

## MICROBIOLOGY OF METALWORKING FLUIDS: WHAT WE KNOW AND LESSONS TO BE LEARNT

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**Abstract:** Water-miscible metalworking fluids are an essential component of many manufacturing processes. During their lifetime they are subject to permanent changes in their physical and chemical characteristics. Due to their high content of water and their chemical composition in use, metalworking fluids (MWF) are prone to microbial life, i.e. the proliferation of bacteria and fungi. The microbial activity leads to significant changes in the chemical composition of the MWF, which can result in the loss of their technical properties. This paper briefly discusses the influences of microbial contamination on the technical quality of MWF and presents common monitoring systems for the detection of microorganisms. Finally, measures are described that can be taken to protect MWF from damage caused by high microbial loads in daily practice. In a short outlook, alternative research approaches are mentioned that aim at sustainable use of MWF.

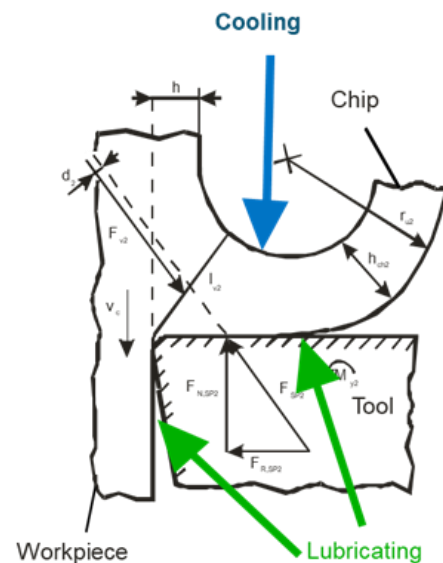
**Key words:** Metalworking fluids, microbiology, biofilms, monitoring

### 1. INTRODUCTION: METALWORKING FLUIDS (MWF)

The use of MWF in metal cutting and forming processes has been state of the art for decades. In machining processes, a large proportion of the machine power input is converted into heat, which can have a detrimental effect on the machined workpiece and the tools. MWF counteract this process by dissipating the heat energy directly at the point of machining and/or reducing the generation of heat by reducing friction (s. Fig. 1). The use of MWF generally leads to better machining results and ensures cost-saving production.

The elementary role that machining and MWF play in everyday life was described by Hans Ernst as early as 1951 as follows: 'Directly or indirectly, it (metal cutting) affects every aspect of our civilization. Every product we use, wear, or eat is related to metal cutting, either directly, in its own manufacture, or indirectly, through the manufacture of the machine that makes it' [1].

For the user, the longest possible service life of the MWF with constant performance is of great relevance. Drag-out losses due to chips and scooping parts require subsequent redosing with MWF. A decrease in technical quality due to physical-chemical processes and in particular microbial activity in water-mixed MWF leads to changes in MWF chemistry and a decline in the working result. Therefore, proper monitoring and surveillance of the MWF are necessary. An optimally coordinated use of MWF can lead to cost savings irrespective of the size of the plant and equipment. In addition, extended change intervals and reduced waste quantities also contribute to environmentally conscious production. The use of water-mixed lubricants thus contributes significantly to the cost structure in a production chain [2, 3].



**Fig. 1.** Primary functions of cooling and lubricating of a MWF. The arrows indicate the areas in which the two functions are effective

### 2. MWF' TASKS AND CHEMISTRY

According to DIN51385, coolant lubricants are divided into three groups: non-water-miscible, water-miscible and water-mixed coolant lubricants. The water-mixed MWF designates the application state of the water-miscible MWF after mixing a MWF concentrate with 90–95% water. Mineral oils, hydrocrack oils, polyolefins and synthetic esters of various origins with different degrees of refinement are used as base oils in these technical fluids. In their

