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## Butyrylcholinesterase as a perioperative complication marker in patients after transcatheter aortic valve implantation?

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3 **Butyrylcholinesterase as a perioperative complication marker in patients after transcatheter aortic**  
4 **valve implantation?**  
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## Abstract

*Objectives:* Transcatheter aortic valve implantation (TAVI) is performed in elderly patients with severe aortic valve stenosis and increased operative risks. We tested the hypothesis that acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) have a predictive value for prevalent complications after TAVI and could serve as indicators of systemic inflammation in the early postoperative period.

*Design:* Prospective observational study.

*Setting:* This study is a secondary analysis of multi-center CESARO-study.

*Participants:* 48 TAVI patients were included, 43 obtained the complete assessment.

*Primary and secondary outcome measures:* Patients clinical parameters, demographic data, peripheral AChE and BChE activities and routine blood markers were assessed throughout the perioperative period using bedside point-of-care measurements for AChE and BChE. Postoperative complications screening was conducted up to the 3<sup>rd</sup> postoperative day and included infections, delirium and heart-rhythm disturbances. After assessment the patients were divided into complication and non-complication group.

*Results:* Of 43 patients, 24 developed postsurgical complications (55.8%). Preoperative assessment showed no significant differences regarding demographic data and laboratory markers, but preoperative BChE levels were significantly lower in patients who developed postoperative complications (complication group  $2589.2 \pm 556.4$  vs. non-complication group  $3295.7 \pm 628.0$ ,  $p < 0.05$ ). In complication group we observed an early, sustained reduction in BChE activity from preoperative to postoperative period. In complication group BChE levels were significantly lower at each time point compared to non-complication group. AChE activity showed no significant difference between both groups. Complication group also had longer stay in hospital overall.

*Conclusion:* BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative complications after TAVI. By using point-of-care measurements the levels of BChE are fast available and can lead to an early targeted therapy. Predicting the length of the hospital stay might play an important role in staff and resources management for these patients.

*Trial registration:* NCT01964284

**Key words:** cardiac surgery, TAVI, inflammation, delirium, butyrylcholinesterase, acetylcholinesterase

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3 Strengths and limitations of this study  
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5 This study is a secondary analysis of the prospective observational multi-centre CESARO-study.  
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7 Our study included 48 cardiosurgical patients with an observation time of three days.  
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9 BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative  
10 complications after TAVI.  
11

12 By using point-of-care measurements the levels of BChE are fast available and can lead to an early  
13 targeted therapy.  
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15 Predicting the length of the hospital stay might play an important role in staff and resources  
16 management for these patients.  
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## Introduction:

Recently, transcatheter aortic valve implantation (TAVI) has become the therapeutic standard for medical treatment in elderly, multimorbid patients with severe aortic valve stenosis and increased operative risks (1, 2). TAVI involves the implantation of a prosthetic valve, which is introduced with a catheter through transfemoral (TF), transapical (TA) or direct transaortic access. Usually, the TF approach is preferred, because thoracotomy and penetration of the myocardium are not needed. The TA approach is common, if severe atherosclerotic disease does not allow retrograde insertion of the catheter. In patients with severe aortic stenosis, who could not undergo a surgical replacement of the aortic valve, TAVI significantly reduced the rates of death at any cause, compared to standard therapy (3). However, previous studies have shown that pneumonia, acute renal failure, indication for a permanent pacemaker and delirium were the most frequently recorded complications after TAVI (4). Covello et. al. reported a pneumonia rate of 7-8 % after TAVI (5). The incidence of delirium after TAVI is reported as 29 % in literature (6).

Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) are a focus of current research. Recent studies have shown that AChE and BChE serve as diagnostic markers of low-grade systemic inflammation (7–9). Rapid changes in cholinesterase activity have also been reported in patients after acute trauma, infections, burns and critical illness (10–14). Both enzymes may serve as indicators of systemic inflammation and have remarkable predictive value for mortality in critically ill patients. Zivkovic et. al. showed that reduced serum activity of BChE indicates severe systemic inflammation in critically ill patients (13). Furthermore, a recent study showed, that a sustained reduction in serum cholinesterase enzyme activity predicts patient outcome following sepsis (15).

Other studies postulate low preoperative plasma cholinesterase activity as a risk marker of postoperative delirium in elderly patients (16). A recently published study on cholinesterase activity in cardiac surgical patients showed no postoperative differences in cholinesterase activities between delirious and non-delirious patients, but showed a perioperative decrease of BChE which was potentially caused by cardiopulmonary bypass (17). However, due to high variability in the etiology and progress of clinical conditions, it was difficult to determine whether the changes in the enzyme activity correlated with the emergence of disease or was affected by concomitant factors such as cardiopulmonary bypass.

This is the first study to investigate the roles of AChE and BChE as inflammatory markers in cardiac surgical patients under standardized perioperative conditions without using cardiopulmonary bypass. Our aim of the present study is to evaluate if there is a predictive association of perioperative determination of AChE and BChE activity and the occurrence of postoperative complications after TAVI.

