Effects of dexmedetomidine, propofol, sevoflurane and S-ketamine on the human metabolome

A randomised trial using nuclear magnetic resonance spectroscopy

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BACKGROUND Pharmacometabolomics uses large-scale data capturing methods to uncover drug-induced shifts in the metabolic profile. The specific effects of anaesthetics on the human metabolome are largely unknown.

OBJECTIVE We aimed to discover whether exposure to routinely used anaesthetics have an acute effect on the human metabolic profile.

DESIGN Randomised, open-label, controlled, parallel group, phase IV clinical drug trial.

SETTING The study was conducted at Turku PET Centre, University of Turku, Finland, 2016 to 2017.

PARTICIPANTS One hundred and sixty healthy male volunteers were recruited. The metabolomic data of 159 were evaluable.

INTERVENTIONS Volunteers were randomised to receive a 1-h exposure to equipotent doses (EC50 for verbal command) of dexmedetomidine (1.5 ng ml⁻¹; n = 40), propofol (1.7 μg ml⁻¹; n = 40), sevoflurane (0.9% end-tidal; n = 39), S-ketamine (0.75 μg ml⁻¹; n = 20) or placebo (n = 20).

MAIN OUTCOME MEASURES Metabolite subgroups of apolipoproteins and lipoproteins, cholesterol, glycerides and phospholipids, fatty acids, glycolysis, amino acids, ketone bodies, creatinine and albumin and the inflammatory marker GlycA, were analysed with nuclear magnetic resonance spectroscopy from arterial blood samples collected at baseline, after anaesthetic administration and 70 min post-anaesthesia.

RESULTS All metabolite subgroups were affected. Statistically significant changes vs. placebo were observed in 11.0, 41.3, 0.65 and 3.9% of the 155 analytes in the dexmedetomidine, propofol, sevoflurane and S-ketamine groups, respectively. Dexmedetomidine increased glucose, decreased ketone bodies and affected lipoproteins and apolipoproteins. Propofol altered lipoproteins, fatty acids, glycercides and phospholipids and slightly increased inflammatory marker glycprotein acetylation. Sevoflurane was relatively inert. S-ketamine increased glucose and lactate, whereas branched chain amino acids and tyrosine decreased.

CONCLUSION A 1-h exposure to moderate doses of routinely used anaesthetics led to significant and characteristic alterations in the metabolic profile. Dexmedetomidine-induced alterations mirror α2-adrenoceptor agonism. Propofol emulsion altered the lipid profile. The inerntness of sevoflurane might prove useful in vulnerable patients. S-ketamine induced amino acid alterations might be linked to its suggested antidepressive properties.

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Introduction
Metabolomics has provided promising results in the fields of early diagnosis, biomarker discovery and stratification of disease risk-groups.\textsuperscript{1–4} Pharmacometabolomics, a relatively new research direction, uses large scale data-capturing methods to uncover shifts in the metabolic profile induced by pharmacotherapy. Further analysis of these changes in biochemical pathways and identification of baseline predictors of patient response via biomarker acquisition may eventually offer an opportunity to enhance early prediction of treatment outcomes, to reveal novel mechanisms of drug action and to identify metabolic pathways contributing to drug response phenotypes.\textsuperscript{2,5–8} Ultimately, pharmacometabolomics might enable the discovery of novel tools for clinical decision-making, the optimisation of personalised medical therapy and precision medicine.

The pharmacometabolomic effects of anaesthetic agents remain insufficiently documented. The effects of dexmedetomidine on insulin secretion, S-ketamine on circulating catecholamines and propofol on triglyceride levels are examples of pharmacological attributes that may have interesting metabolic ramifications.\textsuperscript{9–21} In this exploratory study, the aim was to discover whether four routinely used anaesthetics, dexmedetomidine, propofol, sevoflurane and S-ketamine, cause acute alterations in the human metabolic profile in the absence of confounding factors present in clinical anaesthesia and surgery. An infographic summarising the study design, metabolomic analysis and results is shown in Fig. 1.