application state, MWFs are divided into emulsions and solutions. To convert the two phases of water and oil from the concentrate into a stable emulsion, surface-active substances known as emulsifiers or surfactants are added to the concentrate. These have an amphiphilic molecular structure, i.e. they consist of a nonpolar, hydrophobic part and a polar, hydrophilic part. They reduce the interfacial tension between the two phases by dissolving with their non-polar part in the oil and their polar part in the water. As a result, they form finely dispersed droplets, known as micelles, with the oil. The stability of the emulsion depends on the size of the micelles that form. They represent the degree of dispersity of the emulsion. The average droplet size of a MWF emulsion is between 0.1  $\mu\text{m}$  and 10  $\mu\text{m}$  [4]. Finely dispersed emulsions have smaller micelles and are more stable. MWF emulsions can be demulsified by lowering the pH value, salinisation, evaporation, or energy input using ultrasound. MWF solutions are true solutions in the chemical sense, in which the ingredients are uniformly dissolved in water. They are homogeneous mixtures of inorganic and/or organic substances with water [5–7].

High demands are placed on the machining process when cutting metals. Here, MWF has the task of reducing friction and wear at the contact points of the tool and workpiece, as well as dissipating the developing heat. Due to the use of MWF, changes in friction mechanisms and reduced wear occur, and the quality of the newly created surface in the machining process can be significantly improved. The occurrence of lower cutting forces enables the increase of process performance. Fig. 1 displays the areas of cooling and lubricating in a metal-cutting process. Gottwein et al. [8] wrote a fundamental work on whose content almost all subsequent work was based. A detailed description of the MWF ingredients will not be given here; this has already been done in detail in [9–11]. The turnover rate of in-use MWFs in Germany is about 600.000 tons p.a. [12].

In addition to the primary functions of cooling, lubrication and transport of chips and swarf there are further relevant functions of MWF. Depending on the machining process, there are secondary requirements, such as corrosion protection, oxidation stability and a low tendency to foam formation. Due to the high demands placed on the machining process in manufacturing today, the fields that MWF must cover have expanded significantly. In addition to other literature, in Germany, the rule DGUV R-109-003 of the Employer's Liability Insurance Association defines the requirements that an effective MWF should meet today. The following is a selection of MWF secondary requirements [6, 11, 13]:

- Cooling capacity
- Lubricity
- Flushing capacity
- Long service lifetime
- Corrosion protection
- Anti-foaming properties
- Skin compatibility
- Low hazardous potential
- Environmental friendliness
- Wetting capacity
- Compatibility with other machine tool components
- Low disposal costs

Due to these requirements, a MWF can be formulated with up to 30 different ingredients, and >300 raw materials are available for the formulation of MWFs. The degree of purity of the raw materials used is of technical quality, so in any case admixtures of unknown origin and concentration must be expected [9, 10, 14].

### 3. MICROORGANISMS AND MWF

Microorganisms are ubiquitously present. In MWF bacteria and fungi are the main microbial inhabitants. Microorganisms have special abilities. They represent the oldest form of life, they are involved in all natural cycles, e.g. C-, N-, and they are adapted to live in extreme environments e.g. temperatures >100°C, pressures >100 bars, pH-values 0-14. Bacterial cells usually have a size between about 0.5  $\mu\text{m}$  and about 5  $\mu\text{m}$ , and the single cells of fungi can reach sizes of up to 20  $\mu\text{m}$ . The existence of bacteria and fungi in MWF however, has been described since the early 1920s. The first studies on the interactions between microbial contamination, hygienic aspects and the quality of the MWF date back to the 1920s [15–18]. The significance of the microbial infestation of MWF derives from the microorganism's capability to use the ingredients of a MWF for their metabolism. The degradation led to a decline in the components and consequently altered the technical quality of the MWF [19, 20].

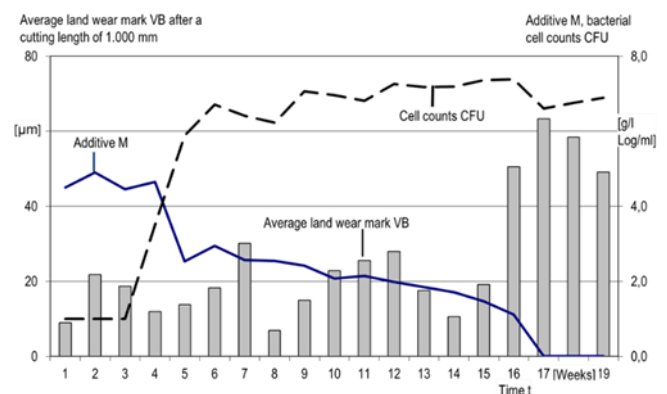


Fig. 2. Course of average tool wear, bacterial cell counts and a selected additive in a drilling process over a run time of 19 weeks