## Material and Methods:

### *Study design and patient population:*

This work is a secondary analysis of the prospective observational multi-center CESARO-study, powered for the detection of postoperative delirium. The CESARO study was initiated at Charité – Universitätsmedizin Berlin, Department of Anesthesiology and Operative Intensive Care Medicine (Clinicaltrials.gov ID: NCT01964284) and approved by the local independent Charité Ethics Committee, Charité – Universitätsmedizin Berlin, Germany (ref.: EA1/220/13). After further approval of the local ethics board of the University of Regensburg a total of 48 patients were included into the study between March 2014, and June 2016 at University Hospital of Regensburg. Written informed consent was obtained from each patient (Figure 1).

Inclusions criteria: minimum age of 18 years, admission to intensive care unit (ICU) following elective TAVI in general anesthesia.

Exclusion criteria: missing consent, patients with a known pseudocholinesterase deficiency, patients with language, visual or hearing impairments.

### *Preoperative variables:*

Preoperative data included demographic data, such as age, sex, height, weight, regular use of alcohol and nicotine, American Society of Anesthesiologists (ASA) class, logistic Euro Score (European System for Cardiac Operative Risk Evaluation), New York Heart Association (NYHA) class, long-term medication and left ventricular ejection fraction (EF). The patients' previous medical history was examined for conditions such as chronic kidney disease, cerebrovascular events, including stroke and transient ischemic attacks, myocardial infarction, chronic obstructive pulmonary disease, diabetes mellitus and pre-existing cardiac arrhythmias. Every patient was screened for preoperative delirium, using the nursing delirium screening scale (NU-DESC). Preoperative assessment of AChE, BChE, CRP, leukocytes, haemoglobin and creatinine were performed (table 1).

### *Intraoperative variables:*

Key elements of intraoperative data included the selected access type, anesthetic procedure, transfusion of erythrocyte concentrates and extubation rate as well as the procedure duration.

### *Postoperative variables:*

Postoperative data included the patient's stay in the ICU and the stay in hospital in general. Next to the sampling of laboratory markers, every patient was screened for delirium with NU-DESC for the first 3 days after surgery. Patients were daily assessed for pain, using the numeric rating scale (NRS score: 0 = no pain – 10 = maximum pain). Furthermore, any complication in recovery time was noticed. Mortality reasons are divided into cardiac, acute kidney injury, cardiovascular events and infections.

Variables:

*Delirium:*



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3 Delirium screening was conducted perioperatively using a validated screening tool (NU-DESC) (18). NU-  
4 DESC assesses five dimensions: orientation, behaviour, communication, illusion/hallucination and  
5 psychomotor retardation. The symptoms are rated on a three-point scale, whereas a score of two or  
6 more cumulative points indicated delirium. Delirium assessment was performed one day prior to the  
7 operation, on admission to ICU and daily up to the third postoperative day. Patients with Richmond  
8 Agitation Sedation Scale (RASS)  $\leq -2$  were excluded for the current testing.  
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#### 11 12 13 *Laboratory parameters:*

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15 Blood samples were taken from every patient at following points: one day before operation  
16 (screening), shortly before anesthetic induction, on admission to ICU, one day after surgery and two  
17 days after surgery (Figure 2). The measurements included the determination of AChE and BChE. Both  
18 were measured in 10  $\mu$ l whole blood, using *ChE check mobile*, a point-of-care testing device (ChE check  
19 mobile®, Securetec Detektions-Systeme AG, Neubiberg, Germany) by following the manufacturer's  
20 instructions. Also, blood count, C-reactive protein (CRP), creatinine were measured at each time point.  
21 Creatine kinase (CK) and heart enzymes (CK-MB) were measured on the first postoperative day in the  
22 normal laboratory control. Brain natriuretic Peptide (NT-proBNP) was measured at the screening day.  
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#### 27 *Postoperative complications:*

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29 Since delirium, pneumonia, heart rhythm disturbances and acute renal failure are the most frequently  
30 reported complications after TAVI, we have screened all patients until the discharge of the hospital.  
31 Infection was defined as an increase in CRP, fever and diagnosed infection-focus (pneumonia, urinary  
32 tract infection, other infections). Delirium was diagnosed by using NU-DESC. Postoperative heart  
33 rhythm disturbances occurred by AV-block and atrial fibrillation. Patients were divided into two  
34 groups: those who did not develop any postoperative complications (non-complication group) and  
35 patients who showed one of these complications within 3 days after TAVI (complication group).  
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#### 40 *Operation procedure:*