Materials and methods

Ethics
Approval for this study (LOC-2016, EudraCT 2015-004982-10, ClinicalTrials.gov Identifier NCT02624401) was provided by the Ethics Committee of Hospital District of Southwest Finland, Turku, Finland on 15 December 2015.

Trial design and participants
This randomised, open-label, controlled, parallel group, phase IV clinical drug trial was conducted at Turku PET Centre, University of Turku, Finland, as a part of ‘The Neural Mechanisms of Anaesthesia and Human Consciousness’ project. A detailed description of the study methods has been published previously in an article comparing the effects of anaesthetic agents on regional cerebral glucose metabolism.\textsuperscript{22}

One hundred and sixty healthy, American Society of Anaesthesiologists class I (ASA I) male volunteers were randomised to receive dexmedetomidine (Dexdor 100 µg ml\textsuperscript{-1}, Orion Pharma, Espoo, Finland; \( n = 40 \)), propofol (Propolipid 10 mg ml\textsuperscript{-1}; Fresenius Kabi, Bad Homburg, Germany; \( n = 40 \)), sevoflurane (Sevoflurane 100%, Abbvie, North Chicago, Illinois, U.S.A; \( n = 40 \)), and placebo (Ringer's acetate; \( n = 20 \)).

The circulating metabolic profile arises from a complex interplay of biochemical pathways reflecting changes on many levels: From gene expression to cell and tissue metabolism.

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Fig. 1 Infographic for the study

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S-ketamine (Ketanest-S 25 mg ml\(^{-1}\), Pfizer, NY, U.S.A; \(n = 20\)) or placebo (Ringer’s acetate, \(n = 20\)). The inclusion criteria have been described earlier. Written informed consent was obtained from all participants in accordance with the Declaration of Helsinki.

Only male participants were studied because of radiation exposure related to a subsequent PET study. Due to the exploratory nature of the study, a formal power analysis was not considered applicable. Randomisation was performed using balanced permuted block sizes of 16 as previously described. Fig. 2 shows the Consort Flow diagram for the study participants.

**Anaesthetic protocol and monitoring**

All participants fasted from midnight until time of anaesthesia. Intake of alcohol, caffeine and medications were prohibited preceding anaesthesia. The duration of the anaesthetic administration protocol was 60 min; details have been published earlier. Intravenous anaesthetics were administered using target-controlled infusion with a

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**Fig. 2** Consort flow diagram for participants in the study

**Enrolment**

- Assessed for eligibility (\(n = 366\))
  - Excluded (\(n = 186\)):
    - Not meeting the inclusion criteria (\(n = 151\))
    - Declined participation (\(n = 35\))
    - Other reasons (\(n = 0\))
  - Randomised (\(n = 180\))
  - Withdrew after randomisation (\(n = 15\))

**Allocation**

- Allocated to dexmedetomidine (\(n = 40\))
  - Received allocated intervention (\(n = 40\))
  - Did not receive allocated intervention (\(n = 0\))

- Allocated to propofol (\(n = 40\))
  - Received allocated intervention (\(n = 40\))
  - Did not receive allocated intervention (\(n = 0\))

- Allocated to sevoflurane (\(n = 41\))
  - Received allocated intervention (\(n = 41\))
  - Did not receive allocated intervention (\(n = 0\))

- Allocated to S-ketamine (\(n = 24\))
  - Received allocated intervention (\(n = 24\))
  - Did not receive allocated intervention (\(n = 0\))

- Allocated to placebo (\(n = 20\))
  - Received allocated intervention (\(n = 20\))
  - Did not receive allocated intervention (\(n = 0\))

**Intervention**

- Discontinued intervention (\(n = 0\))
  - Excessive motor anxiety (\(n = 1\))

- Discontinued intervention (\(n = 0\))

- Discontinued intervention (\(n = 3\))
  - Excessive motor anxiety (\(n = 3\))

- Discontinued intervention (\(n = 0\))

**Analysis**

- Analysed (\(n = 40\))
  - Excluded from analysis (\(n = 1\))
  - Sample lost (\(n = 1\))

- Analysed (\(n = 39\))
  - Excluded from analysis (\(n = 1\))

