.

POPULATION AND LOSS ESTIMATION

There are four forces affecting the size of population:

1) nasality (births)

2) mortality (deaths)

3) immigration movement

4) emigration

In combined form:

$$N_{t+1} = N_t + (B - D) + I - E$$

where:

N_{t+1} = population at time $t + 1$ (final)

N_t = population at time t (initial)

B = births

D = deaths

I = immigrants

E = emigrants

The population growth of birds is characterized by a logistic growth curve:

Characteristics of curve:

- 1. Slow rate of increase to start with**
- 2. Faster rate of increase at greater population size**
- 3. Flattens off to an asymptote**

Thus, a pair of birds can give rise to 1,720 individuals in a span of one year, potentially capable of consuming 927 kg and causing a variety of losses through reduced grain quality due to contamination, damage to structures, etc.

Types of population estimations:

1. relative density-abundant, common, rare

2. absolute density-number per area

1. Measurement of relative density

- indices of abundance such as droppings, nests and amount of food consumed.

Rice hulls that remain after the birds feeding on paddy may be collected. Since sweeping may still contain paddy. these should be removed and the rice hulls weighed. From this, the population and/or loss may thus be calculated:

$$\text{weight of brown rice consumed of loss per day (kg.)} = \frac{\text{ave. wt. ricehulls collected / day}}{0.23}$$

weight of palay loss per day (kg) = wt. of rice hulls collected/day + wt. of brown rice consumed per

$$\begin{aligned} \text{population} &= \frac{\text{weight of brown rice loss / day (kg)}}{0.006 \text{ kg 5 bird / day} \times 0.91} = \\ &= \frac{\text{weight of brown rice loss / day}}{.0055 \text{ kg / bird / day}} \end{aligned}$$

To get a good estimate of loss or population, the collection of sweeping should be undertaken daily for 12 weeks per month and at least 4 months in a year, that is, 2 months for the dry season and 2 months for the wet season:

Example:

An average of 0.5 kg. of rice hulls are swept daily at NFA warehouse.

weight of brown rice consumed or loss per day = $0.500 / 0.23 = 2.174$ kg.

weight of palay loss per day = $0.500 + 2.174 = 2.674$ kg.

$$\text{population} = \frac{2.174}{0.0055 \text{ kg}} = 395 \text{ birds}$$

2. Measurements of absolute density:

1. total counts-ex census of population; visual counting of birds.

2. sampling methods-counts of a small porportion of the population to stimated the total.

3. Capture-recapture (tagging) methods allows estimation of density, birth rate and death rate; when distance between recaptures are known, movement and home range may be determined.

Assumptions:

i) Marked and unmarked animals are caught at the same rate (equal probability of capture)

ii) Marked and unmarked animals are subject to the same mortality rate.

iii) Marks are not lost or overlooked.

Estimation:

Total population size (N) / Total caught in Sample (n)

No. marked in population (M) / No. marked animals in sample (m)

So:

Mn where sampling is continued until (m)

N = m animals are recaptured

Where:

N = population size

n = sample size

M = total marked in population

m = total marked in sample (re-capture)

Removal method (sampling without replacement)

- when animals have to be sacrificed for some reason (economic or health reasons) make it inadvisable to return trapped animals to the population. Assumptions:

i) stationary population (immigration = emigration)

ii) Probability of capture constant for each trapping Hayne's method (1948)-by regression

Example:

Night	Catch (y)	Total previous catch
1	165	0
2	101	165
3	54	266

Zippin's Method (1958)-by multinomial equation

Cleaning the hulls and spillage. Doorways can be provided with curtains or thick PVC strips

suspended from the roof. This deters birds from entering but permits almost unrestricted entry by man, vehicles and commodities. It is a lot easier to prevent birds from entering than to force them to leave with scarers such as explosives, horns and amplified distress cries.

As much as possible, within the limits imposed by other considerations, the exterior of grain warehouses should be free from ledges suitable for roosting. Attractive nearby roosting sites (such as large trees) should be reduced to the minimum, the remaining pruned to lessen the cover available for birds.

Utmost care should be taken to keep the store and surrounding area clear of spillage and food debris. Immediately after intake or discharge of stocks, all spilled residues should be swept up. Residues should never be thrown away near the stores and condemned foodstuffs should not be dumped nearby. Removal of seeding grasses and other plants from areas adjacent to the store is a helpful measure to reduce bird infestation.

Egg collection and pest destruction

Regular attention to the destruction and removal of eggs and nests will help to reduce the resident bird population. This needs to be carried out regularly because birds are very reluctant to change nesting sites, even after disturbance, and they tend to return again to the same place.

Traps

A smaller portable version of the modified Australian crow trap (1 m long x .05 m wide x .75 m

high mesh wire cage with a V-shaped top with slots trough which birds can enter) has been tried and was partially successful. This is a very time-consuming method because the traps need to be attended to twice daily. Cages should be baited with an attractive food material.

Foot Stickers (glue)

Slow drying plastic jellies can be applied to sedges where birds roost. These give the perching bird an insecure sensation. Grease can also be used for this purpose. The jellies are applied to horizontal surfaces only, using a caulking gun. The jelly remains effective until clogged with dust residues.

Shooting

This is of limited value but may help to prevent bird roosting in certain areas.

Noise

Noise, and other forms of explosion are likewise of limited value.

Ultra-sonic disturbances and recorded bird call (warning cries and distress call)

These are not reliably effective indoors because the sounds are reflected off in internal surfaces. This causes a modified signal to which birds eventually become accustomed.

In general, physical methods are capital-and labor-intensive especially if birds stay above the piles.

Biological Methods

In the field, bigger predatory birds and other animals feed on the weavers. Rodents and cats may also be considered as predators inside stores. Even people catch them at their roosting places at night for food or for trade.

Chemical Methods (poison baiting)

These methods should be considered as a last resort if all appropriate alternative methods have failed. Baiting involves the use of avicides which are potentially hazardous materials and these should be introduced only where safety and supervision can be kept under control. The use of bird poison or pesticidal products intended for other purpose is not recommended. It is unnecessary and dangerous. The food bait for the poison has to be very carefully chosen and it must suit the species of bird. For example, pigeons prefer peas, wheat and cracked or whole corn; sparrows prefer small grains such as palay. All these birds will take bread or cake very readily. The bait offered must, however, be, more attractive than available foods. Pre-baiting 34 days increases the chances of success. For pre-baiting, the same sort of food to be used for the poison bait (but lacking the poison) it is put out in the same place each day.

Repellents like methiocarb discourage birds from further feeding on the seeds. Alpha-chloralose is a material which at low doses has the effect of stupefying birds without killing them. Thus, it is possible to immobilize, collect and remove troublesome birds. They can be released at a safe distance from the warehouse, or destroyed if they are not protected species. As an advantage of this it is that if non-target species happen to consume the bait, they too will be only temporarily

affected. When used at a higher dosage rate, alpha-choralose acts as acute poison and there is no recovery. The concentration of poison to be incorporated in the bait to produce either stupefaction or mortality requires careful determination under local conditions.

Neurological frightening agents such as 4aminopyridine prepared in grain baits have been used successfully. The material (4-AP) causes birds to emit distress cries and perform erratic flight displays. The bird showing these symptoms will die but it may be sufficient to kill only few birds because the affected birds serve to scare away the rest of the flock. Passer montanus is more susceptible to 4-AP than methiocarb. This species also gave the most audible and greatest number of distress calls in response to 4AP (Garrison, et. al., 1981). The use of 4-AP against sparrows in paddy fields and storage warehouses shows promise.

Chemosterilants are used to sterilize a pest population of birds thereby controlling its breeding. The method, however, has disadvantages. The less-toxic chemosteritants are too short-term in action whereas those with more persistent effects are much more toxic. Furthermore, since there is no immediate reduction in population, birds continue to cause a nuisance for some time and may even propagate further because additional birds are attracted to the site by the bait.

None of these agents should be placed where poultry and other animals have access to them. If affected birds are eaten by cats or dogs, they cause secondary poisoning of the predator. For all chemical control measures, a high degree of supervision is essential. This must include regular and frequent collection of dead birds (more than once a day), the burying or destruction of the dead birds, the final removal of all treated bait and the proper disposal of unused bait.

CONCLUSION

The primary objective of bird control in storage should be to prevent or reduce grain losses and not merely kill animals. Effective bird-proofing of stores is strongly recommended for use in the first instance. Sanitation, elimination of harbourage, proper stock and warehouse maintenance and removal of bird nests should be standard practices. Quick turn-over of stocks also reduces the exposure time of grain to the pests. If this is done, then the use of other control measures will, in most cases, be found unnecessary. If it becomes essential to introduce other measures, these must be planned and carried out so as to 1) avoid public concern, 2) pose no hazard to non-pest species, man or domestic animals.

