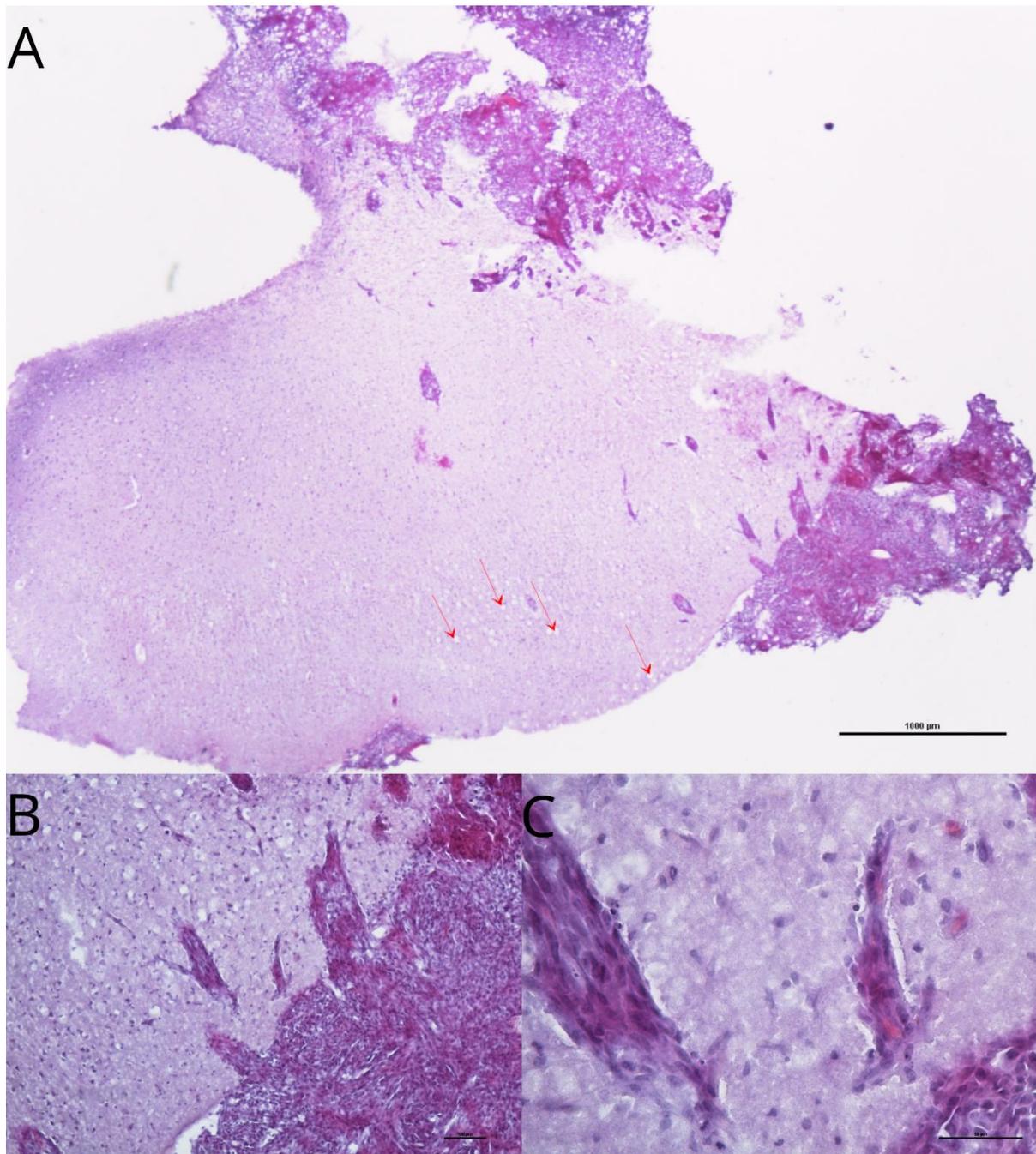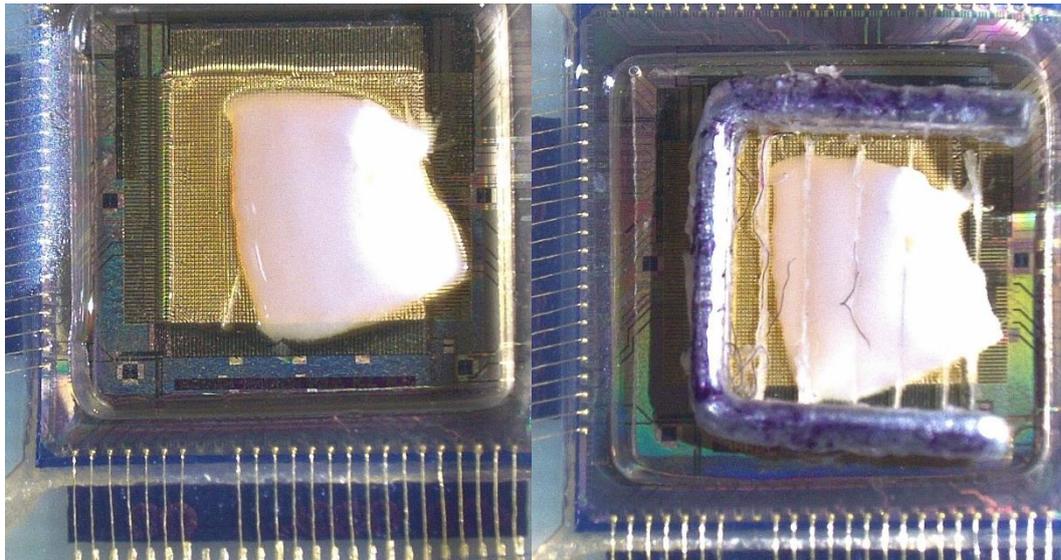Supplementary figure 1. H&E-stained sections confirming the invasive nature of the F98 glioma in Fischer 344 rats. Densely-packed cells represent F98 glioma cells. **(A)** Small vacuolar spaces match electrode penetration. Exemplary vacuolar spaces are marked by red arrows. **(B)** and **(C)** show the same slices at different magnifications.