- Analysed (\(n = 20\))
  - Excluded from analysis (\(n = 1\))
  - Programming error, received only 7% of planned S-ketamine dose (\(n = 1\))

- Analysed (\(n = 20\))
  - Two individual timepoint samples were lost

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CONSORT flow diagram showing each stage of the randomised-controlled-trial, enrolment, allocation, intervention and analysis.
Harvard 22 syringe pump (Harvard Apparatus, South Natick, Massachusetts, USA) and Stanpump software (www.opentci.org/code/stanpump). Previously reported pharmacokinetic parameters were used (Domino, Talke, March-models).\textsuperscript{21–25} Sevoflurane was administered and monitored using a Dräger Primus anaesthesia workstation (Drägerwerk AG & Co KGaA, Lübeck, Germany). The details of participant preparation and monitoring have been described earlier.\textsuperscript{22}

The target concentrations (EC\textsubscript{50} for verbal command) were based on previous studies: 1.5 ng ml\textsuperscript{-1} for dexmedetomidine, 1.7 µg ml\textsuperscript{-1} for propofol, 0.75 µg ml\textsuperscript{-1} for S-ketamine and end-tidal target of 0.9% for sevoflurane.\textsuperscript{26–28} The arterial blood concentrations of the intravenous anaesthetics were measured off-line and the end-tidal concentration of sevoflurane was monitored continuously during anaesthetic administration.

**Blood sampling and metabolomic analysis**

Arterial blood samples for metabolomic analysis were collected before anaesthetic administration (timepoint 1), at the end of 60 min of anaesthetic administration (timepoint 2) and 70 min after the cessation of anaesthetic administration (timepoint 3). A 9 ml whole blood EDTA sample was collected at each timepoint. The blood samples were protected from light and placed on ice immediately after sampling. Within 30 min, plasma was separated using cold centrifugation (+4°C) and divided into amber tubes (Matrix 1.0 mL 2D Screw tubes Amper PP; Thermo Scientific, USA). Amber tube samples were immediately frozen at -20°C and the samples were transferred to -70°C within the same day keeping the temperature under -70°C by means of dry ice. Sampling, storage and transfer of the metabolomics samples were conducted according to the specifications of the company responsible for metabolite quantification (Nightingale Health, Helsinki, Finland).

Metabolic biomarkers were quantified from plasma samples using high-throughput nuclear magnetic resonance (NMR) spectroscopy (Nightingale Health Ltd, Helsinki, Finland). The NMR metabolomics platform is based on proton NMR spectroscopy, wherein each molecule with hydrogen atoms gives a characteristic spectral signal. This signal is of a distinctive shape, which permits the identification of molecules, and the signal area is proportional to the concentration of the molecule in the sample. Metabolites were measured and compared with each added propofol concentration. In addition, the mean changes in very low-density lipoprotein particle (VLDL) markers in the propofol group were compared with the VLDL changes in the in vitro experiment to assess the possibility of soybean oil accumulation during continuous infusion. The results of the in vitro analysis are described in detail in Appendix B, http://links.lww.com/EJA/A604.

**Statistical analysis**

Logarithmic transformation was performed for metabolites with skewness more than 1 (42% of all metabolites). All metabolites were scaled to baseline standard deviation (SD) to enable comparisons across metabolites with different units. The SD was chosen instead of the Z-score to ensure that the lipid formulation of propofol had no direct effect on NMR quantification of metabolite markers in vitro at propofol target concentration of 1.7 µg ml\textsuperscript{-1}. The same NMR instrumentation as in the main analyses was used (Nightingale Health). In vitro analyses were conducted using pooled human serum samples that had been previously prepared from whole blood samples collected in lithium heparin tubes and stored at -80°C, similar to the main analysis. Propofol was added to pooled samples in increamental concentrations (1.75 to 100 µg ml\textsuperscript{-1}). Metabolite concentrations were measured and compared with a blank pooled sample (without added propofol) and with each added propofol concentration. In addition, the mean changes in very low-density lipoprotein particle (VLDL) markers in the propofol group were compared with the VLDL changes in the in vitro experiment to assess the possibility of soybean oil accumulation during continuous infusion. The results of the in vitro analysis are described in detail in Appendix B, http://links.lww.com/EJA/A604.