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42 All patients were admitted and evaluated at least one day before the operation. TAVIs were performed  
43 by the cardiac team (cardiac surgeon, cardiologist, and cardiac anesthesiologist) in a hybrid operating  
44 theatre. All procedures were performed with the patients placed under general anesthesia. In all  
45 patients, monitoring consisted of pulse oximetry, 5-channel electrocardiogram, invasive blood pressure,  
46 central venous pressure, urinary output and bladder temperature. The maintenance of normothermia  
47 was accomplished by a heating blanket placed beneath the patient. The patients received right  
48 ventricular pacemakers for rapid ventricular pacing during balloon aortic valvuloplasty and valve-  
49 expansion. Pre-oxygenation was performed with pure oxygen using a facemask. Anesthesia was  
50 induced with etomidate (Etomidat-Lipuro®, B. Braun Melsungen AG, Melsungen, Germany),  
51 remifentanyl (Ultiva®, GlaxoSmithKline GmbH & Co. KG, Munich, Germany) and rocuronium  
52 (Rocuronium Inresa®, Inresa Arzneimittel GmbH, Freiburg, Germany) and maintained with sevoflurane  
53 (Sevorane®, AbbVie Deutschland GmbH & Co. KG, Wiesbaden, Germany). Piritramide and metamizole  
54 were used as additional pain medication. PONV prophylaxis was used intraoperatively, depending on  
55 the patient's risk. Cardiovascular drugs (e.g. norepinephrine, and dobutamine) were administered, as  
56 needed. A prophylactic antibiotic (1.5 g, Cefuroxim Hikma®, Hikma Pharma GmbH, Gräfelfing,  
57 Germany) was administered to each patient. In the operating theatre, the patient was connected to a  
58 defibrillator, and a TEE probe was introduced. After preparing the access points and anticoagulation  
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3 with heparin (Ratiopharm GmbH, Ulm, Germany; mean given dose  $5293 \pm 2643$  IU), the native valve  
4 was opened under rapid ventricular pacing, and the prothesis was implanted. The position and  
5 function of the prothesis was verified with TEE. Extubation of the patient was the goal at the end of  
6 each procedure. After surgery, patients were monitored for at least 12 hours in the ICU. Following this  
7 period, patient care continued either in the ICU or in the general ward. There was no use of heart lung  
8 machines.  
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### 10 11 12 13 *Patient and public involvement*

14 Patients were not involved in the study.  
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### 17 18 19 *Statistics:*

20 The data were electronically gathered and stored by using Excel. Data analysis was performed by using  
21 SPSS (Version 22.0; SPSS Inc., Chicago, IL, USA). Data are presented as mean with standard deviation.  
22 Shapiro-Wilk test was used to verify Gaussian distribution of the study groups. Statistical significance  
23 between the patient groups was tested using t-test, Mann-Whitney U test, analysis of variance and  
24 chi-quadrat. A p value < 0.05 indicated statistical significance.  
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## Results:

### *Baseline data*

A total of 48 patients were included, and 43 patients completed the assessment battery (figure 1). The mean age was 79.5 +/- 5.71 years and the mean BMI was 27.93 +/- 5.36. There were almost equally men and women (22 (51.2 %) vs. 21 (48.8 %), table 1). All patients received elective TAVI in general anesthesia. TF access was selected for 32 (74 %), with TA chosen for 11 (26 %) patients. There was no use of heart-lung-machines. The demographic data and pre-existing conditions are shown in table 1. 32 (74.4 %) had an ASA-class of three, 11 (25.6 %) of four. Except of four, every patient was extubated immediately after operation and brought to ICU. One high risk patient was still intubated when brought to ICU and died two days after operation by multiorgan failure. Another patient was extubated on the first postoperative day. Two patients were extubated a few hours after brought to ICU. Patients were discharged to a normal ward after one day and left the hospital after 13.28 +/- 6.2 days.

### *Postoperative complications*

24 patients (55.8 %) had postoperative complications as defined above. One multimorbid and high-risk patient died due to multiorgan-failure at ICU two days after surgery.

Of 43 patients, 12 developed postoperative delirium (27.9 %). Most patients developed their delirium on the first day after surgery.

Of 43 patients, 2 developed pneumonia. However, in 3 patients with raised infection markers and suspected infection no focus was found. All of them received antibiotics.

There were 7 patients with postoperative indication for pacemaker (16.3%). Overall 12 patients developed heart rhythm disturbances (27.9%). Some of the patients developed more than one complication, e.g. delirium or infection.

### *Comparison between complication and non-complication group*

#### Preoperative variables

Preoperative assessment showed no significant differences regarding demographic data and laboratory routine markers like haemoglobin, leukocytes, CRP, NT-proBNP and creatinine ( $p = n. s.$ ). Preoperative BChE levels were significantly lower in patients who developed postoperative complications (D 1 complication group  $2589.2 \pm 556.4$  vs. D 1 non-complication group  $3295.7 \pm 628.0$   $p < 0.05$ , table 2). Preoperative AChE enzyme activity in contrast did not show any difference between complication and non-complication group. There was also no difference regarding alcohol ( $p = 0.23$ ) or nicotine ( $p = 0.8$ ) consumption. Men showed a higher incidence of postoperative complications ( $p = 0.09$ ).

#### Postoperative variables

All patients were postoperatively admitted to the ICU extubated and hemodynamic supported by catecholamines. Two patients did not meet the extubation criteria in the operation room and were extubated a few hours later at ICU. One high risk patient died at ICU due to multiorgan-failure. One patient was extubated on the first postoperative day.

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Complication group showed an early, sustained and statistically significant decrease in BChE activity from the preoperative to the first postoperative measurement (D 0:  $2784.0 \pm 534.9$  vs POD 0:  $2379.6 \pm 525.1$ ,  $p < 0.05$ , figure 3). In contrast in patients without postoperative complications we observed a delayed decrease in BChE activity (D 0:  $3072.6 \pm 652.1$  vs POD 2:  $2713.5 \pm 510.6$ ,  $p < 0.05$ ). In all time points a significantly lower BChE activity was observed in patients with complications compared to patients without postoperative complication (figure 3).

Both groups showed a moderate decrease in AChE activity after preoperative screening measurement (figure 4). From anesthesia induction to the second postoperative measurement we observed no significant changes in AChE activity over time in both groups. There were no significant differences in AChE activity between patients with and without complication in any time point (figure 4).

