Macroanalytical Procedures Manual (MPM)

V-3 Whole Grains and Pseudo-Grains

May 2021

Editor: Hans Loechelt-Yoshioka
Co-editor: Gabi Pires

Note: The page number for this chapter in the original 1984 version of the MPM was V-15.

https://www.fda.gov/food/laboratory-methods-food/macroanalytical-procedures-manual-mpm

Table of Contents

A. Method for Wheat, Corn, Popcorn Kernels, and Rice

(1) Scope

The methods below describe procedures for the determination of damage/adulteration in wheat, corn, and rice caused by rodents, insects, molds, and other defects.

(2) Applicable Documents

- CPG Sec. 578.300 Wheat - Adulteration by Insect and Rodent Filth
- CPG Sec. 578.350 Wheat for Human Consumption-Reconditioning
- CPG Sec. 578.400 Treated Grain Seed - Mercury Residue
- CPG Sec. 585.675 Popcorn - Adulteration with Rodent Filth and Field Corn
- IOM Chapter 4, Chart 4-Wheat Carload Sampling, p. 4-102
- 21 CFR 2.25 Grain Seed Treated with Poisonous Substances

(3) Defects

The principal defects found in grains involve damage during storage caused by rodents, birds, insects, and molds. Rodent damage can consist of rodent excreta, gnawing damage and even presence of nesting material (Zimmerman & Friedman, 2000). Birds can also damage grain in a similar manner to rodents (McCarthy, 2003). Insect damage can be caused by primary and secondary pests. Field damage by fungal plant pathogens (including wheat or corn scab fungus, corn blight fungus, and black tip fungus) may become a problem during adverse climatic conditions. Through improper cleaning of transportation equipment and storage facilities grain can become contaminated with seeds. Although grains and seeds may appear similar at times, there are differences. A grain is a small edible fruit usually harvested from plants in the Poaceae. A seed is an embryonic plant covered in a hard seed coat. Seeds are often treated with various pesticides and identified with indicator dyes. These dyes range in color from reds, blues, yellows, and greens (Figures V-3-A-21, 50, B-16). Grains can become contaminated with dyes from bagging materials (Figure V-3-A-33); therefore, one needs to be careful when examining grains with dyes not to confuse them with treated seeds or molds.

Grains and pseudo-grains can be adulterated with other grains. For example, field corn is occasionally present in unpopped popcorn, regular long grain may be mixed with Basmati rice, and Durum wheat (*Triticum durum* Desf.), which is a high-quality wheat commonly used to make pasta, may be mixed with the cheaper common wheat (*T. aestivum* L.).

ARTHROPODS:

The insect and mite pests of grains can be broken down into two broad categories: Field pests and storage pests, which are listed below. Additionally, the insects that are listed below are relatively common in grains. The list is not all inclusive, and other countries may have different pest common to their regions. For more information on common pests see (Appendix B).

Field Pests:

While grains are growing in the field (pre-harvest) they can be damaged by multiple groups of pests. They are exposed to climatic conditions sun, rain, wind, which can affect their appearance. The difference between field and storage damage is not always well defined. It is important to be able to distinguish field damage versus storage damage, when possible.

COLEOPTERA:

Coccinellidae**:** Although not normally a pest of grains, they can be found in grain samples. Ladybird beetles, also known as ladybugs, are beneficial insects feeding on other insects especially aphids. Their colorful elytra can occasionally be found in samples of grain. One exception is the multicolored Asian lady beetle, *Harmonia axyridis* (Pallas)*,* which is an invasive species. This ladybird beetle will harm native insects/biodiversity and is invasive in homes.

DIPTERA:

Cecidomyiidae: The Hessian fly, *Mayetiola destructor* (Say), has done tremendous damage to grain crops over the years. This fly attacks barley, rye and wheat. The larvae will feed on the sap of the plant, weakening it to the point where it cannot support the grain bearing heads.

HEMIPTERA:

Alydidae: The broad-headed bugs include the rice bugs, *Leptocorisa chinensis* Dallas and *L. acuta* (Thunberg), which are serious rice pests throughout Asia. They are one of the major causes of ‘pecky rice’ in Asia. Pecky rice can lead to secondary damage caused by fungal infection.

Aphididae: Aphids feed on the plants in the field. In the process of feeding on the plants, they can spread various plant viruses. The Russian wheat aphid, *Diuraphis noxia* (Mordvilko), introduced into the U.S. in Texas, 1986 (Michaud & Sloderbeck, 2005) has become a serious pest of wheat. It is just one of many species of aphids attacking grains.

Pentatomidae: Stink bugs are a major field pest on rice, especially during the early development of the rice grain. Their feeding damage can cause malformed and shriveled kernels. Their feeding damage can also produce ‘pecky rice’, similar to the Alydidae. Some of the more serious stink bug pests include the rice stink bug, *Oebalus pugnax* (F.), and rice black bug, *Scotinophara coaractata* (F.).

LEPIDOPTERA:

Many types of Lepidoptera larvae attack grain plants, in the field during the growing season. Some of the more important field pests for grains are the Noctuidae and Crambidae moths. The Noctuidae group contains some of the biggest economic pests like the corn earworm, *Helicoverpa zea* (Boddie) (Figure V-3-A-37A), tobacco budworm, *Heliothis virescens* (F.), and various armyworms While the Crambidae contain pests like the European corn borer, *Ostrinia nubilalis* (Hübner), Mexican rice borer, *Eoreuma loftini* (Dyar) and the rice stalk borer, *Chilo plejadellus* Zincken

ORTHOPTERA:

Grasshoppers (Figure V-3-A-37B) and crickets are common field pests in grains. Their bodies and harder structures like their heads, hind legs and mandibles can be seen in grain samples. At times, wing fragments can also be found.

Storage Pests:

After harvest, grains are subjected to different environmental conditions throughout the supply chain originating from the farm. The grains may be attacked by different species at different points of post-harvest production. Occasionally, a field pest can also act like a storage pest depending upon its location in the supply chain. Processing of the grains may reduce some of the defects.

Primary Pests:

Primary insect pests feed and breed in intact grains. Primary insect pests can do considerable damage in stored food products. Some of the common primary insect pest of grains belong to the following insect orders and families:

COLEOPTERA:

Bostrichidae: The two most common members in this group include the lesser grain borer, *Rhyzopertha dominica* (F.) and the larger grain borer, *Prostephanus truncatus* (Horn) (Figure V-3-A-3). The eggs are laid on the outside of the grain and the larvae burrow into the grain forming a circular hole.

Dermestidae: The Khapra beetle, *Trogoderma granarium* Everts, is a USDA/APHIS regulated quarantine pest found in imported grains like basmati rice, wheat, barley, oats, and various other products. As of 2015, there were 31 countries recognized by USDA as having endemic populations of the Khapra beetle (Stibick, 2007).The average life cycle is 4-6 weeks, thus allowing 4-5 generations per year to be produced. The adult females can lay 50-100 eggs, depending on temperature. The adults, larvae and pupae can be confused with the similar appearing species warehouse beetle *Trogoderma variabile* Ballion, *T. versicolor* (Creutzer) and *T. glabrum* (Herbst). If suspected, *T. granarium* Everts is found in a sample, please contact your local/regional USDA/APHIS office.

Curculionidae: The species most commonly encountered include members of the genus *Sitophilus* spp. Schönherr (Figures V-3-A-1, 2, 25, 26, 36) such as the granary weevil, *Sitophilus granarius* (L.), the rice weevil, *S. oryzae* (L.), and the maize weevil, *S. zeamais* Motschulsky. The females make a small impression on the grain, where they lay their egg. The resulting depression is then covered with a glandular secretion which forms a small plug that can be difficult to see. This plug can be made visible through a staining process. The larvae hatch and tunnel into the grain where they complete their development. Another Curculionidae, formerly a Scolytidae, *Pagiocerus frontalis* (F.) is a pest of corn in storage.

LEPIDOPTERA:

Gelechiidae: The Angoumois grain moth, *Sitotroga cerealella* (Olivier) (Figure V-3-A-4), adult has a body length of approximately 9.0 mm with a wing span up to 10.0-15.0 mm. The hindwings narrow to a point and have a long lateral fringe of hair-like scales.

Secondary Pests:

Secondary pests only attack grains which have been previously damaged by a primary insect pest or by some type of mechanical damage.

ACARI:

Acaridae: The grain mite, *Acarus siro* L., the mold mite, *Tyrophagus putrescentiae* (Schrank), are two common storage pest mites on grains. Their diet is not restricted to grains, but includes nearly any food utilized by man (Mason 2018). Grain mites can experience population explosions when the humidity and temperature of grain increases. Grains heavily infested with mites can have a lemon-like odor and display a resulting brown dust-like coating due to live/dead mites and cast skins.

Cheyletidae: Occasionally, predatory mites of the genus *Cheyletus* Latreille, are associated with storage mites. These mites have been used as biological control agents in stored grains (Zdarkova 1998).

Pyemotidae: The straw itch mite, *Pyemotes tritici* (LaGrèze-Fossat & Montané), is normally found in grain and hay. This mite is a parasite of other arthropods and not a pest of grain. There has been intrest in this mite as a biological control agent.

COLEOPTERA:

Anobiidae: The drug store beetle, *Stegobium paniceum* (L.) and the cigarette beetle, *Lasioderma serricorne* (F.) are common secondary pests of grains.

Laemophloeidae: The flat grain beetle *Cryptolestes pusillus* (Schönherr) and the rusty grain beetle *C. ferrugineus* (Stephens) are secondary pests of grains. Larvae have been known to feed upon *Sitophilus* spp. (Schönherr) larvae while still within the grain.

Silvanidae: The sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.), the merchant grain beetle, *O. mercator* (Fauvel), and the foreign grain beetle, *Ahasverus advena* (Waltl), are just a few of the common grain pests in this family. These beetles are cosmopolitan in nature and have been found on other stored products as well.

Tenebrionidae: The confused flour beetle, *Tribolium confusum* Jaq. Du Val and the red flour beetle, *Tribolium casteneum* (Herbst) and other members of this family are considered secondary pests to grains. The larvae typically start feeding on the germ layer of the damaged grain. Developmental times vary and are directly proportional to the average temperature and humidity.

LEPIDOPTERA:

Pyralidae: The Indian meal moth, *Plodia interpunctella* (Hübner), is the most common Lepidopteran grain pest (Solis 2006). The almond moth, *Cadra cautella* (Walker) has been found as a pest on flax seed (Figures V-3-C-3-6).

PSOCODEA (formerly Psocoptera):

Psocids, often called booklice and barklice, are not true lice. Booklice are psocids which tend to be found indoors, while barklice tend to be found outdoors. Adult psocids can be winged or wingless (apterous).The booklice can be found on a wide variety of stored food products and tend to live in environments with a higher humidity. One of the most important psocid species found in stored products is *Liposcelis bostrychophila* Badonnel, which has a worldwide distribution.

VERTEBRATES:

Vertebrates can be pests of grains and pseudo-grains. Their damage includes direct feeding, contamination with hairs or feather material, and direct contamination with bodily fluids like excreta pellets, urine, and saliva. Mammals and birds have been reported as common sources of disease transmission through excreta.

Aves (Birds): There are numerous species of birds which can attack grains and pseudo-grains. Some common species observed on grain material include: The European starling, *Sturnus vulgaris* L., the rock pigeon, *Columba livia* Gmelin, and the house sparrow, *Passer domesticus* (L.). A major Asian pest is the rose-ringed parakeet, *Psittacula krameri* (Scopoli) (Figure V-3-B-7). The main source of contamination concern from birds is excreta pellets (Figure V-3-B-8) and feather material. Common pigeons and house sparrows are commensal in nature and can contaminate product while stored in a warehouse. Birds can defecate on farm equipment used for harvesting, transporting or processing grains, thus becoming a source of contamination for the product.

Cervidae: White tailed deer, *Odocoileus* spp. Rafinesque, and wapati, *Cervus* spp. L., contaminate grains and pseudo-grains through excreta pellets and hairs. As the animals walk and lay in a field, their droppings (Figure V-3-A-23) and hairs can adhere to the plants and become incorporated with the grain during harvest.

