

A microscopic view of microbial products, showing numerous clusters of yellowish, fuzzy, and granular structures against a dark background. A semi-transparent white rectangular box is overlaid on the upper left portion of the image, containing the title and author information. A small yellow rectangular shape is located in the top left corner of the white box.

Safe-handling of Novozymes microbial products

By Novozymes Product Safety

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This handbook is intended for managers and employees in production facilities handling intermediate microbial products. It provides information useful to companies in designing their own production safety program.

Here you can find operational safe-handling procedures for daily work that Novozymes shares as inspiration.

You can also find general information about Novozymes' assessment methods, background about regulatory requirements, exposure limits, etc.

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Introduction

As a world leader in biotechnology, Novozymes produces a variety of naturally-occurring, beneficial microbial products. These products include yeasts, fungi and bacteria, and range from tech-grade to feed- and food-grade. We commonly refer to these diverse products as “microbials”. Microbial products are used to improve the performance of a wide variety of sustainable end-products used in agriculture, industrial and consumer cleaners, human health supplements, and many other useful applications.

Like other substances containing tiny organic particles, microbial products should be handled with care. Our primary concern is the safety and efficacy of our products. Novozymes thoroughly screens each microbial strain to avoid any toxic or infectious organisms. Even so, it is important to handle microbial products safely to prevent health reactions that can occur from aerosolized exposure to microbials or organic dust.

This handbook is designed to give you a good understanding of how to handle Novozymes microbial products in a knowledgeable and safe way.

This handbook has two sections. The first describes background information about why it's important to handle microbial products with care, including how Novozymes assesses its products, the general hazards of inhaling bioaerosols, exposure limits, etc. The second section has practical information, what industrial hygiene experts suggest to ensure safe handling, what respiratory protective equipment is available, how to clean spills, etc.

These guidelines are intended to provide information to managers and employees who are developing a safety program for facilities that handle microbial products. The safe-handling information here does not apply to consumer end-products that contain microbials, as these are designed to be very safe. Neither does the information here address production issues having to do with product quality.

We hope you find this handbook useful. Novozymes is eager to support you in getting the necessary knowledge so you can confidently and safely handle our microbial products and get the most out of them with the smoothest possible process.

If you have questions, please check [Novozymes.com](https://www.novozymes.com) or contact your Novozymes sales representative.

Section one

The background is an abstract composition of organic textures. A large, bright yellow-green area occupies the center and right, while darker green and brownish-green areas are on the left and top. Numerous small, translucent bubbles or droplets are scattered across the surface, particularly in the darker green and brown areas. The overall effect is one of natural, fluid movement and organic growth.

1. Novozymes' approach to safety

Novozymes' aim is to market microbial intermediate products that are assessed and found to be safe to handle and use when the recommended safe-handling guidelines are followed.

Novozymes conducts a thorough risk assessment to evaluate the safety of all our microbial products. This includes classification of microorganisms into one of four biosafety risk groups, based on their likelihood to cause diseases. Novozymes exclusively develops products from microorganisms in Risk Group 1, which are documented to have no harmful effects on humans.¹⁻³

Nonetheless, if the microbial products are not handled correctly, health reactions may arise. Inhalation of bioaerosols or dust of any organic matter (e.g. pollen, bacteria, flour, soils, etc.) above a certain threshold can lead to unwanted health reactions, and can be avoided by correct handling.

The information here is provided to empower users to make informed decisions about their own safety programs and procedures. Our microbial products are described in detail in the Novozymes Safety Data Sheets (SDS) and product labels. In addition, production facilities may need to adhere to country-specific safe-handling requirements, where relevant.

As a manufacturer, Novozymes must comply with safety requirements from regulatory authorities around the world. As an example, here is the legal context in the European Union:

Microbial products fall under the EU Directive on occupational health risks of biological agents. EU Directive 2000/54/EC⁴ regulates the minimum requirements for the protection of workers from health risks related to biological agents. Accordingly, employers (e.g. manufacturers and blenders of microbial products, professional cleaning service companies, other companies using microorganisms for example to produce food and beverage products) are required to conduct a risk assessment, including the classification of the microorganisms used into one of four risk groups based on the pathogenic potential⁵.