Supplementary figure 2. Positioning of a brain slice on the MEA system. The left picture shows a brain slice without an anchor on the MEA system, whereas on the right photograph, the slice was anchored onto the MEA system.
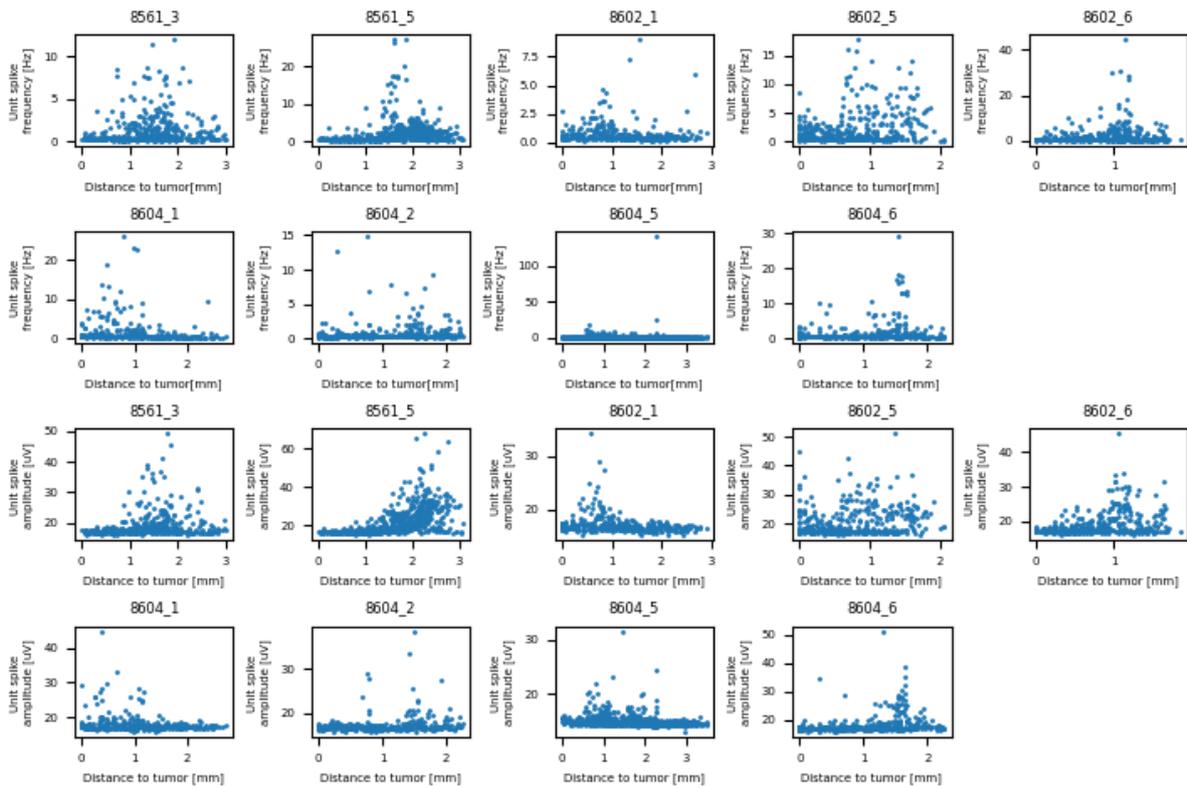
Supplementary figure 3: Unit spike frequency and mean unit amplitude vs. distance from glioma. Only recordings with a visually discernible glioma are depicted. The first and second rows plot the spike frequency of each unit against its distance from the glioma, while the third and fourth rows plot the unit spike amplitude for each unit against its distance from the glioma. Subheadings indicate the recording numbers used for internal reference, with each corresponding to one slice. There are no clear trends across recordings toward increased unit spike frequency or amplitude depending on the distance from the glioma.

