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Short communication

# Remote switching of temperature, gaseous, and aqueous phase in a low-volume interface chamber for brain slices

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ABSTRACT

A new remote-controlled interface-type chamber was designed in order to conduct experiments in brain slices involving gas, fluid, and temperature changes with as little tissue manipulation as possible. The chamber allows for extremely quick changes between different fluid and/or gaseous phases and for active cooling as well as heating by using a set of electromechanical valves and Peltier elements. The design drawings are complemented by exemplary tests of temperature and gas changes, and electrophysiological recordings of slices manipulated with gas and fluid alterations were used to test the efficacy and accuracy of the design. Changing between normoxia and anoxia needs less than 30 s, while the readjustment of the chamber to a new, preset temperature is accomplished in about 1 min. Supplementary data provide a proposal for the electronic circuit diagram. This chamber design should simplify data acquisition in interface environments.



## 1. Introduction

In vitro models of pathophysiological states have been widely used during the last decades, which is especially true for the simulation of clinically relevant conditions such as hypoxia and ischemia, or for tests of pharmacological measures under these circumstances. Apart from an obviously more effective use of animal resources, the brain slice model of ischemia excludes systemic influences, and it provides control over parameters which are not easily accessible in vivo (Wassmann et al., 1996). The possibility of ethically unobjectionable experiments using human brain tissue originating from tumor or epilepsy surgery is even more important, as the species border between model and possible clinical application can be avoided (Wölfer et al., 2006). Compared with submerged-type settings, the interface model has been proven to be superior in anoxia experiments, as it more closely replicates the disturbances of ion homeostasis to be observed in vivo (Hansen, 1985; Croning and Haddad, 1998). These disturbances are correlated to typical changes of bioelectric signals like the loss of evoked potentials (EP), spreading depression, or so-called anoxic depolarization (AD) (Rothstein, 2000; Somjen, 2001; Dehbandi et al., 2008). This is why electrophysiological measurements play an important role in slice models.

In an interface setting, a large range of conditions can be easily created by changing the composition of the aqueous and/or the gaseous phase. Further, most (patho-)physiological processes within the tissue are highly temperature-dependent (Dietrich et al., 1996; Greiner et al., 1998), so temperature is another major parameter to be closely controlled.

Currently used interface chamber designs have the disadvantage that experimental parameters have to be changed by a series of manipulations which may compromise measurement accuracy and tissue integrity, e.g. by slight movements of microelectrodes within the tissue during initiation of substrate depletion. Furthermore, experimental protocols which involve temperature changes are difficult to realize as most designs are rather sluggish to adjust between different temperature levels. On the other hand, a closer simulation of clinical situations like a series of short ischemias (as to be observed, e.g. in cerebrovascular surgery) or of anesthesiologic intervention (e.g. intraoperative hypothermia) requires quick and easy reaction of the setup.

With our new chamber design we aimed at combining the principal advantages of the interface design with a technical setup which allows for a quick and remotely controlled switch between two predefined settings of each aqueous phase, gaseous phase, and temperature.

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## 2. Materials and methods

### 2.1. Chamber design

The actual chamber (upside and front view drawn to scale in Fig. 1A) is based on an interface element originally derived from a design by Haas (Haas et al., 1979), and machined out of an acrylic glass block. Its ground is covered with a coarse hydrophilic nylon mesh, which is fixed at its lateral margins and allows for uniform distribution of the aqueous phase. Condensate traps are used to avoid larger condensate droplets from the gaseous phase being blown over the slice. The chamber is covered by a thin acrylic glass plate with an opening for several electrodes, which confines and directs the flow of the gaseous phase while additionally providing temperature insulation. The acrylic base has lateral openings for reference and temperature electrodes. The fluid volume held within the plastic mesh ranges about 0.3 ml, while the gaseous phase comprises about 1.5 ml.

