Microbial update
environmental yeasts

by Roy Betts, Head of Microbiology, Campden-BRI, UK. Produced as a service to the food industry by Oxoid Ltd, part of Thermo Fisher Scientific.

Yeasts are a unique and important group of micro-organisms. Their microscopic structure is very different to that of bacteria, and their cells are more similar to those of plants and animals. They can be found in a wide variety of locations, and are virtually ubiquitous in any environment.

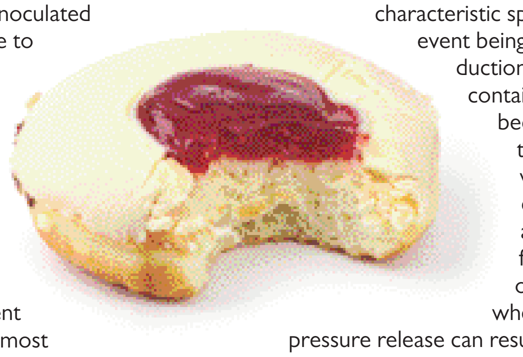
Yeasts are very important within food microbiology as they can create both positive and negative effects. Where would we be without bread, beer and wine, all created by the fermentative action of yeasts.

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

Undoubtedly the origin of the positive effects of yeasts in food production came about by an accidental contamination of some raw materials with an environmental yeast. 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.

Today, 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 (for example high in sugar), then the yeast 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.

Yeast growth

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, conserves and fondants are all susceptible to spoilage by xerotolerant yeasts.

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.

Contamination of water

Yeasts are not commonly reported as a normal part of the groundwater flora and their presence would indicate that a high carbohydrate or organic acid pollution may be occurring. However, where water is abstracted as surface water about 50% of the fungal flora are yeasts. Pichia spp. in particular are found in water supplies. Yeasts may also be present in badly maintained factory water systems.

Processing equipment

A lack of attention to the hygienic design of factory plant and cleaning and sanitation procedures can lead to yeast contamination of products, especially in fruit juice plants where the juices may be stored chilled as part of the manufacturing process.

Care must be taken to ensure that equipment such as proportioning pumps, hose connections and valves, which by their nature may be difficult to clean, are adequately sanitised. The factory 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 a specialist spoilage yeast to create major spoilage problems in these products. Such yeasts are ideally suited to the environmental niches in these production environments, hence the need for good hygiene.

Packaging materials

It is very important 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 contamination.

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 with environmental yeasts.

Cardboard may be highly contaminated with environmental yeast 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.

Contamination of foods

● Dairy products

Yeasts play an important role in the spoilage of dairy products as some are able to grow at low temperatures and low pH. Yeast spoilage is more important in fermented milk products than in fresh milk products. In cheese processing, yeasts may be introduced in the milk, via the rennet or from the production environment. They may spoil soft or fresh cheese by the production of gas and off-flavours. In yoghurt manufacture, yeasts are eliminated in the milk by pasteurisation, but may be reintroduced into the product by contaminated fruit pulps or via the production and packaging environment. The doming of yoghurt pot foils is a characteristic symptom of yeast spoilage, while gas formation in sealed packs of cheese, will result in pack distension and even blowing.

● Meat products

The spoilage of fresh meat by yeasts is limited unless there are unusual conditions and competition from the normal bacterial flora is reduced. A similar selection process occurs in cured or dried meats with the reduced moisture content favouring some yeasts. Natural tolerance to organic acids, usually lactate and acetate, also have implications for meat plants as these are increasingly been used to decontaminate the surfaces of meat and poultry of pathogenic bacteria.

● Fruit products

In the field fruits are exposed to environmental yeasts present in air, soil, wind blown dust, rain and via insects. These normally colonise the fruit surfaces and may be present in numbers of 10^2-10^4/cm of fruit surface. The intact fruit skin provides limited nutrients, but once the fruit is processed, for example as ready to eat fruit pieces, fruit pulps or juices, the yeasts are then in a high sugar, high acid environment and numbers can reach 10^8 yeasts per gram. Spoilage is indicated by gas production and off-flavours.

● Bakery products

Bakery coatings and fillings with a high water content are normally spoiled by bacteria. Spoilage problems associated with yeasts in bakery products occur in lower available water products such as uncooked pastry doughs. Osmophilic yeasts spoil very low water products such as dried fruits, nut paste, marzipan, icings and jams. These organisms may be present in the materials themselves or may contaminate them via poor hygiene. Attention needs to be paid to completely removing sugars from surfaces of production equipment, as a film of sugar water may exacerbate contamination problems.

● Confectionery

There are well known instances of fondant filled chocolate products being contaminated by very specialist yeast. These can grow within the filling producing gas, and causing a blowing of the chocolate shell.

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. This, however, is not as easy as it sounds. Whilst 'normal' yeasts may be easily grown on media such as Rose Bengal Chloramphenicol Agar (RBCA) or Malt Extract Agar (MEA), specialist spoilage yeasts 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 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 spoilage yeasts, expert help is consulted on methods required. ■

FaxNOW +44 1256 329728
✉ val.kane@thermofisher.com

References

● Boekhout, T. and Robert, V. (Eds). Yeasts in Food. Woodhead Publishing 2003.

● Samson, R. A., Hoekstra, E. S. and Filtenborg O. Introduction to Food Borne Fungi. Centraalbureau Voor Schimmelcultures 1995.

● Skinner, F. A., Passmore, S. M. and Davenport, R. R. Biology and Activities of yeasts. 1980 SAB Symposium Series no. 9 Academic Press.

● Suriyarachchi, V. R., and Fleet, G. H. (1981). Occurrence and Growth of Yeast in Yoghurts. Applied and Environmental Microbiology Oct., Vol 42, No.3, p.574-579.

● Campden BRI. (2007). Manual of Microbiological Methods for the Food and Drink Industry. Campden BRI Guideline No. 43.