Leporidae: Rabbits and hares can contaminate grains and pseudo-grains with their excreta pellets and hairs. Grain is often stored outdoors in large mounds. As the rabbits feed on the grains their excreta pellets and hairs can become incorporated with the grain. Their excreta pellets, typically a spherical shape and the size of a pea, also contain hair from the animal (Figure V-3-A-24).

Muridae: Rats and mice are common rodent pests in grains (Gecan,1980; Sarwar, 2015), and pseudo-grains. Their commensal nature and wide distribution make them a serious pest of stored products. They have been known to spread microorganisms such as *Listeria* spp.*, Pseudomonas* spp., *Salmonella* spp.*,* and *Vibrio* spp. Due to their synanthropic nature, Muridae can contaminate food products through direct feeding, shedding of hairs, bodily fluids and excreta. Some cosmopolitan species include the Norway rat, *Rattus norvegicus* (Berkenhout), the black/roof rat, *R. rattus* (L.), and the house mouse, *Mus musculus* L. Additional commensal rodents include the Polynesian rat, *R. exulans* (Peale), the lesser bandicoot rat, *Bandicota bengalensis* (Gray), the ricefield rat, *R. argentiventer* (Robinson & Kloss), and the wood rat, *R. tiomanicus* (Miller). Other murids that feed on grains and pseudo-grains include the common field mouse, *Apodemus sylvaticus* (L.), the wood rat, *Neotoma* sp. Say and Ord, the deer mouse, *Peromyscus maniculatus* (Wagner) and the hispid cotton rat, *Sigmodon hispidus* Say & Ord (Valencia et. al., 1994). The excreta pellets of rats and mice tend to be blackish-brown in color with a slight elliptical shape (Figures V-3-A-5, 27, 38). Rodent excreta pellets are recognized by their size, shape, presence of rodent hairs and the formation of a mucous coating with the addition of a couple drops of water. The approximate size range for rat excreta pellets is 5.0 mm - 20.0 mm, while mice excreta pellets tend to have an approximate size range of 3.0 mm - 6.0 mm. When the rodents have been feeding on toxic colored baits (Figure V-3-A-52), their excreta pellets often take on the color of the bait.

When feeding on grains rats and mice will leave evidence on the grain of the their double incisor marks. This is evident especially on larger grains like corn and popcorn.

The rodents have a preference for the germ portion of the grain, and will leave the rest of the grain (Figure V-3-A-6A). If the rodent population is high or the food supply is small, then the rodents will eat the entire grain or leave just empty hulls. (Smith 1953).

When corn is stored on the cob, the rodents are unable to reach the preferred germ layer. In this case, the tops of the grains are eaten away leaving a “blazed” area, where the white endosperm has been exposed. This may cover a few grains or it may extend over a large part of the cob (Figure V-3-A-7). Usually the marks left by the teeth of the rodent, show in the damaged areas at the top of the grains. (Smith 1953).

Other Rodents: There are other numerous rodents that feed on grains and pseudo-grains, especially in the field. Occasionally, some of these rodents can become almost commensal in nature. They include the squirrels (Sciuridae) and hamsters and voles (Cricetidae).

FUNGAL DISEASES:

Fungi includes molds and yeasts. Mold (or mould), produces multicellular hyphae, a long branching structure with some general characteristics such as parallel cell walls, branching filaments, septation of the cell walls, and the production of fruiting bodies and spores (Skinner 1951). Yeast, on the other hand, are single-celled, non-filamentous and reproduce by a simple budding process. Another group of fungi attacking grains are the smuts, which belong to many different genera (Figure V-3-A-28). One smut attacking maize and teosinte, *Ustilago maydis* (de Candolle) Corda, is commonly known as corn smut or Huitlacoche (Figure V-3-A-18). Huitlacoche, an edible fungus, is considered a delicacy, and has been referred to as the Mexican truffle. *Ustilago maydis* (de Candolle) Corda is not counted as a filth element. The rice false smut, *Ustilaginoidea virens* (Cooke) Takah, during its development, will replace rice grains with ball-shaped fungal mycelia, false smut balls. In the maturing process of the fungi, it will go through a color change from yellowish orange, green, olive green to a black at maturity. The fungus is becoming an increasing threat to rice production. It often produces sclerotia. One other fungus grown on grain and not considered a filth element is *Cordyceps* sp. Fries, which grows into an orange fruiting body on grains (Figure V-3-A-49). The following list of fungi is for informational purposes only. It is beyond the scope of this manual to attempt an identification of the fungi.

Dothideomycetes:

Alternaria sp. Nees-- A very common fungus found in soil, on plants and indoors. It is a black sooty mold, which can also be dark olive-green to brown in color.

Cladosporium sp. Link-- Can range in color from gray to black and even a dark olive-green.

Epicoccum sp*.* Link --This fungus will produce a pinkish stain on the grain. Commonly referred to as pink epicoccum (Figure V-3-A-17). This pink stain can sometimes be mistaken for dyes from packaging or from treated seeds.

Lasiodiplodia theobromae (Patouillard) Griffon & Maubi -- A blackish grey Ascomycetes fungus attacking many commodities. It will cause maize ear rot in corn and black kernel rot (Figures V-3-A-12, 13).

Macrophomina sp. Petrak-- Commonly known as charcoal rot, this fungus favors hot dry weather conditions and has been found on at least 500 different types of plants. The fungus is dark brown to black in color (Figures V-3-A-14, 15).

Eurotiomycetes:

Aspergillus sp. P. Micheli ex Haller --This common fungus ranges in color from tan, sooty-black, green or yellowish-green (Figure V-3-A-6). This genus causes major health issues in humans and animals like Aspergillosis.

Penicillium sp. Link --Tends to be a blue to greenish-blue colored mold (Figure V-3-A-6). This mold is typically found in the soil.

Leotiomycetes:

Sclerotinia sp. Fuckel --This plant fungus is called white mold or even cottony rot, as it somewhat resembles a piece of cotton on the plant stem. This fungus will produce black sclerotia (Figure V-3-A-47), which can look like ergot (Wang, 2007). They also can be mistaken for cockroach excreta pellets or even rodent excreta. They can be separated from ergot in cross-section. The sclerotia in cross-section are white, while ergot is purplish black.

Sordariomycetes:

Claviceps purpurea (Fries) Tulasne--Commonly known as ergot (Figures V-3-A-42, 58), this is a parasitic fungus which attacks grains, especially rye (Ergot, 2019, Wang, 2007). Ergot sclerotia can sometimes be confused with rodent excreta and cockroach excreta pellets (Figures V-3-A-38, 53).
Fusarium sp. Link--A common fungus on barley. It will readily infect other grains like wheat and corn (Figures V-3-A-9, 10). It can range in color from white to yellow, brownish, pinkish and even a slight reddish color.

Gibberella sp*.* Saccardo--One of the fungi which cause corn ear rot.*Gibberella fujikuroi* (Sawada) S. Ito is a major pest of rice, causing foolish seedling disease.This fungus serves as a source for gibberellic acid.The fungus is pinkish to reddish color.

Nigrospora sp Zimmermann--A brownish black fungus which will cause ear rot in corn (Figure V-3-A-11). It can produce an allergy response in humans.

Trichoderma sp. Pearsoon--This mold is one of the causative agents for corn ear rot. It is dark green in color.

OTHERWISE UNFIT:

This category includes grains damaged by environmental factors like heat, weathering, and flood damage. Grains stained from bagging material dyes (Figure V-3-A-33), these stains need to be verified, that they are not caused by mold. Grains may also contain toxic foreign seeds, such as *Crotalaria* spp. (Meyer, 2010) (Figure V-3-A-51A) or grains which are considered allergens when mixed in with other grains (for instance, wheat grains mixed in a rice sample). Grains contaminated with seeds, where the grain is the edible fruit of the plant and the seed is the embryonic plant covered in a seed coat and used for planting and often treated with a pesticide. These seeds can be recognized by their characteristic indicator dyes (Figures V-3-A-21, 50, B-16). If found, testing for mercury/pesticides needs to be considered. If wheat contains an average of 10 or more pink kernels per 500 grams this is rejectable, then it does need to be tested for mercury (CPG Sec. 578.400). Even poison baits for rodents can be observed (Figure V-3-A-52).

(4) Procedure: Determination of Extraneous Materials in Wheat, Corn, Popcorn Kernels, and Rice

a. Sample Preparation --The representative sample consists of at least 6 (454 g) subsamples or consumer size packages of at least 454 g taken from the lot. The examination of popcorn requires 10/225 g subsamples or 6/284g (10 oz) consumer size packages.
Determine the weight of each subsample to the nearest 0.1 g.

b. Visual Examination

Consumer size packages: Examine the exterior of each consumer size package of wheat, corn, popcorn kernels or rice for signs of entrance/exit holes. Insect entrance/exit holes can be confirmed using (AOAC Official Method 973.63 Insect Penetration through Packaging Materials). Also examine the consumer size package for signs of rodent gnawing damage, clumped material and urine stains. The urine stains can be visible under UV light (AOAC Official Method 945.88 Urine Stains on Foods and Containers-Ultraviolet Light Examination) and confirm presence of urea by using the xanthydrol test (AOAC Official Method 959.14 Urine Stains on Foods and Containers-Xanthydrol Test for Urea). Note: The sensitivity of xanthydrol to urea can be variable based on the manufacturere. It is important to test for this variability, in order to produce a positive reaction at 4 µg urea. (Biles & Ziobro, 1998).

Examine the entire content of each subsample of wheat, corn, popcorn kernels or rice for extraneous material by spreading a small amount at a time on a sheet of white paper, or use any other convenient light-colored surface or device that will permit the rapid and accurate visual examination of an appropriate portion of each subsample at a time.

- Rodent excreta pellets: Remove and weigh the rat and mouse excreta pellets and fragments. Include as rodent pellet material any whole pellets or broken pellets, provided they are large enough to be readily identified as such by their size, shape, surface coating, and presence of rodent hairs. Rodent pellets have a surface/mucous coating when wetted with a few drops of water. Refer to the (AOAC Chapter 16 Subchapter 16 Animal Excretions) to confirm animal excreta in grains. Some of the more specific methods (AOAC Official Method 972.41 Urine on Grain and AOAC Official Method 990.10 Mammalian Feces in Grain Products).

- Feather material and bird excreta pellets: Test the bird excreta pellets for uric acid in the white chalky material only (AOAC Offical Method 962.20 Excrement (Bird) on Food and Containers-Microchemical Test for Uric Acid).

- Insect and insect material: Check for presence of live/dead insects, insect excreta, webbing material. Note that some cockroach and grasshopper excreta pellets can resemble rodent excreta. Ergot and *Sclerotinia* can also resemble rodent excreta pellets.

- Examine product for other extraneous materials: sticks, strings, hard/sharp objects like rocks, glass, plastic, metal shavings, etc.

- Examine popcorn for presence of field corn: Field corn is also called dent corn. Popcorn has a higher moisture content from field corn and comes in a range of colors. Popcorn also tends to be more round with a glossy appearance. Field corn has a low moisture content. This lower moisture content in the field corn causes the characteristic dorsal indentation or dent. Field corn is a relatively flat grain with a dull appearance. It usually comes in a yellow or white (Figure V-3-A-22A). Visually separate popcorn from field corn by their physical differences in appearance. After removing the field corn, record the weight found to the nearest (g).

- Sweet canned corn may be adulterated with field corn. Refer to AOAC: Official Method 928.09 Field Corn in Canned Mixtures of Field and Sweet Corn.

Do not discard product after examination in 4(b), as the product may be analyzed again in subsequent sections.

c. Report -- For each subsample, report identification, total count, and size range of the recovered extraneous materials. If whole insects are found, state if alive or dead.

Record also, any insect fragments or related material such as webbing or insect excreta pellets recovered.Rodent excreta pellets, whole & broken, report as follows:

- Wheat: Rodent excreta pellets are based on weight: mg of pellets per kilograms of wheat.
- Popcorn: Total number of rodent excreta pellets needs to be recorded by count per sub. Also the number of rodent hairs needs to be counted per sub.
- Other grains and pseudograins: Rodent excreta pellets are based on weight: mg of pellets per kilograms of other grains.
- Give a count and measurement of all whole insects, their fragments and other extraneous material recovered. Identify all to the lowest level of identification.