All technical fluids containing water are susceptible to microbial infestation. Due to their water content of >90% MWF are prone to microbial infection. Microorganisms find very favourable conditions for their growth and cell reproduction in MWF. They utilise all biological usable components of the MWF as a source of food and energy. The temperatures in the cooling system ranging from 20°C to 45°C also ensure optimum conditions for mesophilic microorganisms. In the course of the degradation processes, changes occur in the chemical composition of the MWF and the structure of ingredients. This results in a loss of the desired technical properties of the MWF. The microbial utilisation of MWF ingredients is accompanied by the formation of new, unknown intermediates and end products and a drop in the pH value. The microbial degradation of MWF ingredients as emulsifiers, corrosion inhibitors and/or performance additives causes increased wear and corrosion of tools and workpieces as well as the formation of foul odours and discolouration of the MWF. The increase in average land wear mark in a drilling process due to microbial degradation of ingredients (e.g. additive M) and the increase in bacterial cell counts in a MWF is given in Fig. 2. The process parameters were a drilling process of 16MnCr5, tool had a diameter of 10.0 mm, land wear mark VB was determined after a cutting length of 1.000 mm, MWF was a 5% mineral oil-based emulsion, bacterial cell counts detected by colony forming units

(CFU). Microbial infestation of MWF leads to increased consumption of concentrate, biocides and additives, as well as a shift in the chemical composition. The microbial degradation of a MWF is complex and does not affect a single ingredient or property alone. Therefore, the initial condition of the MWF cannot be restored by the addition of additives. The damaged MWF must be replaced and disposed of prematurely. This results in costs for the user that can be avoided. Careful selection, proper care and maintenance, and proper personal protection during use can significantly improve productivity and worker safety [9, 14, 19, 20].

In summary, the microbial degradation processes can have the following effects on the MWF system [13, 21]:

1. Effects on the MWF
  - Degradation of the ingredients
  - Decrease in the pH-value
  - Decrease in corrosion protection
2. Effects on the circulation system
  - Oil separation, emulsion splitting
  - Clogging of lines by biofilms
  - Occurrence of foam
  - Hydraulic difficulties during filtration
3. Effects on the environment
  - Risk of infection for employees
  - Accumulation of skin irritation
  - Occurrence of odours
4. Effects on downstream systems
  - Interference with ultrafiltration during disposal of cooling lubricants
  - Carryover into downstream systems
  - Increase in dissolved organic carbon/chemical oxygen demand (DOC/CSB) in the wastewater disposal stream

In the production process, the aim should be to achieve the lowest possible microbial load in MWF to keep the technical quality of the MWF at a high level.

A particular problem of contamination of MWF is the development of biofilms present in the system. The term biofilms refers to the extensive growth of bacteria and fungi on interfaces. Biofilms are found on almost all interfaces that are in contact with water. On clean surfaces in contact with an aqueous phase, a conditioning film of macromolecules forms spontaneously within a few minutes, on which bacteria grow to form a biofilm. According to Costerton et al. [23] bacterial biofilms represent the most successful form of life regarding the entire biomass as well as the diversity, type and expansion of populated habitats.

