MAC (CP50i) is defined as ‘minimum alveolar concentration’ of an inhaled anaesthetic (minimum plasma concentration of i.v. anaesthetics) that prevents movement in response to skin incision in 50% of a test population. MAC1 and CP50i2 are well-established measures to compare potencies of anaesthetics. The underlying clinical endpoint immobility reflects mainly effects of anaesthetics on the spinal cord, which limits the use of this measure for comparison of effects on the main target organ of general anaesthesia—the brain. The present study determines the median concentration of sevoflurane, isoflurane, and propofol that induce the onset of electroencephalogram (EEG) suppression (‘silent second’): MACBS and CP50BS.

Methods. Fifty-five unpremedicated patients (ASA physical status of I or II) undergoing elective surgery were randomly assigned to receive general anaesthesia with sevoflurane, isoflurane, or propofol. A two-channel EEG was continuously recorded to identify ‘silent second’. Independent cross-over pairs were analysed using the ‘Dixon’s up-and-down’ method, and MACBS/CP50BS values were calculated by logistic regression.

Results. CP50BS was 4.9 \mu \text{g} \text{ml}^{-1} for propofol. MACBS was 2.9 vol% for sevoflurane and 1.5 vol% for isoflurane. CP50BS of propofol was less than one-third of CP50i, whereas MACBS of sevoflurane was >1.4-fold of MAC; MACBS of isoflurane was 1.3-fold of MAC.

Conclusions. Immobility and cerebral effects reflect different entities of anaesthetic action. The median concentration of anaesthetic drug (volatile or i.v. agent) required to induce ‘silent second’ might be a more useful metric than the median concentration required to prevent movement in response to a surgical stimulus in order to compare relative potencies of anaesthetic agents on the brain. Advantage of the ‘silent second’ is an easy identification of this endpoint, while such a deep level is not required for clinical anaesthesia.

Keywords: anaesthetics, inhalation; anaesthetics, intravenous; electroencephalography

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Methods

Patient selection and randomization

After informed written consent and approval of the ethics committee of the Technische Universität München, Munich, Germany, 55 unpremedicated patients with an ASA physical status of 1 or II undergoing elective surgery under general anaesthesia were randomly assigned to the groups SEVO (sevoflurane, Abbott GmbH, Wiesbaden, Germany), ISO (isoflurane, Abbott GmbH, Wiesbaden, Germany), and PRO (propofol, Disopivran, GlaxoSmithKline, Middlesex, UK).

Patients with contraindications to the study drugs; a history of psychiatric or neurologic diseases, drug abuse, or medication known to affect the central nervous system; pregnancy; or indication for emergency procedures and rapid sequence induction were excluded from the study.

Monitoring

Standard vital parameters (including non-invasive arterial pressure, heart rate, oxygen saturation, end-tidal carbon dioxide concentration, anaesthetic concentration, and respiratory parameters) were monitored with a Datex AS/3 compact monitor (Datex Ohmeda, Helsinki, Finland). A two-channel referential EEG was continuously recorded from ZipPrep electrodes (Aspect Medical Systems, Newton, MA, USA) at positions Fp1-Cpz; Fp2-Cpz (electrode positions according to the international 10–20 system) with an Aspect A-1000 EEG-Monitor (bispectral index (BIS)-Version 3.3, Aspect Medical Systems, Inc., Newton, MA, USA) using a 256 Hz digitizing rate and an analogue band-pass from 0.25 to 100 Hz. Electrode impedances were kept below 5 kΩ. EEG and standard monitoring parameters were stored on a personal computer.

Anaesthetic procedure

General anaesthesia was induced through mask inhalation with flow rates of 8 litre min⁻¹ for the sevoflurane and isoflurane group or with target-controlled infusion of propofol. Atracurium (0.6 mg kg⁻¹) was administered for neuromuscular block. Tracheal intubation was performed in all patients in order to maintain end-tidal CO₂ constantly between 4.3 and 4.8 kPa.