Identification of other animal excreta pellets like bat, bird, deer and other animals give the size of the pellet. If found in a grain or pseudograin then report the pellet as mg/ kilogram of grain. If found in corn report excreta pellets based on count. All identifications should be identified to the lowest taxonomic level possible. An aid in the identification of the animal excreta pellets can be found in (Zimmerman and Friedman, 2000). See example of reporting format (Table V-3-1).

(5) Procedure: Determination of Insect-Damaged Wheat Kernels

a. Sample Preparation – Use subsamples from 4(a). Using one of the three recommended dividers (rotary splitter, stationary splitter (Jones Divider), or fractional shoveling), mix and reduce each subsample to about 200 g. If the weight of the subsamples of the consumer size package is less than 454 g, product does not need to go through a divider. Instructions for using the various dividers can be found in Chapter II Apparatus for Macroanalytical Methods. Please note that the product needs first to be examined for extraneous material as described in 4(b). Examine wheat either visually 5(b) or by X-ray 5(d).

b. Visual Examination -- Use subsamples from 5(a). Weigh out 100 g of each reduced subsample for visual examination. Weigh to the nearest 0.1 g. Examine wheat by spreading a small amount at a time on a sheet of white paper or other convenient device, like a Seedburo Picking Tray, that will permit the rapid and accurate visual examination of an appropriate portion of the sample at a time and examine for insect damaged kernels. Figure V-3-A-34 gives specifications and visual reference of reject or passable insect damage in wheat kernels. Kernels that are only slightly eaten by insects or from which the germ has been removed by insects are considered acceptable. Figures V-3-A-35 and 36 show examples of rejected wheat kernels due to insect damage. Remove and count insect-damaged kernels. Specify if whole insects are alive or dead the insect stage of development (for example, adult, larva, etc.), and provide insect identification to the lowest level possible (for example, species, genus, family, etc.).

c. Report of Visual Examination -- Report the number of insect-damaged kernels per 100 g of wheat for each subsample. See example of reporting format (Table V-3-1).

d. X-ray Examination --Take slightly more than 100 g of each reduced subsample from 5(a) and brush the wheat, a small amount at a time, onto a 5- or 8-inch No. 12 sieve. Use a stiff bristle brush to work any surface insects, dust, dirt, or broken kernels through the sieve. Weigh 100 g of the brushed wheat from each subsample as the test portion for X-ray examination***.*** Spread the test portion of wheat in a single layer, on a support interposed between a source of X-rays and a radiographic film, if a digital X-ray machine is not available . Expose to soft X-rays using a standard wedge according to the instructions for obtaining maximum contrast as described in (Chapter IV, "Special Techniques). Examine the radiograph for insect-damaged kernels using any suitable commercially available X-ray film viewing screen or directly on a monitor if the image was digitally captured. In general, the wheat kernel appears white or gray on the negative. Any cavity within the grain appears as a dark region and an insect within the cavity appears light in color (Figure V-3-A-46). Mark the insect-damaged kernels and count the total per 100 g.

e. Report of X-Ray Examination – Report the number of insect-damaged kernels per 100 g of wheat for each subsample. See example of reporting format (Table V-3-1).

(6) Procedure: Determination of Decomposition in Wheat Caused by Molds and Other Factors

a. Visual Examination -- Use subsamples from 5(a). Weigh out 50 g of the reduced sample from the second approximate 100 g portion, and examine the kernels using the naked eye or with low power magnification. If the magnification exceeds 10X, this should be stated in the method section of the report.

b. Classification of Decomposed Wheat Kernels—Classify decomposed wheat kernels as follows. Note: identification of type of mold is not required:

(i) *Moldy*: Classify as moldy, those kernels which fit any of the following categories:

- Kernels which are affected by scab to the extent that the bran coat is broken open and there is evidence of mold or other disease or damaged condition.
- Kernels which have a dull, lifeless and chalky appearance (so-called "tombstone") because of disease.
- Kernels which have mold in the germ or have mold in the crease of the kernel. *Note*: Carefully remove the bran coat covering the germ; scraping the bran coat too deeply could remove the mold.
- Kernels with Bunt disease, where the grain is filled with teliospores (Figures V-3-A-40, 41).

- Kernels which are affected by black-tip fungus to the extent that the fungus growth is on the germ and extends into the crease of the kernel (Figure V-3-A-39).
- Kernels affected by white, pink, gray, black, or green mold (Figures V-3-A-39, 43, 44).
- Kernels with ergot are black or dark purple sclerotized grains on the inside (Figures V-3-A-42, 48), while *Sclerotinia* sp. Fuckel is white on the inside (Figure V-3-A-47).

(ii) *Otherwise Unfit*- Classify as otherwise unfit those kernels which fit any of the following categories:

- Kernels which have been materially discolored on the surface and damaged by external heat. (Figure V-3-A-45).
- Kernels damaged as a result of heating caused by excessive respiration or fermentation.
- Flood damaged grains, tend to be discolored. The grain will rapidly spoil due to the increased moisture. In warm summer conditions flooded grain can spoil in 1 to 2 days (USDA 2019).
- Kernels which are unfit by other causes (describe the condition).
- Kernels coated with dirt.
- Kernels with fumigant, insecticide type odors, or strong rancid-like odor by smelling the entire portion in the subsample.

c. Report -- Report percent of decomposed kernels by weight for each category. Also report the total percent of decomposed kernels for each subsample. Describe principal type(s) of damage found. See example of reporting format (Table V-3-1).

(7) Procedure: Determination of Insect and Rodent-Damaged Corn and popcorn

a. Sample Preparation -- Use subsamples from 4(a). Important note: first examine product for extraneous material using protocol in 4(b) before proceeding. Prepare sample as in 5(a) except reduce each subsample to slightly more than 250 g corn. Examine corn either visually 7(b) or by X-ray 7(d).

b. Visual Examination – Weigh out 200 g of the reduced product from each subsample for visual examination. Weigh to the nearest 0.1 g. Examine corn kernels by spreading a small amount at a time on a sheet of white paper or use any other convenient light-colored surface or device, like a Seedburo Picking Tray, that will permit the rapid and accurate visual examination of an appropriate portion of the sample at a time. Examine for insect damaged kernels. Classify as a reject any kernel showing insect tunneling and/or insect feeding punctures (Figures V-3-A-1-4).

While examining for insect damaged grains, also look for rodent damage. When it is suspected that a damaged grain of corn or popcorn has been gnawed, it is sometimes possible to locate the marks made by the upper incisor teeth.These marks are made as the upper incisors are braced against the outer edge of the grain, while the lower incisors cut out the bite from the grain (Figiure V-3-A-6B). The finding of these marks is conclusive evidence that the grain has been gnawed by rodents and not damaged by some other means. At times the parallel grooves made by the lower incisors are also visible in the damaged section of the grain (Smith 1953).

c. Report of Visual Examination -- Report the number of insect-damaged kernels per 200 g of corn for each subsample. Report the number of rodent gnawed grains per 200 g of corn for each subsample as well. See example of reporting format (Table V-3-1) for corn and (Table V-3-2) for popcorn.

d. X-ray Examination -- Weigh 200 g of each reduced subsample and follow examination instructions as shown in 4(d). Look for tunneling (Figure V-3-A-20) and other feeding damage. Count and report the number of insect damaged kernels per 200g of sample.

e. Report of X-Ray Examination -- Report the number of insect-damaged kernels per 200 g of corn for each subsample. See example of reporting format (Table V-3-1) for corn and (Table V-3-2) for popcorn.

(8) Procedure: Determination of Decomposition in Corn and Popcorn Kernels Caused by Molds and Other Factors

a. Visual Examination -- Weigh out 50 g of the reduced sample from 7(a). Weigh to the nearest 0.1 g. Examine the kernels using the naked eye or with low power magnification. If the magnification exceeds 10X, this should be stated in the method section of the report.

b. Classification of Decomposed Corn and Raw Popcorn—Classify decomposed corn and raw popcorn as follows. Note: identification of type of mold is not required.:

(i) *Moldy* -- Classify as moldy, those kernels which fit any of the following categories:

- Kernels and pieces of kernels infected with mold on exposed endosperm. When a kernel of corn has been broken exposing the starch, it becomes susceptible to mold. Check broken pieces carefully for mold. Do not confuse kernels that have dirt on them with kernels containing mold. Mold is usually blue or green in color.
- A kernel germ infected with blue-eye mold (*Aspergillus* sp. P.Micheli ex Haller or *Penicillium* sp. Link) (Figure V-3-A-6). If the mold is distinct, it is not necessary to open or scrape the kernel. Otherwise, lift the germ cover carefully to avoid destroying the evidence of mold. *Note:* do not confuse purple plumule with blue-eyed mold. Purple plumule (Figure V-3-A-7) is not damage but it is a genetic or varietal characteristic and will have no signs of mold hyphae.

- Kernels showing damage from cob rot (Figures V-3-A-9-15). Cob rot can be caused by various fungi including (*Gibberella* sp. Saccardo, *Fusarium* sp. Link and *Aspergillus* sp. P. Micheli ex Haller).
- Silk-cut kernels. Kernels and pieces of kernels with mold in silk cuts. Kernels with clean silk cuts are sound and are not considered as being damaged.
- Pink Epicoccum (Figure V-3-A-17). Kernels and pieces of kernels with a dark pinkish discoloration in the region of the germ. *Note*-do not confuse pink epicoccum with the reddish genetic characteristics found in some hybrid corns.
- Kernels which have mold in the germ or on the grain surface. *Note*: Carefully remove the bran coat covering the germ; scraping the bran coat too deeply could remove the mold.
- Kernels materially affected by white, pink, gray, black, or green mold (Figures V-3-A-6,8,16).

(ii) *Otherwise Unfit* -- Classify as otherwise unfit those kernels which fit any of the following categories:

- Kernels which have been materially discolored and damaged by external heat through drying.
- Kernels damaged as a result of heating caused by excessive respiration or fermentation.
- Kernels coated with dirt.
- Kernels with fumigant, insecticide type odors, or strong rancid-like odor, by smelling the entire portion in the subsample.
- Flood damaged kernels (Figure V-3-A-19).
- Rodent gnawed popcorn.

c. Report -- Report percent of decomposed kernels by weight for each category. Also report the total percent of decomposed kernels for each subsample. Describe principal type(s) of damage found. Rodent gnawed popcorn needs to be recorded by total count of rodent gnawed popcorn in 454 g of product for each subsample. See example of reporting format (Table V-3-1) for corn and (Table V-3-2) for popcorn.

(9) Procedure: Determination of Insect-Damaged Rice

a. Sample Preparation -- Use subsamples from 4(a). Important note: first examine product for extraneous filth using protocol in 4(b) before proceeding. Prepare sample as in 5(a). Reduce the subsample to slightly more than 200 g rice. This procedure applies to all rice, including basmati rice. Examine rice either visually 9(b) or by X-ray 9(d).

b*. *Visual Examination -- Weigh out 100 g of each reduced subsample for visual examination. Weigh to the nearest 0.1 g. Examine rice by spreading a small amount at a time on a sheet of white paper or use any other convenient light-colored surface or device, like a Seedburo Picking Tray, that will permit the rapid and accurate visual examination of an appropriate portion of the sample at a time. Examine for insect damaged rice (Figures V-3-A-25, 26). Classify as reject, those kernels which have been bored by insects.

c. Report of Visual Examination -- Report number of insect-damaged kernels for each subsample and the average for the subsamples, based on count in 100 g of sample. See example of reporting format (Table V-3-1).

d. X-ray Examination --Take slightly more than 100 g of each reduced subsample and brush the rice, a small amount at a time, on a 5- or 8-in. No. 12 sieve. Use a stiff bristle brush to work any surface insects, dust, dirt, or broken kernels through the sieve. Weigh 100 g of the brushed rice from each subsample as the test portion for X-ray examination. Look for tunneling (Figure V-3-A-32) and other feeding damage. Additional instructions on X-ray examination can be found under 4(d).

e. Report of X-Ray Examination -- Count number of insect-damaged kernels per 100 g of rice. See example of reporting format (Table V-3-1).