Life in biofilms offers microorganisms a series of advantages over existence as free-floating organisms in the water zone. The biofilm has to be understood as a three-dimensional habitat that is neither shaped evenly in a temporal nor in a spatial sense. It presents its inhabitants with protection from hydraulic loads, fluctuation in pH, osmotic stress, dehydration and pollutants such as biocides. At the same time, the biofilm is in a way decoupled from the water body. That makes it impossible to read from data from the water body where biofilms are, what extent they have and which organisms live within them. The extracellular polymeric substance (EPS) is the base frame for biofilms; it is traversed by water-filled channels in which signalling and messenger substances and nutrients are transported. It serves as a storage space for nutrients and is the compartment for chemical-physical reaction processes. The coexistence of different species of microorganisms forms a pool of genetic information in a narrowly defined space leading to the development of micro-consortia. This

results in the possibility of horizontal gene transfer and the ability to degrade molecules in co-metabolic processes, which cannot be degraded by single microbes. With increasing thickness of the biofilm and decreasing oxygen content, more ecological niches emerge for anaerobic organisms. These local habitats do not emerge in the free water zone, as anaerobic bacteria could not be active physiologically there. Fig. 3 displays a schematic of a biofilm structure [23–27].

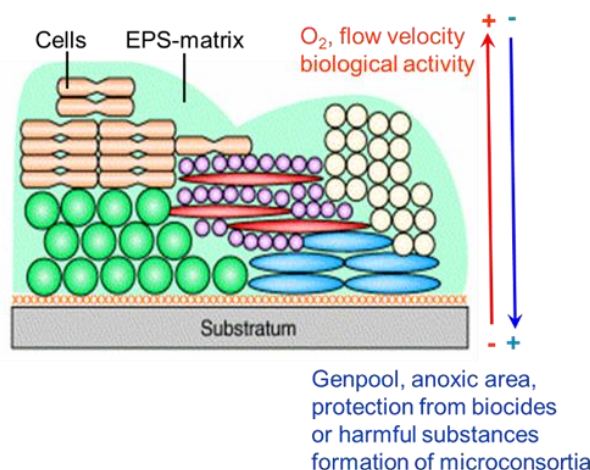


Fig. 3. Schematic representation of a biofilm and selected gradients over the depth of the biofilm. Natural biofilms usually consist of >90% water and in the EPS matrix, the proportion of microorganisms is very low [acc. [28] modified]

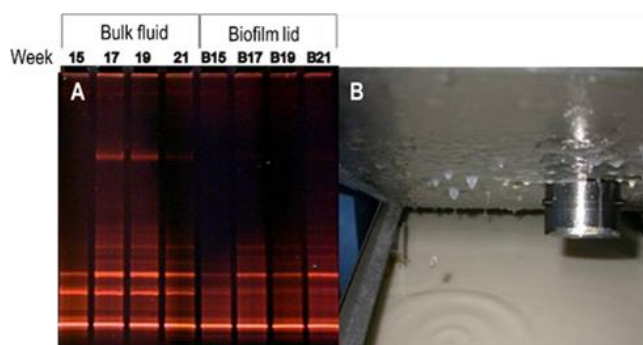


Fig. 4. DGGE-analysis of the bacterial community in Biofilms in a MWF system, A = Plot of the DGGE-analysis, B = Backside of the lid containing a visible biofilm dripping in the bulk fluid

Biofilms ranging in thickness from a few micrometres to a few centimetres are regularly found in MWF systems. In a MWF system, the microbial community of a biofilm is in exchange with the bulk liquid in terms of the predominant species spectrum. Fig. 4 shows the analysis of denaturing gradient gel electrophoresis (DGGE) of the bulk fluid and the biofilm on the backside of a lid in a MWF system, and the testing interval was six weeks. Every single bar in the lanes of the gel plot represents a specific bacterial strain. It is clearly visible that the species composition is nearly identical in the bulk fluid and the biofilm. In pipelines and filter systems, the reduction of the cross section or the complete blocking of the lines can lead to hydraulic difficulties in the MWF system. Due to their good adhesion and persistence to chemicals, biofilms can only be removed from a MWF system by a combination of system cleaning and mechanical post-treatment of the surfaces [25].



#### 4. MONITORING SYSTEMS

In Germany, the monitoring of MWF is regulated in DGUV-R-109-003. Once a week the noticeable alterations in the MWF, pH value, MWF concentration, and nitrite content of the MWF have to be measured and documented. The measurement of the microbial load in MWF is not provided. In case of an increased microbial occurrence in the MWF, reference is made to the rule DGUV-I 051 (BGI762) and the documents available in the appendix of the rule [13, 31].

