

# Microbial update

## yeasts & moulds

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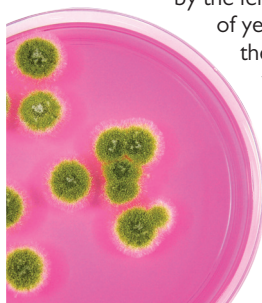
It is quite probable that fungi cause more losses within the world food supply chain than any other group of micro-organisms. As such they should be of major interest to growers, processors, suppliers and retailers of foods.

Fungi tend to be divided into two groups, the yeasts and the moulds. Yeasts are characterised by their single celled nature. They reproduce by a process known as budding, where 'daughter' cells bud off the parent cell forming new single cells. Yeasts tend to grow within food and drink matrices in planktonic form and they tend to ferment sugars growing well under anaerobic conditions. Moulds on the other hand tend to grow on the surface of objects in the form of a visible 'mycelium' made up of many cells. Moulds exist in this multicellular format, usually known as a hyphal mat, and tend to grow on the surface of food and drinks and not within them as yeasts do. They are aerobic organisms and cannot grow well under conditions in which oxygen is limited.

Yeasts and moulds form an important group of organisms of great importance to the food industry. These groups are very different from the bacteria that are commonly associated with food, being eukaryotic organisms similar to cells found in plants and animals. The yeasts and moulds are widely dispersed, found in a variety of locations and are virtually ubiquitous in any environment.

### Yeasts

Yeasts are very important within food microbiology as they can create both positive and negative effects. Where would we be without bread, beer and wine, all created by the fermentative action of yeasts? Conversely, the ability of some yeasts to survive and grow at low pH, low water activity, and in the presence of some common chemical preservatives, makes them



potent food spoilage organisms responsible for large economic losses of some food products.

The origin of the positive effects of yeasts in food production probably came about by an accidental contamination of some raw materials with environmental yeasts. Mead, a fermented honey drink, is the oldest alcoholic beverage known to man and is believed to have been discovered during the Stone Age.

A chance occurrence of honeycomb becoming wet from rain and then airborne yeasts fermenting the mixture is thought to have led to its discovery. Leavened bread first appeared in Egypt about 5,000 years ago, when flat bread dough became contaminated with wild yeasts which would have produced carbon dioxide, and 'raised' the bread. No doubt an 'accidental' contamination of various fruit juices would have caused the production of wines.



Contamination of raw materials with wild environmental yeasts is still used to produce some foods such as specialist sourdough breads and lambic beers. However, most food production that uses yeasts will now utilise specialist strains obtained from culture collections that are cultured and deliberately inoculated into their growth substrate to create the food required.

Today in food production, yeasts are more usually linked with food spoilage. Yeasts are slow growing organisms when compared to bacteria. If yeasts and bacteria were placed in the same optimum environment and both could grow, it is most likely that the faster growing bacteria would quickly outgrow and outcompete the slower growing yeast, becoming the dominant flora. However, if we move outside the 'optimum' growth conditions of most bacteria, into environments that are acidic, or of low water activity (high in sugar), then the yeasts have the advantage and would rapidly overtake the growth of bacteria. It is in these specialist niches in foods that the spoilage yeasts become a problem.

Yeasts are generally associated with the fermentation of sugars such as glucose and sucrose, but they are able to utilise a variety of other compounds, such as alcohols, organic acids, hydrocarbons and aromatic compounds. Some yeasts are also capable of utilising certain acid-based preservatives such as benzoic acid, propionic acid and sorbic acid, and this can make them a major issue in foods and drinks that rely on these preservatives for stability.

Other general environmental factors that influence growth are temperature and the concentration of oxygen. The temperature range for yeast growth is about 0-47°C, with yeasts from Antarctic soils, for example, having a maximum growth temperature of 17°C, whilst some from tropical environments will grow at greater than 40°C.

Some yeast species are strict aerobes whilst others also have a fermentative metabolism. In the case of a contaminated fruit juice for example, fermentative yeasts will cause alcoholic fermentation in the bulk of the product, whilst aerobic yeasts will produce a film or pellicle on the surface of the liquid. In most food spoilage it is the anaerobic fermentative yeasts that cause the major issue. In sealed food containers, any oxygen will be rapidly consumed creating an anaerobic environment, and it is here that the spoilage organisms will grow, the most characteristic spoilage event being gas production. Flexible containers will become distended, whilst rigid containers appear unaffected until opened when rapid pressure release can result in a forcible ejection of the contents.

The osmophilic or xerotolerant yeasts specialise in growing in environments of high osmotic pressure, due to the presence of salt or sugar. Many of the yeasts isolated from high salt environments will grow readily under salt-free conditions, but this is not the case for those organisms isolated from high sugar environments, which grow poorly or not at all on standard growth agars. Syrups, jams, preserves, fondants are all susceptible to spoilage by xerotolerant yeasts.

### Moulds

Like the yeasts, moulds can also produce both positive and negative effects in foods. Their negative effects are well known – mould contamination of products containing high sugar or of low pH is obvious with the organisms tending to grow as colonies on the surface of such products. Their positive

effects are also well known. Moulds will be seen on the surface of mould ripened cheeses such as brie and camembert, and within blue veined cheeses like Stilton and Danish Blue.