**In vitro analyses**

Because of the lipid emulsion formulation of propofol and due to extensive changes observed in the lipoprotein markers during propofol infusion in vivo, as a quality control measure, in vitro analyses were carried out to ensure that the lipid formulation of propofol had no direct effect on NMR quantification of metabolite markers in vitro at propofol target concentration of 1.7 µg ml\textsuperscript{-1}. The same NMR instrumentation as in the main analyses was used (Nightingale Health). In vitro analyses were conducted using pooled human serum samples that had been previously prepared from whole blood samples collected in lithium heparin tubes and stored at -80°C, similar to the main analysis. Propofol was added to pooled samples in increamental concentrations (1.75 to 100 µg ml\textsuperscript{-1}). Metabolite concentrations were measured and compared with a blank pooled sample (without added propofol) and with each added propofol concentration. In addition, the mean changes in very low-density lipoprotein particle (VLDL) markers in the propofol group were compared with the VLDL changes in the in vitro experiment to assess the possibility of soybean oil accumulation during continuous infusion. The results of the in vitro analysis are described in detail in Appendix B, http://links.lww.com/EJA/A604.

From each sample, 146 metabolite markers and nine ratios were analysed or calculated. The measured biomarkers included 101 lipoprotein measures, 37 lipidrelated markers (including 16 fatty acid, nine cholesterol, nine glycerides and phospholipids and three apolipoprotein measures), concentrations of eight amino acids, three glycolysis-related metabolites, three ketone bodies, creatinine and albumin and an inflammatory marker glyco-protein acetylation. The metabolite markers within each of the aforementioned metabolite subgroups are illustrated in the forest plots in Appendix A, http://links.lww.com/EJA/A603.
outcome and time as a within factor and group as a between factor. Because all metabolites were analysed using separate ANOVA models, there are no assumptions concerning the dependency between metabolites. The mean differences in SD changes (95% CI) between groups for all metabolites were estimated from a repeated measures model using group by time interaction effect.

The anaesthetic-placebo and anaesthetic-anaesthetic group differences in SD changes were estimated between timepoints 1 vs. 2 and 1 vs. 3. To account for multiple testing (155 metabolites, 10 pairwise group comparisons and two timepoint comparisons) the observed p values were Bonferroni corrected by factor 3100 such that an alpha threshold of 0.05 remained. Data are expressed as SDS (95% CI) between timepoints 1 vs. 2, if not otherwise stated. Statistical analyses were carried out with SAS software (version 9.4; SAS Institute Inc., Cary, North Carolina, USA).

For data visualisation, forest plots and line graphs were created using R (Version 1.1.383, https://www.R-project.org/) ggpplot2 function (Version 3.2.1, https://ggplot2.tidyverse.org). The infographic was created using Microsoft PowerPoint (version 16.16.18). The cross-correlations of the analytes were calculated based on the current data of the analytes in the dexmedetomidine, propofol, sevoflurane and S-ketamine groups, respectively. The majority of these changes remained significant in one or more anaesthetic-anaesthetic comparisons (Appendix A, http://links.lww.com/EJA/A603). Due to the large number of significant changes, up to three metabolites in each anaesthetic group showing the largest statistically significant SDS change (timepoints 1 vs. 2) are shown in detail in Table 2, Fig. 4 and Fig. 5. All analysed biomarker changes in all inter-group comparisons are shown in Appendix A, http://links.lww.com/EJA/A603. A heatmap containing cross-correlations of measured metabolites is provided in Appendix C, http://links.lww.com/EJA/A605.

In the dexmedetomidine group, strongest observed changes were the elevation of glucose and concomitant decrease in ketone bodies (Table 2). Changes in HDL composition and a decrease in very large HDL concentration of -0.29 SDS (95% CI -0.42 to -0.17), P < 0.02 were observed. The anaesthetic-placebo changes in glucose, 3-hydroxybutyrate and very large HDL phospholipid and free cholesterol content were significant in all inter-group comparisons.