Due to the high relevance of microbial loads in MWF, this chapter will give an overview of the common techniques for the detection of bacteria and fungi in MWF displaying their advantages and disadvantages. It should be noted that all methods described below evaluate the cell counts in the bulk fluid. Cell count determination in microbial biofilms is more complex and is not performed in MWF monitoring practice.

##### 4.1. Dip-Slides

Dip-Slides are still the most widely used method in operational practice to monitor microbial cell counts in MWF. They are plastic strips coated with a growth media for the identification of bacteria and fungi/yeasts on each side respectively. For cell enumeration, the strips are dipped into the MWF to be wetted with the fluid. Afterwards the strips are sealed in a tube. After an incubation time of mostly 48 h for bacterial and 72 h for fungi/yeast growth respectively at a given temperature, the slides can be evaluated. The addition of a dye to the medium serves the colouring of the bacteria colonies and is supposed to simplify the reading. The classification of the results is conducted with a colour scale supplied by the manufacturer, which displays the cell number estimation in intervals of orders of magnitude (s. Fig. 5).

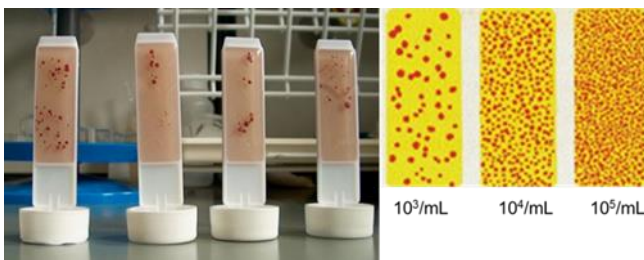


Fig. 5. Left: Dip-Slides after 48h incubation time. MWF were taken at different sample sites of the machine tool. The photograph shows the side with the media for bacterial growth. The red dots are coloured colonies that have arisen from a single cell. Right: Example of a colour scale for the cell count enumeration of the Dip-Slides

Unfortunately, the results of the cell count estimation via Dip-Slides are overrated in operational practice, and a nonexistent growth is interpreted as sterility. The results are more an estimation of cell counts than a reliable determination due to the following inaccuracies:

- The volume of MWF remaining on the Dip-Slide after dipping is dependent on viscosity and temperature and is therefore not constant
- By dipping the slide directly into the MWF, the oily surface film is brought onto the Dip-Slide

- Bacterial strains that are not coloured by the dye are often overlooked in the reading
- Bacterial strains with a low rate of cell division do not form colonies large enough to be identified by eye inspection
- The method is time-consuming (48–72 h)

The main advantages of Dip-Slides are their low price and their easy handling, which does not require a laboratory [32].

##### 4.2. CFU/plate counts

The culturing of microorganisms on solid media has been a proven technique to determine microbial cell counts for decades. The reproductive rate of microorganisms is used to make them visible to the human eye and therefore accessible for quantification. A defined volume (normally 0.1 mL) of fluid/MWF out of a dilution series is applied to a solid medium and evenly spread to separate the organisms. A numerical evaluation of the forming colonies takes place after a predetermined incubation period of normally between 24 h and 48 h. Fig. 6 shows as an example a section of a CFU plate with bacterial cultures of different sizes. The result is represented in cell count per mL. The determination of the CFU offers the possibility to cultivate bacterial strains selectively by using specific media and thus differentiate species within bacteria populations. Single strains or bacteria groups from one mixed culture can be isolated and verified selectively. The determination of the cell counts employing CFU requires laboratory equipment and requires qualified personnel [33, 34]. Therefore, this technique is mainly used in research on MWF microbiology. The advantages and disadvantages of using CFU for the determination of microbiology in MWF can be summarised as follows:

- Lab required, therefore costs are higher
- Reliable results when using the same cultivation media in routine control
- Selective growth conditions can be set up
- Routine required for reliable assessment
- Time consuming (>48 h)

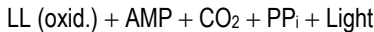
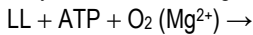


Fig. 6. Section of a CFU plate incubated with MWF. The dots are bacterial colonies that have grown from a single cell. The different sizes of the colonies are due to the different growth rates of the respective bacterial strains and indicates the presence of different species in the sample

