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# Suitable and unsuitable microorganisms

The following lists give selected microorganisms which present minimum risk given good practice.

As a result of changes to the hazard categorisation of certain microorganisms by the Advisory Committee on Dangerous Pathogens (ACDP), this table supersedes the list found in the CLEAPSS *Laboratory Handbook* (1992), the CLEAPSS *Shorter Laboratory Handbook* (2000), *Microbiology: An HMI Guide for Schools and Further Education* (1990), *Topics in Safety* (1988 and 2001) and *Safety in Science Education* (1996). As well as naming suitable organisms, the list gives points of educational use & interest and comments on the ease with which organisms can

# be cultured and maintained. The list of microorganisms is not definitive; other organisms may be used if competent advice is obtained.

It should be noted that strains of microorganisms can differ physiologically and therefore may not give expected results. Where possible, fungi that produce large numbers of air-borne spores should be handled before sporulation occurs, so that the spread of spores into the air and possible risks of allergy or the triggering of asthmatic attacks are minimised. This is particularly important for some species, such as *Aspergillus* and *Penicillium*, which produce very large numbers of easily-dispersed spores. It should be noted that certain species of these two fungi, previously listed as unsuitable for use in schools, are now not thought to present such a serious risk to health, given good practice in culture and handling.

 Updates to the categories of hazard are published periodically in *The Approved List of Biological Agents*, which is available online at www.hse.gov.uk/pubns/misc208.pdf
Organisations which can be consulted about the suitability of microorganisms include: the ASE (Association for Science Education)\*, CLEAPSS\*, MiSAC (Microbiology in Schools Advisory Committee), NCBE (National Centre for Biotechnology Education) and SSERC (Scottish Schools Equipment Research Centre)\*. (\*Members only.)

Bacterium	Educational use/interest/suitability	Ease of use/maintenance
Acetobacter aceti	Of economic importance in causing spoilage in beers and wines. Oxidises ethanol to ethanoic (acetic) acid and ultimately to carbon dioxide and water.	Needs special medium and very frequent subculturing to maintain viability.
Agrobacterium tumefaciens	Causes crown galls in plants; used as a DNA vector in the genetic modification of organisms.	Grows on nutrient agar, but requires 2-3 days' incubation.
Alcaligenes eutrophus	In the absence of nitrogen, it produces intracellular granules of poly-β- hydroxybutyrate (PHB); was used in the production of biodegradable plastics.	Grows on nutrient agar.
Azotobacter vinelandii	A free-living nitrogen fixer, producing a fluorescent, water-soluble pigment when grown in iron-limited conditions.	Grows on a nitrogen-free medium.
Bacillus megaterium	Has very large cells; produces lipase, protease and also PHB (see <i>Alcaligenes</i> ); Gram-positive staining.	Grows on nutrient agar.
Bacillus stearothermophilus	Thermophilic species which grows at 65 °C; produces lipase & protease. Used to test the efficiency of autoclaves.	Grows on nutrient agar.
Bacillus subtilis*	General-purpose, Gram-positive bacterium. Produces amylase, lipase and protease.	Grows on nutrient agar.
Cellulomonas sp.	Produces extracellular cellulase.	Grows on nutrient agar but also used with agar containing carboxymethylcellulose.
Chromatium sp.	A photosynthetic, anaerobic bacterium.	Requires special medium and light for good growth.
Erwinia carotovora (= Pectobacterium carotovorum)	Produces pectinase which causes rotting in fruit and vegetables. Useful for studies of Koch's postulates.	Grows on nutrient agar.
Escherichia coli*	K12 strain: general-purpose, Gram-negative bacterium. B strain: susceptible to T4 bacteriophage.	Grows on nutrient agar.