Extensive changes in the lipid profile were observed in the propofol group. Strongest increase was observed in saturated fatty acids to total fatty acids ratio (SFA/FA), very large VLDL free cholesterol content (XL_VLDL_FC) and very large HDL triglyceride content (Table 2). Unsaturated fatty acids decreased by -0.45 SDS (95% CI -0.60 to -0.30), P < 0.001 and saturated fatty acids (SFA) increased by 0.54 SDS (95% CI 0.37 to 0.70), P < 0.001. Significant changes in lipoprotein concentration and content were observed in a range of lipoprotein particles sizes (Appendix A, http://links.lww.com/EJA/A603). Only a modest increase in serum total triglyceride level (Serum_TG) 0.32 SDS (95% CI 0.21 to 0.42), P < 0.001 was observed. The inflammatory marker glycoprotein acetylation (GlycA) increased by 0.37 SDS (95% CI 0.21 to 0.53), P = 0.023. Thirty-seven metabolites showed significant changes in all inter-group comparisons in metabolite subgroups of lipoproteins, fatty acids and glycerides and phospholipids (Appendix A, http://links.lww.com/EJA/A603).

In the sevoflurane group, only the increase in alanine was significant and was 0.6 SDS (95% CI 0.4 to 0.81), P < 0.001.

Table 1 Baseline characteristics of volunteers

|               | n   | Height (cm)       | Weight (kg)    | BMI       | Age (years) |
|---------------|-----|-------------------|----------------|-----------|-------------|
| Dexmedetomidine| 40  | 179.1 ± 6.5       | 77.3 ± 10.8    | 24.1 ± 2.9| 24.7 [20 to 30]|
| Propofol      | 40  | 180.5 ± 6.0       | 77.6 ± 11.1    | 23.6 ± 3.3| 23.4 [18 to 28]|
| Sevoflurane   | 39  | 179.7 ± 7.2       | 79.5 ± 8.7     | 24.6 ± 2.8| 24.4 [19 to 30]|
| S-ketamine    | 20  | 182.7 ± 5.4       | 79.9 ± 10.6    | 23.9 ± 3.0| 23.4 [20 to 30]|
| Placebo       | 20  | 182.4 ± 8.8       | 82.9 ± 14.2    | 24.8 ± 2.6| 23.1 [20 to 28]|

Data are given as mean ± SD or mean [range].

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In the S-ketamine group, strongest changes were observed in glucose, lactate and leucine. Glucose and lactate increased, while leucine decreased (Table 2). Isoleucine decreased by $-0.89$ SDS (95% CI $-1.25$ to $-0.54$), $P = 0.006$, tyrosine by $-0.89$ SDS (95% CI $-1.27$ to $-0.52$), $P = 0.017$ (timepoints 1 vs. 3). The decrease in leucine was significant in all inter-group comparisons.

In vitro analyses
Briefly, the in vitro analysis showed that propofol emulsion, when directly added to baseline samples at concentrations corresponding to the target concentration of $1.7 \mu g \text{ml}^{-1}$, caused only minor changes in the metabolic profile. Interestingly, the in vivo propofol infusion with a target concentration of $1.7 \mu g \text{ml}^{-1}$ resulted in a change in VLDL markers corresponding to the levels observed at propofol concentration of $17.5 \mu g \text{ml}^{-1}$ in vitro. The detailed results of in vitro analyses are described in Appendix B, http://links.lww.com/EJA/A604.

Discussion
In this study, we evaluated the metabolic profiles of anaesthetic exposure to equipotent doses (EC$_{50}$ for verbal command) of dexmedetomidine, propofol, sevoflurane and S-ketamine in healthy men using NMR.

Fig. 3 Individual measured concentrations of the four anaesthetics: Dexmedetomidine, propofol, sevoflurane, S-ketamine

The study drugs were administered for 60 minutes, but their concentrations were measured at 20, 30, 40 and 50 minutes. Targeted concentrations are depicted with red lines. Mean measured concentrations have been published earlier.

