High information-content screenings based on whole-cell current patch-clamp recordings have become available for ion channel pharmacological research with the development of the QPatch automated patch-clamp technology. A significant increase in system throughput was recently achieved by a tripling of the number of parallel operating patch-clamp sites (from 16 to 48) by the introduction of QPatch HT. In a series of hERG screening studies we have subsequently aimed at increasing the throughput further by reducing the average experimental time consumption associated with each IC₅₀ determination, and by employing alternative calculational algorithms. We have investigated the effect of the following three procedures on effective QPatch throughput:

1. Application of multiple drugs per cell
2. Reduction of the duration of the experimental protocol
3. Estimation of drug potency (IC₅₀) based on a single inhibitor concentration

2. - Reduction of the protocol execution time

In the standard hERG screening protocol (Figure 2A), Panel (1) the cell is first clamped to -70 mV to establish holding potential (Vh) for 40 s, after which a 50 ms depolarization to +40 mV for test current determination is followed by a depolarization to a test potential (Vt) of +140 mV for 500 ms. Subsequently, Vh is switched to -70 mV for 500 ms for current determination before clamping to -70 mV for 3000 ms. The total duration of the complete voltage protocol amounts to 1000 s. Between each voltage protocol the seal is reversed to a new cell. Consequently, the standard voltage protocol can be executed five times per minute.

The whole-cell current is recorded in response to the voltage protocol as shown in Figure 2B. Panel (2) shows the resulting current time (I-t) plot throughout the course of the experimental procedure. If each cell is exposed sequentially to each of the 48 compounds, we conducted a series of experiments, in which an obvious way of increasing the throughput is to apply several concentrations of the same compound per cell. To allow many compound additions to each cell, we used a ‘ramp’ protocol in which we applied increasing concentrations of the same compound sequentially to each cell. Figure 1 shows seal and whole-cell resistances for the 48 channels of a single QPlate 48 used for a screening job. In this experiment, 286 compound tests were initiated. Out of these 251 were completed successfully. The screening was completed in 43 minutes, corresponding to 5.0, 9.8 and 14.6 protocol executions per minute.

In addition we tested the two alternative short lasting protocols of Figure 2B1 and 2B2, which have been described in the literature. First, we tested an ‘interrupt’ protocol (Figure 2B2) in which a 20 ms hyperpolarization to -120 mV were conducted during the depolarization step (1400 ms duration) followed by a 800 ms repolarization to -50 mV. This protocol proved advantageous because it led to a significant reduction in the overall throughput to ~2000 compounds per day (corresponding to 5.0, 9.8 and 14.6 protocol executions per minute).

In conclusion, the single concentration IC₅₀ method increase the throughput provided the protocol execution times for the compounds tested in this study.