(10) Procedure: Determination of Decomposition in Rice Due to Molds and Other Factors

a. Visual Examination -- Weigh out 50 g of the reduced sample from the second 100 g portion of 9(a). Weigh portion to the nearest 0.1 g. Examine the kernels using the naked eye or with low power magnification. If the magnification exceeds 10X, this should be stated in the method section of the report.

b. Classification of Decomposed Rice—Classify decomposed rice as follows. Note: identification of type of mold not required.:

(i) *Moldy* -- Classify as moldy, any rice grains showing any of the following characteristics. This list representative only.

- Kernels which are affected by scab to the extent that the bran coat is broken open and there is evidence of mold or other disease or damaged condition.
- Kernels which have a dull, lifeless and chalky appearance (so-called "tombstone") because of disease.
- Kernels smut or bunt, (*Tilletia barclayana* (Brefeld) Saccardo & P.Sydow), where the endosperm of the grain is attacked by the fungus, and the grain starch is replaced with smut spores.
- Kernels materially affected by white, pink, gray, black, or green mold (Figure V-3-A-29).
- Pecky Rice (Figure V-3-A-30), a water-stained surface discoloration which can lead to mold growth.There are at least 5-6 types of fungi which can cause this condition. A similar discoloration can also be produced by the feeding action of the rice stinkbug, *Oebalus pugnax* (F.) and the broad headed bug, *Leptocorisa chinensis* Dallas.This damage is especially noticeable in brown rice varieties. The Hemiptera feeding damage, often leads to a fungal infection of the rice grain.

(ii) *Otherwise Unfit* -- Classify as otherwise unfit those kernels which fit any of the following categories:

- Kernels which have been materially discolored and damaged by external heat (Figure V-3-A-31).
- Kernels damaged as a result of heating caused by excessive respiration or fermentation.
- Kernels which have been materially decomposed by other causes (describe the condition).
- Kernels coated with dirt.
- Kernels with fumigant or insecticide type odors or strong rancid-like odor, by smelling the entire portion in the subsample.
- Flood damaged grains.

c. Report -- Report percent of decomposed kernels by weight for each category. Also report the total percent of decomposed kernels for each subsample. Describe principal type(s) of damage found. See example of reporting format (Table V-3-1).

Figures

CORN

Figure V-3-A-1. Corn showing feeding damage and tunneling by *Sitophilus* sp. Schönherr, by the presence of exit holes as seen through stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-2. Insect feeding damage and tunneling on popcorn by *Sitophilus* sp. Schönherr, as seen through a stereomicroscope (Source: H. Loechelt-Yoshioka, FDA).

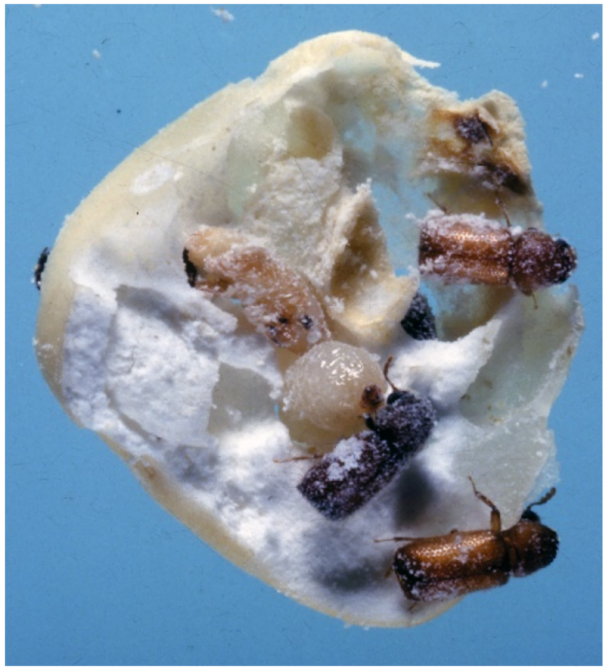

Figure V-3-A-3. Larger grain borer *Prostephaneus truncatus* (Horn), showing extensive damage to corn kernel. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-4. Angoumois grain moth *Sitotroga cerealella* (Oliver). Adult moth with larval damage to the corn kernels. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-5. Popcorn kernels with rat, *Rattus* sp. Fischer excreta pellets, as seen through a stereomicroscope (6.3x). (Source: H. Loechelt-Yoshioka, FDA).

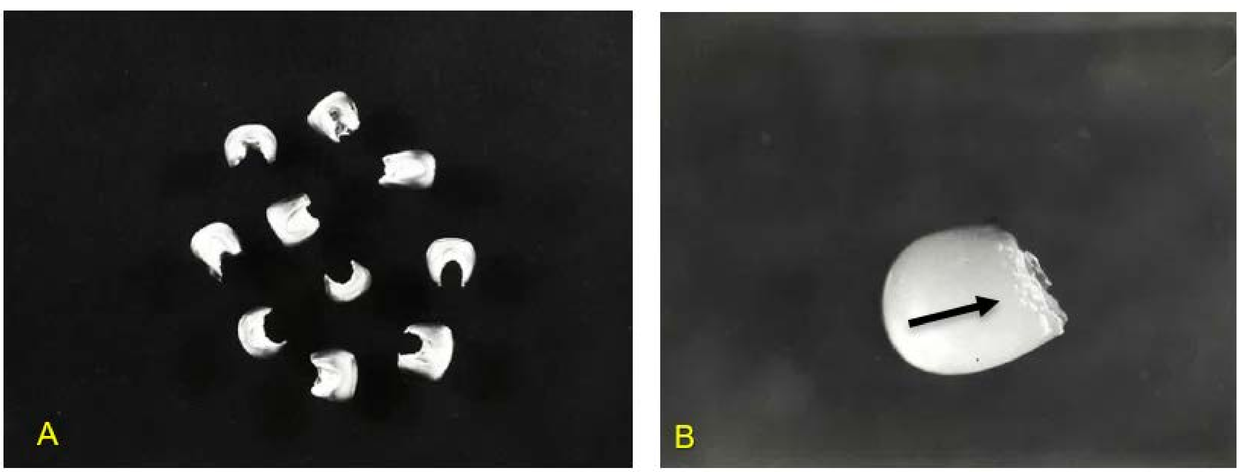

Figure V-3-A-6. A. Dent corn degerminated by rodents (Natural size). (Source: F. Smith, FDA). **B.** Gnawed popcorn showing marks left by the rodents' upper incisor teeth, indicated by the arrow (5x). (Source: F. Smith, FDA).

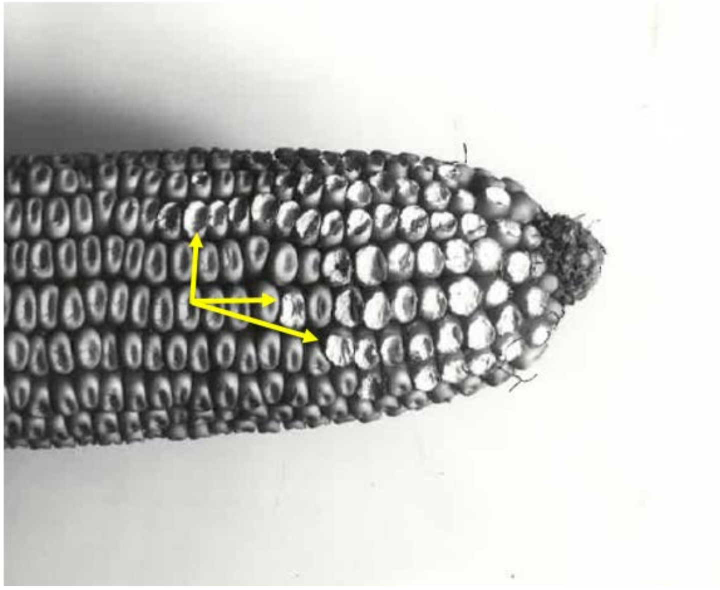

Figure V-3-A-7. Dent corn on the cob, "blazed" by rodents. Arrows showing some of the marks left by their incisors (Natural size). (Source: F. Smith, FDA).

Figure V-3-A-8. Corn with damage by blue-eyed mold. Blue-eyed mold is typically caused by *Aspergillus* sp. P.Micheli ex Haller or *Penicillium* sp. Link. Mold hyphae will be present. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-9. Purple plumule is not damage, but it is a genetic or varietal characteristic. It can be confused with blue-eyed mold, but it will not have hyphae. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-10. Corn kernel with mold damage, from an unidentified mold. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-11. Fusarium mold infected cobs of maize, cobs have extensive damage from the mold. (Source: Photo courtesy of © State of New South Wales through NSW Department of Primary Industries, 2007).

Figure V-3-A-12. Maize kernels with fusarium cob rot. Rot indicated by the greyish borwn discoloration. (Source: Photo courtesy of (c) State of New South Wales through NSW Department of Primary Industries, 2007).

Figure V-3-A-13. Nigrospora ear rot on corn kernel, it is caused by the fungus, *Nigrospora oryzae* (Berkeley & Broome) Patch.The Kernels become discolored with their ventral end covered in a mass of dark spores. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-14. Black kernel rot, *Lasiodiplodia theobromae* (Patouillard) Griffon & Maubl., on a corn cob. The diseased kernels have a dark, shiny appearance. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-15. Black kernel rot, *Lasiodiplodia theobromae* (Patouillard) Griffon & Maubl. The diseased corn kernels have a dark, shiny appearance. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-16. Charcoal ear rot, *Macrophomina phaseoli* (Maubl.) S. F. Ashby. Normally occurs in hot, humid areas that have had some dry periods. The mold on the kernel forms small, round, black, pinhead-shaped sclerotia. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-17. Corn kernels with extensive damage by charcoal ear rot, *Macrophomina phaseoli* (Maubl.) S. F. Ashby. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-18. Unidentified mold on the surface of the corn kernel. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-19. Corn with Pink Epicoccum, *Epicoccum* sp. Link, is a type of mold. Do not confuse the pink coloration with a reddish color due to genetic characteristics. Verify presence of mold hyphae. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016)

Figure V-3-A-20. *Ustilago maydis* (de Candolle) Corda, commonly known as corn smut or Huitlacoche. Huitlacoche is an edible fungus, considered a delicacy. It has been referred to as the Mexican truffle. It is not considered a defect. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-21. Discolored corn kernels caused by flood damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

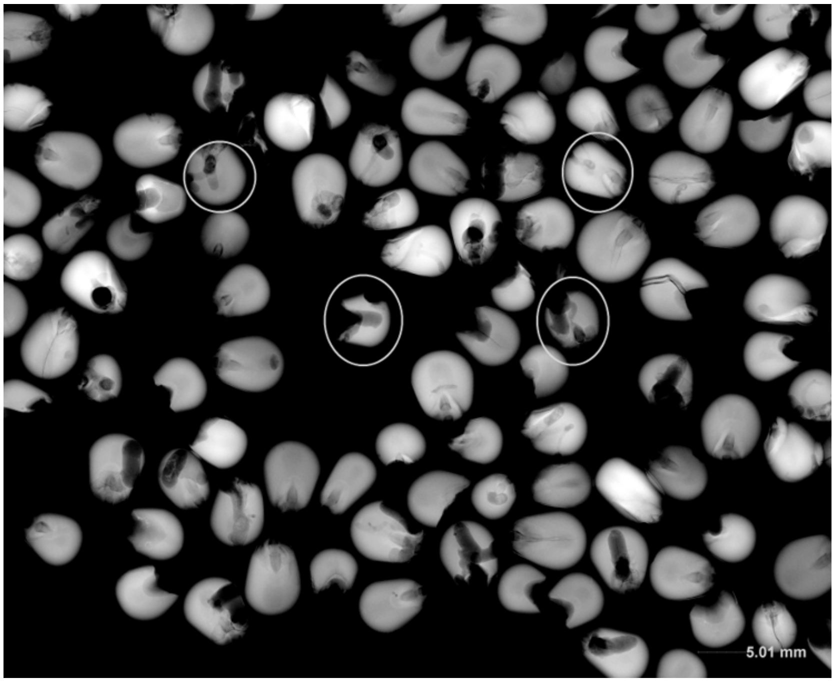

Figure V-3-A-22. X-ray image of Sitophilus sp. Schönherr-damaged popcorn. A few damaged grains identified by circles. (Source: H. Loechelt-Yoshioka, FDA).