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spectroscopy. Interestingly, marked alterations in the metabolic profiles of anaesthetics vs. placebo were revealed, regardless of moderate dosing and a relatively short anaesthetic exposure. Furthermore, the metabolic profiles of four commonly used anaesthetics were quite different.

The changes in the metabolic profile of dexmedetomidine were mainly seen in glucose and ketone bodies. Dexmedetomidine increased glucose concentration 29% from baseline. Changes in HDL composition and a decrease in very large HDL concentration were observed. Increased glucose and concomitantly decreased ketone bodies in fasted individuals reflect dexmedetomidine-induced inhibition of insulin secretion in response to α2-adrenoceptor agonism.15,16 It is possible that the alterations in HDL markers too (Appendix A, http://links.lww.com/EJA/A603) are linked to (α2-adrenoceptor agonism, as selective alpha adrenoceptor blockade is known to increase HDL levels.31

Propofol induced a subtle increase in the inflammatory marker GlycA, the concentration of which was 3.8% higher in the propofol group at timepoint 2 when compared with placebo. GlycA is a complex NMR signal arising from the N-acetyl groups in circulating glycoproteins that are involved in the acute phase response. Chronically elevated GlycA has been associated with a gene network enriched for neutrophil functions, elevated levels of inflammatory cytokines, long-term risk of serious infection and all-cause mortality.32,33 However, little is known about acute alterations in GlycA.34 Our interpretation of the results is that even though GlycA elevation seems mechanistically interesting, the magnitude of GlycA increase in response to short-term propofol administration is small and unlikely to have clinical implications.34

Propofol induced marked changes in the lipid profile, and surprisingly, only a modest increase in total triglycerides was observed. In the propofol group, serum total triglycerides increased 8.1% (timepoints 1 vs. 2). As a
## Table 2 Summary of absolute and SDS changes in selected metabolites

| Timepoint | Timepoint | Difference in SDS | P        | Timepoint | Timepoint | Difference in SDS | P        |
|-----------|-----------|-------------------|----------|-----------|-----------|-------------------|----------|
| 1         | 2         |                   |          | 1         | 2         |                   |          |
| Dexmedetomidine | Placebo | /C6 /C6 | 0.61 | 4.6 | /C6 /C6 | 0.25 | 4.11 | /C6 /C6 | 0.001 |
| 0.001     | <0.001    |                   |          | 0.001     |          |                   |          |
| Propofol | Placebo   | /C6 /C6 | 1.74 | 35.2 | /C6 /C6 | 1.62 | 35.2 | /C6 /C6 | 0.009 |
| 0.009     | <0.001    |                   |          | 0.009     |          |                   |          |
| Sevoflurane | Placebo | /C6 /C6 | 1.62 | 35.2 | /C6 /C6 | 0.06 | 0.28 | /C6 /C6 | 0.06 |
| 0.06     |          |                   |          | 0.06     |          |                   |          |
| S-ketamine | Placebo  | /C6 /C6 | 0.34 | 0.82 | /C6 /C6 | 0.44 | 0.59 | /C6 /C6 | 0.05 |
| 0.05     |          |                   |          | 0.05     |          |                   |          |

Data are given as mean ± SD or difference (95% CI).

Timepoint 1 refers to the samples drawn before anaesthetic administration at baseline, timepoint 2 samples were drawn after 60-min anaesthetic administration and timepoint 3 samples were drawn 70 min after the cessation of anaesthetic administration. Changes between timepoints 1 vs. 2 and 1 vs. 3 are in SDS units, P values are shown for statistically significant changes. Before the calculation of the SDS, logarithmic transformation for skewed variables was performed, all metabolites shown here were logarithmically transformed before calculation of SDS. AcAce, acetoacetate; Ala, alanine; bOHBut, 3-hydroxybutyrate; Glc, glucose; Lac, lactate; Leu, leucine; SFA_FA, ratio of saturated fatty acids to total fatty acids; XL_HDL_TG, triglycerides in very large HDL particles; XL_VLDL_FC, free cholesterol in very large VLDL particles.