# Selected microorganisms

Bacterium	Educational use/interest/suitability	Ease of use/maintenance
Janthinobacterium (= Chromobacterium) lividum <sup>†</sup>	Produces violet colonies. Grows best at 20 °C.	Needs frequent subculture and is best grown on glucose nutrient agar and broth.
Lactobacillus sp.	Ferments glucose and lactose, producing lactic acid; <i>L. bulgaricus</i> is used in the production of yoghurt.	Requires special medium containing glucose and yeast extract and frequent subculturing to maintain viability.
Leuconostoc mesenteroides	Converts sucrose to dextran: used as a blood plasma substitute.	Requires special medium as for Lactobacillus.
Methylophilus methylotrophus	Requires methanol as energy source; was used for the production of 'Pruteen' single-cell protein.	Requires special medium containing methanol.
Micrococcus luteus (= Sarcina lutea)	Produces yellow colonies; useful in the isolation of the bacterium from mixed cultures. General-purpose, Gram-positive bacterium.	Grows on nutrient agar.
Micrococcus roseus	Produces red colonies; useful in the isolation of the bacterium from mixed cultures. Grows more quickly than <i>M. luteus</i> . General-purpose, Gram-positive bacterium.	Grows on nutrient agar.
Photobacterium phosphoreum	Actively-growing, aerated cultures show bioluminescence; grows in saline conditions.	Requires a medium containing sodium chloride.
Pseudomonas fluorescens	Produces a fluorescent pigment in the medium.	Grows on nutrient agar.
Rhizobium leguminosarum	A symbiotic, nitrogen fixer; stimulates the formation of nodules on the roots of legumes. Only fixes nitrogen in plants.	Grows on yeast malt agar; some authorities recommend buffering with chalk to maintain viability.
Rhodopseudomonas palustris	A photosynthetic, anaerobic, red bacterium. Also grows aerobically in the dark.	Requires light and a special medium, growing atypically on nutrient agar
Spirillum serpens	Of morphological interest.	May grow on nutrient agar but needs very frequent subculturing to maintain viability.
Staphylococcus epidermidis (albus)**	A general-purpose, Gram-positive bacterium, producing white colonies.	Grows on nutrient agar.
Streptococcus (= Enterococcus) faecalis	Of morphological interest, forming pairs or chains of cocci.	Nutrient agar with added glucose can be used but grows better on special medium, as for <i>Lactobacillus</i> .
Streptococcus (= Lactococcus) lactis	Of morphological interest, forming pairs or chains of cocci. Commonly involved in the souring of milk; also used as a starter culture for dairy products.	Can grow on nutrient agar with added glucose; some authorities recommend buffering with chalk to maintain viability.
Streptococcus thermophilus	Ferments glucose and lactose, producing lactic acid; used in the production of yoghurt. Grows at 50 °C.	Can grow on nutrient agar with added glucose; some authorities recommend frequent subculturing to maintain viability.
Streptomyces griseus	Responsible for the earthy odour of soil. Grows to form a fungus-like, branching mycelium with aerial hyphae bearing conidia. Produces streptomycin.	Grows on nutrient or glucose nutrient agar but better on special medium which enhances formation of conidia.
Thiobacillus ferrooxidans	Involved in the bacterial leaching of sulfur-containing coal. Oxidises iron(II) and sulfur. Demonstrates bacterial leaching of coal samples containing pyritic sulfur.	Requires special medium.
Vibrio natriegens (= Beneckea natriegens)	A halophile, giving very rapid growth. Prone, however, to thermal shock with a sudden drop in temperature.	Requires a medium containing sodium chloride.

\* Some strains have been associated with health hazards. Reputable suppliers should ensure that safe strains are provided.
\* Can be chosen for investigations that once required the use of *Chromobacterium violaceum* or *Serratia marcescens*.
\*\* This organism has been known to infect debilitated individuals and those taking immunosuppressive drugs. Some authorities advise against its use.

Fungus	Educational use/interest/suitability	Ease of use/maintenance
Agaricus bisporus	Edible mushroom; useful for a variety of investigations on factors affecting growth.	Grows on compost containing well- rotten horse manure; available as growing 'kits'.
Armillaria mellea	The honey fungus; causes decay of timber and tree stumps. Produces rhizomorphs.	Grows very well on malt agar. Some authorities recommend carrot agar.
Aspergillus nidulans*	For studies of nutritional mutants. Produces abundant, easily-dispersed spores - may become a major laboratory contaminant!	Grows on Czapek Dox yeast agar. Special media required for studying nutritional mutants.
Aspergillus niger*	Useful for studies of the effect of magnesium on growth & the development of spore colour. Used commercially for the production of citric acid. Produces abundant, easily-dispersed spores - may become a major lab contaminant!	Requires special sporulation medium for investigations.
Aspergillus oryzae*	Produces a potent amylase; useful for studies of starch digestion. Also produces protease. Used by the Japanese in the production of rice wine (saki).	Grows on malt agar; add starch (or protein) for investigations.