Potential allergenic or toxigenic effects (especially the former) are not reflected by the risk group scheme, but these effects also must be considered⁶. EU Directive 2001/95/EC⁷ on general product safety also applies and requires a safety assessment and risk-related information be made available to consumers by manufacturers and importers of these products.

1 WHO Laboratory Biosafety Manual, 4th edition, 2020

2 EU Directive on Risk Group: 2000/54/EC

3 CDC Quick Learn Lesson: Recognizing the Biosafety Levels

4 European Union, 2000, Directive 2000/54/EC, Doc 32000L0054

5 European Union, 2000, Directive 2000/54/EC Annex III

6 European Union, 2000, Directive 2000/54/EC Articles 3, 3(d)

7 European Union, 2001, Directive 2001/95/EC, Document 32001L0095

2. Novozymes' safety assessment of strains & applications

Novozyymes' safety assessment is a structured process that ensures our microbial products undergo an accurate and consistent analysis to ensure that:

1. Novozymes microbes cannot cause disease or toxicity (strain safety assessment)
2. Novozymes microbes are not likely to be used or applied in a way that results in bioaerosols with high concentrations of microbes above a certain limit (application safety assessment, i.e., a bioaerosol exposure assessment)

Strain safety assessment

During the risk assessment of a microbial strain, taxonomic identification, and characterization of the potential hazard are based on both genotypic analyses (e.g., bioinformatics/whole genome sequence-based searches for genes of concern) and phenotypic analyses (e.g., antimicrobial susceptibility testing, in vitro cytotoxicity testing). The risk assessment also includes an exhaustive scientific literature review as well as in vivo toxicological data, if available.¹

Application safety assessment

Assessments of exposure routes and exposure levels are based on available information about current known use patterns, proposed uses or other potential uses (e.g., misuses). Assessments may involve realistic exposure measurements representing a worst-case scenario under simulated use conditions.

The risk is characterized depending on the hazard severity (e.g., production of toxins) and the potential for exposure above a certain limit, taking into consideration the weight of evidence and scientific uncertainties.²

Definition of bioaerosol

A general industry rule of thumb describes inhalable bioaerosols as tiny (100 microns or less in diameter) organic dust particles or aerosol microdroplets that float in the air and can be inhaled. Bioaerosols can be generated from liquid or dry microorganisms, dead or alive, or including raw materials like plant or microbial-based hydrolysates, or even peat or yeast.³⁻⁴

The safety assessment can be expressed as a simple formula:

Risk is proportional to Hazard × Exposure

¹ EFSA, 2018, "Guidance on the characterisation of microorganisms..."

² Louter et al. 2012, "Beneficial Microorganisms In Agriculture..."

³ OSHWiki, definition of bioaerosols

⁴ CDC, The National Institute for Occupational Safety and Health (NIOSH)

3. Respiratory health effects of inhaling bioaerosols and organic dust

Inhalation of bioaerosols or dust containing any organic matter (e.g. pollen, bacteria, flour, soils, etc.) can lead to unwanted health reactions, and should be avoided by correct handling. Inappropriate handling can lead to exposure levels that deserve attention. Some microbial products are supplied as freeze-dried or spray-dried products, which may include an organic carrier as well as the microbe. These may present a higher exposure risk than a liquid formulated product.