Patients, who developed postoperative complications had a significantly longer stay in hospital in general (complication-group:  $15.2 \pm 6.3$  vs. non-complication-group:  $11.1 \pm 5.5$  days,  $p < 0.05$ ). There was no difference regarding the stay on ICU (complication group vs. non-complication group  $p = n.s.$ ) Complication-group also showed a higher Nu-DESC score on the first postoperative day (complication-group:  $2.1 \pm 2.4$  vs. non-complication group:  $0.58 \pm 0.51$ ,  $p < 0.05$ ). Patients with postoperative delirium showed highest NU-DESC score on the first postoperative day (delirium:  $3.3 \pm 2.6$  vs. non-delirium:  $0.27 \pm 0.79$ ). The preoperative score of NU-DESC was  $0.42 \pm 0.67$  within patients, who developed postoperative delirium. Routine laboratory markers like haemoglobin, leukocytes, CRP, CK, CK-MB and creatinine did not show any difference (complication group vs. non-complication group  $p = n. s.$ , table 2).

Furthermore, there was no difference in Euroscore regarding on complication (complication group vs. non-complication group  $p = n. s.$ , table 1)).

#### TA vs. TF

Patients, who underwent TA approach declared postoperative higher pain levels measured by NRS ( $p < 0.05$ ). They also showed higher CRP levels on POD 2 ( $88.8 \pm 44.5$  vs.  $161.6 \pm 70.2$ ,  $p < 0.05$ , table 2) and higher levels of CK ( $110.8 \pm 134.5$  vs.  $398.7 \pm 139.0$ ,  $p < 0.05$ ) and CK-MB ( $8.3 \pm 11.8$  vs.  $29.8 \pm 14.7$ ,  $p < 0.05$ ) on the first postoperative day. There were no further differences between patients with TF and TA approach, especially regarding on complications or BChE and AChE enzyme levels.

### Discussion:

TAVI has become the therapeutic standard for medical treatment in elderly patients with severe aortic valve stenosis and increased operative risks. The primary objective of the present investigation was to evaluate the roles of AChE and BChE as predictive markers for prevalent complications in cardiosurgical patients after TAVI.

Previous studies assumed an interaction of the immune and cholinergic system (19) and identified AChE and BChE as useful biomarkers for early detection of patients with emerging inflammation (16). Rapid changes in cholinesterase activity have been reported in patients after acute trauma, infections, delirium and critical illness (10–14). Both enzymes may serve as indicators of systemic inflammation and have a remarkable predictive value for mortality in critically ill patients. Zivkovic et al. showed that bedside-measurement of BChE activity predicts patient morbidity and length of ICU stay following major traumatic injury (20). Another study with patients undergoing venoarterial extracorporeal membrane oxygenation therapy after cardiac surgery revealed BChE as a strong predictor of all-cause and cardiovascular mortality (10).

In our present study patients with postoperative complications after TAVI had significant lower preoperative levels of compared to the non-complication group. This finding suggests that BChE activity, used in combination with common preoperative evaluation procedures, could serve as a useful predictive indicator to identify high-risk patients.

Due to high variability in the onset, aetiology and progress of clinical conditions among patients, determining whether changes in the enzyme activity are correlated with the emergence of disease or are affected by concomitant factors is difficult. John et al. tested the hypothesis that AChE and BChE have an impact on patients after cardiac surgery with postoperative delirium. They showed that AChE increased and BChE decreased within the first 3 days after surgery but did not discern between patients with and without delirium. The authors supposed that the perioperative change of AChE and BChE activity might possibly be explained by an interaction of AChE and BChE and the use of a cardiopulmonary bypass (17). In our present study we evaluated the role of AChE and BChE activity in cardiosurgical patients after TAVI, as a standardized operative procedure. We could show that complication group shows a significantly perioperative decrease of BChE within the first 3 days after TAVI, despite the fact that there was no use of heart-lung machines in our patients. Furthermore, there was no use of blood products in the present study, so we can rule out a possible interaction of AChE and BChE with blood products as well.

The pathophysiologic mechanism behind the BChE activity change and the systemic inflammation presumably involves the non-neuronal anti-inflammatory activity of the cholinergic system. Conventional markers like CRP and leucocytes did not differ in both groups.

Delirium is a complex symptom which is very common in operative and non-operative disciplines in the course of hospital stay. The incidence is especially high among patients undergoing heart surgery (21). The incidence in this patients population has been described to be from 30 up to 80 % (22). The incidence of delirium after TAVI is reported as 29 % in literature (6). Delirium occurred significantly more frequently following TA procedures (23). In the present study 26,7 % of the patients were diagnosed with delirium overall. There was no difference depending on TA or TF approach. Perioperative measurement of AChE and BChE did not discern between patients with and without delirium, which is in accordance with the findings by John et al.