[Table 1. A Guide to the Identification of Philippine Weavers](#)

[Table 2. Some notes on the biology and behaviour of Philippine weavers](#)

Post-harvest microbial infection of cereal grain

by Lina L flag*

Spoilage of grains after harvest is due to the interaction of prevailing environmental conditions and the various organisms that attack and contaminate the grains. The environment factors are temperature, moisture, light, gases, and chemicals that may be present. The organisms responsible

for grain deterioration are rodents, insects, mites and microorganisms. Changes in the non-living environment provide conditions that may either stimulate or inhibit the activity of the living organisms and thus enhance or prevent spoilage as the case may be.

Our main concern here is the spillage of grains by microorganisms.

Grain spoilage is brought about by any of the following microorganisms:

- 1. Bacteria, which morphologically appear as a single cell or groups of single cells that may be spherical or rod-shaped. They reproduce by binary fission or simple cell division. They need a natural opening or wound to gain entrance to the host as they cannot penetrate the intact grain.**
- 2. Actinomycetes, which are closely related to the bacteria but with elongated cells and form branches. These are usually saprophytic. Many form antibiotics (such as neomycin and streptomycin) which inhibit or kill other microorganisms in a storage bin.**
- 3. Yeasts, generally appear as single cells and multiply by budding. They predominate in sealed silos where the oxygen supply is low and the moisture content is high. They may impart a fermentation or yeasty odor to the grains.**
- 4. Molds, fungi that have filamentous vegetative structures called mycelium. Reproduction is complicated. They form sexual spores (after the fusion of two compatible cells) as well as asexual spores (without previous fusion of cells). The spore is more resistant than the mycelium to adverse conditions. When a spore lands on a substrate (such as grain of rice) it germinates and sends out**

thread-like structures (mycelium) which grow and branch and soon colonize the entire grain, and later produce more spores to complete the cycle. Spores are continuously present in the soil and air, and are spread by air currents, insects and other agents.

Deleterious Effects of Microbial Contamination

1. Loss of Seed Viability:

The germ or embryo is a preferred site of microbial attack because it is delicate, thin-walled and it contains the nutrients needed for microbial growth. Once the scutellum is affected, the food supply for the germinating plant is destroyed.

2. Altered Nutritional Value:

The formation of free fatty acids, lowered protein digestibility and vitamin changes have been associated with deteriorative changes in grains brought about by microorganisms.

3. Health Hazards:

Grain dust contains fungal spores and bacterial cells along with a number of substances that cause a variety of symptoms and ailments such as respiratory disorders, eye irritations, skin itchiness, allergies, etc. *Aspergillus fumigatus* can cause lung diseases in man when spores are inhaled. *A. flavus* produces aflatoxin which is carcinogenic and poisonous to several animals, possibly including man.

- 4. Spoiled grain causes a reduction in the milling yield and milling quality. Such grains are very friable.**
- 5. Contaminated grains are often unpalatable with a musty or sour flavor.**
- 6. Microbial infection may cause of granular products.**
- 7. Molds may cause deterioration of packaging and sealing materials which often result in grain spillage.**
- 8. Microorganisms cause heating of the grain. As the microorganisms respire, they emit heat and moisture which usually make the micro-environment more favorable for further microbial growth. The formation of hot spots in bulk-stored grains is mainly due to the metabolic heat from microbial respiration.**
- 9. Certain microorganisms bring about grain discolorations such as blackening, reddening and yellowing. Yellow grain has become a major problem in the humid tropics. The percentage of yellow grains is high in wet palay that had been left unthreshed for a few days. The high moisture in the unthreshed palay allows for rapid microbial growth. This results in the release of metabolic heat from the respiring molds, causing an increase in the grain temperature in the unthreshed pile. The high temperatures, along with the various secondary metabolites released by the microorganisms, are the likely causes of grain yellowing.**

Factors Affecting Microbial Growth and Development

1. Temperature

Temperatures from 20 to 40C favor the growth of most microorganisms. However, some may grow at as low as -90C (psychrophilic) and at as high as 80C (thermophilic). Temperatures below 10C generally inhibit microbial growth but is not lethal in most cases. Thus, a return to favorable temperature for growth, after a period of cold storage, often results in resumption of luxuriant growth.

2. Moisture Content of Grains

Microorganisms are typically moisture loving and a certain amount of moisture in the substrate is required for growth. The amount of moisture determines the type of organism that is capable of growing in the grain, and the rate at which it grows. Among the various microorganisms, bacteria and actinomycetes are the most hydrophilic followed by the yeasts, and finally the molds which are more droughtresistant. The microorganisms are inhibited at moisture contents in equilibrium with below 70% relative humidity. Fungal spores are not destroyed by dry conditions; their germination and growth are merely inhibited until the moisture in the grain rises to a favorable level.

3. Length of Time the Grain is Stored and Prevailing Storage Conditions:

The expected storage life of the grain is inversely related to temperature and moisture content. The lower the storage temperature and moisture content, the longer the storage life. Cleanliness in the storage bin and warehouse is a prime requisite in prolonging grain storage life.

4. General conditions of the Grain:

Microorganisms can easily penetrate and initiate infection in damaged or cracked grains. Grain that had been previously invaded by bacteria and fungi is more easily prone to damage than grain that has had no contamination.

5. Presence of Insects and Mites:

Insect infestation usually accompanies mold invasion. Insects and mites serve as vehicles for the spread of molds and other microorganisms. Insects are attracted to grains that harbor molds because the insects often feed on the molds as well as on the grain.

6. Oxygen:

The biochemical oxidation of food materials to obtain energy for life processes (respiration) is carried out by some microorganisms only in the presence of oxygen in amounts close to those present in air at atmospheric pressure. Other microorganisms can do with less oxygen and still others grow without oxygen. The molds are strongly aerobic so they grow profusely on the surface of the commodity where atmospheric oxygen is in ample amounts. Generally, yeasts and bacteria can do with less oxygen than the molds (microgerophilic).

7. Light:

The ultra-violet portion of the spectrum is lethal to microorganisms, but due to its low penetrating

power, only those organisms present on the surface of the commodity are killed. Many microorganisms grow best in the. dark although some require light for sporulation.

CONTROLLING MICROBIAL SPOILAGE OF GRAINS

Since microorganisms are present everywhere, there is no practical means of totally eliminating them. However, they may be kept under control by providing conditions that will innibit growth. The following are recommended:

- 1. Dry harvested crops promptly to a safe moisture level (13-14% for maize and rice). This is by far the most practical means of preventing microbial spoilage. Prevention is the key control measure because once invasion occurs, with the subsequent deteriorative changes such as aflatoxin contamination, practically nothing can be done to improve the quality of the grain or to remove the toxin. After the grain has been dried to the desired moisture level, rewetting or moisture absorption should be prevented.**
- 2. Provide aeration in bulk-stored grain to provent moisture condensation which is highly favorable for microbial growth. Aeration cools the mid-portion of the stored bulk and lowers temperature uniformly throughout the stored grain. This prevents moisture migration. Aeration should, however, not be done at night or when the relative humidity is high because the grains may take up moisture. Increased moisture content will then favor the growth of microorganisms.**

3. Prevent insect and mite infestation. In addition to the direct damage they cause, these pests disseminate fungal spores and activate the growth of molds.

4. Low temperature storage (at 10C or lower) inhibits microbial growth but this method may be expensive and impractical for bulk stored cereals.

5. Storage in air-tight structures has been studied for the control of mold growth. However, bacteria and yeasts may develop under these conditions which often results in off-odors to the commodity. This renders the grain unfit for human consumption because the objectionable odor may be carried over to the processed products.

6. Fungicidal treatment to keep the molds in check could be expensive and there is the added problem of toxic residues that may render the food or feed unsafe for consumption. Grains intended for seeding purposes may be chemically treated. Fungicides may also be difficult to apply and may adversely affect the processing of grain.

Mold growth can be controlled by 1-2% ammonia in high moisture corn (22-28% moisture content) for 6 months. However, bacterial growth may continue after an initial drop in their number. Calcium propionate or sodium propionate at 5000 ppm can lengthen the storage life of rough rice.

Drying, coupled with proper ventilation or aeration appears so far to be the most effective, the safest, and the most practical means of maintaining the quality of grains. Periodic sampling and testing for moisture content, temperature and presence of microorganisms will allow the detection of potential trouble before anything serious occurs.

Commodities that yield toxin-forming fungi may not necessarily contain toxin(s) because morphologically identical strains vary in their ability to form a toxin and the fungi observed may just be superficially present on the commodity. On the other hand, grains that appear free of molds may be contaminated with a toxin if the toxin-forming mold had been eliminated through desiccation, high temperature or by some other means.

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I. INTRODUCTION

Despite the increase in food production due to technological advances in agriculture, enormous quantities of harvested food are wasted due to inadequate protection of stored products. According to are FAO estimate, the losses due to insect infestation is about 10 per cent or more in developing countries. The losses can be potentially greater now with the increasing attention being given to the establishment of national and international buffer stocks of foodstuff to guard against irregularities in production due to the unpredictable climatic conditions.