Exposure to high and low concentrations

If a worker inhales high concentrations of bioaerosols or dust from organic materials, the person may develop organic dust toxic syndrome (ODTS). ODTS is an acute, temporary and non-infectious illness that is caused by temporarily overloading the lungs with bioaerosols. It is often described as producing flu-like symptoms, including weakness, headache, fever, chills, body aches, coughing or shortness of breath. ODTS can occur within several hours after inhalation. It typically resolves within 24 hours to a few days. Medical treatment is usually not required for otherwise healthy individuals.¹

The same causative factors contributing to ODTS can also lead to a rare, chronic, and often irreversible lung disease known as hypersensitivity pneumonitis (HP). Examples of working environments with risk exposures for HP include farmers working with mouldy hay, poultry handlers breathing in particles from feathers and bird droppings, or bakers working with contaminated flour.

Where ODTS may be linked to high and intermittent exposures, HP can develop after numerous or continuous lower levels of bioaerosol exposure. Therefore in a health risk assessment, repeated or daily respiratory exposure - even at low concentrations - should also be considered.

Detection of ODTS is a valuable indicator that the working environment is not in good control and needs improvement. Importantly, mitigation efforts to prevent ODTS simultaneously and proactively also reduce the risk of HP.

1 P.A. Genevois and P. Vuyst, 2006, "Imaging of Occupational and Environmental Disorders of the Chest"

Table 1:
Summary of the inhalation/respiratory health effects of bioaerosols and organic dust

	Organic Dust Toxic Syndrome (ODTS)	Hypersensitivity Pneumonitis (HP)
Exposures	Single, very high exposures. Relevant in industrial settings.	Frequent repeated exposure
Consequence of re-exposure	Transient	Can develop into chronic illness
Physical signs at examination	Absent	Chest X-ray abnormalities, decreased pulmonary function and blood gases

Stay on the safe side

Novozymes’ microbes used in microbially-based products are non-pathogenic and non-toxigenic (Risk Group 1 according to the WHO classification). However, because microbes contain proteins and all non-human proteins can theoretically become respiratory or dermal sensitizers under certain conditions, there is a potential risk of microbes acting as sensitizing agents.

Due to the lack of suitable testing methods, all microorganisms are therefore considered to be potentially sensitizing by both the dermal and the inhalation routes until further guidance becomes available or scientific evidence shows otherwise.

It is therefore recommended to use the same industrial hygiene precautions as you would use with any other material of proteinaceous nature.¹⁻⁵

For any new application, Novozymes performs an assessment of the risks from exposure by airborne/ respiratory and other routes (e.g., dermal, oral, etc.). For companies who need information on safe-handling of enzymes as well as microbials, please visit Novozymes’ homepage.

Assessing respiratory sensitization

It should be noted that there is no agreed-upon test for assessing respiratory sensitization due to varied responses to microbial exposure. For example, bacteria are not likely to elicit IgE-mediated responses (type 2 immunity / hypersensitivity type I). This is in contrast to fungi. Therefore, Novozymes follows the current methodologies and safety guidelines recommended by international authorities, including the European Food Safety Authority for the risk assessment of microbial additives and consumer products.⁶⁻⁹

1 WHO Laboratory Biosafety Manual, 4th edition, 2020
2 US EPA, 2018, “Label Review Manual Chapter 10”
3 EFSA, 2012, “Guidance on studies...”
4 ECHA, 2017, “Guidance on the Biocidal Products Regulation”
5 Federal Register, 2007, Vol. 72, No. 207
6 NIH, National Library of Medicine, 2014, Steiling et al.

7 CDC, NIOSH Manual of Analytical Methods (NMAM), 5th Edition, Lindsley et al.
8 EFSA, 2012, “Guidance on studies...”
9 EFSA, 2018, “Guidance on the characterisation...”

4. Exposure limits and monitoring

No globally harmonized exposure limits to bioaerosols have been defined by authorities. Based on national guidance references, Novozymes has developed an exposure limit for safe-handling that we use in our own production facilities.