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3 The present study highlights the validity of BChE measurements for early detection of high-risk patients  
4 after TAVI. Surprisingly, the BChE assessment proved more effective than the EURO-Score in  
5 discriminating between the patient groups making it a valuable biomarker for the early detection of  
6 high-risk patients. Euro-Score is a well-established clinical assay for the patient mortality analysis-(24)  
7 and requires documenting multiple and diverse datasets. The datasets are in most cases readily  
8 available; however, in some cases, a particular set of data might not be accessible, delaying or making  
9 the scoring impossible. By using a POCT system for a single BChE measurement, the results of an  
10 equally efficient outcome assessment tool are readily available at the bedside.  
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13 Prompt assessment of the systemic immune response with an immediate, rapid and affordable  
14 bedside measurement of the BChE activity might improve risk evaluation and help optimize  
15 postoperative management and therapy of patients after TAVI. Predicting the length of the hospital  
16 stay might play an important role in staff and resources management for these patients.  
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### 23 Limitations

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26 Limitations of the present study might be the short duration of 3 days' measurement. Blood was taken  
27 from each patient; in case the analysis could not be performed immediately (during anesthesia  
28 induction), the sample was cooled down in a refrigerator. Maybe values of AChE and BChE changed in  
29 combination with lower temperature. Furthermore, it was only one measurement performed with  
30 each sample, so no control values could be achieved.  
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33 A further limitation of this study is the low number of included patients. However, even with the low  
34 sample, the described test demonstrated high sensitivity, particularly in the initial time period, as  
35 compared to the benchmark methods, suggesting a rapid, effective, and simple patient outcome assay.  
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38 There was no comparison between the aetiology of inflammation. A larger, possibly multicentre study  
39 would be needed to validate our findings.  
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<b>Characteristic</b>	<b>Total sample (n = 43)</b>
<b>Age (years) [M (SD)]</b>	79.47 (5.71)
<b>Sex [n (%)]</b>	
Male	22 (51.2)
Female	21 (48.8)
<b>BMI [M (SD)]</b>	27.93 (5.36)
<b>ASA – PS [n (%)]</b>	
3	32 (74.4)
4	11 (25.6)
<b>Operative procedure [n (%)]</b>	
Transapicale TAVI	11 (25.6)
Transfemorale TAVI	32 (74.4)
<b>Relevant comorbidities [n (%)]</b>	
hypertension	37 (86)
diabetes	16 (37.2)
Congestive heart failure	36 (83.7)
Congestive kidney failure	20 (46.5)
Coronary heart disease	31 (72.1)
Cardiac arrhythmias	25 (58.1)
Stroke/TIA	8 (18.6)
nicotine	19 (44.2)
alcohol	8 (18.6)
hypothyreosis	10 (23.3)
hypercholesterinemia	14 (32.6)
<b>Euroscore [n (%)]</b>	
low	15 (34.9)
middle	18 (41.9)
high	10 (23.3)

Table 1 Description of baseline data

Variables	<u>complication</u>	N	M	SD	p-value
Euroscore	Yes	24	21.8	15.4	0.82
	no	19	22.8	13.5	
Weight [kg]	Yes	24	78.3	15.5	0.50
	no	19	74.9	18.0	
BMI [kg/m <sup>2</sup> ]	Yes	24	28.1	4.7	0.86
	no	19	27.7	6.2	
Age [years]	Yes	24	79.9	5.3	0.57
	no	19	78.9	6.3	
NT-proBNP [pg/ml]	Yes	24	6244.8	6773.1	0.56
	no	19	4806.6	7809.4	
Hemoglobin D 0 [g/dl]	Yes	24	12.1	1.9	0.92
	no	19	12.2	1.4	
Hemoglobin POD 0 [g/dl]	Yes	24	10.7	1.5	0.57
	no	19	10.9	1.4	
Hemoglobin POD 1 [g/dl]	Yes	24	10.1	1.4	0.99
	no	19	10.1	1.1	
Hemoglobin POD 2 [g/dl]	Yes	24	10.1	1.1	0.67
	No	19	10.3	1.2	
Hemoglobin POD 3 [g/dl]	Yes	24	9.6	1.0	0.27
	No	19	10.0	1.3	
Creatinine D 0 [mg/dl]	Yes	24	1.4	0.7	<b>0.09</b>
	no	19	1.1	0.4	
Creatinine POD 1 [mg/dl]	Yes	24	1.2	0.4	0.35
	No	19	1.1	0.4	
Creatinine POD 2 [mg/dl]	Yes	24	1.5	0.8	0.19
	no	19	1.2	0.4	
Creatinine POD 3 [mg/dl]	Yes	24	1.5	1.0	0.24
	No	19	1.1	0.4	
Leukocytes D 0 [/nl]	Yes	24	7.8	2.1	0.38
	no	19	7.3	1.6	
Leucocytes POD 1 [/nl]	Yes	24	9.6	4.3	0.50
	No	19	8.8	2.1	
Leukocytes POD 2 [/nl]	Yes	24	9.9	2.7	0.62
	No	19	9.4	3.5	
Leukocytes POD 3 [/nl]	Yes	24	8.8	3.1	<b>0.08</b>
	No	19	7.0	1.5	
CRP D 0 [mg/l]	Yes	24	<b>16.3</b>	17.8	0.26
	no	19	<b>9.7</b>	11.9	
CRP POD 1 [mg/l]	Yes	24	31.8	21.1	0.18
	No	19	22.0	18.9	
CRP POD 2 [mg/l]	Yes	24	116.3	52.9	0.52
	No	19	101.3	78.2	
CRP POD 3 [mg/l]	Yes	24	115.3	68,5	0.11
	No	19	72.7	76,0	
BChE D 0 [U/l]	Yes	24	2784.0	534,9	0.12
	no	19	3072.6	652,1	
BChE D 1 [U/l]	Yes	24	2589.2	556,4	<b>0.001</b>
	no	19	3295.7	628,0	