The popular outcry against the use of synthetic pesticides is agriculture cannot overshadow the necessity of their use nor can any speedy decline in their use be foreseen, so that protection of the consumers of treated produce and education of the users of the chemicals is imperative. Chemical pest control methods, if carried out intelligently and knowledgeably, can be both effective and safe. It is extremely important for users to have a knowledge of the classification, mode of action, properties, metabolism and residues of the pesticides, to enable them to make proper appraisal of the benefits and potential hazards of the pesticides. Thus, they should be able to choose insecticides judiciously and formulate efficient control measures in any particular set of circumstances.

II. CLASSIFICATION OF INSECTICIDES

Insecticides are classified according to their mammalian toxicity, chemical origin or composition, mode of entry, and formulation.

A-1. Mammalian Toxicity

Toxicological studies are conducted to determine the threshold limit of a chemical which an animal or human is capable of handling without significant biological effects. The usual beginning in any toxicological evaluation is the assessment of the acute toxicity, i.e. the effects of a single dosage of the chemical. The general technique is the determination of the LD₅₀ (the dosage necessary to produce death or reproducible effect in 50% of the animal population tested). The compound is administered on a weight/weight basis (milligram or gram of compound per kgm of body weight of test animals) in a suitable solvent or suspension system. This is evaluated by acute tests, orally (AO) or dermally (AD); chronic oral tests (CO), Vapor toxicity tests (VA) and chronic vapors tests (VC) or inhalation tests (IT).

Insecticides can be classified according to their toxicity based on the LD₅₀ values:

1. Highly toxic

AO LD₅₀ = 0-50 mg/kg

AD LD₅₀ = 0-200 mg/kg

IT LD₅₀ = 0-2000 ug/l

Danger, skull and crossbones and poison on label.**2. Moderately toxic****AO LD₅₀ = 51-500 mg/kg****AD LD₅₀ = 201-2000 mg/kg****IT LC₅₀ = 2,001-20,000 ug/l****Warning on label.****3. Slightly toxic****AO LD₅₀ = 501 -5000 mg/kg****AD LD₅₀ = 2000-20,000 mg/kg****IT LC₅₀ = more than 20,000 ug/l****4. Relatively non-toxic****AO LD₅₀ = 5000 + mg/kg****AD LD₅₀ = 20,000 + mg/kg**

Generally, the insecticide used in stored product treatment is of low mammalian toxicity (Table 1) in a formulation that is likely to be effective against the species involved, persistent for the

required period of time under given storage conditions and will not alter the flavor, color and odor of the stored commodity.

A-2. Chemical Origin of Composition

A-2.1. Inorganic compounds - The toxicity of these compounds (arsenicals, flourides) are usually associated with the concentrations of elements. They are highly toxic to man, domesticated animals, and plants. They accumulate in the soil.

A-2.2. Organic Insecticides - These groups are characterized by organic carbon bonding (i.e. c-c; c = c).

a. Botanicals - The toxic principles are extracted from plants such as pyrethrum from the flowers of *Crysanthemum cinerariaefolium* and *C. cocciniu*. It has remarkable low toxicity to mammals but toxic to insects.

b. Synthetic insecticides - They are syntihesized in the laboratory and are classified into three main groups: the organochlorine, organophosphates, and carbamates.

b-1. Organochlorines - These are chlorine (C1) - containing compounds further subdivied into DDT type, Hexachloro-cyclohexane type and Cycloclines. Most of them are quite toxic to man and are not used on stored food commodities.

c. Organophosphates - This is a generic term for all pesticides containing phosphorus which can be

an ester of phosphoric acid (P = O) or phosphorothioate acid (P = S) and can be represented by this formula:

Formula

The formula implies that sulphur (S) or oxygen (O) is directly linked to phosphorus. R1 and R2 may be alkyl or aryl groups or amine radical whereas X is the acyl or leaving group which may be radical of an inorganic acid. This is a very large class of compounds which features a greatly varying activity despite the very uniform mechanism of action. The organophosphorus compounds, however, decompose rapidly and recent advances in understanding the mechanisms of selective toxicity of insecticides such as malathion have led to safer insecticides such as malathion, bromphos, pirimiphos methyl, chlorpyrifos methyl, etc.

d. Carbamates - These are esters of carbamic acid, HOC (O) NH₂ and can be represented by this formula:

Formula

R1 can either be an aliphatic or aryl radical. This contains compounds of high to low mammalian toxicity such as carbaryl and have so far shown limited application in stored product pest control.

e. Insect Growth Regulators - These groups of compounds are synthetic analogues of naturally occurring hormones in insects, ecdysone and juvenile hormones. Ecdysone regulates metamorphosis by initiating the moulting process while juvenile hormones regulate growth and

development under normal concentrations. These growth regulators (IGRs) also control other developmental processes in insects such as sexual maturation, colour differentiation and reproduction. Examples are difludenzuron and methoprene.

A-3. Mode of Entry

Insecticides can be divided into three main groups depending upon the way they penetrate into the body of the insect.

A-3.1. Contact insecticides - These insecticides are applied in such a manner that they come in contact with some part of the body of the insect; the compound is able to penetrate the exoskeleton and is transported to the tissues via the circulatory system. Most of the insecticides used in storage belong to this group. The inert insecticidal dusts which disrupt the thin epicuticle leading to the dessication and death of the insect are included in this group.

A-3.2. Stomach insecticides - These materials exert their toxic action only when they are consumed and absorbed through feeding on treated surfaces through the guts.

A-3.3. Systemic insecticides - These are translocated to the untreated parts of plants or animals in concentration that makes the final translocation site toxic to insects. These are not used in stored product pest protection.

A-3.4. Fumigants - These are insecticidal gases at normal temperatures penetrating through the tracheal system into body tissues and are used in enclosed spaces. Examples are phosphine and

methyl bromide. Some contact insecticides like dichlorvos, vaporize partially in warm ambient conditions, thus having fumigant and contact actions.

A-4. Mode of Action

A-4.1. Physical insecticides - These materials such as the heavy mineral oils and inert dusts, characteristically exert a physical rather than a biochemical. Mineral oils exert a purely asphyxiant effect and dusts affect loss of body moisture by abrasion (aluminum oxide) or by absorbing moisture (charcoal).

A-4.2. Protoplasmic insecticides - The action of these compounds is associated with the cellular destruction of the midgut epithelium such as inorganic insecticides.

A-4.3. Respiratory poisons-These include the fumigants and those that block cellular respiration such as rotenone. Rotenone inhibits the catalytic action of cytochrome oxidase and other Fecontaining oxidase.

A-4.4. Cholinesterase (CHE) inhibitors - The OPs and carbamates, inactivate the cholinesterase, consequently causing sickness or if applied at correspondingly high dosages, death of the affected organisms.

The CHE are a group of esterases which are capable of hydrolyzing acetylcholine, a chemical transmitter of nerve impulse. Normally, acetylcholine is rapidly hydrolyzed to acetate and choline in insect or in vertebrates in the presence of cholinesterase. But, with an inhibitor (either OP or

carbamates) there is an accumulation of acetylcholine, which will cause an excessive stimulation (twitching) and finally complete block of the system (paralysis). Depression of the cholinesterase of the central nervous system results in restlessness, discomfort, giddiness and anxiety, followed by a headache, sleeplessness, ataxia, tremors, ultimately resulting in coma, generalized spasms and disappearance of reflexes.

A-4.5. Neurotoxicant - These materials act on the central and peripheral nervous system by changing the required balance between input and events. There is an excessive stimulation which later will cause excitation, convulsion, paralysis and death - these are characteristic symptoms of nerve poisoning. To this group belong the organochlorine insecticides, nicotinoids and pyrethroids.

A-4.6. Enzyme inhibitors - These compounds (fluorides, arsenates) inhibit enzymes necessary for normal metabolism. Fluoroacetate blocks the tricarboxylic cycle by combining with acetyl CoA forming fluorocitrate which inhibits aconitase which convert citrate to succinate. The blockage leads to reduced energy production and O₂ utilization resulting in respiratory disorders causing death. Arsenates and nitrophenols kill primarily by inhibiting respiratory enzymes which block the production of AIP (energy).

A-4.7. Insect growth regulators - There are two types of IGRs: the chitin synthesis inhibitors such as diflubenzuron and the antijuvenile hormone such as methoprene that disrupts the moulting process. Chitin which is essentially a polymer of N-acetyl glucosamine is a structural component of the insect cuticle essential for proper insect development. Diflubenzuron interferes with the larval cuticle deposition and disrupts the moulting process by inhibiting the synthesis of chitin. The function of hormone is to maintain the larval tissues at a moult. The presence of JH mimics such as

methoprene in the insect when the natural hormone level is low would be expected to disturb normal morphogenesis.

