INTRODUCTION: In recent years there has been a growing interest in the development of microelectronic biosensors where the electrical activity of neurons and the metabolic activity of tumor cells can be measured in-vitro and non-invasively. Important uses for such devices include pharmaceutical screening systems and further understandings of signal propagation in the brain.

Over the past 10 years, it has become possible to use the ISFET device as a cellular probe. The advantages of the ISFET, as opposed to conventional microelectrodes are (i) the possibility of measuring metabolic activity (via small pH changes in the culture media); (ii) the local amplification of the signal yielding less interference and noise and (iii) the possibility to multiplex transistor arrays. The latter is essential for designing large-scale sensor arrays. Electrical activity in cells has successfully been measured with ISFETS in the past (Fromherz et al, 1991, Offenhäusser et al, 1997) where a neuron sits on top of the non-metalized gate and induces an electrical signal into the ISFET channel. Metabolic activity has also been measured using ISFETS (Baumann et al, 1999) where the H+-sensitive layer of the ISFET, in proximity to a cell population, detects variations in cell metabolism via small (local) pH changes of the weakly buffered culture media.

Due to the high potential of FET-based biosensor arrays, we systematically measured and compared the electrical characteristics of Silicon-On-Insulator (SOI) FET-based devices. This will provide the basis for further performance optimizations, with regard to the recording of neural electrical signals as well as metabolic signals.

METHODS: The ISFET-array design utilizes 16 and 32 gates at different sizes and pitches. P-channel FETS are used for better stability. Each ISFET shares a common source and has individual drain connections for multi-channel read-out. Fabrication of the ISFET array is performed on SOI substrates to eliminate any shorting problems between transistors. Six masks are used in the process: The first mask defines the active-area where the transistors will be formed. A 20nm dry oxide is the grown followed by a V_t-adjust implant. Next the source and drain areas are defined followed by a Boron implant to dope these regions. This is followed by the deposition of 10nm silicon nitride (for the sensing layer) and 0.5um BPSG oxide (for the isolation layer). Next the ISFET gates and contacts are defined simultaneously by wet etching and after the contact regions are etched clear using a fourth mask. Following this etch, 0.6um Aluminium is deposited, patterned and wet-etched. Finally 0.5um of passivation is deposited, patterned and wet-etched to the gate areas and the bond pads. Prior to stripping the resist after the final pattern, 10nm chrome and 50nm Gold is deposited on one of the wafers. A lift-off process is then used to clear the metal everywhere except over the gate areas and bond pads. On another wafer, 10nm of Tantalum is deposited over the entire wafer by CVD technique and oxidized, resulting in Ta2O5 over the Si3N4, which has been shown to be superior to Si3N4 in terms of pH sensitivity and drift.

Figure 1: Schematic cross-section of an SOI ISFET

We have fabricated the following device variants:
1. Gate sensing layers: 10nm Si3N4 (over the SiO2); 50nm Au./Pt (over the Si3N4) and 10nm Ta2O5 (over the Si3N4).
2. Gate sizes: 30x10, 21x7, 15x5 and 10x1 um² at pitches of 100 and 75 um.
3. Passivation layers: 0.5um TEOS oxide and 1.2um Polyimide.

The fabricated chips were mounted in DIL ceramic chip-carriers, wire-bonded and partially encapsulated. A square polystyrene cuvette was attached to the chip carrier to contain 400μl of solution. Electrical measurements with the FETS were carried out using a Ag/AgCl reference electrode (defines gate potential) and a PBS saline solution (pH 7.4).

