

# Multi-Feed Plate – User Manual

AppNote by **Kuhner shaker**

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The Multi-Feed Plate is a versatile tool to imitate fed-batch conditions in microplates via controlled glucose release. This Application Note will guide you through the plate's features and provide best practices for its optimal use. It highlights the influence of different release rates and helps you select the optimal Feed Plate® and cultivation conditions for your specific process. Click [here](#) to watch the application video or scan the QR code above. Let's get in touch after testing to select your preferred Feed Plate®: [feeding.de@kuhner.com](mailto:feeding.de@kuhner.com)

## Feed Plate® for continuous nutrient release

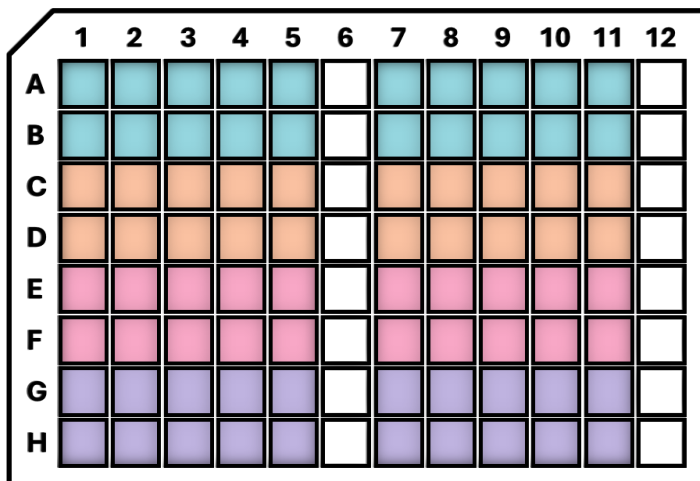
The Feed Plate® is a polymer-based slow-release system, which enables fed-batch conditions in microplates. It is routinely used in biological cultivations in high-throughput screening and process development, especially for microbial and cell culture applications with controlled nutrient supply. Different biological systems require specific substrate availability, which is why the Feed Plate® is offered in multiple, optimized release rates.

The Multi-Feed Plate combines four different substrate release rates in a single plate (Fig. 1) allowing direct comparison under identical experimental conditions. This design enables a rapid and reliable choice of the most suitable release rate for a given process without the need for multiple separate experiments, saving time and resources while improving reproducibility.

## Functionality of the Multi-Feed Plate

To start the substrate release, no additional equipment is required. Once filled with an aqueous solution (i.e. cultivation medium), substrate begins to diffuse from the polymer matrix into the liquid. This process starts immediately upon contact and continues at a defined rate determined by the properties of the matrix and the surrounding solution. The release rate depends primarily on the composition of the medium, the pH value, and the osmolality.

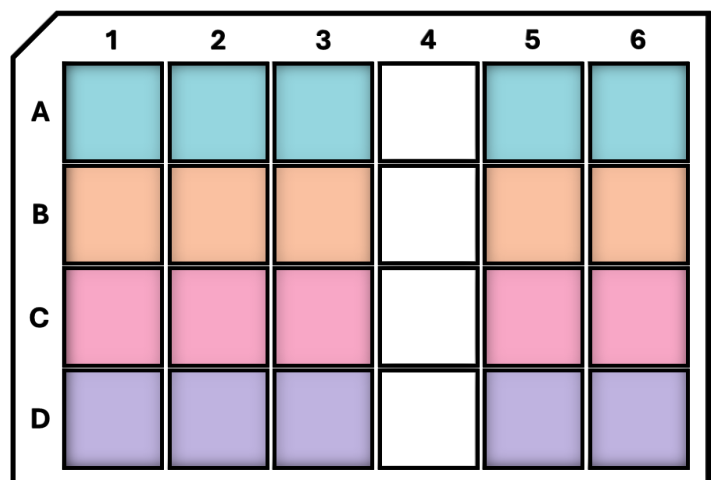
To ensure optimal testing conditions, there are a few important aspects to consider when working with the Multi-Feed Plate. For this reason, we have summarized the most important observations and recommendations in a table on the next page.



| Release profile |        |      |           |
|-----------------|--------|------|-----------|
| low             | medium | high | very high |

96-well: ~ 1-10 mg glucose per day  
24-well: ~ 4-40 mg glucose per day

= empty wells for "batch controls" without substrate feed



available in 96- &  
24-well format

**Figure 1. Schematic Multi-Feed Plate**

The four different substrate release rates are color-coded. Empty wells (without matrix) remain white and serve as batch references.