##### 4.3. Adenosine triphosphate (ATP)-measurement

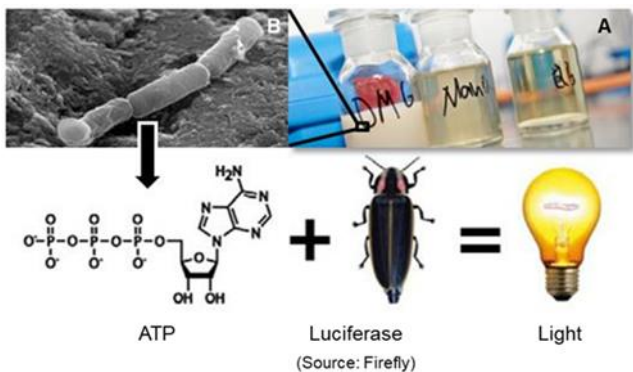
ATP is the overall energy supply used in cells, which is traceable in all living beings. The measurement of the ATP content is therefore an appropriate method for the detection and monitoring of microbial loads in MWF. The basis of the ATP determination is

the reaction of luciferin with the enzyme luciferase. In the presence of ATP and oxygen, the luciferin-luciferase-complex is oxidised by the emission of light (s. Fig. 7).



LL = luciferin-luciferase-complex; AMP = adenosine monophosphate; PP<sub>i</sub> = inorganic phosphate (acc. [35]).

In the detection process according to ASTM E2694 [36] (American Society for Testing and Materials) at first, the microbial cells were disrupted, and the released ATP is bound to a filter. After a washing step, the ATP is resolved and can react with the added luciferin-luciferase complex. The emitted light is detected and from these data, microorganism equivalents can be calculated. Depending on the ATP measuring device in use, the time needed for an ATP content measurement is between 5 min and 20 min. Thus, it offers fast results and allows intervention in the MWF system if necessary. It can be carried out without any special laboratory equipment. Furthermore, the method is well suited to show changes in the microbial load in monitoring over time. If changes in the measured values occur the process can be flanked by further methods (e.g. determination CFU). Passman et al. [37] confirm this statement and emphasise the high reproducibility of the measurement results. They conclude that ATP measurement is a powerful tool to improve microbial monitoring in MWF.



**Fig. 7.** Scheme for the measurement of adenosine triphosphate (ATP). Upper part: A = Microbial contaminated MWF samples. B = Bacterial cells containing ATP. Below: Reaction of the extracted ATP with the luciferin-luciferase-complex from firefly lead a light emission

#### 4.4. Molecular biological techniques

The permanent development of chemical-analytical methods has allowed for new procedures for the fast determination and exact identification of microorganisms. Molecular biological methods, such as the amplification of DNA with the polymerase chain reaction (PCR), are standard in daily work in microbiological laboratories. Based on this, further methods have been established in the most scientific reflection on MWFs microbiology. The PCR technique requires laboratory and qualified personnel and therefore have currently no role in daily MWF surveillance. But they have been contributing significantly to the scientific understanding of the microbiology of MWF. Developments such as Lab on Chip, which are already state of the art in other fields of application, will certainly also find their way into MWF monitoring shortly and contribute to fast and reliable test results.

Fig. 8 compares the methods presented here for their characteristics in terms of cost, the time required between sampling and result presentation, routine, validity and laboratory requirements. The methods Dip-Slides and ATP measurement are, due to the easy handling and the possibility to be used on-site, the methods that have the widest spread in practice.

	Costs	Time	Standardized	Validity	Lab/On-site
Dip-Slides	Green	Red	Green	Yellow	On-site
CFU	Yellow	Red	Green	Green	Lab
ATP	Green	Green	Green	Yellow	On-site
Molecularbiol.	Red	Green	Green	Green	Lab

**Fig. 8.** Comparison of the presented methods for the determination of microbial cell counts in MWF. The evaluation of the characteristics costs, the time between sampling and result presentation, standardisation, validation and necessity of a laboratory is done by a traffic light system

### 5. METHODS FOR COUNTERACTING

This section briefly presents physical methods and chemical counteracting that exert a biocidal effect in MWF and have achieved relevance in practice.