In order to measure the performance in amplifying neural activity, we applied a test pulse ...
(amplitude=400uV, width=1ms, frequency=100Hz) to the reference electrode. The FET’s drain-source current was then measured and amplified by a one-stage OPA with automatic offset-compensation and low-pass filtering (5kHz cut-off frequency). The drain and gate voltages were adjusted accordingly so that the (small-signal) amplification:noise ratio was maximized. Our main performance indicators have been the peak-to-peak voltage ($V_{pp}$) and the RMS of noise ($V_{NRMS}$) (without the test signal) of the filter output. A useful measure for spike detection is the ratio $V_{pp}/V_{NRMS}$. In order to compare the performance of the FETs with standard microelectrode arrays, we used a 10x10 array of 10x10um$^2$ platinum electrodes. For pre-amplification, we used a discrete p-channel MOSFET (BS292). The amplifier and filter circuit is the same as that described above.

For the pH sensitivity measurements, we used DU-145 prostate carcinoma cells, cultured on our SOI ISFETS (gate area 30x10um$^2$) for 2 days in an incubator. For the measurements, we used weakly-buffered (10mM, pH=7.4) RPMI cell-culture medium and recorded the change in source-drain current over time at driving conditions in which the gate-source voltage was −2.5 V and the drain-source voltage was −0.5 V (approx Ids = -35uA)

RESULTS: Table 1 contains the average measurements of 30x10um$^2$ FETS, and 10x10um$^2$ Pt microelectrodes. Table 2 contains averages for Si$_3$N$_4$-gate FETs with different gate sizes.

Table 1. Average FET/Pt electrode performance.

<table>
<thead>
<tr>
<th>Device type</th>
<th>$V_{pp}$ [mV]</th>
<th>$V_{NRMS}$ [mV]</th>
<th>Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Si$_3$N$_4$ Gate FET</td>
<td>101.8</td>
<td>0.31</td>
<td>340</td>
</tr>
<tr>
<td>Au-Gate FET</td>
<td>96.3</td>
<td>0.26</td>
<td>375</td>
</tr>
<tr>
<td>Pt-Electrode</td>
<td>5750</td>
<td>15.50</td>
<td>384</td>
</tr>
</tbody>
</table>

Table 2. Si$_3$N$_4$ gate FETs with different gate sizes.

<table>
<thead>
<tr>
<th>Device type</th>
<th>$V_{pp}$ [mV]</th>
<th>$V_{NRMS}$ [mV]</th>
<th>Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>30x10um$^2$</td>
<td>101.8</td>
<td>0.31</td>
<td>340</td>
</tr>
<tr>
<td>21x7um$^2$</td>
<td>108.13</td>
<td>0.46</td>
<td>281</td>
</tr>
<tr>
<td>15x5um$^2$</td>
<td>106.25</td>
<td>0.46</td>
<td>230</td>
</tr>
</tbody>
</table>

The result of the metabolic measurement is shown in Fig.2. The pH-measurements showed a pH sensitivity of approximately 45mV/pH (equivalent to a 3uA change in Ids).

DISCUSSION & CONCLUSIONS: There was no major difference between different gate layer-materials, except for the Au-layer which showed slightly better noise performance. However, the metal-layer FETs showed fabrication problems which caused reliability problems. Pitch size and passivation layer have no effect on electrical characteristics but are relevant for cell-studies. The 30x10um$^2$ FETS showed the best signal-to-noise ratio, which was comparable with that of the micro-electrode. The microelectrodes signals are bi-phasic (in comparison to mono-phasic FET signals) and are more sensitive to interference. We observed higher noise levels for smaller gate areas, in particular for the 10x1um$^2$. Regarding the pH-measurements, the obtained sensitivity is fairly typical for Si$_3$N$_4$-gate ISFETS.

In this initial work, we have systematically characterized various FET transistor types with regard to signal pick-up from neurons, in particular signal-to-noise performance. However, more detailed analyses are necessary to fully exploit the potential of FET-based biosensors.

REFERENCES:  
ACKNOWLEDGEMENTS: We are grateful to Prof. A. Offenhäusser (Inst. Thin Films & Interfaces, Research Center Jülich, Germany) for assisting us with the encapsulation of the devices and to Prof. Tom Cotter for the use of his cell culture facility.