Fungus	Educational use/interest/suitability	Ease of use/maintenance
Botrytis cinerea	Causes rotting in fruits, particularly strawberries. Useful for studies of	Can be grown on malt agar or agar
	Koch's postulates with fruit, vegetables and <i>Pelargonium</i> spp. Important in the production of some dessert wines ('noble' rot). Used in ELISA protocols.	with oatmeal.
Botrytis fabae	Causes disease in bean plants.	Requires agar with oatmeal.
Candida utilis (= Lindnera jadinii)	Simulates behaviour of pathogenic <i>Candida</i> spp. in investigations of fungicidal compounds.	Grows on malt agar or glucose nutrient agar.
Chaetomium globosum	Useful for studies of cellulase production; thrives on paper.	Can be grown on V8 medium but survives well just on double thickness wall paper, coated with a flour paste.
Coprinus lagopus	For studies of fungal genetics.	Grows on horse dung.
Eurotium repens (= Aspergillus pseudoglaucus)	Produces yellow cleistocarps (cleistothecia) embedded in the medium and green conidial heads in the same culture.	Requires special medium.
Fusarium graminearum	Causes red rust on wheat; used in the manufacture of 'Quorn' mycoprotein.	Can be grown on V8 medium.
Fusarium oxysporum	A pathogen of many plants. Produces sickle-cell-shaped spores, a red pigment and pectinase.	Grows well on several media: malt, potato dextrose and Czapek Dox yeast agar.
Fusarium solani	Digests cellulose; macroconidia have a sickle shape.	Grows on potato dextrose agar.
Helminthosporium avenae	A pathogen of oats.	May not grow easily in laboratory cultures.
Kluyveromyces lactis	A yeast, isolated from cheese and dairy products. Ferments lactose and used to convert dairy products to lactose-free forms. Genetically-modified strains are used to produce chymosin (rennet).	Grows on malt agar or glucose nutrient agar.
Leptosphaeria maculans	For studies of disease in <i>Brassica</i> plants.	Requires cornmeal agar or prune yeast lactose agar to promote sporulation in older cultures.
Monilinia (= Sclerotinia) fructigena	For studies of brown rot in apples. Useful for studies of Koch's postulates.	Grows on malt agar or potato dextrose agar.
Mucor genevensis	For studies of sexual reproduction in a homothallic strain.	Grows on malt agar.
Mucor hiemalis	For studies of sexual reproduction between heterothallic + and - strains and zygospore production.	Grows on malt agar.
Mucor mucedo	Common black 'pin mould' on bread. For sporangia (asexual), mating types and amylase production.	Grows on malt agar.
Myrothecium verucaria	For studies of cellulose decomposition, but <i>Chaetomium globosum</i> is preferred.	Grows on malt agar.
Neurospora crassa*	Red bread mould. Produces different coloured ascospores. Can be used in studies of genetics. Beware - readily becomes a major laboratory contaminant!	Grows on malt agar.
Penicillium chrysogenum*	Produces penicillin; useful for comparative growth inhibition studies in liquid media or when inoculated on to agar plates seeded with Grampositive and -negative bacteria. Produces yellow pigment.	Grows on malt agar, though some authorities indicate that it thrives better on liquid media.
Penicillium digitatum*	Does not produce penicillin; causes spoilage of citrus fruit.	Grows on malt agar.
Penicillium expansum*	Does not produce penicillin; causes disease in apples. Useful for studies of Koch's postulates.	Grows on malt agar.
Penicillium notatum*	Produces penicillin; useful for comparative growth inhibition studies in liquid media or when inoculated onto agar plates seeded with Grampositive and -negative bacteria.	Grows on malt agar.
Penicillium roqueforti*	Does not produce penicillin; the familiar mould of blue-veined cheese.	Grows on malt agar
Penicillium wortmanii*	Produce wortmin rather than penicillin.	Grows on malt agar.
Phaffia rhodozyma	A fermenting red yeast. Used to colour the food supplied to fish-farmed salmon.	Grows on yeast malt agar.
Phycomyces blakesleanus	Produces very long sporangiophores which are strongly phototropic.	Grows on malt agar.
Physalospora obtusa	An ascomycete fungus that grows on apples. Thought to produce pectinase.	Grows on potato dextrose agar.
Phytophthora infestans†	Causes potato blight. Produces motile zoospores.	Can be grown on V8 medium.
Pichia anomala	Produces a fragrant ester when grown on a carbohydrate- rich medium. Can be used in a microbial fuel cell because it can donate electrons directly to the electrode.	Grows on GYEP (glucose, yeast extract, peptone) broth or agar. High levels of glucose are needed to encourage ester production.
Plasmodiophora brassicae	For studies of disease in <i>Brassica</i> plants, particularly club root. Useful for studies of Koch's postulates.	May not grow easily in culture.
Pleurotus ostreatus	Edible oyster cap mushroom.	Can be grown on rolls of toilet paper!