Somewhat surprisingly, the ratio of saturated fatty acids to total fatty acids (SFA_FA) was increased in the propofol group. This is interesting, as soybean oil and Intralipid contain mostly unsaturated fatty acids. The unsaturated fatty acids decreased and saturated fatty acids increased, and were relatively unaffected by addition of propofol (1.75 to 17.5 mg l⁻¹) in vitro analysis (Appendix B, http://links.lww.com/EJA/A604). Previous in vitro studies on human skeletal and heart muscle cell bioenergetics have shown a profound inhibitory effect of propofol on fatty acid oxidation (FAO). On the basis of previous research, after an overnight fast, FAO may account for up to 70% of total body energy expenditure. In theory, a disruption of fatty acid utilisation could lead to changes in the circulating lipid profile. However, as free fatty acid availability exceeds the needs for FAO by a substantial amount, one could argue that changes in fatty acid ratios would take time to emerge. Whether propofol induced changes in fatty acid utilisation or FAO could affect the elevation of SFA_FA remains unknown.

On the basis of our results, the lipid load induced by propofol could be reflected by the increase of SFA_FA during a 60-min propofol infusion, whereas triglyceride levels remained relatively unchanged. It is worth contemplating whether the cumulation of propofol emulsion constituents or the alterations observed in the lipid profile could, in prolonged infusions and with vulnerable patients, be associated with the known complications of propofol infusion: hypertriglyceridaemia, hypertriglycerideraemia associated pancreatitis and the propofol infusion syndrome. Currently, triglyceride levels of ICU patients are routinely screened for early diagnosis of propofol-induced hypertriglyceridaemia, which tends to develop after a median of 4 to 7 days in vulnerable...
First measurement at baseline before anaesthetic administration, second after 60 min of anaesthetic administration and third 70 min after the cessation of anaesthetic administration. The timepoint means are depicted with red lines. Ala, alanine; AcAce, acetoacetate; bOHBut, 3-hydroxybutyrate; Glc, glucose; Lac, lactate; Leu, leucine; SFA_FA, ratio of saturated fatty acids to total fatty acids; XL_HDL_TG, triglycerides in very large HDL particles; XL_VLDL_FC, free cholesterol in very large VLDL particles. Line graphs of all metabolites are shown in Appendix A, http://links.lww.com/EJA/A603.
patients. Whether changes in SFA_Fa levels precede the rise in circulating triglycerides remains an open question.

S-ketamine increased glucose and lactate. Lactate concentration was 53.8% higher than the corresponding concentration in placebo group at timepoint 2. This increase in lactate seems biologically significant, as the concentration of lactate at timepoint 2 in the S-ketamine group was 53 and 10.6% of the lactate levels observed during moderate and high-intensity exercise, respectively. A hyperadrenergic state caused by S-ketamine offers a possible explanation for the observed increase in lactate and glucose. The gradual decrease in tyrosine may also be explained by the hyperadrenergic state induced by S-ketamine, as tyrosine is funneled to noradrenaline synthesis. Interestingly, previous studies have shown that subanaesthetic doses of racemic ketamine increased cerebral blood flow (CBF) and regionally elevated cerebral glucose metabolic rate (GMR) without affecting the cerebral metabolic rate of oxygen (CMRO₂). Anaesthetic doses of S-ketamine increased GMR in the thalamus, with only minor alterations in the CMRO₂. Increased GMR in the absence of increased CMRO₂ suggested a role of nonoxidative glucose metabolism in the CNS during ketamine infusion, possibly related to a ketamine-induced increase in glutamate cycling. However, on the basis of current findings, it is not possible to identify the origins of the observed lactate release. The moderate increase in circulating lactate during isolated S-ketamine administration is nevertheless an interesting finding. Whether S-ketamine-induced lactataemia exaggerates lactataemia during critical illness in comparison to other anaesthetics is worth investigating.