Insects exposed experimentally to large concentrations of IGRs during the life cycle when they are not normally active, inhibit various developmental and morphological abnormalities including a juvenilizing effect.

A.5. Usage in Stored Grain

Insecticides used on or around grain may be classified based on usage:

- 1. Knock down agents - i.e. Dichlorvos**
- 2. Surface sprays - i.e. Bioresmethrin**
- 3. Structural treatment - i.e. Fenitrothion**
- 4. Grain protectants - i.e. Malation**
- 5. Fumigants - i.e. Aluminum Phosphide**

III. PROPERTIES OF SOME INSECTICIDES

The selection of insecticides for treatment of edible commodities is based mainly on the toxicological data (low mammal fan toxicity), effectiveness and persistence under certain storage conditions and absence of side effects such as discoloration, flavor alternation and odor. FAO and

WHO collect toxicological information and give advice on overall tolerance figure. However, the maximum levels of pesticide residue acceptable in grain consumption must be calculated by local authorities who have relevant information on the inert feeding habits and agricultural practices.

To qualify for selection as possible candidate material for use on or around grain, the insecticide must fulfill the following requirements (FAO, 1982):

- 1. it must be effective at economic rates of use;**
- 2. it must be effective against a wide variety of insect pests;**
- 3. it must present no hazards to consumers of grain and grain products and to users or applicators;**
- 4. it must be acceptable to health authorities;**
- 5. it must not give rise to unacceptable residues;**
- 6. legal maximum residue limit must be established;**
- 7. it must not affect the quality, flavour, smell or handling of grain;**
- 8. it must be acceptable in international grain trade;**
- 9. it must not be flammable, explosive or corrosive; and**
- 10. its method of use must be compatible with established grain handling procedures.**

Of the many insecticides reported to be toxic and/or effective against stored product pests, the number which have been cleared for application are stored grain and for which maximum residue limits are established is limited.

The common insecticides used in stored product pest control belong to four groups: the

pyrethroids, the organophosphorus; the organochlorines, and the carbamates.

A. The Pyrethroids

The Pyrethroids are either isolated from plants or synthesized.

A.1. Natural Pyrethrins

Pyrethrins remain one of the more widely used materials in stored product pest control. The rapid knockdown effect, a wide spectrum of activity against insect pests, a general acceptance of their use associated with foodstuff and an established codex of tolerance have been the principal reasons for their use. Their use has not been intensive as their cost has been prohibitive.

The disadvantage of high cost, poor stability, inadequate toxicity to some species and lack of ovicidal and acaricidal action have been offset somewhat by the use of synergists, i.e. piperonyl butoxide, sesamin, piperonyl cyclonene, propyl isomer, sesamex and sulfoxide. Synergists are usually present at ratios 1:3 or 1:10 (insecticide: synergist).

A.2. Synthetic Pyrethroids

The synthetic pyrethroids such as biores-methrin, deltamethrin, permethrin, fenvalerate and phenothrin are becoming acceptable over pyrethrins because of their high levels of activity against a wide range of pests and a cost advantage. Bioresmethrin appears to be the most commonly used. Several other synthetic pyrethroids showing promise against storage pests are under

development.

There have been deficiencies as with pyrethrins. For example, resistance is known although not extensive, and breakdown to malodorous decomposition products has presented a problem where repeated applications have been made on the same surface. In addition, like pyrethrins, control of *T. castaneum* has been less than desirable. Pyrethroids are highly summarized with piperonyl butoxide.

A-2.1. Bioresmethrin - It is one of the most potent broad spectrum insecticides currently available and has a good knockdown performance against insects. Bioresmethrin, at low concentrations, is an effective killing agent against most insect pests attacking households, industrial premises and food storage. Bioresmethrin is currently being used as:

- a. household aerosols and sprays formulated in combination with pyrethrum, bioallethrin, tetramethrin and piperonyl butoxide;**
- b. an insecticide for the control of pests in food premises; and**
- c. in grain disinfestation and protection.**

Bioresmethrin has an exceptionally high potency against some insect species, particularly *Rhizopertha dominica* and it has proved useful when applied at the rate of 1 mg/kg in conjunction with selected organophosphorus insecticides, enabling the amount of OP to be reduced considerably without loss of effectiveness. It has lower toxicity against *Sitophilus granarius*,

Tribolium castaneum and moderately toxic against mites. The toxicity of bioresmethrin can be improved to a significant extent with piperonyl butoxide, the factor of synergism ranging from 2 to 9 fold. Bioresmethrin at 1 mg/kg plus fenitrothion at 12 mg/kg controls typical malation-resistant strains of *S. oryzae*, *R. dominica*, *T. castaneum* and *Ephestia cautella*.

A-2.2. Fenvalerate - It is an ester related and in many ways similar to pyrethroids. It is a highly active broad spectrum insecticide with adequate stability and relatively low mammalian toxicity. It has been shown to be effective at low doses against *R. dominica* and at higher doses against most species such as *Sitophilus* and *Tribolium*. Fenvalerate is an effective alternative to bioresmethrin. It combines well with OP and its potency is synergized by the addition of piperonyl butoxide. Deposits on grain are stable, though the bulk of the deposit is removed with bran or hulls. Those residues which carry through the white flour or milled rice remain substantially undiminished following cooking. Fenvalerate at 1 mg/kg along with fenitrothion at 12 mg/kg and piperonyl butoxide at 8 mg/kg control common field strains of *S. oryzae* and *R. dominica* and completely prevent progeny production in *T. castaneum*, *T. confusum* and *E. cautella*. The same combination of fenvalerate control typical malathion-resistant strains of above -mentioned species.

Table 1. Acute oral LD50 (mg/kg body wt. rat) of insecticides used for storage pest control.

INSECTICIDE	ORAL (RAT)	DERMAL
1. Malathion***	1375-2800	4000-4800
2. Pirimiphos methyl***	2050	2000
3. Chlorpyrifos methyl*	1650-2100	3000

4. Tetrachlorvinphos*	4000-5000	5000
5. Bromophos*	4000-8000	2188
6. Dichlorvos* *	80	107
7. Fenitrothion* *	250-500	3000
8. Diazinon*	300-850	2150
9. Iodofenphos*	2100	-
10. Phoxim*	1845	7100
11. Etrimfos*	1800-2040	-
12. Lindane*	88	1000
13. Methoxychlor*	5000-7000	2820-6000
14. Carbaryl *	400-850	4000
15. Pyrethrum * *	1500	1800
16. Bioresmethrin***	9000	10,000
17. Deltamethrin	1290	2940
18. Fenvalerate*	450	3700-5000
19. d-Phenothrin*	5000	5000
20. Resmethrin*	1500	3040
21. Permethrin *	4000	4000

22. Methoprene*	5000	relatively nontoxic
23. Piperonyl butoxide	relatively nontoxic	relatively nontoxic

***Occasional Use**

****Moderate Use**

*****intensive Use**

a Used as synergist

B. Organophosphorous compounds

There are severe, organophosphorous compounds (OP) used in stored product pest control. The most common are malathion, dichlorvos, fenitrothion, pirimiphos methyl and chlorpyrifos methyl.

B-1. Malathion - Malathion is the only OP that has been widely used for over 20 years for the routine protection of stored products especially cereals in practically every country in the world. It is effective against the many destructive pests of stored products at 5 to 20 mg/kg. It is virtually ineffective against stored product moths and requires higher dosage to control non-resistant R. dominica.

Malathion is intensively used in developed exporting countries such as Australia, Argentina and USA. Whereas before, malathion (premium or deodorized grade) was the most important material for admixture with grain, increasing use of dichlorvos was evident. Due to development of insert

resistance, there appeared to be a significant move away from malathion for disinfestation of storage facilities and a less marked but noticeable change to use alternative materials for treatment of bags.

The amount of the malathion deposit which penetrates the individual grains is relatively small and therefore most of the deposit is removed in the milling of wheat and rice. More than 95% of the deposit on the raw cereal grain is removed or destroyed before the cereal food reaches the consumer.

Malathion is used as admixture in dust or spray form; for building and fabric treatment; floor wash; and surface treatment of bags and grains and associated buildings and fabrics.

B-2. Dichlorvos - It is the most commonly used material next to malathion. It is an extremely effective stored product insecticide due to its high vapour pressure and is used as fumigant rather than as contact insecticide. Enclosed spaces such as warehouses, storage and grain bins allow build-up of air concentrations of vapours toxic to most flying and crawling stored product insects. The vapour will not penetrate into grain masses, other commodities or the fabric of buildings. The principal uses for dichlorvos are in aerosol-dispensing units which could be programmed for automatic daily release, usually at dusks; space sprays or fogs; slow release formulations in which dichlorvos is dissolved in solid strips (or beads) of polyvinyl chloride plastic suspended in the free space of storages; surface application of concentrates on wooden floors; surface treatment for protection of bagged commodities; and direct application to grain either as a surface treatment for moth control or for admixture with grain as disinfestation treatment alone or mixed with residual protectants.

