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International Standard



6639/1

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**Cereals and pulses — Determination of hidden insect infestation —  
Part 1: General principles**

*Céréales et légumineuses — Détermination de l'infestation cachée par les insectes — Partie 1: Principes généraux*

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## Foreword

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Draft International Standards adopted by the technical committees are circulated to the member bodies for approval before their acceptance as International Standards by the ISO Council. They are approved in accordance with ISO procedures requiring at least 75 % approval by the member bodies voting.

International Standard ISO 6639/1 was prepared by Technical Committee ISO/TC 34, *Agricultural food products*.

Users should note that all International Standards undergo revision from time to time and that any reference made herein to any other International Standard implies its latest edition, unless otherwise stated.

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# Cereals and pulses — Determination of hidden insect infestation —

## Part 1: General principles

### 0 Introduction

This International Standard describes methods of determining hidden insect infestation in cereals and pulses. It consists of the following parts:

*Part 1: General principles.*

*Part 2: Sampling.*

*Part 3: Reference method.*

*Part 4: Rapid methods.*

### 1 Scope and field of application

This part of ISO 6639 establishes the general principles of methods of determining hidden insect infestation in cereals and pulses.

### 2 Definitions

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

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

**2.2 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.)

**2.3 grain:** Cereal grains and/or seeds of pulses.

### 3 General

Some species of insects are especially adapted to attack whole grains and normally spend a considerable part of the life cycle, including the entire larval feeding period, inside grains. Other

species also take advantage of holes or cracks in grains to enter and feed inside them. These insects constitute a *hidden infestation* that cannot readily be seen in consignments or samples.

Most insect pests of stored grains are very small, under 5 mm in length, cryptic in behaviour and dull in colour. Those that fly do so mostly in dull light or at high temperatures. Thus, even when they are living freely outside the grains and do not constitute a hidden infestation as described above, they are not easy to detect unless populations are large enough to induce noticeable activity.

Being mobile, at least in the free-living form, insects are capable of moving through a bulk of grain and tend to concentrate in those parts of the bulk most favourable for feeding and breeding. Such centres of insect activity are not necessarily static; they may expand, contract or move for many complex reasons. The most important reasons are changes in the physical condition of the grain (for example temperature and moisture content) and overcrowding of the insects due to rapid breeding. Thus, the distribution of insects in a bulk of grain is rarely random, and their detection requires specialist knowledge and techniques.

### 4 Sampling

The methods of sampling cereals and pulses specified in ISO 950<sup>1)</sup> and ISO 951<sup>2)</sup> are not appropriate for sampling for hidden infestation in cereals and pulses, because of the non-random distribution of insect populations, particularly after a period of prolonged storage or transportation.

Special techniques, not relevant or desirable for the purposes of determination of grain quality, for which ISO 950 and ISO 951 were developed, include selecting samples from the top and outer layer of bags or packages in a stack and sampling from the surface layer and warmer regions of bulk grain. These are locations in which insect infestation is generally most likely to be found, and therefore justify the departure from the basic principles of representative sampling. Despite these differences, it will be possible in most instances for the same personnel to use the same equipment to obtain samples for either the determination of grain quality or the determination of hidden insect infestation, or for both purposes, during the same operation.

1) ISO 950, *Cereals — Sampling (as grain)*.

2) ISO 951, *Pulses in bags — Sampling*.

If information on the distribution of insects within a lot is required, the increments taken shall not be combined, but each shall be considered as a laboratory sample. In other cases, increments can be combined to form a bulk sample, which is then reduced, by an appropriate method, to a laboratory sample.

**5 Methods of determining hidden insect infestation**

There are two types of methods for the determination of hidden insect infestation, i.e. rapid and reference methods. Rapid methods, such as the X-ray, the flotation, the carbon dioxide production, the ninhydrin, and the acoustic method are specified in ISO 6639/4. A reference method against which these rapid methods may be assessed is specified in ISO 6639/3.

The reference method for true grain insects that spend the larval stage, and usually the pupal stage, within the grain is to incubate the sample at a standard temperature and relative humidity and to examine the sample at regular intervals. This

method is slow to yield results, because grain insects need several weeks to complete their life cycle.

Rapid methods of determining hidden infestation have been developed to avoid the need to wait at least 6 weeks for an estimate of population size, during which period the population may have increased many times. All methods are likely to detect the advanced stages of development with ease, but, in most cases, it is not possible to detect eggs or young larvae with certainty. In rapidly growing populations, developing insects may account for a large proportion of the numbers of individuals. If a continuing rise in temperature is recorded or an expected fall in temperature does not happen, infestation should be suspected and samples taken. The choice of method depends on the time available, cost, and on whether the user prefers an answer related to number of insects or to the mass of damaged grain. The principal characteristics of the various methods are summarized in the table.

During handling of samples, particularly when sieving is involved, there is a risk that some insects will be killed. Thus, there is no certainty that all insects at all stages will be retained intact on samples of grain.

**Table – Summary of the principal characteristics of methods of determining hidden insect infestation**

Method	Speed	Destruction of sample	Effectiveness* of detecting			Comments	Capital cost
			eggs	larvae	pupae		
Reference	Slow	Yes	XXX	XXX	XXX	Very accurate	Moderate
Carbon dioxide	Rapid	No	—	XXX	XXX	Good laboratory method	Rather high
Ninhydrin	Rapid	Yes	X	XX	XX	Field or laboratory method	Moderate
Flotation	Rapid	Yes	—	X	X	Seriously underestimates insect populations	Low
X-ray	Rapid	No	X or XX	XXX	XXX	Very accurate laboratory method, permanent record	High
Acoustic	Rapid	No	—	XXX	—	Good laboratory method requiring sound-proof facilities	Rather high

\* Scale of effectiveness

- nil
- X fair
- XX good
- XXX very good