The target anaesthetic concentration was determined by Dixon’s up-and-down method. The initial concentration was set to 3.8 vol% sevoflurane (Group SEVO), isoflurane 2.4 vol% (Group ISO) and propofol 6.0 µg ml⁻¹ (Group PRO), according to clinical expertise. Target sevoflurane, isoflurane, and propofol concentrations were maintained constant for 15 min. In the 16th min, EEG was analysed to identify the onset of burst suppression pattern. Burst suppression was defined as EEG suppression lasting at least 1 s (‘silent second’, Fig. 1). ‘Silent second’ has been suggested as the endpoint for a threshold test of anaesthetic potency in animals. In Group PRO, an additional blood sample was obtained at the end of the 16th minute to determine plasma concentration of propofol. Then, surgery was performed and general anaesthesia was administered according to standard clinical practice.

Dixon’s up-and-down technique

Dixon’s up-and-down technique is a commonly used method to define MAC. Anaesthetic concentration applied to the first patient is estimated by clinical experience. If ‘silent second’ occurs, anaesthetic concentration in the subsequent patient is decreased by 10% (Fig. 2). If the EEG does not show suppression pattern, the concentration is increased by 10%. In each series of patients, independent cross-over pairs in the order of measurements are identified. A cross-over pair consists of two consecutive patients, where either EEG suppression occurred at the first but not the second patient or vice versa. Each patient was considered only for one single cross-over pair in order to assure independent test data.

Statistical analysis

Six independent cross-over pairs and ~10 changes in direction are required to obtain reliable results. Logistic regression (LR) was used to determine MACBS and CP₅₀BS. The probability of BS (p) is modelled in terms of log odds by a linear function of the independent explanatory variable ‘measured concentration’ X by a logistic regression: log[p/(1−p)]=A+B×X, where A (intercept) and B (regression slope) are the model parameters. The solution of 0.5=exp (A+B×X)/1+exp(A+B×X) results in the MAC value estimation. To quantify uncertainty of the MAC values, 95% percentile bootstrap confidence intervals (CIs) were used. Statistical evaluation and figures were performed using R (R Development Core Team, http://www.r-project.org/percentile).

Statistical differences between results of LR calculated for target and measured anaesthetic concentrations were analysed for all three groups. The ratio between concentrations...
of MAC/CP50i values and the corresponding MACBS/CP50BS values were calculated.

**Results**

Each study group included six independent cross-over pairs (Fig. 3). Patient characteristic data and haemodynamic parameters did not show significant differences between groups (Table 1).

MACBS and CP50BS calculated with logistic regression are illustrated in Figure 4. For sevoflurane MACBS was 2.93 vol% (95% CI 2.80–3.15) and for isoflurane 1.51 vol% (95% CI 1.45–1.60). The corresponding CP50BS for propofol was 4.85 µg ml⁻¹ (95% CI 4.25–5.40).

There were no statistical differences in results of logistic regression calculated for measured and target anaesthetic concentrations in all three groups. Results for MACBS were additionally set in relation to age-related MAC/CP50i values (MAC is 2.05 vol% for sevoflurane²⁰ and 1.15 vol% for isoflurane,²¹ CP50i 15.2 µg ml⁻¹ for propofol)² as recommended by manufacturers, previous research standards, or both. MACBS sevoflurane was >1.4-fold MAC. MACBS of isoflurane was >1.3-fold MAC and CP50BS of propofol was less than one-third of CP50i.

**Discussion**

We have found interesting differences in the ratio between concentrations required for EEG burst suppression and for suppression of movement. For sevoflurane the MACBS (2.9 vol%) was >1.4-fold MAC, for isoflurane the MACBS (1.5 vol%) was >1.3-fold MAC, whereas for propofol the CP50BS (4.9 µg ml⁻¹) was less than one-third of CP50i. These differences underline current research results: immobility and unconsciousness represent different entities of anaesthetic action. Immobility during general anaesthesia reflects mainly anaesthetic effects on the spinal cord and may be mediated by particular substance-specific mechanisms.³ ⁴ ⁷ Despite this, traditional concepts for comparison of anaesthetic potencies have used immobility as clinical endpoint. But improved concepts should rather be based on measures that reflect effects on the main target organ of anaesthesia—the brain—in a reproducible manner.