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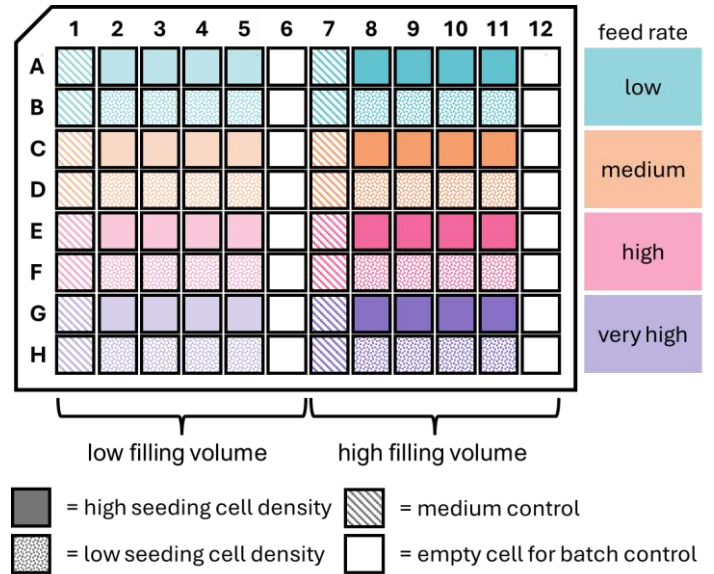
## Application of the Multi-Feed Plate (96-well)

**General:** Substrate release starts right after contact with liquid. Carefully plan the start of the experiment. Multi-Feed Plate is a single use item. An exemplary experimental layout is presented in Figure 2. Please check general limitations and recommendations in Table 1 before setting up your experiment.

**Filling volume:** We recommend testing at least two different filling volumes, resulting in different release rates. Low filling volume in columns 1–6 and high filling volume in columns 7–12. See recommendations for upper and lower limit in Table 1.

**Seeding cell density:** We recommend testing two different seeding cell densities. “Normal” in rows 1, 3, 5 and 7 and “high” in rows 2, 4, 6 and 8. A too low seeding density can result in prolonged “batch-like” growth when substrate release exceeds consumption. This way, the effect of the Feed Plate® cannot fully be exploited.

**Blanks and controls:** Use columns 1 and 7 for medium-only controls to monitor the release of the substrate in the absence of cells (for glucose analysis). Rows 6 and 12 contain no matrix, providing space to run a batch culture as a reference.



**Figure 2.** Colored representation of the proposed pipetting scheme of the Multi-Feed Plate for biological experiments.

**16 different fed batch conditions** are imitated in a **single plate** – check results and **pick your favorite condition**

**Table 1. Recommended use of the Multi-Feed Plate.** The table lists critical process parameters and states upper and lower limits.

| What to consider                      | Recommendation  |   | Explanation   |
|---------------------------------------|---|---|---|
| shaking conditions and filling volume | 300 rpm, 50 mm orbit  | 96-well: 300-750 µL,<br>24-well: 1,5-3 mL       | Optimal shaking conditions for good mixing and oxygen supply. Keep in mind that the Feed Plate® soaks media (~50 µL in 96-well plates and ~200 µL in 24-well plates after 72 h) |
|                                       | 350 rpm, 25 mm orbit  | 96-well: 300-750 µL,<br>24-well: 1,5-3 mL       |   |
|                                       | 1000 rpm, 3 mm orbit  | 96-well: 300-1200 µL<br>24-well: not applicable |   |
| immediate nutrient release            | Different seeding cell densities should be tested, to find the optimal starting point                       |   | substrate can accumulate if release > consumption, leading to batch-like conditions   |
| substrate concentration               | Try high and low filling volumes to achieve different nutrient concentrations                               |   | Plate has a constant substrate release (mg/day); concentration depends on filling volume  |
| media influence                       | Use blanks to determine nutrient release under your specific conditions measuring the glucose concentration |   | release slightly varies with pH, media components, and osmolality   |
| Blanks and controls                   | Compare the Feed Plate result to your state-of-the-art batch process (not mandatory)                        |   | Use wells without matrix for background control and batch references  |