Physical methods:

UV-Radiation in the wavelength 254 nm destroys chemical bonds in the microbial DNA. This method can only be used in bypass, as the penetration depth is low and in addition, shadowing effects occur.

Ultrasonic treatment is a method that is mainly used in the food industry to get liquids free of microbes. When used in MWF, there is a risk that the energy input will destroy emulsion droplets and macromolecules.

Contact catalysts as e.g. AGXX® disrupt the electric field of a cell, causing damage to the cell membrane that can lead to cell death. It has a limited range and produces the effect only in passing liquids.

The physical methods mentioned here, only act in the bulk fluid and have no biocidal effect on biofilms.

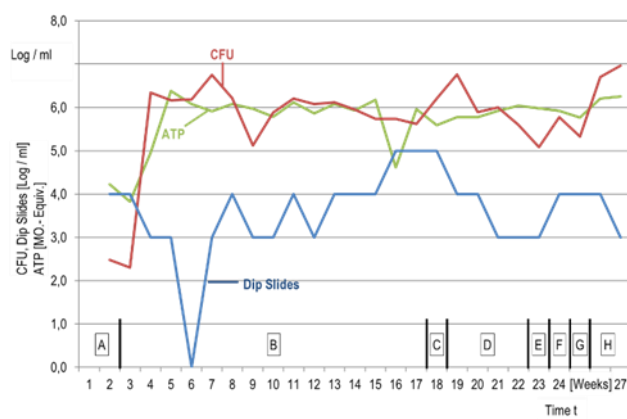
Chemical counteracting:

Common substance classes of chemical agents are formaldehyde releasing agents, isothiazolinone, chlorocresol and iodocarbamate. The main acting site of chemical agents is also within the bulk fluid. In biofilms, the impact of chemical biocides is decreased due to reactions of the substances with the EPS matrix and a significantly lower flow velocity. With the entry into force of the Globally Harmonised System GHS regulation, the number of chemicals available on the market that may be used as biostatics or biocides in the EU has been significantly reduced. Currently, 11 substances are approved for storage preservation and 6 substances as protective agents for machining fluids [24, 29, 30].

### 6. RESEARCH RESULTS

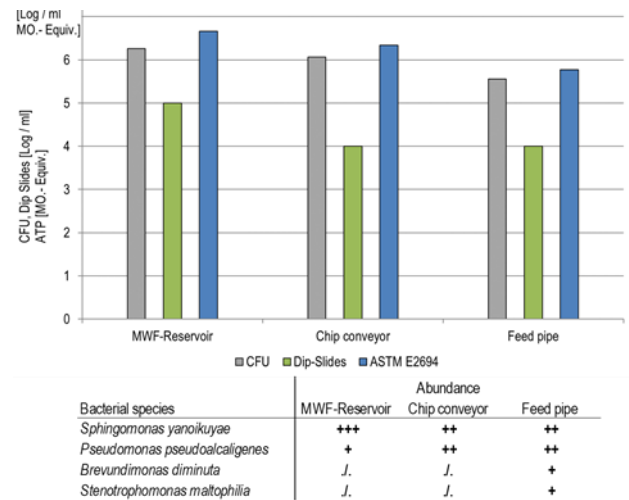
Microbial life in MWF is dynamic regarding the cell counts and the diversity of the microorganisms. Fig. 9 displays the development of microbial cell counts in a MWF system detected over a

period of 27 weeks. CFU plate counts, ATP measurement according to ASTM E2694, and Dip-Slides were used as techniques for the enumeration of the bacterial cell counts. Dip-Slides lead to an underevaluation in the range of one to more than two orders of magnitude compared to CFU and ATP measurement. Therefore Dip-Slides could give only a hint of bacterial life in MWF and should not be used for a reliable enumeration of cell counts. For the description of the changes in bacterial diversity patterns over time, the dominating strains on CFU plates were identified by molecular biological techniques. The identified bacterial strains are displayed in the text below Fig. 5. *Pseudomonas pseudoalcaligenes* was the strain which could be detected in the longest time range in the MWF, starting two weeks after the start until the end of the investigation time of 27 weeks. The changes in diversity are due to the varying rate of degradation of the MWF ingredients and thus the varying nutrient supply for the bacteria.