S-ketamine decreased the levels of branched chain amino acids (BCAA) isoleucine, leucine and valine. The decrease in the aromatic amino acid (AAA) tyrosine reached statistical significance after the cessation of anaesthetic administration. At timepoint 3, the concentrations of isoleucine, leucine and tyrosine were 12.1, 2.5 and 7.4% lower in the S-ketamine group as compared with the placebo group. To offer a biological framework for these changes, a 35-min running exercise in healthy individuals decreased isoleucine by 9.0%, leucine 10.3%, valine 6.7% and tyrosine 4%, and the decrease in tyrosine was nonsignificant. In this respect, the magnitude of changes in BCAA may have biological significance. It seems tempting to consider that reduction of BCAA could be linked to increased muscle activity. However, the energy demands of working skeletal muscle during a 35-min running exercise probably outweigh an adrenergic stimulus caused by S-ketamine at rest. Thus, the changes in muscle cell bioenergetics in response to adrenergic stimuli at rest seem insufficient to account for the observed decrease in BCAA levels. Changes in BCAA concentrations are especially interesting, as S-ketamine has been approved for the treatment of “treatment-resistant” depression. BCAAs decrease brain uptake of AAAs due to a shared competitive transport mechanism across the blood-brain barrier. AAA tryptophan is a precursor of serotonin while AAA tyrosine, via transformation to dopamine, serves as a precursor for noradrenaline synthesis. Increases in circulating BCAAs lead to decreases in serotonin and catecholamine synthesis and release in the brain. Thus, decreases in circulating BCAAs could, in theory, increase the effects of serotonin and noradrenaline in the brain, providing a plausible mechanism for the antidepressant effects of S-ketamine. It is noteworthy, that the mechanism of action of commonly used antidepressants rely on selective serotonin and serotonin-noradrenaline reuptake inhibition. In addition, an earlier study revealed that the reduction in symptoms of major depressive disorder induced by sertraline was associated with decreases in BCAAs valine, leucine and isoleucine in serum samples. Therefore, the current results may help explain the underlying mechanisms of the antidepressant effects of S-ketamine.

On the basis of the current results, the most inert anaesthetic seems to be sevoflurane. Observed increases in motor restlessness may explain the increase in alanine concentration. Sevoflurane sedation in ICU has been suggested as a viable alternative to propofol or midazolam sedation. The current findings on the relative inertness of sevoflurane during short-term administration is a promising finding, which could prove useful in vulnerable patients with critical illness. However, the effects of prolonged sevoflurane sedation on the human metabolome remain unknown.

A few limitations must be addressed. First, the doses were moderate and the duration of anaesthetic exposure relatively short when compared with clinical anaesthesia. Second, only healthy male ASA I volunteers were studied, without the accompanying medications or comorbidities generally present with general anaesthesia. However, our aim was to discover and compare the direct effects of anaesthetic agents on the metabolic profile without the confounding factors usually present during the peri-operative period. This compromise leads to a relatively pure and comparable depiction of metabolic response to specific anaesthetics, yet limits our ability to offer clinically relevant predictions based on metabolite change. Further pragmatic studies are needed to assess the predictive value of metabolic changes associated with specific anaesthetics. Third, on the basis of the current results, we are unable to differentiate the effects of propofol from those of the accompanying lipid emulsion, as there was no control group for isolated lipid emulsion. However, as propofol is exclusively administered in lipid emulsion, the combined metabolic effects are of interest. Lastly, the metabolites that are closely related and share metabolic routes, are to some extent cross-correlated; this
has been illustrated in the heatmap in Appendix C, http://links.lww.com/EJA/A605.

In conclusion, this exploratory study revealed that, in absence of peri-operative confounding factors, a 1-h exposure to moderate doses of four routinely used anaesthetics induced unique alterations in the metabolic profile in healthy individuals as measured by NMR spectroscopy. Dexmedetomidine induced metabolic changes mirroring α2-adrenoceptor agonism and the inhibition of insulin secretion. Propofol emulsion influenced the lipid profile and elevated slightly the inflammatory marker GlycA. The inertness of sevoflurane might prove useful in vulnerable patients. Changes in amino acid profiles could be linked to the suggested antidepressant properties of S-ketamine. A need for further pragmatic studies on the clinical impact of the suggested antidepressant properties of S-ketamine. A need for further pragmatic studies on the clinical impact of the suggested antidepressant properties of S-ketamine.

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