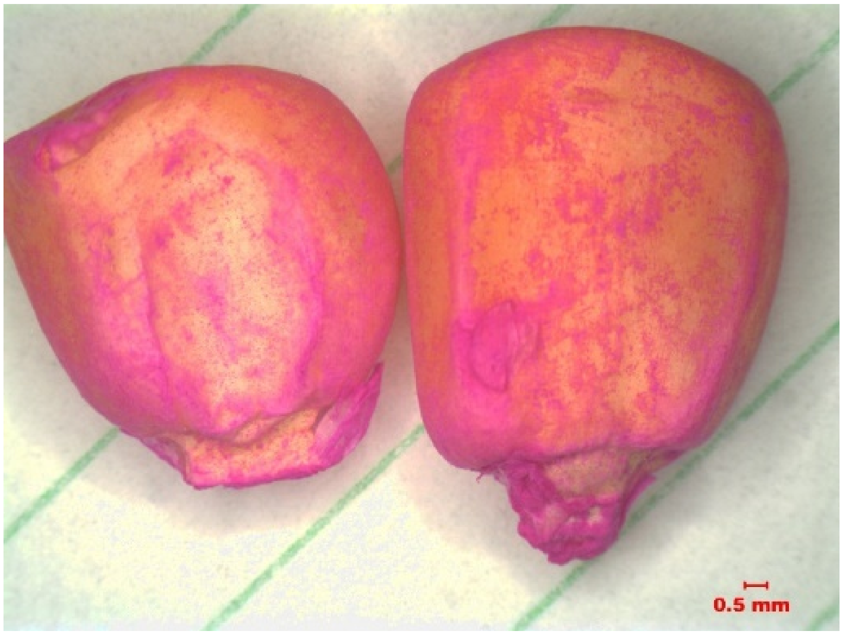

Figure V-3-A-23. Treated corn seed with a fungicide as seen through a steromicroscope. (Source: H. Loechelt-Yoshioka, FDA).

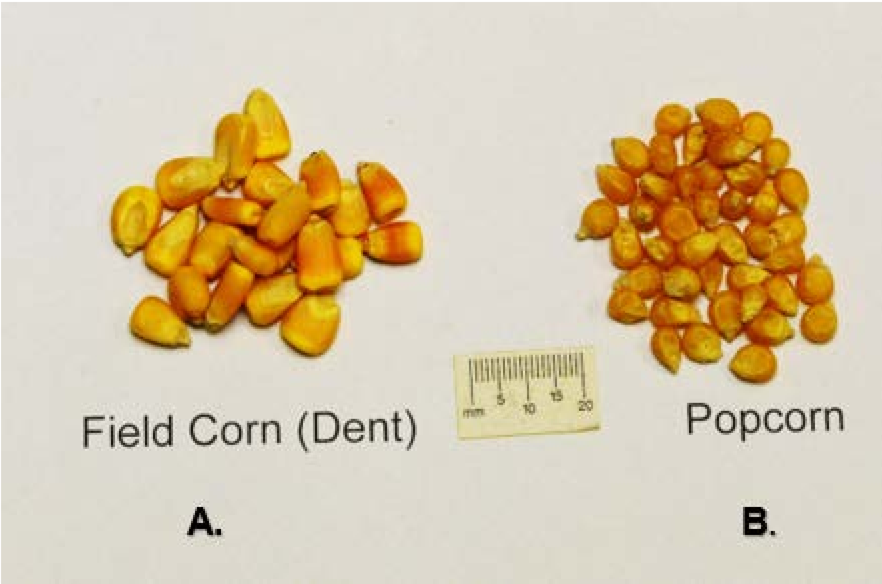

Figure V-3-A-24. A. Field corn is flat and with a dent on the broad end. **B.** popcorn is rounded and without a dent on the broad end. (Source: H. Loechelt-Yoshioka, FDA).

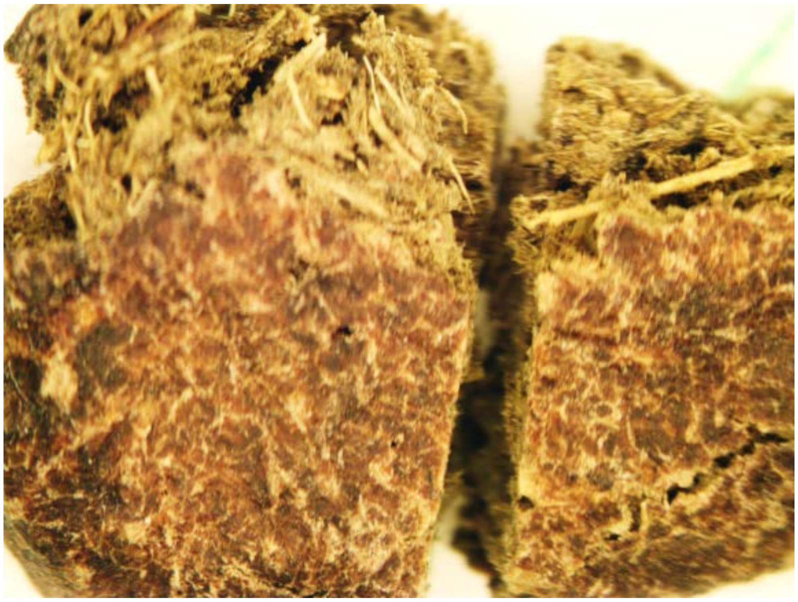

Figure V-3-A-25. White-tailed deer, *Odocoileus* sp. Rafinesque excreta pellet recovered from a corn shipment as seen through a steromicroscope (6.3x). (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-26. Eastern cottontail, *Sylvilagus floridanus* Allen, excreta pellets as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

RICE

Figure V-3-A-27. Insect damaged basmati rice. Damage caused by weevils, *Sitophilus* sp. Schönherr as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-28. Rice weevil, *Sitophilus oryzae* (L.) damaged brown rice with mold. Adult rice weevil found inside grain of rice as seen through a steromicroscope. (Source: H. Loechelt-Yoshioka, FDA).

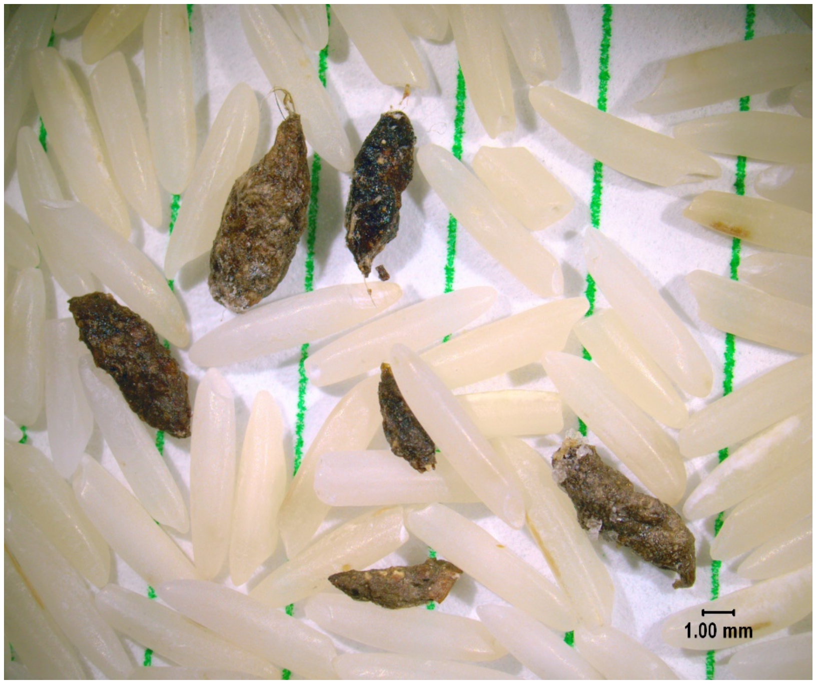

Figure V-3-A-29. Rice with the presence of mouse *Mus* sp. L. excreta pellets, as seen through a stereomicroscope. (Source: H. K. Loechelt-Yoshioka, FDA).

Figure V-3-A-30. Brown rice with smut (a fungus) damage. Smut consists of chlamydospores, which tend to be black in color. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016)

Figure V-3-A-31. Unidentified mold in brown rice as seen through a steromicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-32. 'Pecky' brown rice. Feeding damage by the rice stink bug, Oebalus pugnax (F.), leading to fungal infection as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA)

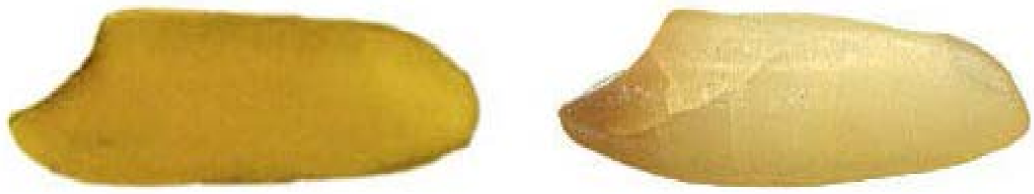

Figure V-3-A-33. Otherwise decomposed-rice kernels with discoloration caused by heat damage. (Source: Photo courtesy of USDA-FGIS Visual REference Images 2016).

Figure V-3-A-34. X-ray of insect, *Sitophilus* sp. Schönherr, damaged basmati rice. Damage indicated by circles (3x). (Source: H. Loechelt-Yoshioka, FDA).

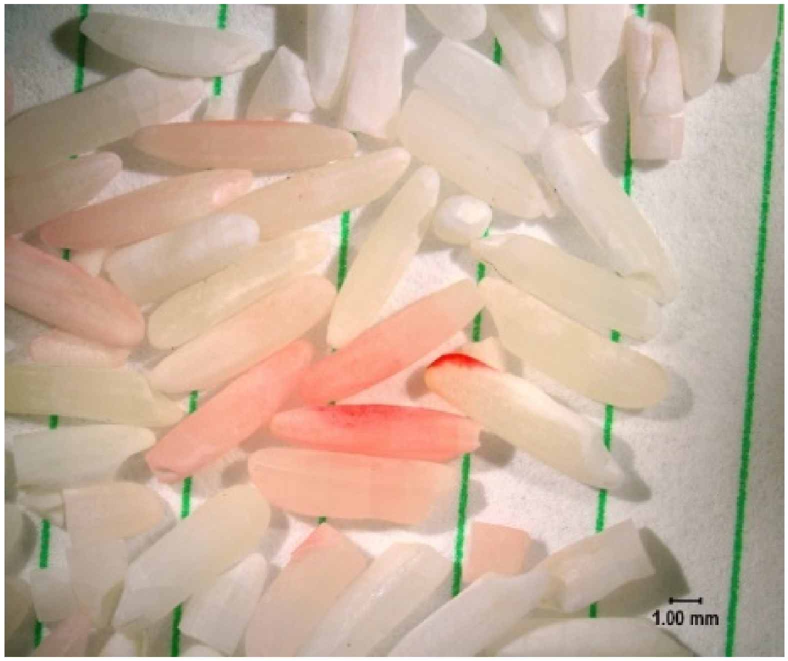

Figure V-3-A-35. Rice which has been stained by red lettering on burlap bagging as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

WHEAT

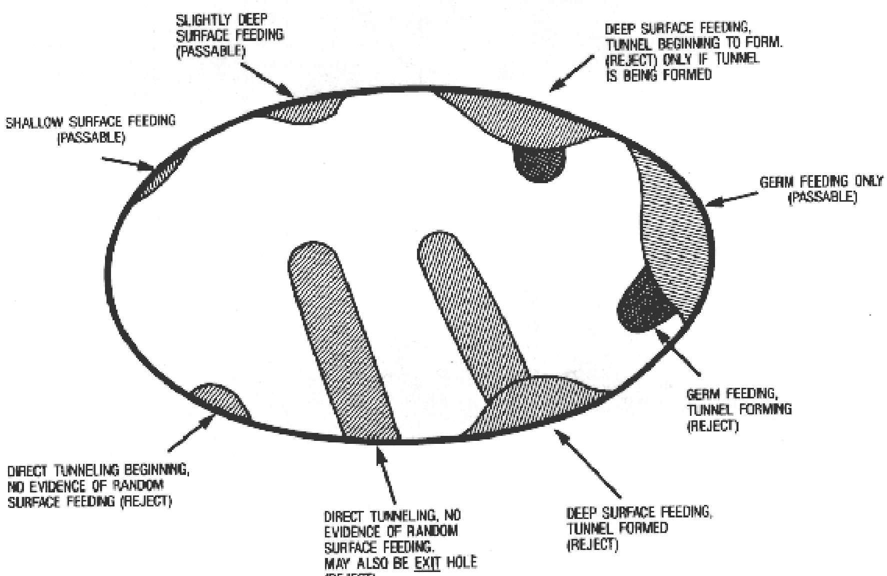

Figure V-3-A-36. Classification of insect damage in wheat kernels (cross-sectional view). (Source: MPM 1984, electronic version 1998).