Dichlorvos has the advantage over other residual insecticides in that it is considerably more active against immature stages of pests that develop within individual grains. Whereas malathion, for example, will kill only first instar larvae of *S. oryzae*, dichlorvos will give a significant kill of all larval stages except final instar larvae and the pupal stage. Resistance from *R. dominica*, been detected in low levels.

B-3. Fenitrothion - It is a broad spectrum insecticide with a much lower acute mammalian toxicity than many similar insecticides. It is widely used for the control of pests of many crops and principally as a residual spray in houses for the control of mosquitoes; pests of forest trees and for the management of locust swarms.

Fenitrothion has been used for a considerable time for structure, treatment and for surface treatment of bag stocks particularly where malathion resistance was present. Fenitrothion is considerably more effective than malathion against *Sitophilus* spp. and *Lepidoptera* and of comparable effectiveness against *Tribolium* spp. but not fully effective against *P. dominica*. High potency and good stability mean that deposits in the region of 5-10 mg/kg are sufficient under most storage conditions to give complete protection for 9 to 12 months. When combined with pyrethrum or synthetic pyrethroids, the effectiveness of fenitrothion is increased and the dosage level can be reduced. Fenitrothion is more effective than malathion for conditions where it is generally applied in the form of a very dilute dust. There is minimal penetration into the grain so that the deposit is mostly removed in bran of wheat and husks of rice.

B-4. Phosphoromethidate - It is a fast-acting broad spectrum OP with both contact and fumigant action. It gives long lasting control of insect pests of inert surfaces such as wood, sack and

masonry. It retains its biological activity when applied to stored agricultural commodities including raw grain, nuts, pulses, dates and cheese.

Pirimiphos-methyl has been used in many situations against stored product pests. The minimum effective dose against a wide range of insects is lower than most other OP on use or under development as grain protectant. It is potent against beetles, weevils, moths and mites, but not sufficiently effective against some strains of *R. dominica*. It is useful against immature stages within the individual grains and it appears quite effective against many lindane-malathion-resistant strains. In the Philippines, pirimiphos-methyl was found to be an effective protectant of corn grains against a variety of pests especially *Sitophilus* spp. for 6 months. Pirimiphos is more persistent in maize than in sorghum. Pirimiphos methyl-impregnated sacks are more effective than malathion for the control of storage pests of shelled corn.

B-5. chlorpyrifos-methyl - It is a broad spectrum organophosphorous insecticide of relatively low toxicity and moderate persistence. It shows reasonably good stability in stored products such as grain and dried fruits. In these products it controls a wide spectrum of beetles, weevils, moths and mites including several species which may have developed resistance to insecticides.

Chlorpyrifos-methyl is potent against all storage pests except resistant *B. dominica*. It is effective against moths which are not readily controlled by malathion. Deposits on grains and sacks are stable under most storage conditions. Studies in the Philippines show that it is a grain protectant more potent than tetrachlorvinphos, pirimiphos-methyl, malathion, MIPC against *Sitophilus* spp. for grain use. However, it was not as effective as tetrachlorvinphos against *B. dominica*. In general, the residual toxicity increased with higher concentration. Chlorpyrifos methyl was the most stable

among the five OPs, evaluated in corn and sorghum.

B-6. Tetrachlorvinphos - This compound has been used in the field for control of non-storage pests for considerable time and it is only recently that it is being evaluated as grain protectant

Tetrachlorvinphos has a very low level of toxicity and is a suitable compound for the protection of foodstuff. It has been shown to be effective against many species of stored product pests, both the immature and adult stages. It was also found to be highly stable in dry grain and posed no odour problem. In the Philippines, it was found to be effective as preharvest spray in sorghum at 2% for controlling *S. zeamais* and *Rhizopertha* in the field. It is more effective than malathion and pirimiphos-methyl and equally effective as chlorpyrifos ethyl against *Rhizopertha*.

Tetrachlorvinphos deposits do not penetrate the individual grains to any extent and appear to be removed on husks and bran.

B-7. Metacrifos - The compound acts as a contact, vapour and stomach poison against all important arthropod pests of stored products. It is also highly effective against major malathion and lindaneresistant insects. It is one of the few compounds that is effective against *R. dominica*. It is particularly useful where grain temperature can be regulated and where aeration of the grain mass can take advantage of the high potency of methacrofos vapour. It is marketed under the trade name Dam Fine.

Metacrifos penetrates the individual grains fairly rapidly and is therefore effective against larval stages within the grain. It is extremely potent at lower temperature and has a pronounced vapour action, but it degrades rapidly at high temperature and humidity.

B-8. Bromophos and others - Bromophos is used in limited scale as grain protectant and residual spray of warehouse facilities and bagged grain.

Other OPs that have been subjected to extensive study are diazinon, etrimphos, phoxim and iodafenphos. These compounds have been used as residual sprays but are currently registered as grain protectant

C. Organochlorines

As insecticides, many of the organochlorines are cheap and have excellent insecticidal property against many insects. These insecticides are photostable, and are resistant to degradation both in the environment and in biological systems. However, their resistance to environmental degradation and their stability after entering biological systems have led to general contamination of the world ecosystem. Also, because of its low cost and effectiveness, its widespread use has resulted in high levels of resistance in many insects. In addition, many of these chemicals exert profoundly deleterious long-term effects in animals, effects that were not known until after a long time that these chemicals were put into extensive use. Many organochlorines, including DDT and several to the cyclodienes have been shown to induce the formation of tumors in laboratory animals fed with a low dietary dose. Aside from tumorigenicity, it has been shown to upset reproduction in birds, and mammals.

Organochlorines that have been used for postharvest treatment were DDT, lindane and methoxychlor. DDT was used in foodstuffs since its introduction in the early 1940s. When its persistence and tumorigenicity effects were uncovered, its use has been stopped. Methoxychlor is

a DDT analog where the chlorine is substituted with methoxy (CH₃O) groups which render it more rapidly degraded by sunlight but is slowly converted to methoxy-DDE. But unlike DDT, it does not accumulate in fatty tissues. Although it has a very low mammalian toxicity (5000-7000 mg/kg rat) its use is limited because of its high cost.

Lindane (gamma-BHC) replaced DDT which found a limited use in the immediate post-war years in postharvest treatment. It has many of the desirable traits required especially for stored pest control; a wide spectrum of insecticidal activity in both beetles and moths and reasonable effectiveness against mites; stability under a wide range of conditions; a significant vapour pressure allowing some fumigant effect; repellency in some circumstances; and a comparatively low mammalian toxicity. The major uses for lindane apart from seed dressings have been in surface treatment of bagged stocks of grains, coffee and cacao beans and particularly, in treatment of warehouse and transport facilities.

It loses much of its effective residual life through evaporation and is very susceptible to dehydrochlorination when applied to alkaline surfaces.

D. Carbamates

Many of the properties of carbamates such as mode of action, lack of toxicity, lack of environmental persistence and lack of safety to beneficial insects are similar to organophosphates. However, the inhibition of carbamates is less permanent than with organophosphates. This means that at least for man, these insecticides are less dangerous than organophosphates as they have had a better safety record under practical use conditions.

For carbamates have been proven useful for storage pest control in the past, and the one that has carbaryl - has been largely replaced by the synthetic pyrethroids for controlling Rhyzopertha. Carbaryl, like other carbamates, is rapidly degraded into watersoluble, hydroxylated products which are easily conjugated to glucosides.

E. Others

Methoprene - This compound is the most commonly advanced example of the class of IGRs with juvenile hormone activity. This is available under the trade name Altosid. Methoprene is effective as protectant at 5 mg/kg against *Plodia interpunctella*, *Lasioderma serricorne*, *R. dominica*, *Oryzaephilus surinamensis* and *mercator*, and at 10 mg/kg against *Ephestia cautella* and *T. castaneum*. The juvenile hormone analogue is relatively non-toxic and has moderate stability under storage conditions but is rapidly decomposed by light. It is expensive and effective against few insect species.

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Insecticide formulations

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INTRODUCTION

In the commercial development of insecticides, one of the first steps that a manufacturer enplays is to chemically produce the compound in a form which is called the "technical grade material". This technical grade material or toxicant may be sold in solid form such as crystals, or powder, or in a liquid or gas forms.

Since it is not normally used in these forms for insect control, the technical grade material must be formulated. Formulation involves processing the technical grade material by any method that will improve its effectiveness, storage, handling, safety and ease of application

A. Formulation Process

This is usually accomplished by grinding the material to a powder or dissolving it in a petroleum solvent. The toxicant may then be diluted with other substances to make the desired formulation and is then known as the active ingredient, which is often abbreviated to "Ai"

The other substances which are added to the formulation are called adjuvants. An adjuvant is anything that enhances the physical properties of an active ingredient, are by itself it may have no killing properties. Some examples are xylene, talc, flur and bran.