For determination of MACBS/CP50BS, we chose the methodology described by Dixon and colleagues.¹³ This methodology works best when the starting point is near the ED₅₀. The respective initial agent level was determined according to clinical expertise. Analysis revealed that the chosen initial anaesthetic concentrations were higher than the population MACBS/CP50BS. As a consequence, the CIs of the respective MACBS/CP50BS values may be inadequately large. In particular, the propofol logistic regression graph (Fig. 4c) illustrates how wide the CIs are. This can be considered as a potential weakness of the study. On the other hand, this is similar to other propofol Cp (blood concentration) metrics, e.g. for the ‘MAC’ equivalent as reported by Smith and colleagues.² The authors argued that variability in sensitivity to propofol may be further influenced by pharmacodynamic and pharmacokinetic differences (e.g. attributable to varying volumes of the central compartment and of total body clearance). These effects may also have influenced the results in our study population.

The large CIs of the respective MACBS/CP50BS values may also be attributable to the small sample size required by Dixon’s up-and-down method.¹³ This limitation was accepted to maintain comparability to studies, which determined MAC concentrations to other endpoints (e.g. movement to skin incision).²² Still, this may be seen as a limitation of the current study.

The choice of our endpoint ‘silent second’ during burst-suppression EEG has to be carefully discussed. For lighter levels of anaesthesia, clinical endpoints for anaesthetic action (e.g. loss of consciousness) have been recommended. A burst suppression pattern represents a pseudo clinical endpoint. But there is no other clinical assessment during very deep anaesthesia which can be easily identified and is based on effects of anaesthetics on the main target organ of anaesthesia—the brain. All drugs with a hypnotic effect—volatile and i.v. anaesthetics except ketamine—lead to a dose-dependent suppression of electric brain activity with high concentrations of the drug: ‘silent second’ of EEG burst suppression.

Korkmaz and colleagues¹⁴ introduced ‘silent second’ for a threshold test to quantify and compare cerebral effects of anaesthetic drugs in an animal model.²³ For the threshold test, drugs are administered continuously until an EEG criterion indicating deep anaesthesia is reached: the ‘silent second’, defined as burst suppression lasting at least 1 s. The respective...
threshold (drug) dose needed to induce ‘silent second’ is a dependent variable. In the intact animal, it depends only on the potency of the drug and the dose administration rate of the infusion. This method is considered to be superior to commonly used specifications of burst suppression pattern, for example, burst suppression ratio (the ratio between bursts and suppression in a certain time window) as quantitative time domain measure. An endpoint of EEG burst suppression with duration for >1 s involves increasing equilibration between anesthetic concentration at the effect site, the brain, and plasma concentration. Furthermore, it may be considerably influenced by pharmacokinetic properties. As a consequence, the anesthetic dose to induce burst suppression for >1 s does not allow to exclusively quantify anesthetic potency to reach the threshold burst suppression but it may be biased by intra- and inter-individual differences in metabolism and redistribution. The apparently simple definition of ‘silent second’ not only allows easy monitoring, it has also served as a well-defined pharmacological endpoint for threshold testing of anesthetic potency in an animal model. This approach of ‘silent second’ was transferred to human EEG in the current study.

Justifications of choice of an appropriate EEG-based endpoint should be based on the underlying physiology. Mechanistic causes of anesthetic-induced burst suppression have not been elucidated. But current research data provide very fascinating considerations. Most prevailing features of burst suppression are the assumptions of synchrony of burst onset (stereotypic response simultaneously across the entire cortex) and of its parametric sensitivity to the level of general anesthesia. Burst suppression has been described as occurring on a continuum as general anesthesia deepens, probably guided by a changing of the underlying physiological process. Anesthetics are most commonly supposed to alter neuronal activity in a dose-dependent manner: at intermediate concentrations neurons begin to oscillate between a depolarized up-state and a hyperpolarized down-state. With increasing anesthetic depth, up-states transform to short bursts and the length of down-states increases progressively. Finally, an EEG burst-suppression pattern with increasing length of suppression periods is observed.