Fungus	Educational use/interest/suitability	Ease of use/maintenance
Pythium de baryanum†	Causes 'damping off' of seedlings; cress is best to use.	Grows on cornmeal agar.
Rhizopus oligosporus	Used in the fermentation of soya beans to make 'tempe', a meat- substitute food in Indonesia	Grows on potato dextrose agar, Czapek Dox yeast agar and other fungal media.
Rhizopus sexualis	Produces rhizoids and zygospores. Useful for studies of the linear growth of fungi.	Grows on potato dextrose agar and other fungal media.
Rhizopus stolonifer	Produces rhizoids. Produces lipase.	Grows on potato dextrose agar, potato carrot agar, Czapek Dox yeast agar and other fungal media.
Rhytisma acerinum	An indicator of air pollution: less common in industrial areas. On sycamore leaves, it forms 'tar' spot lesions, the number or diameter of which can be compared at different sites.	Difficult to maintain but laboratory cultures are not likely to be needed.
Saccharomyces carlsbergensis	Useful for demonstrating how different species can metabolise different sugars. ( <i>S. carlsbergensis</i> can use raffinose for anaerobic fermentation while baker's yeast, <i>S. cerevisiae</i> , cannot.)	Grows on malt agar or glucose nutrient agar.
Saccharomyces cerevisiae	Valuable for work in baking and brewing, showing budding, for spontaneous mutation and mutation-induction experiments, and for gene complementation using adenine- and histidine-requiring strains.	Grows on malt agar or glucose nutrient agar.
Saccharomyces diastaticus	Able to grow on starch by producing glucoamylase.	Grows on malt agar and nutrient agar +1% starch.
Saccharomyces ellipsoideus	Used in fermentations to produce wine; can tolerate relatively high concentrations of ethanol.	Grows on malt agar.
Saprolegnia litoralis†	Parasitic on animals. Produces zoospores. Good illustration of asexual and sexual stages.	Culture by baiting pond water with hemp seeds.
Schizosaccharomyces pombe	Large cells, dividing by binary fission. Good for studies of population growth, using a haemocytometer for cell counts. Prone to thermal shock.	Grows on malt agar. For studies of population growth, a malt extract broth can be used.
Sordaria brevicollis	For studies of fungal genetics, including inheritance of spore colour and crossing over in meiosis.	Requires special medium for crosses between strains.
Sordaria fimicola	For studies of fungal genetics, including inheritance of spore colour and crossing over in meiosis.	Grows on cornmeal, malt and other agars but may not transfer readily from one medium to another. White- spore strain may not always grow normally on standard cornmeal agar.
Sporobolomyces sp	Found on leaf surfaces. Spores are ejected forcibly into the air from mother cells.	Grows on malt, yeast malt and glucose nutrient agar but laboratory cultures may not be needed.
Trichoderma reesei	Commercial production of cellulase.	Grows on malt agar.

\* Possible risk of allergy/asthma if large numbers of spores are inhaled.

† Now classed as a protoctist, so may not be listed under fungi by some suppliers.

# **Viruses**

These are rarely studied in schools & colleges but a selected list of those which might be used is given below.

Cucumber Mosaic Virus

Tobacco Mosaic Virus

Bacteriophage (T type) (host *E. coli*) Potato Virus Y (not the virulent strain) Potato Virus X Turnip Mosaic Virus

### Algae, protozoa (including slime moulds) and lichens

Though some protozoa are known to be pathogenic, the species quoted for experimental work in recent science projects and those obtained from schools' suppliers or derived from hay infusions, together with species of algae and lichens, are acceptable for use in schools.

#### Unsuitable microorganisms

A number of microorganisms have in the past been suggested for use in schools but are no longer considered suitable; these are listed below.

#### Bacteria

Chromobacterium violaceum Clostridium perfringens (welchii) Pseudomonas aeruginosa Pseudomonas solanacearum Pseudomonas tabaci Serratia marcescens Staphylococcus aureus Xanthomonas phaseoli **Fungi** *Rhizomucor (Mucor) pusillus* 

Some fungi previously considered unsuitable have been reinstated in the list of selected organisms now that it is thought that they do not present a major risk, given good practice.

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