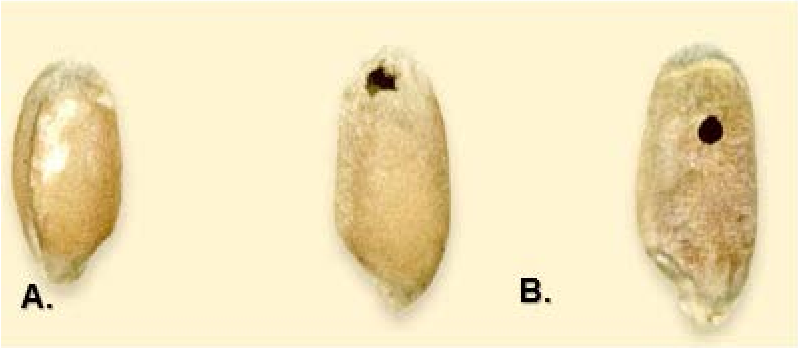

Figure V-3-A-37. Insect damaged wheat, **A. s**howing surface feeding damage **B.** showing insect boring damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-38. Insect damage wheat by *Sitophilus* sp. Schönherr as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

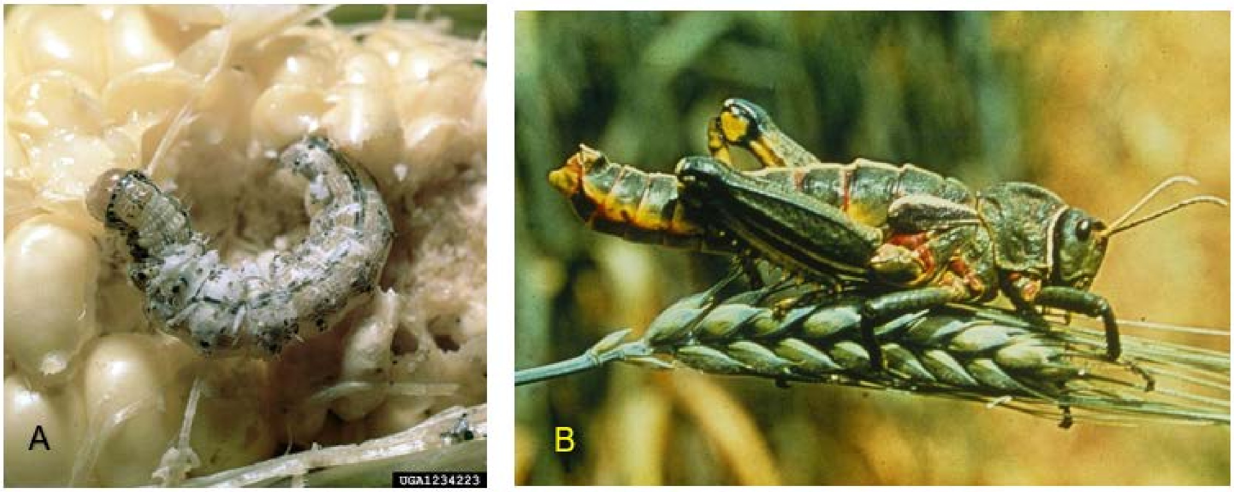

Figure V-3-A-39. Field pest, **A. c**orn earworm feeding on corn, *Helicoverpa zea* (Boddie). (Source: Photo courtesy of CLemson University-USDA Cooperative Extension Slide Series, Bugwood.org). **B.** grasshopper found on a head of wheat. Their fragments can be found mixed in with grains. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-40. Wheat contaminated with rat *Rattus* sp. Fischer excreta pellets and their feeding damage, as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-41. Wheat showing black tip mold damage on the germ end. (Source: Photo courtesy of USDA-FGIS VIsual Reference Images 2016).

Figure V-3-A-42. Common or Dwarf Bunt, *Tilletia* sp. Tulasne & C. Tulasne. Typical characteristic for a bunt disease is the presence of 'bunt balls'. They resemble the grain kernel but are filled with dark teliospores. The diseased kernels give off a fishy type odor, when smashed or crushed. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-43. Karnal Bunt in wheat, *Tilletia indica* Mitra. The endosperm of the grain gets replaced by telispores and the pericarp can stay intact or rupture. The diseased kernels give of a fishy type of odor, when smashed or crushed. (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-44. Head of wheat with ergot, *Claviceps purpures* (Fries). (Source: Photo courtesy of C. R. Beaver, III, CIMMYT).

Figure V-3-A-45. Wheat kernels with mold damage. The mold is indicated by the dark areas on ther kernels. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-46. Wheat kernels with mold damage in the crease and germ area. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-47. Otherwise decomposed-wheat kernels discolored from heat damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

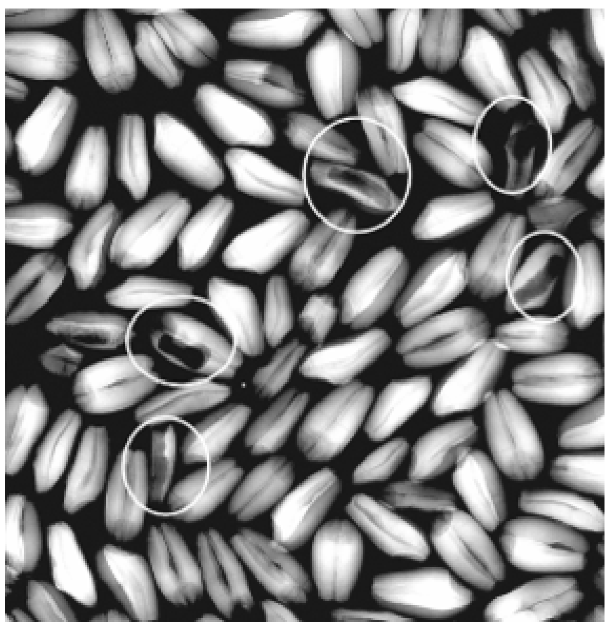

Figure V-3-A-48. X-ray image of damaged wheat kernels by *Rhyzopertha dominica* (F.). Note-the circled kernels show tunneling damage (3x). (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-49. Sclerotina, *Sclerotinia sclerotiorum* (Lib. de Bary), a fungal disease on the stems of plants. Cross section of sclerotinia revels a white center. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-50. Ergot, *Claviceps purpures* (Fries) Tulasne, a fungal disease affecting the flora tissue of many grains. Cross section of ergot revels a greyish-white discoloration with a purplish tinge. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016)

Figure V-3-A-51. Flos Cordyceps, *Cordyceps* sp. Fries, fungus growing on mixed grains. Not a defect, unless grown on grain that contains gluten, and product label does not show allergen warning for gluten. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-52. Treated wheat seed with a fungicide as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-53. A. Crotalaria, *Crotalaria* spp. L. seeds and seed pods. The seeds come in various colors and have a glossy appearance. **B.** Non-toxic velvet leaf weed, *Abutilon theophrasti* Medikus seeds. They have a similar size and shape, but have a dull, rough appearance. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-A-54. An example of toxic colored bait used for rodent control as seen through a stereomicroscope (6.3x). (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-A-55. Cockroach excreta pellets as seen through a stereomicroscope (8.0x). (Source: H. Loechelt-Yoshioka, FDA).

B. Method for Other Grains

(1) Scope

Other grains refer to the other food crops belonging to the Poaceae, which includes but is not limited to barley, oats, millet, sorghum, and wild rice (Matz 1959). The following methods describe procedures for the determination of damage/adulteration in other grains.

(2) Applicable Documents

None

(3) Defects

The defects listed in section A(3) are applicable for this section as well. Figures below show some examples of defects caused by insect damage (Figures V-3-B-1, 3, 9, 11), mold (Figures V-3-B-2, 4, 5, 10, 12, 13), and otherwise unfit due to heat damage and weather (Figures V-B-6, 14, and 15) on barley, oat, rye, and sorghum.

(4) Procedure: Examination of Other Grains

Sample Preparation, Visual Examination, Report for Visual Examination, X-ray Examination, and Report for X-ray Examination.

A representative sample consists of at least 6 (454 g) subsamples or consumer size packages of at least 454 g taken from the lot. Examine other grains either visually or by X-ray. Select procedures as appropriate from A4(a) through A10(a), exluding A7 and A8. See example of reporting format (Table V-3-1).

Figures

BARLEY

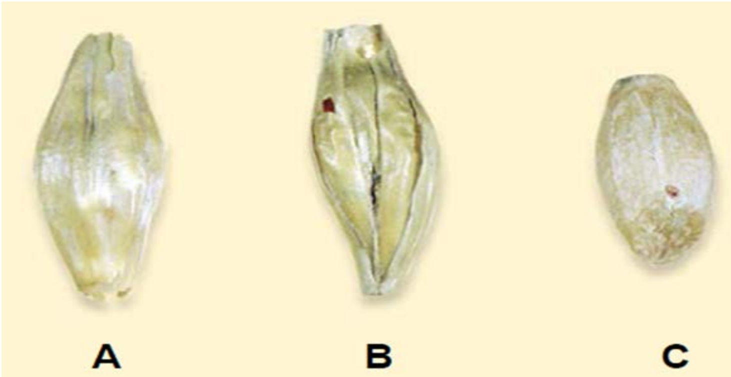

Figure V-3-B-1. Insect damaged barley: **A.** damage by insect tunneling **B.** damage by insect boreing **C.** damage by insect boreing. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-2. Barley showing evidence of mold (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

OATS

Figure V-3-B-3. Insect damaged oats. Damage evidence by presence of insect boreholes. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-4. Hull-less oat kernel showing mold damage in the area of the germ. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-5. Oat Kernel with mold damage. Mold covering apical half the kernel. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-6. Otherwise decomposed-oat kernel with heat damage indicated by a reddish discoloration in the germ layer. Only visible when hulls are removed. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-7. A group of rose-ringed parakeets *Psittacula krameri* (Scopoli), one Alexandrine parakeet *Psittacula eupatria* (L.) (lower circle) and one rock pigeon *Columba livia* J. F. Gmelin, (upper circle), feeding on stored grain. (Source: Photo courtesy of Vivek rathod 17-Own work, CC BY-SA 3.0, https://commons.wikimedia.org/w/index.php?curid=33456851).