BChE POD 0 [U/l]	Yes No	24 19	2379.6 2972.5	525.1. 599.2	<b>0.001</b>
BChE POD 1 [U/l]	Yes No	24 19	2300.3 2936.2	561.0 523.1	<b>&lt; 0.005</b>
BChE POD 2 [U/l]	Yes No	24 19	2166.7 2713.5	537.0 510.6	<b>0.002</b>
AChE D 0 [U/gHb]	Yes no	24 19	45.0 43.3	8.1 6.0	0.45
AChE D 1 [U/gHb]	Yes no	24 19	42.0 39.2	10.0 4.7	0.26
AChE POD 0 [U/gHb]	Yes No	24 19	42.9 37.6	10.0 6.4	<b>0.051</b>
AChE POD 1 [U/gHb]	Yes No	24 19	41.5 38.4	9.2 5.2	0.2
AChE POD 2 [U/gHb]	Yes No	24 19	41.2 36.5	8.2 6.8	<b>0.058</b>
CK POD 1 [U/l]	Yes No	24 19	189.6 186.1	186.8 190.5	0.95
CK-MB POD 1 [ng/ml]	Yes No	24 19	15.5 15.9	18.2 14.3	0.95
CK-Index POD 1	Yes No	24 19	7.2 8.5	3.6 3.8	0.31
Stay in hospital [days]	Yes no	24 19	15.2 11.1	6.3 5.5	<b>0.03</b>

Table 2 Perioperative laboratory markers

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### **AVAILABILITY OF DATA AND MATERIAL**

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

### **CONFLICT OF INTERESTS**

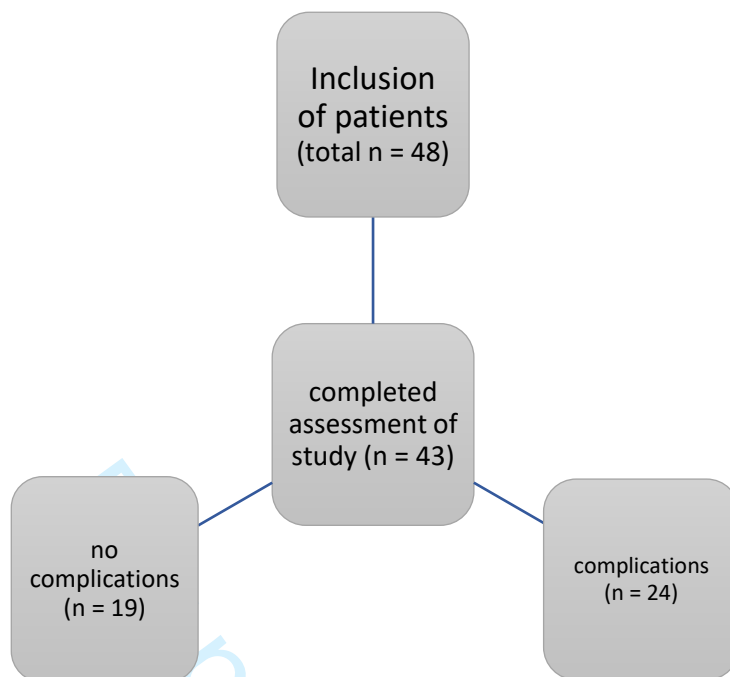
The authors declare no conflict of interests.

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### **CONTRIBUTIONS**

DB, YZ, AM and BM were responsible for study design, statistical analyses and drafting of the manuscript. WP, AB, BM and BG performed the experiments and drafted the manuscript. BM and AM were responsible for statistical analysis. All authors read and approved the final manuscript.



*Figure 1 Study design: a total of 48 patients were screened. 5 patients were not included. 43 patients completed the assessment of the study. 19 patients showed no complications. 24 patients developed postoperative complications.*

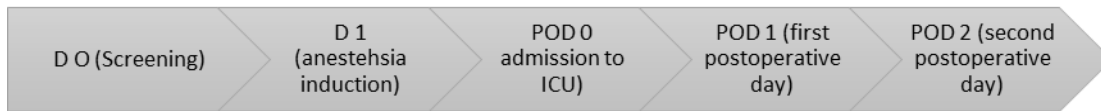


Figure 1 Timeline of measurements of BChE and AChE: blood samples were taken one day preoperative (D O), shortly before anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery (POD 1) and two days after surgery (POD 2). If the measurements could not be conducted immediately, the samples have been cooled in a refrigerator and the measurement was performed up to 2 hours later.