There is a common notion that different anesthetics produce burst suppression when they reduce cerebral metabolic rate (CMR) to the same residual rate: for example, GABAergic anesthetics lead to a dose-dependent reduction of CMR and suggest a link between reduced CMR and burst suppression. All commonly used i.v. and inhalation anesthetics reduce CMR. In contrast, the N-methyl-D-aspartate (NMDA)-receptor antagonist ketamine increases cerebral metabolism with small doses and does not induce burst-suppression pattern in clinically relevant concentrations. Thus, the CP50BS cannot be applied to ketamine as a measure of anesthetic potency.
Fig 4 (A–C) Calculation of burst-suppression MAC (MAC_{BS}) and CP_{50BS} with logistic regression for the sevoflurane (A), isoflurane (B), and propofol (C) groups. Blue dot: Burst suppression EEG; green ring: no burst suppression EEG.
EEG burst suppression seems to be reached gradually and thus may serve as an appropriate endpoint for threshold tests of anaesthetic potency. This is in contrast to endpoints associated with loss of consciousness. Current research data suggest that when a critical anaesthetic concentration is reached, abrupt loss of consciousness occurs as a result of a nonlinear collapse of a repertoire of neuronal states rather than a complete switch in sensory relay.27

Current evidence suggests that anaesthetic agents do not cause global suppression of neuronal activity but rather exert differential effects on cortico-cortical and (specific and non-specific) thalamo-cortical dynamics: there is increasing evidence for residual neuronal activity during general anaesthesia, even during burst suppression.26 27 31–37 The transfer of these functional magnetic resonance imaging (fMRI) findings to burst suppression remains hypothetical: burst suppression pattern might be associated with an underlying clinical state with interruption of information integration, rather than with a complete suppression of information transfer from the periphery to the sensory cortex. Unfortunately, there are no results of imaging studies combined with EEG data during burst suppression available. But there are studies, which analyse cortical electrophysiology during anaesthetic-induced burst suppression.24 33 Breshears and colleagues33 used electrocorticography to integrate spatial, temporal and spectral features of cortical dynamics during induction and emergence from propofol anaesthesia. Their results support the assumption that a larger stable neuronal architecture is maintained, even during burst suppression: large-scale functional networks (e.g. correlated fluctuations of the slow cortical potential < 0.5 Hz over the somatomotor cortex) remained stable regardless of the level of anaesthesia, despite sequential changes in cortical and thalamo-cortical dynamics with increasing anaesthetic depth. There seems to be an ordered interplay between cortical, thalamo-cortical, and reticular system during loss and return of propofol-induced consciousness. Superimposed on a maintained functional architecture, dynamic electrophysiological changes were observed: for example, linear decline of gamma-rhythm (cortical suppression) with anaesthetic depth, then early increase of delta-band (early hyperpolarization of thalamus) with transition from tonic to bursting activity pattern.

The mechanisms of anaesthetic-induced loss and return of consciousness have not been elucidated so far. But heterogeneous effects on different neuronal receptor-mediated mechanisms have been investigated. One might expect that these varying mechanisms result in differences in global or regional electrophysiology. Indeed, there exist agent-specific characteristics of EEG burst suppression.37 38 Akrawi and colleagues37 found substantial electrophysiological differences among the burst suppression states achieved with propofol, isoflurane, etomidate, or thiopental: for example, there were significant differences between duration of bursts, peak-to-peak-voltage and area under the curve. Furthermore, there was a considerable residual electrophysiological activity during suppression periods. These results support the view that the different burst suppression pattern achieved with various anaesthetics may reflect differences in the underlying neuronal dynamics. Furthermore, their findings are consistent with previous observations of agent-specific regional differences in anaesthetic-induced reduction of CMR, in particular of limbic or cerebellar metabolism.28 30 39 40 The previously described similarities in global reduction of CMR may predominantly reflect the most uniform cortical depression. A potential explanation may be provided by the underlying supratentorial measurement technique in the superior sagittal sinus.41