Figure V-3-B-8. Bird excreta in hull-less oats as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

RYE

Figure V-3-B-9. Rye Kernels with insect damage, indicated by the presence of tunneling and bore holes. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-10. Broken/cracked rye kernels with exposed mold growth. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

SORGHUM

Figure V-3-B-11. Insect damaged sorghum indicated by presence of bore holes. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-12. Sorghum showing unidentified mold damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

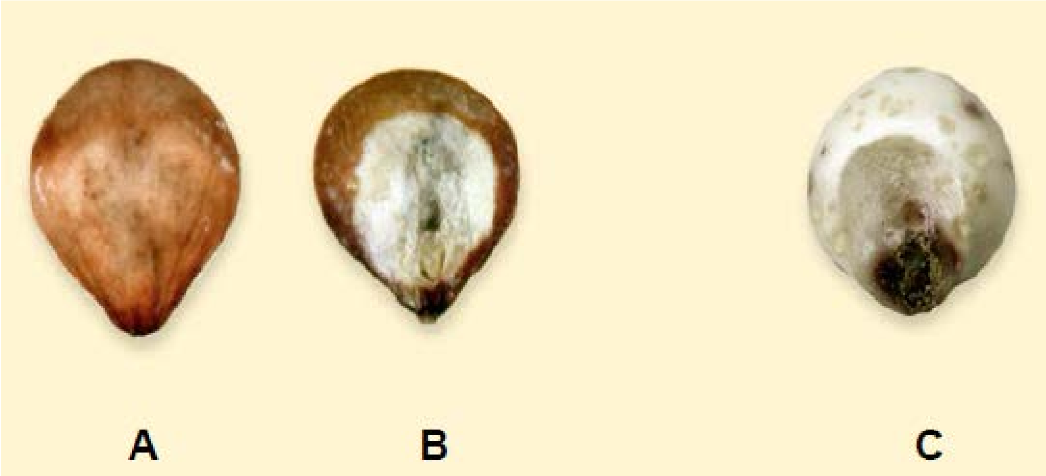

Figure V-3-B-13. Bleaching turns penetrating molds dark. **A.** suspected mold, slight blue discoloration in germ area **B.** confirmed mold damage **C.** bleached sorghum grain showing mold damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-14. Otherwise decomposed sorghum-ground and/ or weather damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-15. Otherwise decomposed sorghum-heat damage. Note-must perform a cross section through middle of germ to confirm damage. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-B-16. Treated sorghum seed with a fungicide as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

C. Method for Pseudo-Grains

(1) Scope

The method below describe procedures for the determination of damage/adulteration in pseudo-grains/pseudo-cereals caused by rodents, insects, molds, and other defects. Pseudo-grains (also knows as pseudo-cereals) belong to dichotomous plants, which are used like grains. They include, but are not limited to, the following plant families: Amaranthaceae, Asteraceae, Brassciaceae, Chenopodiaceae, Fabaceae, Lamiaceae, Linaceae, and Polygonaceae. Representatives of each of these families include: amaranthus, sunflower, canola, quinoa, kañiwa, wattleseed (Acacia), chia, flaxseed, buckwheat. (Oregon State University 2021, Wikipedia, 2021)

(2) Applicable Documents

None

(3) Defects

The defects listed in section A(3) are applicable for this section. Additionally, pseudo-grains/pseudo-cereals can be attacked by host specific pests. Figures below show examples of defects caused by insect damage (Figures V-3-C-3-5, 8-10), mold (Figures V-3-C-1, 11), and otherwise unfit (Figures V-3- C-6, 12) in pseudo-grains/pseudo-cereals. Flax can have mechanical damage, which looks similar to insect damage (Figures V-3-C-2). Do not count the mechanical damage as a reject. Example of insect damage viewed through X-ray examination of sunflower achenes are shown in Figure V-3-C-13.

(4) Procedure: Examination of Pseudo-Grains/Pseudo-Cereals

Sample Preparation, Visual Examination, Report for Visual Examination, X-ray Examination, and Report for X-ray Examination.

Examine pseudo-grains/pseudo-cereals either visually or by X-rays. A representative sample consists of at least 6 (454 g) subsamples or consumer size packages of at least 454 g taken from the lot. Choose appropriate methods from A4(a) to A10(a), excluding A7 and A8. *Note*: in step A5(a) for grain preparation for X-ray examination, the No.12 sieve may need to be reduced in size to retain the smaller pseudo-grains/pseudo-cereals on the sieve. Due to a thick pericarp on sunflower achenes, an X-ray examination is the preferred method of analysis. The pericarp can hide the damage to the sunflower achenes, which is revealed by the X-ray (Figure V-3-C-12). See example of reporting format (Table V-3-1).

Figures

CANOLA

Figure V-3-C-1. Canola seed with white surface mold. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

FLAX

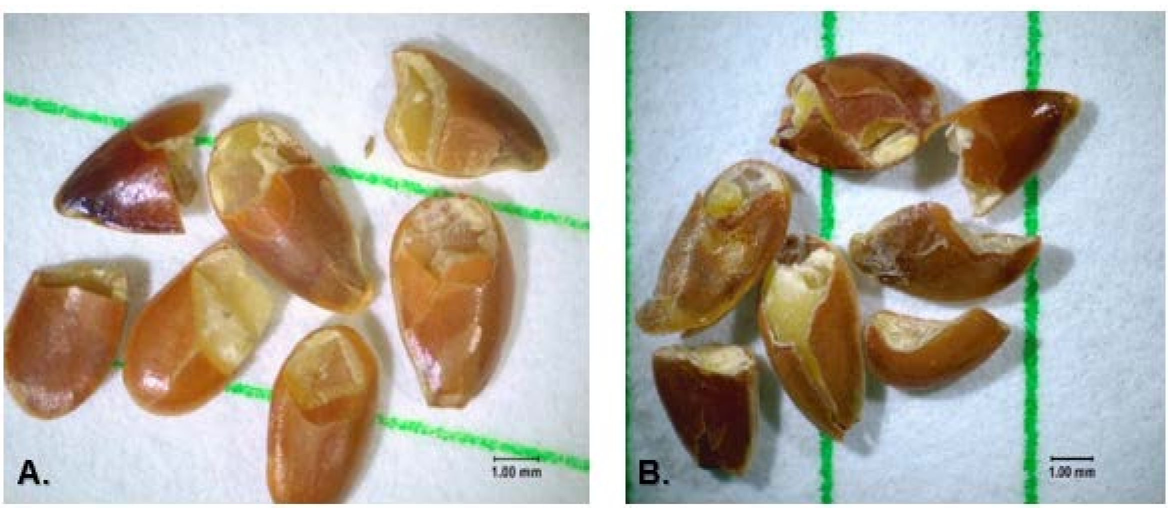

Figure V-3-C-2. A. Flax seeds with mechanical damage. Seeds are broken and usually have smooth, straight edges, where the damage occurred. Not a defect. **B.** Flax seeds with feeding damage by larvae of the almond moth, *Cadra cautella* (Walker). Note how the scalloped edges differ from the straight edges of the mechanical damage. Viewed through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-C-3. Damage of flax seed by the almond moth, *C. cautella* (Walker), larvae A. excreta pellets and webbing material covering flax seeds. B. excreta pellets, webbing material and feeding damage. Viewed through a stereomicroscope. (Source: H. Loechelt-Yoshioka,FDA).

Figure V-3-C-4 Almond moth, *C. cautella* (Walker), adult found in flax seed. Feeding damage by the larvae can be seen in the photo as well. Viewed through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-C-5. Otherwise unfit. **A.** flax seed-heat damage. **B.** flax seed-heat damage. The damaged seeds develop a dark brown to black discoloration. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

SUNFLOWER

Figure V-3-C-6. Insect damaged sunflower achenes. Damage indicated by presence of internal damage and bore holes as seen through a stereomicroscope. (Source: H. Loechelt-Yoshioka, FDA).

Figure V-3-C-7. Banded sunflower moth, *Cochylis hospes* Walsingham, **A.** larval feeding damage inside sunflower seed **B.** image of adult. (Source: Photo courtesy of Frank Peairs, Colorado State University, Bugwood.com).

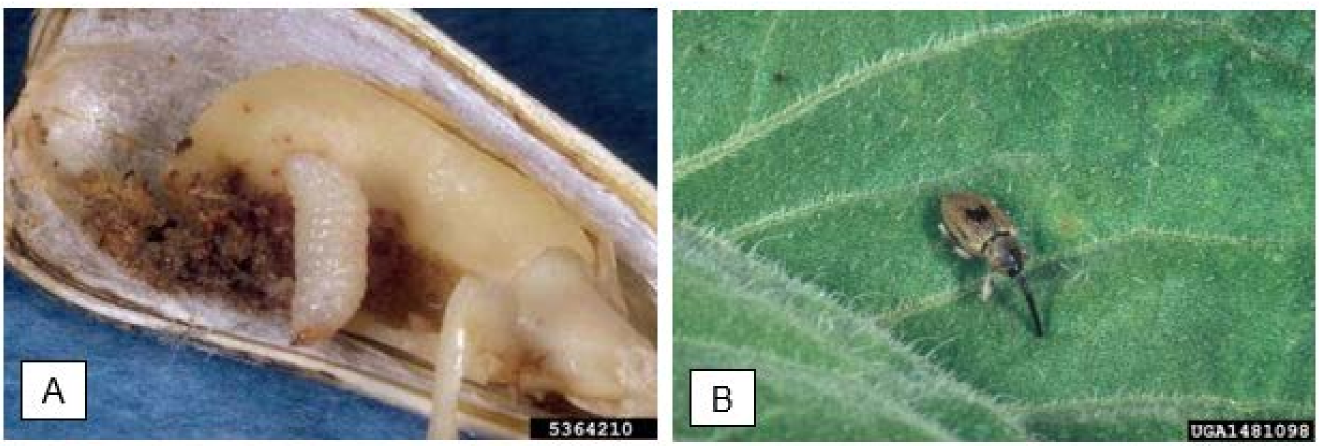

Figure V-3-C-8. Red sunflower seed weevil, *Smicronyx fulvus* LeConte, **A.** larval feeding damage with larva inside sunflower seed **B.** image of adult weevil. (Source: Photo courtesy of Franki Peairs, Colorad State University, Bugwood.com).

Figure V-3-C-9. Sunflower seed midge*, Neolasioptera murtfeldtiana* (Felt), internal feeding damaged sunflower seed. (Source: Photo courtesy of Frank Peairs, Colorado State University, Bugwood.com).

Figure V-3-C-10. Sunflower achenes showing surface mold on the pericarp. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

Figure V-3-C-11. Otherwise unfit-sunflower seed with heat damage, indicated by a reddish brown discoloration. (Source: Photo courtesy of USDA-FGIS Visual Reference Images 2016).

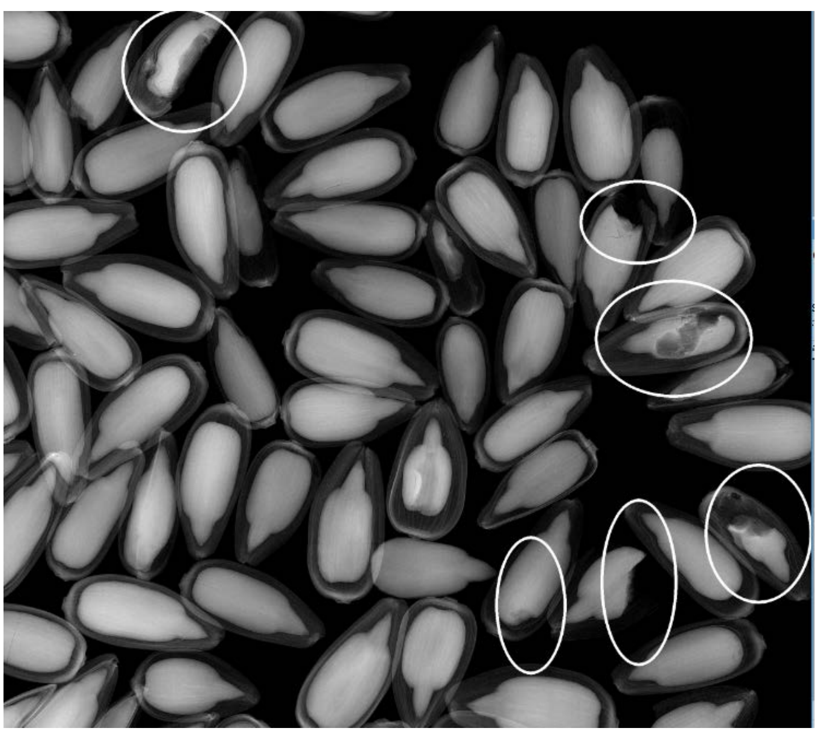

Figure V-3-C-12. X-ray image of sunflowers with hulls. Insect feeding damage indicated by circled seeds (3x). (Source: H. Loechelt-Yoshioka, FDA).

Reporting Tables

The below reporting tables are suggested for use.