For peer review only

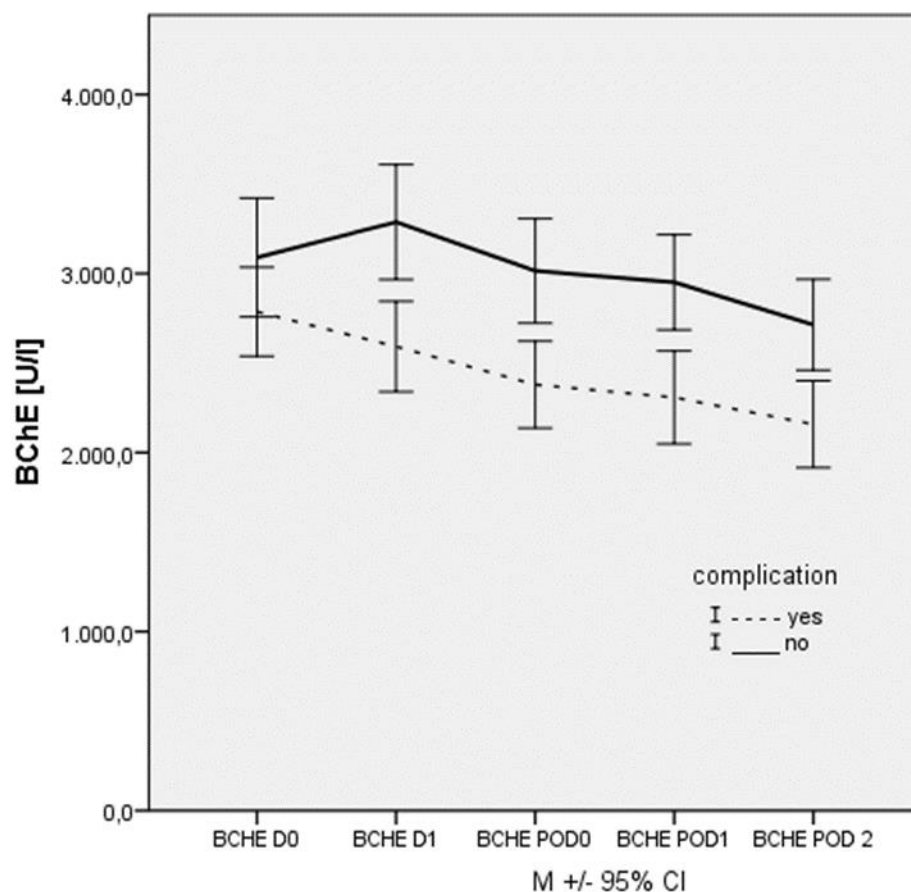


Figure 3 Perioperative levels of BChE activity in complication and non-complication group: Complication group showed an early, sustained and statistically significant decrease in BChE activity from the preoperative to the first postoperative measurement (analysis of variance; D 0:  $2784.0 \pm 534.9$  vs POD 0:  $2379.6 \pm 525.1$ ,  $p < 0.05$ ). Patients without postoperative complications showed a delayed decrease in BChE activity (analysis of variance D 0:  $3072.6 \pm 652.1$  vs POD 2:  $2713.5 \pm 510.6$ ,  $p < 0.05$ ). In all time points a significantly lower BChE activity was observed in patients with complications compared to patients without postoperative complication (t-test).

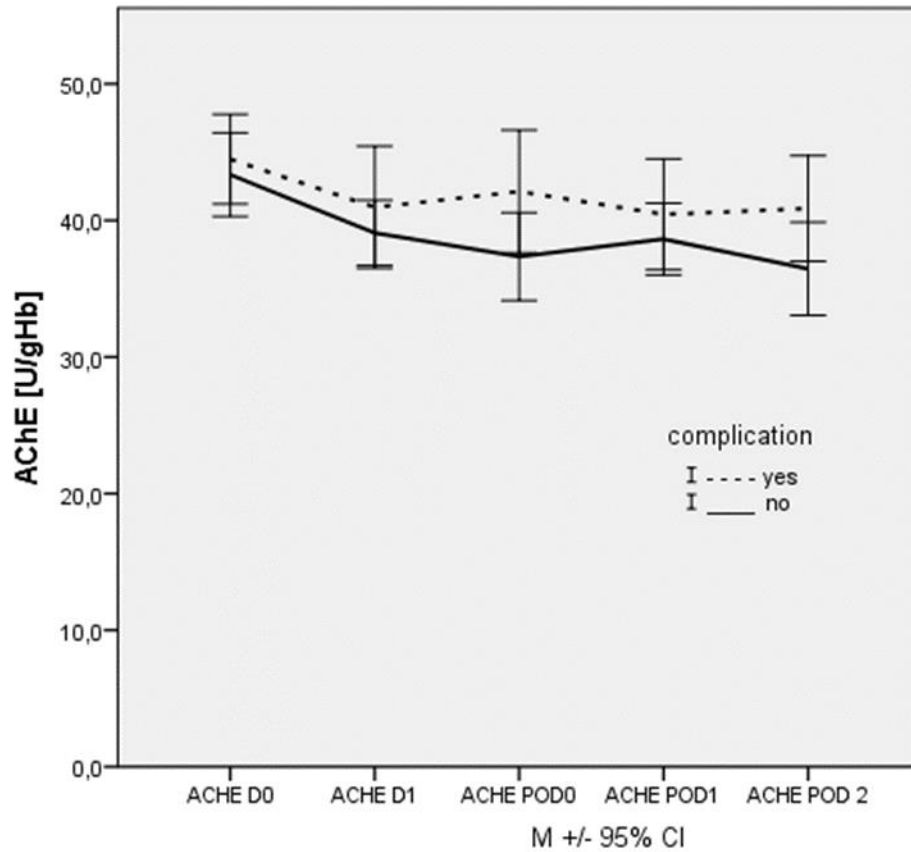


Figure 4 Perioperative levels of AChE activity of complication and non-complication group: From anesthesia induction to the second postoperative measurement we observed no significant changes in AChE activity over time in both groups (analysis of variance,  $p = n.s.$ ). There were no significant differences in AChE activity between patients with and without complication in any time point (t-test,  $p = n.s.$ ). However, analysis of variance showed a significantly decrease over time referred to the first measurement on D 0.