National authorities have proposed various guidance values for maximum acceptable airborne levels of fungi and bacteria (expressed as colony-forming units [CFU]/m³), but these differ between countries. Examples of guidance values can be found in the scientific literature for non-sensitized workers in occupational environments.¹⁻⁴



Novozyymes' internal exposure limit and monitoring

Novozyymes has evaluated a number of sources to help establish our own internal exposure limit based on the potential risks that we have identified within our own organization. Novozymes internal guidance is that employees stay within an 8-hour time-weighted average (TWA) limit of 1×10^4 CFU/m³ for bioaerosols generated during manufacturing processes including microbial products. We use this limit in our own production, and when we assess a new application.

The exposure of workers by inhalation is best measured by personal air monitors, or by measurements of dust in the work environment. Such data are not always available in every production facility, so instead it is common to make a conservative estimate of worker exposure from the dusting potential (g/m³), as measured by the Stauber-Heubach method and the calculations recommended by EFSA.⁵⁻⁷

Novozyymes recommends that you conduct a risk assessment based on your own production setup, employees and local regulations. The exposure limit appropriate for your situation may therefore be different from that chosen by Novozymes.

1 OSHwiki, Section 7, "Threshold limit values for occupational bioaerosols"

2 IRSST, 2001, Goyer et al., "Bioaerosols in the workplace..."

3 Critical Reviews in Toxicology, 2009, Volume 39, Issue 10, Eduard

4 BMC Microbiology, 2010, Article #233, Barfod et al.

5 American Industrial Hygiene Association Journal, 1992, Vol 53 Issue 7, Carlson et al.

6 EFSA, 2012, "Guidance on studies..."

7 Stauber D and Beutel R. (1984). Bestimmung und Kontrolle des Staubpotentials von Futtermittelvormischungen. Fresenius' Journal of Analytical Chemistry 318(7), 522-524.

Section two

The background of the image is an abstract, ethereal composition of fine, wispy textures. It features a mix of deep purple and dark blue tones, with bright white and light lavender highlights that create a sense of depth and movement. The textures appear soft and fibrous, resembling smoke, mist, or perhaps delicate fibers caught in light. The overall effect is dreamlike and atmospheric.

5. Respiratory Protective Equipment

When handling microbial products under enclosed conditions in an area with a good ventilation system, respiratory protective equipment (RPE) may not be needed.

However, if high levels of bioaerosols are suspected, respiratory protective equipment can be considered. The information below is offered as examples for your consideration. Novozymes recommends that you conduct a risk-assessment based on your own production setup, employees and local regulations.



When to use respiratory protection

Situations where respiratory protective equipment should be used:

- When there has been a significant spill of a microbial product
- When equipment used for handling microbial product is being cleaned, for example before repair or maintenance
- During open handling processes, in non-ventilated areas
- During manual disposal of packaging materials
- If an enclosed section of a process line is breached, for example when a process breakdown necessitates opening an enclosed system
- If the exhaust ventilation system is not working properly
- When handling material collected by the local exhaust ventilation system
- If ordered by your supervisor, your safety officer, or medical staff

Choosing personal protective equipment

When choosing respiratory protective equipment (RPE), employers can select appropriate RPE or consult a qualified health and safety professional for guidance. The consultant will be able to provide the necessary Assigned Protection Factor (APF), which is the level of protection that a respirator (or class of respirators) is expected to provide, based on the expected microbial bioaerosol exposure at the workplace.¹⁻²

It is important that respiratory protection is clean and checked for proper fit at all times to ensure optimal protection. Masks should be stored away in a non-production area and away from potential microbial exposure.

Examples of respirators

Below, you can see types of respiratory protective equipment shown here for illustration purposes. They all protect against ODTs potentially caused by microbial exposure in production facilities or microbial pesticides, etc.

Examples of respiratory protection



N95 or P2
filtering facepiece respirator



N100/P100 or P3
filtering facepiece respirator



P100 or P3
elastomeric particulate respirator

1 IRSST, 2007, Lavoie et al., "Guide on respiratory protection against bioaerosols"

2 ICM, respiratory protection guide in Danish

6. How to use microbial products safely in production

The general best-practice elements below are provided as inspiration to support production facilities in setting up their own safety programs. Novozymes recommends that you conduct a risk assessment based on your own production setup, employees and local regulations.