Table V-3-1. Example of Reporting Macroanalytical Findings in Wheat, Corn, Grains, Pseudo-grains - Excludes Popcorn

NET WEIGHT DETERMINATION: Balance used: ☐ ☐	SUB NUMBER						TOTAL	AVERAGE
	1	2	3	4	5	6		
Gross Wt., g.								
Tare Wt., g.								
Net Wt., g.								

FILTH ITEMS RECOVERED:	SUB NUMBER						TOTAL	AVERAGE
	1	2	3	4	5	6		
Whole or W/E insects/mites, etc.: (Describe Below)								
Rodent Excreta Pellets: (Record as mg /Kg) (Describe Below)								
Bird Excreta Pellets: (Record as mg/Kg) (Describe Below)								
Insect Damaged (Visual or X-Ray): (Count Damaged Grains/100 g or damage corn/200 g)								
Moldy: (Weight of Moldy Grains/50g) (Confirm microscopically, presence of septate filaments/mycelia and/or spores)								
Otherwise Unfit: (Weight of Damaged Grains/50g) (Describe Below)								
Hard and or sharp objects/Other extraneous materials: (Describe Below)								
Total Rejects:								
Total % Rejects (Applicable to moldy/otherwise unfit):								

Notes:

Control Blank (open wetted Petri dish) Results:

☐ Sieves and scopes inspected/fit for use prior to use.

☐ Visual exam-Supplemental Magnifier X with Confirmation Scopes used:

W/E= Whole or equivalent, based on head count.

Unless noted otherwise, all recovered specimens were dead, and all sizes are in mm.

Table V-3-2. Example of Reporting Macroanalytical Findings in Popcorn*

NET WEIGHT DETERMINATION: Balance used: ☐ ☐	SUB NUMBER						TOTAL	AVERAGE
	1	2	3	4	5	6		
Gross Wt., g.								
Tare Wt., g.								
Net Wt., g.								

FILTH ITEMS RECOVERED:	SUB NUMBER						TOTAL	AVERAGE
	1	2	3	4	5	6		
Whole or W/E insects/mites, etc.: (Describe Below)								
Rodent Excreta Pellets: (Record Pellet Count/Sub) (Describe Below)								
Rodent Gnawed Popcorn: (Record Count Gnawed/ 454 g) (Describe Below)								
Rodent Hairs: (Count Hairs/ 454 g) (Describe Below)								
Bird Excreta Pellets: (Record Pellet Count/Sub) (Describe Below)								
Insect Damaged (Visual or X-Ray): (Count Damaged Grains/100 g) (Describe Below)								
Moldy: (Weight of Moldy Grains/50 g) (Confirm microscopically, presence of septate filaments/mycelia and/or spores)								
Otherwise Unfit: (Weight of Damaged Grains/50 g) (Describe Below)								
Field Corn: (Weight Field Corn/Weight Popcorn) x 100%								
Hard and or sharp objects/Other extraneous materials: (Describe Below)								
Total Rejects:								
Total % Rejects (Applicable to moldy/otherwise unfit):								

Notes:

Control Blank (open wetted Petri dish) Results:

☐ Sieves and scopes inspected/fit for use prior to use.

☐ Visual exam-Supplemental Magnifier X with Confirmation Scopes used:

W/E= Whole or equivalent, based on head count.

Unless noted otherwise, all recovered specimens were dead, and all sizes are in mm.

*Commercial popcorn can consist of 10 subs

References Cited in Section

Biles, B. V. and G. C. Ziobro (1998). *Identification of the Source of Reagent Variability in the Xanthydrol/Urea Method.* jAOAC, 81(6), pp.1155-1161.

Ergot, Canadian Grain Commission. (2019). Retrieved from https://www.grainscanada.gc.ca/en/grain-quality/grain-grading/grading-factors/grading-factors-wheat/ergot.html

Gecan, J. S., J. J. Thrasher, W. Eisenberg and P. M. Brickey, Jr. (1980). *Rodent Excreta Contamination and Insect Damage of Wheat.* Journal of Food Protection, 43(3), pp. 203-204.

Grain Trade of Australia. (2019). *Visual Recognition Standards Guide for Grain Commodity Sampling & Assessment.* Retrieved from https://www.graintrade.org.au

Mason, L. J. (2018). *Stored Product Pests: Grain Mite Acarus siro (L.).* Retrieved from https://extension.entm.purdue.edu/publications/E-222/E-222.html

Matz, S. A., (1959). *The Chemistry and Technology of Cereals as Food and Feed*, AVI Publishing Co., Inc.

McCarthy, P. (2003). *Bird Management in Grain Storage Facilities.* Australian Postharvest Technical Conference, pp.108-109.

Meyer, D. J. L. and J. Effenberger. (2010).*California Noxious Weed Disseminules Identification Manual.* California Dept. of Food and Agriculture

Michaud, J. P. and Sloderbeck, P. E. (2005). *Russian Wheat Aphid: An Introduced Pest of Small Grains in the High Plains.* Kansas State University, MF-266, pp.1-4.

Oregon State University (2021). *Whole Grain.* Retrieved from https://lpi.oregonstate.edu/mic/food-beverages/whole-grains. This link leads to a website provided by the Linus Pauling Institute at Oregon State University. U.S. FDA is not affiliated or endorsed by the Linus Pauling Instiute or Oregon State University.

Sarwar, M. (2015). *Pattern of Damage by Rodent (Rodent: Muridae) Pests in Wheat in Conjunction with Their Comparative Densities Throughtout Growth Phase of Crop.* International Journal of Scientific Research in Environmental Science. 3(4),159-166.

Shroff, A. P. (1988) *Guidelines for Sampling Basmati Rice from India*, FDA memo.

Skinner, C. E., C. W. Emmons, and H. M. Tsuchiya. (1951) *Henrici's Molds, Yeasts, and Actinomycetes-a Handbook for Students in Bacteriology,* 2nd ed., John Wiley & Sons, Inc

Smith, F. R. (1953). *Detection of Rodent Damage in Corn*. U.S. FDA Microanalytical Items 8: 2-3.

Solis, M. A. (2006). *Key to Selected Pyraloidea (Lepidoptera) Larvae Intercepted at U.S. Ports of Entry: Revision of Pyraloidea in "Keys to Some Frequently Intercepted Lepidopteraous Larvae" by Weisman 1986.* Retrieved from https://www.ars.usda.gov/ARSUserFiles/12754100/PyraloideaKey.pdf

Stibick, J.(2007). *New Pest Response Guidelines: Khapra Beetle.* USDA-APHIS-PPQ-Emergency and Domestic Programs, Riverdale, Maryland.
Retrieved from https://www.inhs.illinois.edu/files/3313/4013/2723/nprg-khapra.pdf

U.S.D.A. (2019). *FGIS-ON-19-0 Inspection of Flood Damaged Grain.* Retrieved from https://www.ams.usda.gov/sites/default/files/media/FGISpn1904.pdf

U.S. FDA (2017). *Natural Toxins and Mycotoxins*. Retrieved from https://www.fda.gov/food/chemicals-metals-pesticides-food/natural-toxins-and-mycotoxins

Valencia, D., Elias, D. J., Ospina, J. A. (1994). *Rodent Pests in Colombian Agriculture.* Proceedings of the 16th Vertebrate Pest Conference. pp. 92-94.

Visual Reference Library-Rice: Rice Objectionable Seeds. USDA (2016).
Retrieved from https://www.gipsa.usda.gov/vri/rice_8.0.aspx

Wang, R. (2007). *Identification of Disease Bodies: Ergot and Sclerotia.* Canadian Food Inspection Agency. Retrieved from https://seedanalysts.ca/assets/csaac_files/pdf/meetings/2014/EN/Identification_of_Disease_bodies.pdf

Wikipedia (2021). *Pseudocereal.* Retrieved from https://en.wikipedia.org/wiki/Pseudocereal

Zdarkova, E. (1998). *Biological Control of Storage Mites by Cheletus eruditus.* Integrated Pest Management Reviews. 3,111-116.

Zimmerman, M. L. and S. L. Friedman, (2000) *Identification of Rodent Filth Exhibits.* Journal of Food Science,65(8), pp.1391-1394.

Additional Information

Informational articles not cited in the above section, but still useful:

Christenson, C. M., and H. H. Kaufman, (1982). *Storage of Cereal Grains and their Products*. American Association of Cereal Chemists.
Dennis, N. M. *A* (1953). *Technique of Grain Orientation for Radiographic Analysis* (pp. 1-4) USDA Bureau of Entomology and Plant Quarantine, ET-310.

Grain Grading Primer. (2016)._Retrieved from https://www.ams.usda.gov/sites/default/files/media/GrainGradingPrimer11272017.pdf

Grain Inspection Handbook: Book I-Sampling, Federal Grain Inspection Service, (2006). Retrieved from https://www.ams.usda.gov/sites/default/files/media/Book1.pdf.

Grain Inspection Handbook: Book II-Grain Grading Procedures, (2013). Retrieved from https://www.ams.usda.gov/sites/default/files/media/Book2.pdf

Grain Inspection Handbook: Book III-Inspection Procedures, (2006). Retrieved from https://www.ams.usda.gov/sites/default/files/media/Book3.pdf

Harris, K. L. (1947). *Recovery of Hairs from Food Containers and Factory Equipment.* U.S. FDA Microanalytical Items: pp.4-5.

Harris, K. L., Nicholson, J. F., Randolph, L. K., and Trawick, J. L. (1952). *An Investigation of Insect and Rodent Contamination of Wheat and Wheat Flour.* Journal of the Association of Official Agricultural Chemists, 35 (1), pp.115-158.

Kim, S. S., Rhy, M., Kim, J. M., Lee, S. (2003). *Authentication of Rice Using Near-Infrared Reflectance Spectroscopy.* Cereal Chemistry, 80(3), pp. 346-349.

Milner, M. , Farrell, E. P., Katz, R. (1953) *Use of a Simple Blowing Device to Facilitate Inspection of Wheat for Internal Infestation.* Journal of the Association of Official Agricultural Chemists, 36, pp. 1065-1070.

Russell, G. E. (1988). *Evaluation of Four Analytical Methods to Detect Weevils in Wheat: Granary Weevil, Sitophilus granarius (L.), in Soft White Wheat.* Journal of Food Protection, 51(7), pp. 547-553.

Seed Impurities of Grain an Identification Kit. Grain Trade of Australia. Retrieved from https://www.graintrade.org.au

U.S.D.A (1997) *Implementation of the FGIS-FDA Memorandum of Understanding: Grain Inspection.* Retrieved from https://www.ams.usda.gov/sites/default/files/media/FGIS9060_2.pdf

U.S. FDA, (1980) *CPG Sec 555.450 Foods-Adulteration Involving Infestation and 1080 Rodenticide.* Retrieved from https://www.fda.gov/regulatory-information/search-fda-guidance-documents/cpg-sec-555450-foods-adulteration-involving-infestation-and-1080-rodenticide

U.S. FDA, (2000). *MOU 225-80-2000 Memorandum of Understanding Between the Federal Grain Inspection Service United States Department of Agriculture and the Food and Drug Administration Department of Health and Human Services.* Retrieved from https://www.fda.gov/about-fda/domestic-mous/mou-225-80-2000

Visual Reference Library-Rice: Rice Objectionable Seeds. USDA (2016). Retrieved from https://www.gipsa.usda.gov/vri/rice_8.0.aspx

Winton, A. L. & Winton, K. B. (1932). *The Structure and Composition of Foods: Vol. I Cereals, Starch, Oil Seeds, Nuts, Oils, Forage Plants.* John Wiley & Sons, Inc.: New York. 710pp.

Revision History

Version No.	Purpose of change	Date
V0	New process	1984 version
V1		1998 electronic version
V2	1. Title: removed grain products and added pseudo-grains 2. Updated table contents 3. Added more applicable documents 4. Detailed information on defects 5. A (3) added section on loose filth 6. A (4, 5) clarified sections 7. A (6) added additional molds and otherwise decomposed elements 8. A (7) removed pint 8. A (8) clarified section 9. Added descriptions of mold and otherwise decomposed 10. A (10-12) added section with rice 11. B clarified section. 12. C. added section on pseudo-grains 13. Added more references 14. Included more photos and made them 508 compliant with Alt Text 15. Added figure tables16. Added example of report tables.	May 2021