Supplementary table 1: Mean distance of network burst (NB) onsets from the glioma

Animal recording ID	Mean NB onset distance from glioma [mm]	Std [mm]
8561_3	1.557	0.370
8561_5	1.972	0.237
8602_1	0.850	0.104
8602_5	0.968	0.494
8602_6	1.159	0.158
8604_1	0.659	0.250
8604_2	1.638	0.475
8604_5	0.974	0.188
8604_6	1.556	0.063
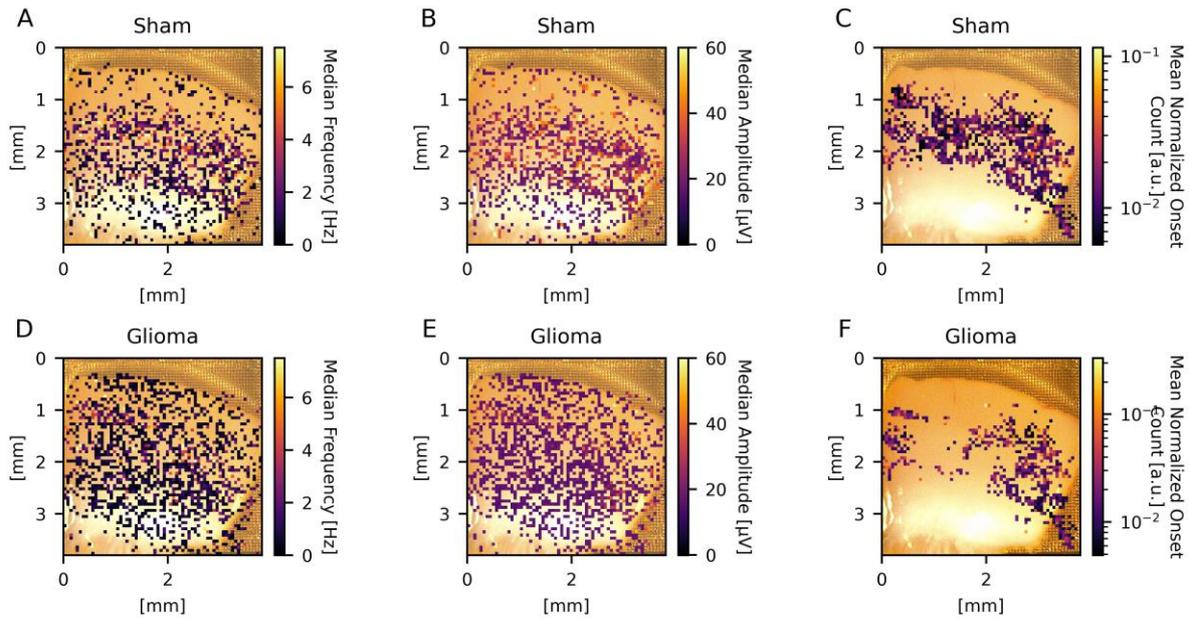
Supplementary figure 4. Spatially aligned unit-based metrics. (A,D) Slices were aligned and well-isolated single units were projected onto a common coordinate system. The slice there were aligned to is plotted as the background for each subplot. It should be noted that the size of the slices tended to vary between recordings, therefore perfect alignment was not always possible. The median frequency for each coordinate was calculated and is illustrated by the color. **(B,E)** The same was performed for the amplitude. **(C,F)** The relative frequency ∈ (0,1) of event occurrences at each coordinate on the 64x64 aligned coordinate system was calculated. If a negative burst always started at one location for a given slice, that coordinate was assigned a value of 1. If it never occurred, the point was assigned a frequency of 0 and neglected for subsequent calculations. The averages for all coordinates in all recordings in each group were calculated. The results are visualized in C and F. No clear trend is apparent, aside from the fact that the onset was always within the gray matter. The reference slice is shown in Supplementary figure 5.