Preventing microbial exposure

Industrial hygiene experts recommend that production facilities be designed, equipped and operated in such a way that, wherever possible, employees can perform normal work using a minimum of personal protective equipment. The use of personal protective equipment (e.g., respiratory protection) should be necessary only during cleaning of spills, maintenance work, and open handling of materials that can generate bioaerosols.

The workplace should minimize visible dust, aerosols, or spills e.g. by:

- Choosing proper process equipment
- Having appropriate ventilation systems, including local exhaust ventilation where applicable
- Encouraging high standards of personal and workplace hygiene
- Paying attention to maintenance programs and process changes

Safe handling

Adding microbial intermediate products as late as possible in the production process will reduce the risk of exposure to organic bioaerosols and contamination of the workplace. It is important that microbial products are handled (mixed, transported, and dispensed) using equipment that minimizes creation of bioaerosols.

Please keep in mind:

- When adding microbial intermediate products to a process, it may be appropriate to install local exhaust ventilation or enclose the production line to reduce the risk of exposure
- Mixers for formulating products containing microbes should provide agitation without shear or crushing forces that could generate bioaerosols
- If a dedicated microbial line is not used, then cleaning with a detergent is typically sufficient for worker safety. If there are concerns about cross-contamination, cleaning should be done with a sporicidal agent
- For dry products, avoid high-velocity pneumatic transporters and drag conveyors
- For liquids, avoid splashing or spraying without proper engineering controls

Packing finished products

If not properly designed, filling machines may trap or generate bioaerosols. Additionally, large quantities of product spillage can build up. It is recommended that filling machines be encapsulated and have an efficient local exhaust.

Equipment design

All equipment containing or transporting microbial intermediate products should be evaluated in terms of their potential to create bioaerosols. In general, be aware of equipment with moving parts.

When handling liquids, pay attention to mixing and dispensing systems that might generate bioaerosols. All leaks and openings in equipment containing microbial intermediate products should be closed or encapsulated.

Ventilation systems

Exposure to bioaerosols can be minimized by constantly renewing the air in the workplace, and by capturing bioaerosols at the processing points where they are likely to be generated by means of local exhaust ventilation.

A production plant can be divided into smaller, enclosed and separated sections. This makes it possible to contain possible contamination within a relatively small area. It also makes it possible to ventilate each section with a different air-change rate where appropriate.

General recommendations:

- Keep equipment containing microbial products in well-ventilated, enclosed areas and possibly also under negative pressure (i.e., air should flow inwards through all openings where the product could escape)
- Contain the points where microbial products are added, and provide a local exhaust unit to remove any airborne microbes
- Make sure that the design takes account of the effect of opening doors and windows on the ventilation system
- Draw up a program for maintaining the ventilation system
- Use a HEPA (high efficiency particulate air) vacuum for any cleaning operations
- Consult a ventilation expert for guidance when establishing or changing facilities

Storing and handling microbe containers

It is good practice to keep microbial products in their original packaging for as long as possible before use. Follow product instructions for storage temperature and humidity. Any open containers that are partially used can be resealed to prevent any possible exposure.

Emptying containers

Emptying packages is an operation that requires the handling of concentrated microbial products. Industrial hygiene experts suggest the following: Make sure that the area is well ventilated and demarcated from the rest of the plant.

- Employees should receive training to perform this operation
- Wear appropriate protective equipment when opening and emptying microbial product containers
- The emptying procedure and the equipment should be designed to avoid and contain any spillage
- Specially-designed emptying stations are available. For emptying smaller containers, downflow air booths are safe and convenient solutions
- Local exhaust ventilation should be provided at points where there is a risk of microbe-release into the working environment

Removal of empty microbial product containers

Residues in empty containers can be a source of airborne microbes and dust if the containers are handled inappropriately. Packaging waste must be safely contained until further processing.