The preparation and use of pesticide formulations also involve the use of various accessory agents

such as dust diluents, solvents, emulsifiers, wetting and dispersing agents, stickers, deodorants, and masking agents. Accessory agents are given names that denote their specific action or enhancement of the formulation. For example, a "spreader" would help spread the pesticide over a surface.

B. Properties and Ingredients

The physical form in which insecticides are purchased may be either a dry or a liquid formulation. One of the most important components of a formulation is the carrier. This is the substance which carries the active ingredient of the target surface. Carriers also may be either dry or liquid according to the formulation.

B-1. Dry Formulations

The commonly used dry formulations are: dust, granules, baits, wettable powders and soluble powders.

Dusts (D). They are in a dry, powdered form and usually contain from one-half to ten percent se. Most of the material in this formulation is an inert clay diluent or carrier. Dusts do not always adhere well to plants or animals or structures; and they are extremely subject to drift by the wind, therefore posing a greater toxic hazard to the applicator and the environment than many other types of formulations. For these reasons, dusts are usually recommended only for localized application, home control programs and storage system. Dilute dusts are used mainly as grain protectants.

Granular (G) These are large particles, dry formulations that usually contain two to twenty percent active ingredient. Various types of inert clay pellets, peanut hulls and corn cobs are often used as the diluent or carrier. The toxicant is applied to, and adheres to these granules. Granular formulations are easy to apply and do not drift as readily as other formulations. Granules are used widely for spot and broadcast soil applications and are often applied at planting time to protect the roots from soil insects.

Baits (B). Baits contain a low percentage of active ingredient ranging from 1/4 to 5%. The toxicant is mixed with various carriers or attractants such as bran, orange pulp, corn cobs and sugars. The bait is placed or scattered where it will be consumed by the target pests. Baits are commonly used for subterranean soil pests such as ants, mole crickets and cutworms.

Wettable Powder (WP). The AI in a wettable powder usually ranges from 25 to 75%. The AI is essentially a concentrated dust which has been finely ground to a powder mixed with a fine claylike diluent or carrier. While a WP is a dry formulation, it is mixed with water which acts as the secondary carrier. Most AI of a WP formulation are immiscible with water. This incompatibility is overcome by adding a bipolar compound such as a wetting agent. The wetting agent ties the AI and diluents with the water carrier to form a suspension. However, this WP suspension is unstable and it must be constantly agitated to prevent settling and ensure its desired effectiveness.

Soluble Powder (SP) - These are finely ground, highly concentrated powders containing 75 to 90% se. The powder is soluble in water and needs no wetting agent. Simple mixing of powder with water forms the spray. The potentially hazardous concentrated material is packaged in a soluble bag, and the entire package is placed into spray tank with water.

B-2. Liquid Formulations

Most insecticides that are applied as liquid or spray use water as the carrier. The following liquid formulations will be discussed: emulsifiable concentrate, oils, solutions, fumigants, aerosols and ultra-low volume.

Emulsifiable concentrates (EC). - EC are available from 20% to 80% AI/gal. (2 to 8 pds AI/gal.). The AI is dissolved in a petroleum solvent such as xylene and an emulsifier which allows the material to mix with water. The percentage of the emulsifying agent which is present in the insecticide is indicated by the inert ingredients. Emulsifiers and wetting agents are bipolar in that one end is hydrophobic and other end is hydrophilic. EC when mixed with water, forms a milky colored emulsion. This is generally stable for a period of several hours without agitation and should only be mixed in quantities that will be used immediately. EC are used for treating storage structure or fabric, external bag surfaces and disinfecting transport facilities.

Oil. These are sprays in which oil itself is the active ingredient. The oil may be refined to reduce phytotoxicity to plants and is usually mixed with an emulsifier so water may be used as the carrier. The percentage of oil in this type of formulation may range from 1 to 99%.

Fumigants. They are gaseous poisons which boil at room temperature. Application is generally limited to plants or products in tight enclosures or those that can be enclosed in gas-tight tents. Most are highly toxic and must be applied by trained, certified applicator.

Flowable Suspensions (F). - Flowable suspensions are an ingenious solution to a formulation

problem. Earlier it was stated that some insecticides are soluble in neither oil nor water, but are soluble in one of the exotic solvents, making the formulation quite expensive. To handle the problem, the technical material is blended with one of the dust diluents and a small quantity of water, leaving the insecticide-diluent mixture finely ground but wet. This "wet blend" mixes well with water and can be sprayed with the same tanksettling characteristic as wettable powder.

Liquified Gas Aerosols. - Liquified gas aerosols or bombs are the common method for producing small amounts of aerosols for indoor use. The aerosol is formed by the release of a solution of insecticide in a liquified gas through a capillary tube with very small diameter.

Ultra-low-Volume (ULV). - ULV is both a formulation and an application technique. These are usually sold as technical grade materials. They are not further diluted before application by special spray equipment. The extremely fine spray is applied at rates as low as one-half pint to one-half gallon per acre.

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[Contents](#) - [◀ Previous](#) - [Next ▶](#)

Methods of insecticide application for grain protection

[Contents](#) - [Previous](#) - [Next](#)

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APPLICATION METHODS

The choice of insecticide concentration, type and frequency of application depend on a variety of factors such as the species of pest present, the type of storage facilities, length of storage, local insecticide regulations, etc. Therefore, only certain generalized recommendations could be made.

Tables 2 and are a suggested guide for insecticide application (FAO World Food Program 1970; Bengston 1970; Morallo-Rejesus 1976). Dosage rates may be revised depending upon the local regulations, especially on the admixture treatment for food grain. The recommendation only serves as a guide, but the person in-charge is the one who will assess the requirements of any specific case, in order for him to choose the insecticide, dosage and method of application which best meet the situation.

RESIDUAL (STRUCTURAL) TREATMENTS

A residual spray is usually applied to inside surfaces of warehouses, storage bins, transport vehicles or other structural surfaces. A good residual spray should not only kill the insects should a deposit on the treated surface to kill walking Insects. Application may be made during the cleaning of storage facilities before intake of new stocks or to fit in with fumigation or spraying of the stock in storage. It is important to make sure that corners, ledges, cracks and other places difficult to get at are treated. The effectiveness of the residual deposit will decrease with time. The effective life of the deposit depends on the insecticide used, the climatic conditions prevailing and the type of surface sprayed.

Examples are fenitrothion, malathion, synthetic pyrethroids, tetrachlorvinphos, and chlorpyrifos methyl. WP are generally more persistent than EC but are less easy to apply. For most surfaces it is preferable to use the dispersible WP formulations of insecticides especially for absorbent surface such as cement, brick, stone or white washed surfaces. EC may be used on non-absorbent surfaces like metal or painted wood. Insecticides are much more persistent on wood. Malathion is not very satisfactory on alkaline surfaces, e.g. whitewash, bare, concrete or cement, but it has been shown to be effective for over 20 weeks on plywood and fiber board and has remained active through 16 up to 52 weeks.

Frequency of treatment depends partly on the insecticide used and partly on the infestation to be controlled. For example, Lindane and malathion treatments should be repeated at least every three weeks in tropical climates.

Pyrethroid and carbaryl are more effective than OP against *R. dominica*, while OP are more effective against *Sitophilus* spp. Azamethifos has been found effective against malathion-resistant

strains of stored product insects. Azamethifos applied on wooden, galvanized sheets and concrete surfaces at concentration ranging from 0.25 gm/m or 0.5 gm/m is stable for 32 weeks. It is also effective against resistant *Oryzaephilus surinamensis*.

SPACE TREATMENT

Space spraying or fogging of the warehouse is used to control infestations of flying insects that are not controlled by residual treatments, and of flying pests migrating from outside. It has to be carried out frequently and at a time of day when the pests are most active which is generally at dusk. The insecticides used are those with knockdown action. Examples are pyrethrin sprays and aerosols with or without synergists, synthetic pyrethroids with or without synergists, lindane smoke or fog, dichlorvos aerosols and strips.

Aerosols may be dispensed from the household type canister, containing a mixture of liquified gas and insecticide. The internal pressure "blasts" the insecticide into aerosol-sized droplets as the mixtures leave the nozzles. The sophisticated machines which produce aerosol droplets of insecticides are available in more advanced countries and will not be discussed here. Slow release dichlorvos plastic strip hung inside the warehouse at a density of 1 strip/30 cu m. space is also recommended to kill flying moths.

GRAIN PROTECTANTS

Grain protectants are defined as pesticides which are incorporated directly into the grain mass to protect it against insect and mite attack. This is also known as admixture treatment. The

insecticides used as grain protectants are of low mammalian toxicity and are generally safe to use and need only simple equipment for their application.