Using Teflon® screws, this setup is screwed upon a thin aluminum block, which in turn rests upon two Peltier elements (RO4.6-10, Conrad Electronic, Hirschau, Germany; combined maximum heating/cooling power 20 W). These are pressed upon a larger

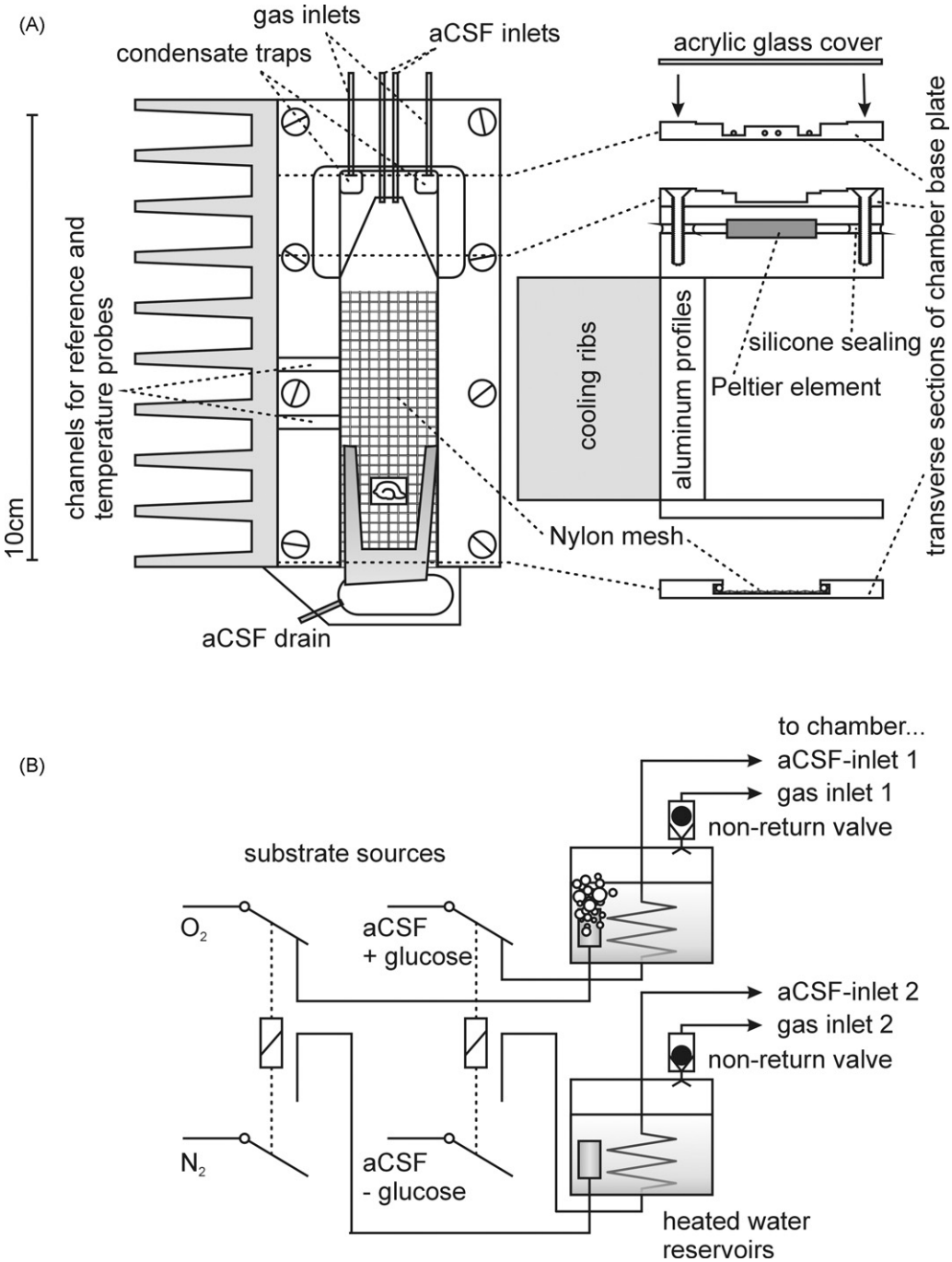


**Fig. 1.** Design and connections. (A) In-scale drawing of the chamber, view from above with a slice in situ, and three orthogonal cuts to demonstrate the profile of the chamber base plate. (B) Connections of substrate sources via coupled electromechanical valves. The water reservoirs are heated independently from the actual chamber. Functional state shown = normoxia at normal fluid glucose. ACSF = artificial cerebrospinal fluid.