Respiratory protection and local exhaust ventilation should be used when handling waste material. Empty containers may be incinerated or washed and recycled, depending on the type of container and local regulations.

Damaged containers

Damaged microbial product containers or packages can cause spills and generate airborne organic dust and bioaerosols during transport and shipping. It is therefore necessary to handle such containers and packages carefully.

7. Spills and housekeeping

Maintaining high standards of cleanliness in the production environment is an efficient safeguard against exposure to bioaerosols. This makes it much easier to quickly detect spills and leaks, and to facilitate a safe working environment.

Routine cleaning

Working facilities should be easy to clean. A general recommendation is that cleaning be performed by washing with a soft stream of water from a hose, wet mopping, or vacuum cleaning to avoid bioaerosols. Vacuum cleaners should be equipped with High Efficiency Particulate Air (HEPA) filters.

Do not sweep with brooms or use high-pressure flushing with air or water, as these can create high levels of bioaerosols.

Spills

Spills of microbial-containing material in the workplace should be cleaned up immediately and in compliance with your local procedures. In general, industrial hygiene experts suggest that you pay attention to the following steps in case of a significant spill or exposure to airborne bioaerosols:

- Leave the area (in case of a major spill with dust emission, the entire area should be completely evacuated)
- Notify the supervisor
- Fetch the appropriate personal protective equipment, safety equipment, and cleaning tools
- Contain the spill
- Clean up the spill by using wet methods or an adequate vacuum cleaning with HEPA filter. If these are not available, carefully shovel the bulk of the product into a plastic-lined container suitable for enclosure and safe disposal
- Safely clean or dispose of all equipment, tools, and clothes used in the cleaning operation



For minor dry spills, use a vacuum cleaner with HEPA filter to remove the bulk of the product and finish up with a soft stream of water from a hose.



For minor wet spills, use a soft stream from a water hose (not jets or high pressure).

Discharge of water containing microbial products should be performed according to local legislation, where such exists.

8. Personal hygiene and first aid

Personal hygiene is important. As a general rule of thumb, industrial hygiene experts recommend the following standards when working with materials that can generate bioaerosols:

✓ REMEMBER

- Wash your hands before eating, smoking or using the restroom
- Use water and a mild soap, and dry your hands thoroughly afterwards
- Change your work clothes frequently
- Take a shower at the end of each work day

X AVOID

- Direct contact with the product
- Contamination of clothes, as microbial residues can be carried to other parts of the plant or to your home
- Taking work clothes home for cleaning

+ First aid recommendations

If microbial products are swallowed:

- Rinse your mouth and throat thoroughly with water and spit out
- Notify your manager and seek medical attention if irritation occurs

If bioaerosols are inhaled:

- Move to an area with fresh air
- If symptoms of irritation occur (for example shortness of breath, wheezing, or severe coughing), notify your manager and seek medical attention if symptoms persist

If your skin or eyes come into contact with a microbial product:

- Rinse skin with plenty of water
- Hold your eyelids open while rinsing with plenty of water (use an eye fountain or an eye-rinsing bottle if available)
- Notify your manager and seek medical attention if irritation occurs

If clothes are contaminated with a microbial product:

- Remove and wash contaminated clothing before re-use
- Avoid taking work clothes home
- Wash affected body parts or take a shower before you change into clean clothes

We hope you've found this handbook useful.

For more information about our products and company, please consult Novozymes' website or your sales representative.

www.novozymes.com/en/biology/microbes

About Novozymes

Novozymes is the world leader in biological solutions. Together with customers, partners and the global community, we improve industrial performance while preserving the planet's resources and helping to build better lives. As the world's largest provider of enzyme and microbial technologies, our bioinnovation enables higher agricultural yields, low-temperature washing, energy-efficient production, renewable fuel and many other benefits that we rely on today and in the future. We call it Rethink Tomorrow.

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