Grain protectants are usually applied as sprays directed into the grain stream during the movement of the grain or at the beginning of long-term storage. The most suitable equipment for spraying grain on a moving belt is a machine with an electrically operated pump, feeding in insecticide through a pressure regulating valve and a precision nozzle. Where a power supply is not available, a pressure retaining knapsack sprayer, equipped with a pressure regulating valve and precision nozzle, may be used. EC are normally used but the volume must not exceed 2.5 liters per 1000 kilos of grain to avoid any appreciable increase in moisture content. It is better applied through a precision apparatus. This is most efficient in bulk handling but could be practical in bag handling system. The use of small pumps with coarse spray nozzles and low pump pressure produce large spray droplets and gives good results while minimizing spray drift.

In the drip feed system, tiny quantities of the concentrate are dripped directly into the grain stream through microcapillary tubes.

For a small scale treatment of bulk or bagged grain dusts it is best to be admixed with simple mechanical aids (rotating drum, shovelling, etc.) but adequate mixing is difficult to achieve.

FUMIGATION

Fumigation is a widely used procedure particularly for the control of stored product insects because the fumigants diffuse and penetrate into places where other forms of control are

impractical or impossible. A fumigant is defined as a chemical which, at a required temperature and pressure, can exist in a prescribed period of time. Fumigation is the process of applying the gas under appropriate conditions to control the target organisms.

Materials with suitable characteristics for fumigation are limited. These vary widely in chemical composition and properties. consequently, the types of formulations, methods of handling, methods of analysis, and the purposes for which they are used vary considerably. The choice of fumigant depends largely on its ability to give effective and economical control of insects without adversely affecting the commodity.

All of the fumigants used to control pest organisms are toxic to human beings. They also may have other adverse properties - they may be highly flammable or corrosive; they may produce offensive odors; they may be phytotoxic; or they have leave harmful residues in food materials. Usually, adverse effects can be eliminated by choosing the most suitable fumigant for the particular treatment in question and by applying the proper methods of handling and use. A few of the fumigants are known to have or are suspected of having the potential for producing longterm chronic effects on human health and some are listed as carcinogenic. Appropriate precautions should be taken to avoid exposure to all fumigants and additional measures should be taken to prevent any contact carcinogenic compounds.

In applying a fumigant to a commodity, it is particularly important to carry out the operation in such a way that the insects are controlled without damaging the commodity and without creating any hazard for personnel. Effective methods of detection and analysis of the fumigant are especially important. Where any possibility of chemical exposure of personnel exists the

atmosphere should be monitored with appropriate equipment and threshold limit values (TLV) such as those recommended by the American Conference of Government Industrial Hygienists (1983-84) and should be strictly adhered to. Misuse or abuse of fumigants can lead to accidents that endanger human life or property, and consequently may give adverse publicity to the practice of applying chemicals to food commodities for insect control.

The most common fumigants that are extensively used throughout the world are methyl bromide and phosphine. According to Champ and Dyte (1976), methyl bromide fumigation in general was the most efficient pest control operation. The greatest single deficiency was its integration into pest control programmes. In many instances, methyl bromide fumigation was heavily relied upon that hygiene and stock management were neglected. As a result, reinfestation occurred immediately after fumigation was completed. Thus, frequent fumigation was necessary and the bromide residue was in excess of the permissible tolerance limit. Most usage involved is fumigation of bag stacks under gasproof sheets and in bulk with recirculation of air. It is used where short exposure periods only are practical and grain has a moisture content less than 11 %. FAO recommends 10 mg/kg body weight as acceptable daily intake.

Phosphine is a very efficient fumigant and its use complements that of methyl bromide. It is preferred in horizontal bulks of grain that can be probed with tablets and verticle storage where methyl bromide cannot be used. Exposure during fumigation must be of adequate duration and at minimum concentration of gas.

The other fumigants used occasionally are hydrogen cyanide, carbon disulfide, ethylene dibromide, chloropicrin, methallyl chloride and carbon tetrachloride. For cereals in international trade a

tolerance of 0.1 ppm expressed as PH3 is recommended.

SURFACE SPRAYS

Surface sprays are treatments applied to the surface of bulk grain or to the outer surfaces of bags. Examples are pyrethrum synergized with piperonyl butoxide, dichlorvos, malathion, pirimiphos methyl; chlorpyrifos methyl and tetrachlorvinphos.

Treatment of bag stacks. There are two methods of treatments: layer by layer (Sandwich method) and external stack treatment. The former method is also known as "sandwich" method where sprays or dusts are applied to each layer of bags during construction of a bag stack.

The external stack treatment usually consists of a spray application to the four sides and the top surface of a bag stack. This method is used to prevent reinfestation. Spraying is usually made immediately following a fumigation, or prior to sheeting in order to minimize the risk of cross infestation. Insecticides most commonly used for this purpose are malathion, primiphos-methyl and fenitrothion at 1-2%. Treatment of bag stacks are only capable of reducing infestations.

SACK TREATMENT

Sacks may either be sprayed (50 ml/m) or dipped in insecticide solution before filling with the grains. Treated sacks should be airdried after treatment. Malathion, pirimiphos-methyl, and chlorpyrifos methyl at 2-4% were found effective. At 2% malathion is only effective for 2-4 months while pirimiphos-methyl and chlorpyrifos methyl were effective for 4-6 months. At 4% these two

compounds can control infestation for 9-12 months.

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ANNEX I

Pesticides and growth regulators

(i) Use

Pesticides and growth regulators are used extensively in the world to improve the yield and/or

quality of crops, including grains. In most countries, legal restrictions as to the use of pesticides are in effect and different kinds of action can be started against possible violators of the regulations.

The general philosophy behind the use of pesticides is laid down in the concept of Good Agricultural Practice (GAP), which includes the following statements:

- **pesticides shall only be used when needed and only in the minimum quantities necessary;**
- **at no moment (neither at application, nor at consumption) public health shall be jeopardised by the use of pesticides;**
- **the environment and non-target organisms shall be protected as much as possible against adverse effects of pesticides;**
- **when alternatives are available, the pesticide with the least side-effects will have preference;**
- **advice to users of pesticides shall be available and regular checks shall be carried out as to the adherence to the advices;**
- **foods for human consumption (home produce as well as imported products) shall be regularly tested for the undue presence of pesticides and if necessary action shall be taken to improve the situation.**

As climatic conditions differ from one region to another, GAP is not identical over the world. It is implied that goods produced under GAP should move freely in international trade, even if it carries residues of pesticides not in use in the importing country. This is only possible if enough information is available as to the toxicity of the pesticide involved.

International bodies can help to make this information available to local authorities. International

work on pesticides in coordinated by the FAO/WHO Codex Alimentarius Committee on Pesticide Residues (CCPR). The secretariat of the CCPR is located in the FAO headquarters in Rome.

(ii) Incidence

The incidence of pesticide residues depends on the way the pesticide has been applied and on the nature of the pesticide. The treatment history of a lot of grain on the market is often unknown, even when the grain has been produced locally, and therefore only a few generalisations can be made.

Pesticides can be applied at several stages of the growth of the grain:

- **before harvest in or on the soil (e.g. against soil insects or weeds);**
- **before harvest on the crop (e.g. against fungi);**
- **after harvest on the grains for protection during storage or transport (e.g. against insects and rodents).**

A special case is the treatment of grains intended for sowing: these seeds should be kept carefully apart from grain for consumption by men or animals involved in animal husbandry (cattle, pigs, poultry etc.)

Pesticides used for protection of grains during storage have a larger chance to be found in the grains than pesticides used in an earlier stage of the growth. It can also be anticipated that herbicides will occur less frequently in objectable concentrations than the other pesticides, as

overdosage of a herbicide sooner leads to phytotoxicity than an overdosage of other pesticides.