Some species are a key part of some fermented food products from Japan, whilst the meat replacement known as Quorn, is produced from a *Fusarium* mould that is cultured and treated to form a 'meat-like' texture. On the negative side, mould like yeasts can affect low pH and low water activity foods. Indeed this group can grow at very low water activities (*Aw*) causing spoilage problems in products with *Aw* values below 0.75.

Moulds are slow growing organisms and will be rapidly out competed by bacteria and even yeasts in normal conditions. They come into their own when the pH and/or *Aw* is reduced and other groups are unable to grow. Then the moulds can take over, forming hyphal mats or colonies on the surface of food products. It is important to realise that moulds are aerobic organisms, they need oxygen to grow, so they will usually be confined to the surfaces of foods, forming easily visible, often coloured colonies.

## Airborne yeasts

Yeasts which are present in the air must be capable of surviving very harsh conditions. Airborne yeasts can be present in the form of ascospores and although these are more resistant than vegetative yeast cells, the majority of yeast particles present in the air will not be viable. Soil, dust, drains, equipment surfaces, raw materials and ventilation ducts can all release yeasts into the air.

Viable counts from settle plates reflect a dynamic situation of yeast particles becoming airborne and then settling again. The size of the yeast particle determines how far it will travel before settling, with most airborne particles being between 2-20µm in size. As a source of product contamination, airborne yeasts are of most concern for such operations as aseptic filling plants and, in these cases, control of the air quality by means of air filtration will be necessary.

## Processing equipment

A lack of attention to the hygienic design of factory plant and cleaning and sanitation procedures can lead to yeast and mould contamination of products. This is particularly problematic in plants producing high sugar/low water activity/low pH products. Factories producing fruit products, baked goods, confectionary, fermented dairy products etc can be at real risk from yeast and mould contamination.

In such plants, care must be taken to ensure that equipment such as proportioning pumps, hose connections and valves, storage chillers, and other product contact

surfaces are adequately sanitised. Factories should be designed in such a way as to reduce the risk of cross contamination of the finished product by the raw product. In many cases it is assumed that low pH products are quite stable due to an inability of bacteria and many yeasts to grow in such conditions.

However it only takes very low levels of specialist spoilage yeasts and moulds to create major spoilage problems in these products. Such organisms are ideally suited to the environmental niches in these production environments, hence the need for good hygiene in such areas.

## Packaging materials

It is of considerable importance to consider the hygienic status not only of the product and production equipment, but also of any containers and packing materials, as these too can be a source of spoilage yeast and moulds.

Any purchased containers, for example jars and bottles and container closures (lids/caps), have to be assessed for their level of contamination and any need for decontamination before use. The storage of such containers/closures before use is also important. On a number of occasions the storage of opened pallets of containers in open/semi-open environments drastically increases the potential for contamination.

Cardboard may be highly contaminated with environmental yeast and mould spores and even foils and plastics may show low levels of contamination. Yeast numbers may increase substantially during storage, unrolling and moulding due to static electricity attracting dust from the environment.

## Resistance and illness

Moulds and some forms of yeast are able to produce spores, and these can be heat resistant. This makes the determination of any heat process used on food products very important. It is not unknown for heat-processed, low pH/low *Aw* products to be spoiled by moulds whose spores have survived heat processes able to destroy vegetative bacteria. Generally, yeast and mould spores are not as resistant as bacterial spores, but are much more resistant than vegetative bacterial or yeast cells.



Companies wishing to establish heat processes able to destroy such organisms are advised to contact specialist advisors who can help to establish suitable heating processes.

Whilst yeasts are not usually acknowledged to cause foodborne illnesses, some moulds can produce a range of mycotoxins that can have chronic effects. Unacceptable levels of many mycotoxins are detailed in legislation, and this should be consulted if moulds are considered a potential problem in particular foods types.

Yeasts and moulds can be major problems within certain sectors of the food industry. Any food or ingredient that has a reduced pH, a low water activity, or has been chemically preserved may be open to attack by yeasts or moulds.

They are able to grow in pH and *Aw* conditions that will prevent all bacterial growth, they can be resistant to many chemical preservatives, and some fungal spores can exhibit heat resistance. Some yeasts have even been seen growing as small visible colonies on the surface of chilled proteinaceous products such as chilled sausages or chicken portions giving rise to a 'white spot' spoilage.

## Testing methods

It is of great importance to ensure raw material, the production environment and the final product are subject to testing for potential spoilage yeasts and moulds. This, however, is not as easy as it sounds.

Whilst 'normal' yeasts and moulds may be easily grown on media such as Rose Bengal Chloramphenicol Agar (RBCA) or Malt Extract Agar (MEA), specialist spoilage organisms present in low water activity or low pH environments may not. It is usual for low water activity foods to be tested using media such as DG18, and even the initial dilution of the foods may need to be done using a specialist diluent, containing levels of sugar.

This will prevent the yeasts and moulds being injured by osmotic stress. Injured cells may not grow, will not be detected and thus a possible problem will remain hidden, until product begins to spoil! It is recommended that when setting up testing regimes for such organisms, expert help is consulted on methods required.

Identification of yeasts and moulds is a specialist area, requiring specialist knowledge. The development of molecular methods has been moving the basis for identification away from fermentation based and morphological assessment, towards the analysis of the DNA within the organism. Again if identification is required, specialist laboratory help should be requested. ■

✉ [food.testing.admin@thermofisher.com](mailto:food.testing.admin@thermofisher.com)

*References are available from the author on request*