Supplementary figure 5. All slices were aligned with this slice for comparison purposes.

Supplementary table 2. **Correlation of NB duration and rate with time passed during each recording.** Significance was reached for only 4 out of 22 recordings.

	Spearman r - NB duration [s], recording time	Spearman r - NB interval [s], recording time
Sham (mean±std)	-0.0861 ± 0.2327	-0.1312 ± 0.1456
Glioma (mean±std)	0 .1470 ± 0.1853	0.0772 ± 0.0854

Supplementary data 1: Correlation of NB duration with graph-based metrics for slices from sham-operated animals and glioma bearing slices.

Sham

- Spearman correlation between NB duration and global efficiency: -0.139 (p-value: 0.701)
- Spearman correlation between NB duration and average node strength: 0.297 (p-value: 0.405)
- Spearman correlation between NB duration and average clustering: -0.576 (p-value: 0.082)
- Spearman correlation between NB duration and modularity: 0.370 (p-value: 0.293)

Glioma

- Spearman correlation between NB duration and global efficiency: 0.552 (p-value: 0.063)
- Spearman correlation between NB duration and average node strength: 0.329 (p-value: 0.297)
- Spearman correlation between NB duration and average clustering: 0.287 (p-value: 0.366)
- Spearman correlation between NB duration and modularity: -0.343 (p-value: 0.276)

Supplementary table 3: Comparison between the network bursts identified by our custom pipeline and those found by BrainWave6 (3Brain, Swiss). The network bursts identified by BrainWave6 were additionally merged or removed according to the same criteria as those from our custom pipeline. The recordings were analyzed using the parameters specified at:

https://github.com/Ferdinand23457/HD_MEA_IPHYS_Rostock/blob/main/brainwave_settings/brainwave_settings.json

Accuracy, precision, and recall are calculated based on whether a sampling point was correctly labelled as part of a network burst.

Rec. No.	N NBs found our custom pipeline	N NBs Brain Wave6	Mean NB duration our pipeline [s]	Mean NB duration BrainWave6 [s]	Accuracy of Brainwave6 compared to our pipeline	Precision of Brainwave6 compared to our pipeline	Recall of Brainwave6 compared to our pipeline
1	53	51	0.45	0.25	0.98	0.98	0.53
2	150	117	0.35	0.36	0.97	0.98	0.77
3	202	158	0.39	0.39	0.96	0.97	0.77
4	59	59	0.45	0.312	0.98	0.92	0.63
5	58	60	0.84	0.87	0.99	0.92	0.98
6	119	140	0.82	0.65	0.97	0.94	0.89
7	152	162	0.91	0.712	0.95	0.97	0.81