A further indication as to the incidence of pesticide residues on grain can be obtained from the documents issued by the CCPR. In this body, the following pesticides are being discussed in connection to their use on grains:

(A = acaricide; F = fungicide; GR = growth regulator; H = herbicide; I = insecticide; M = molluscicide; N = nematocide; R = rodenticide; S = synergist)		lindane	I/R
		malathion	I/A
		metalaxyl	F
		methacrifos	I
aldicarb	I/A/N	methidation	I/A
aldrin/dieldrin	I	methiocarb	I/M
azinphos-methyl	I/A	methoprene	I
benomyl	F/A	methyl bromide	I/N/F/H/AIR
bromophos	I	monocrotophos	I/A
bromophos-ethyl	I/A	oxamyl	I/A/N
captafol	F	paraquat	H
carbaryl	I/GR	permethrin	I
carbendazim	F	phonothrin	I

carbofuran carbon disulphide	I/A/N	phenthoate phorate	I/A
carbon tetrachloride	I	phosmet	I/A
cartap	I	phosphamidon	I/A
chinomethionate	F/A	phoxim	I
chlordane	I	piperonyl butoxide	S
chlorfenvinphos	I/A	pirimicarb	I
chlormequat	GR	pirimiphos-methyl	I/A
chlorothalonil	F	prochloraz	F
chlorpyrifos	I	propargite	A
chlorpyrifos-methyl	I/A	propoxur	I
cypermethrin	I	pyrethrins	I
2, 4-D	H	2,4,5-T	H
DDT	I	thiabendazole	F
deltamethrin	I	thiodicarb	I
demeton-S-methyl	I/A	thiometon	I/A
diazinon	I/A/N	thiophanate-methyl	F/A
1,2-dibromoethane	I	triadimefon	F
1,2-dichloroethane	I	triazophos	I/A/N

dichlofluamid	F/A	trichlorfon	I
dichlorvos	I/A	triforine	F
diquat	H	vamidotion	I/A
disulfoton	I/A	<p>Inclusion of a pesticide in a Codex list means that the pesticide involved is used or found in commodities moving in international trade. The list is therefore especially useful when testing imported grains.</p> <p>(iii) chemistry</p> <p>Pesticides and growth regulators belong to very diverse types of chemical compounds. It is convenient to divide them into groups having common characteristics used in the analysis. Thus the pesticides mentioned above can be grouped as follows:</p> <ul style="list-style-type: none"> - electron-captive compounds (i.e. compounds giving a signal in the electroncapture detector (ECD) used in gas chromatography (GLC); most organochlorine 	
dithiocarbamates	F		
edifenphos	F		
endosulfan	I/A		
endrin	I/R		
ethiofencarb	I		
ethion	A/I		
fenitrothion	I/A		
fensulfothion	I/N		
fenthion	I		
fentin	F/M		
fenvalerate	I/A		
flucythrinate	I/A		
heptachlor	I		
hexachlorobenzene	F		

hydrogen cyanide	I/R/A	compounds fulfill this requirement, as do most organobromine and -iodine compounds; nitrogroups, aromatic rings and conjugated unsaturated systems also contribute to electron-captivity): aldrin/dieldrin, captafol, chlordane, chlorothalonil,
hydrogen phosphide	I/R	
imazalil	F	
inorganic bromide	metabolite	
isofenfos	I	

cypermethrin, DDT, deltamethrin, dichlofluanid, endosulfan, endrin, fenvalerate, flucythrinate, heptachlor, hexachlorobenzene, lindane, permethrin, phenothrin, propargite, pyrethrins, as well as the fumigants carbon disulphide, carbon tetrachloride, 1,2-dibromoethane, 1,2-dichloroethane and methyl bromide.

- **organophosphorus compounds: azinphosmethyl, bromophos, bromophos-ethyl, chlorfenvinphos, chlorpyrifos, chlorpyrifos-methyl, demeton-S-methyl, diazinon, dichlorvos, disulfoton, edifenphos, ethion, fenitrothion, fensulfothion, fenthion, fenthion, isofenfos, malathion, methacrifos, methidation, monocrotophos, phenthoate, phorate, phosmet, phosphamidon, phoxim, pirimiphos-methyl, thiometon, triazophos, trichlorfon, vamidothion, as well as the fumigant hydrogen phosphide.**
- **carbamates: aldicarb, carbaryl, carbofuran, ethiofencarb, methiocarb, oxamyl, pirimicarb, propoxur, thiodicarb.**
- **carboxylic acids: 2, 4-D, 2,4,5-T.**

- **benzimidazole derivatives: carbendazim, thiophanate-methyl, thiabendazole.**
- **quaternary ammonium compounds: diquat, pataquat, chlormequat.**

The other pesticides mentioned in (ii), do not belong to any particular group and must therefore be determined separately.

A limited number of pesticides form metabolites of toxicological importance, e.g. thio-ethers which form the equally toxic sulphoxides and sulphones, and some organochlorine pesticides which form epoxides of toxicological importance such as dieldrin (epoxide of aldrin) and heptachlor-epoxide. In these cases it is mandatory that the metabolites are determined as well.

(iv) Analysis

As very often the treatment history of a lot of grain is unknown, it is important to have methods available which cover as many pesticides as possible. These methods are "multi-residue methods" and will be discussed here in more detail, as they are most often the analyst's first choice.

The discussion will follow the same order as above.

- **electron-captive compounds: gas chromatography (GLC) with electron-capture detection (ECD) is the method of choice. This method is sensitive, specific and applicable to many compounds. The detector is however sensitive to many interferences as well, so that extracts must often be cleaned prior to analysis. Careful maintenance of the detector is mandatory as well.**
- **organophosphorus compounds: GLC with flamephotometric detection (FPD) and alkali**

flameionisation (AFID) is the method of choice. Both detectors shall be present in a well-equipped laboratory, as they are complementary in their properties.

- **carbamates: although some carbamates are amenable to GLC (sometimes after derivatisation), highpressure liquid chromatography (HPLC) is often to be preferred. Organophosphorus compounds and carbamates can also be detected by thin-layer chromatography (TLC) based on the inhibition of the enzyme cholinesterase. This procedure can not be used however for quantitation and sensitivity can differ from one pesticide to another by a factor of 104.**
- **carboxylic acids: HPLC is nowadays the method of choice. GLC procedures are available, but they are cumbersome and subjected to interferences.**
- **benzimidazole derivatives: HPLC is the most advisable method. For thiabendazole GLC procedures using FPD in the sulphur-mode are also widely used. A TLC procedure based on the inhibition of fungi growth is practical for confirmatory purposes.**
- **quaternary ammonium compounds. diquat and paraquat can be determined by one spectrophotometric method; chlormequat must be determined separately however.**
- **fumigants: due to their volatile nature, fumigants are often treated as a separate group. Their volatility requires special attention during sampling, transport and analysis. GLC with ECD, FPD and normal flameionisation detection (FID) is used for the determination. In some cases, head-space analysis is possible, which produces exceptionally clean chromatograms and therefore low limits of determination.**

Special methods (i.e. methods which cover only one pesticide) are necessary for the pesticides not covered by a multi-residue method, viz.:

- **cartap**: the compound is converted to nereistoxin which can be determined by GLC.
- **dithiocarbamates**: this group of compounds (including maneb, zineb, mancozeb, ferbam, ziram, thiram and propineb) is always determined as carbon disulphide after acid hydrolysis. Carbon disulphide can be determined by head-space GLC, but in the case of grains better yields are obtained with the classical spectrophotometric method.
- **fentin**: the compound is converted to its methyl derivative with methyl magnesium bromide and determination is carried out by GLC/FPD, which is also sensitive to tin-compounds.
- **hydrogen cyanide**: this compound is converted to bromo-cyan and determined as such by GLC/ECD.
- **inorganic bromide**: HPLC-, GLC- and spectrophotometric procedures are available. HPLC is the quickest procedure and can be used in most practical cases.
- **methoprena**: determination is carried out by GLC/FID.
- **piperonyl butoxide**: this compound has to be determined separately by GLC/FID or by HPLC.
- **prochloraz**: derivatisation is necessary prior to gas chromatography.
- **triforine**: this compound is converted to chloroform, which can be determined by GLC/ECD.

As can be seen from the data presented above, pesticide residue analysis requires modern chromatographic equipment: at least two gas chromatographs, equipped with ECD, FPD, AFID and FID are necessary, as well a one HPLC-system equipped with UV-detection at 216 and 254 nm. A third gas chromatograph equipped with capillary columns is required for confirmatory purposes. A UV/VIS-spectrophotometer is also necessary for a number of analyses, but this instrument can often be shared with other departments, as it is seldom 100% in use for pesticide residue analysis. High purity gases, solvents and reagents must be available, and spare parts, service kits and

maintenance manuals are indispensable. Clean-up of extracts can be carried by classical column chromatography, but gel permeation chromatography (GPC) is making rapid progress for this purpose. Automatic injection systems and electronic data processors are useful for improving laboratory efficiency, but they can only work to full satisfaction if the chromatography are up to standard.

Just as in agriculture, the general philosophy of pesticide residue analysis has been laid down in a document (Codex Alimentarius Commission, document CAC/PR-7-1984), describing Good Analytical Practice in pesticide residue laboratories. The document is especially useful when a new pesticide residue laboratory has to be set up or when an old laboratory must be up-dated. Special attention is drawn to the chapter on confirmation in the document, as confirmation is a crucial, but often neglected aspect of pesticide residue analysis.

Analytical Quality Assurance (AQA) is another important aspect of Good Analytical Practice.

A bibliography with selected references to analytical methods for pesticides is also available through FAO (document CAC/PR 8-1984).

ANNEX II

TABLE 1. The Geological History of the Insects General history indicated by continuous lines; spots are records of occurrences in Australia. Recent orders in bold face type.

06/11/2011

Towards integrated commodity ... - Se...

[Contents](#) - [◀ Previous](#) - [Next ▶](#)