To sum up, the characteristics of burst suppression pattern (e.g. burst pattern and suppression duration) are agent-specific and influenced by pharmacokinetic differences in distribution and metabolism. Thus, there exist limitations for the use of burst suppression characteristics as uniform endpoint for anaesthetic potency. The overview of imaging and electrophysiologic data supports the view of a linear dynamical EEG frequency progression from start of anaesthetic agent to onset of burst suppression.33 42 43 The endpoint ‘silent second’ is reached with the onset of burst-suppression pattern and may serve as a suitable threshold test for anaesthetic potency in humans. The onset of burst suppression is achieved without agent-specific EEG characteristics. Thus, ‘silent second’ may further be appropriate as a single measure for different anaesthetics. Previous studies on measures for anaesthetic potency used, for example, endpoints from EEG frequency analysis, processed EEG or surrogate markers. They might be of limited value. Changes in the electroencephalographic power spectrum may not primarily reflect the level of hypnosis or general anaesthesia, but rather show a specific reaction to a particular anaesthetic drug. As an example, benzodiazepines induce activation of EEG beta activity, opioids increase activity of the EEG delta band.46 Deep propofol anaesthesia is also reflected by increased delta activity, but this time the level of hypnosis is also increased. During induction of anaesthesia, the behaviour of spectral EEG parameters is affected by a characteristic biphasic reaction.45 Kuizenga and colleagues45 demonstrated this for EEG amplitude and SEF95 in relation to loss of consciousness during induction. Thus, there was no consistent relationship between the time of occurrence of the peak EEG effect or the value of the EEG variable (SEF95 and BIS) and the moment of loss of consciousness. As a consequence, drug-specific EEG changes during induction and light levels of anaesthesia weaken the utility of EEG-based endpoints as comparators for anaesthetic potency.

Previous attempts were made to improve the traditional MAC concept with more complex parameters derived from processed EEG (e.g. MACBISSO).56–58 MACBISSO was defined as the minimal alveolar concentration to maintain BIS values < 50. The utility of the BIS and other derived EEG indices is limited by several factors. They are not physiological measures of cortical activity but represent probability measures of anaesthetic depth: each parameter was developed on the basis of a training data set with different patients or volunteers and different anaesthetic regimens. As a consequence, the performance of an index can only be as good as the underlying database.49 The algorithm of BIS computation remains proprietary.

Glass and colleagues49 analysed the ability of BIS to measure sedation effects of various anaesthetics (propofol,
isoflurane, and midazolam) in a volunteer study. They determined corresponding BIS values where 50 and 95% of all volunteers lost consciousness. Index values varied between the different anaesthetic agents. As the BIS value at the same clinical endpoint is not identical in different drugs, it should not be used as a comparative measure of anaesthetic effects.

To sum up, the comparison of anaesthetic potency based on processed indices is limited because of drug-specific effects, arbitrary index thresholds and the intervariability of index values.45 46

Clinical endpoints, such as loss of righting reflex and changes in the autonomic vegetative system (arterial pressure, sweating, or heart rate), are surrogate markers, which reflect indirect and non-specific effects of anaesthesia.50 51 MACBAR evaluates the potency of anaesthetics to prevent cardiovascular responses by blunting sympathetic responses.52 53 This endpoint may also be non-specific and not related to the main effect of general anaesthesia. In addition, it is affected by medication (e.g. β-blocker), underlying diseases (e.g. cardiovascular instability), and may show an inter-patient variability.

Loss (or return) of consciousness may be a suitable clinical endpoint for a comparison between drugs, as it reflects the main effect of anaesthetics. Therefore, MACawake has been suggested as a measure of anaesthetic potency.54 –56 Unfortunately, there are many methodological problems, which may impede analysis of MACawake.54 –56

In summary, we believe that clinical assessment provides weak endpoints for the comparison of anaesthetic potency.

Summary
The median concentration of anaesthetic drug (volatile or i.v. agent) required to induce burst suppression (‘silent second’) might be a more useful metric than the median concentration required to prevent movement in response to a surgical stimulus in order to compare relative potencies of anaesthetic agents on the brain. Even if a ‘silent second’ in the EEG represents an inadequately deep level for clinical anaesthesia, it can be easily identified and is based on the effects of anaesthetics on the main target organ of anaesthesia—the brain.

Authors’ contributions
S.P. wrote the manuscript, helped to design the study, conduct the study, and analyse the data. She has seen the original study data, reviewed the analysis of the data, approved the final manuscript, and is the author responsible for archiving the study files. D.J. helped to design the study, analyse the data and write the study. He has seen the original study data, reviewed the analysis of the data, and approved the final manuscript. M.K. helped to design the study, analyse the data, and write the manuscript. He has seen the original study data, reviewed the analysis of the data, and approved the final manuscript.


declaration of interest
None declared.

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