**Fig. 9.** The graph displays microbial cell counts in a MWF over a time period of 27 weeks, detected by CFU, Dip-Slides and ATP measurement according to ASTM E2694. The underestimation of cell counts by Dip-Slides is evident. The dynamic in diversity is given by the names of the predominant bacterial species identified per measurement date from the CFU plates. A = *Brevundimonas diminuta*, *Ochrobactrum rhizosphaerum*, unidentified strain; B = *Pseudomonas pseudoalcaligenes*; C = *Arthroabacter sulfureus*, *Ps. pseudoalcaligenes*, unid. strain; D = *Sphingomonas yanoikuyae*, *Ps. pseudoalcaligenes*, unid. strain; E = *Acinetobacter sp.*, *Sp. yanoikuyae*, *Ps. pseudoalcaligenes*, *Brev. diminuta*; F = *Sp. yanoikuyae*, *Ps. pseudoalcaligenes*; G = *Sp. yanoikuyae*, *Ps. pseudoalcaligenes*, unid. strain; H = *Sp. yanoikuyae*, *Ps. pseudoalcaligenes*. (acc. [38] modified)

The bacterial diversity also varies in one MWF at the same time in one machine tool at different places. In Fig. 10 the results of measuring the microbial cell counts using the methods CFU, Dip-Slides and ATP-measurement according to ASTM E2694 are compared in the graph. In addition, the dominating strains on CFU plates were identified by molecular biological techniques. These results are given in the table above the graph. The samples were taken at the same time from different sample sites in the machine tool. From these results, the underestimation using Dip-Slides compared to the other techniques can be seen. In addition, the cell counts as well as the abundance of the bacterial species are different, depending on the site samples were taken. For MWF surveillance in daily practice, this means that routines concerning the sampling site are necessary to achieve valid results.



**Fig. 10.** Comparison of the results of measurement of microbial cell counts by CFU, Dip-Slides and ATP measurement according to ASTM E2694. The samples were taken from one machine tool at different sample sites. Cell counts differ, depending on the sample site and method used. The bacterial diversity differs concerning the sample site. (acc. [38] modified)

## 7. CONCLUSIONS

The life and proliferation of microorganisms are an integral part of almost all aqueous technical fluids. The degradation of fluid ingredients by microbial activity led to a loss of the required technical properties of the fluid, hygienic problems and, in the case of biofilm formation, disturbances concerning fluidic and thermal processes.

The microbiology of MWF has a history of decades, with only little evolution regarding monitoring and prevention techniques. Nevertheless, it still represents a relevant problem in manufacturing technology today, which should be brought back into focus. Due to legal regulations, fewer biocidal agents are available for use as preservation of MWF. This results in improved health protection for employees, but also in the problem of an increasing spread of microorganisms in MWF.

The following points should be realised and implemented in practice and research to enable an extended service life and thus a resource-saving use of MWF in the future:

- Microbial growth in MWF and other water containing technical fluids cannot be avoided.
- Besides the regular examinations of the physical-chemical parameters, the determination of microbial cell counts is necessary for successful monitoring and surveillance of the MWF.
- Molecular biological surveillance techniques should be state of the art in MWF surveillance.
- The result of an individual measurement of the microbial cell counts is not meaningful, a time course of measurements must always be recorded.
- Only suitable methods for cell count determination should be used.
- Compliance with industrial hygiene and machine cleaning standards must be maintained.
- The development of new, alternative biocidal agents or technical processes should be the focus of scientific research.



- The knowledge about biofilms in MWF systems is only in its infancy and also requires more intensive research.
- Legislation such as Biocidal Products Regulation (BPR) and REACH/GHS can be a driver for new developments regarding new biocidal products with a lower hazardous potential.
- Research is needed for the development of alternative and renewable ingredients, e.g. plant-based emulsifiers, antioxidants, performance additives or biocidal substances.

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