# BMJ Open

## Butyrylcholinesterase as a perioperative complication marker in patients after transcatheter aortic valve implantation: a prospective observational study

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Secondary Subject Heading:	Anaesthesia, Cardiovascular medicine
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3 **1 Butyrylcholinesterase as a perioperative complication marker in patients after**  
4 **2 transcatheter aortic valve implantation: a prospective observational study**  
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**Abstract**

*Objectives:* Transcatheter aortic valve implantation (TAVI) is performed in elderly patients with severe aortic valve stenosis and increased operative risks. We tested the hypothesis that acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) have a predictive value for prevalent complications after TAVI and could serve as indicators of systemic inflammation in the early postoperative period.

*Design:* Prospective observational study.

*Setting:* This study is a secondary analysis of multi-center CESARO-study.

*Participants:* 48 TAVI patients were included, 43 obtained the complete assessment.

*Primary and secondary outcome measures:* Patients clinical parameters, demographic data, peripheral AChE and BChE-activities and routine blood markers were assessed throughout the perioperative period using bedside point-of-care measurements for AChE and BChE. Postoperative complications screening was conducted up to the 3<sup>rd</sup> postoperative day and included infections, delirium and heart-rhythm disturbances. After assessment the patients were divided into complication and non-complication group.

*Results:* Of 43 patients, 24 developed postsurgical complications (55.8%). Preoperative assessment showed no significant differences regarding demographic data and laboratory markers, but preoperative BChE-levels were significantly lower in patients who developed postoperative complications (complication group  $2589.2 \pm 556.4$  vs. non-complication group  $3295.7 \pm 628.0$ , Cohen's  $r = 0.514$ ,  $p < 0.001$ ). In complication group we observed an early, sustained reduction in BChE-activity from preoperative to postoperative period. In complication group BChE-levels were significantly lower at each time point compared to non-complication group. AChE-activity showed no significant difference between both groups. Complication group also had longer stay in hospital overall.

*Conclusion:* BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative complications after TAVI. By using point-of-care measurements the levels of BChE are fast available and can lead to an early targeted therapy. Predicting the length of the hospital stay might play an important role in staff and resources management for these patients.

*Trial registration:* NCT01964274

**Key words:** cardiac surgery, TAVI, inflammation, delirium, butyrylcholinesterase, acetylcholinesterase

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3 59 Strengths and limitations of this study  
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5 60 This study is a secondary analysis of the prospective observational multi-center CESARO-study.  
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7 61 Our study included 48 cardiosurgical patients with an observation time of three days.  
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9 62 BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative  
10 63 complications after TAVI.

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12 64 By using point-of-care measurements the levels of BChE are fast available and can lead to an early  
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72 Introduction:

73 Recently, transcatheter aortic valve implantation (TAVI) has become the therapeutic standard for  
74 medical treatment in elderly, multimorbid patients with severe aortic valve stenosis and increased  
75 operative risks (1, 2). TAVI involves the implantation of a prosthetic valve, which is introduced with a  
76 catheter through transfemoral (TF), transapical (TA) or direct transaortic access. Usually, the TF  
77 approach is preferred because thoracotomy and penetration of the myocardium are not needed. The  
78 TA approach is common, if severe atherosclerotic disease does not allow retrograde insertion of the  
79 catheter. In patients with severe aortic stenosis, who could not undergo a surgical replacement of the  
80 aortic valve, TAVI significantly reduced the rates of death at any cause, compared to standard therapy  
81 (3). However, previous studies have shown that pneumonia, acute renal failure, indication for a  
82 permanent pacemaker and delirium were the most frequently recorded complications after TAVI (4).  
83 Covello et. al. reported a pneumonia rate of 7-8 % after TAVI (5). The incidence of delirium after TAVI  
84 is reported as 29 % in literature (6).

85 Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) are a focus of current research. Recent  
86 studies have shown that AChE and BChE serve as diagnostic markers of low-grade systemic  
87 inflammation (7–9). Rapid changes in cholinesterase activity have also been reported in patients after  
88 acute trauma, infections, burns and critical illness (10–14). Both enzymes may serve as indicators of  
89 systemic inflammation and have remarkable predictive value for mortality in critically ill patients.  
90 Zivkovic et. al. showed that reduced serum activity of BChE indicates severe systemic inflammation in  
91 critical ill patients (13). Furthermore, a recent study showed, that a sustained reduction in serum  
92 cholinesterase enzyme activity predicts patient outcome following sepsis (15).

93 Other studies postulate low preoperative plasma cholinesterase activity as a risk marker of  
94 postoperative delirium in elderly patients (16). A recently published study on cholinesterase activity in  
95 cardiac surgical patients showed no postoperative differences in cholinesterase activities between  
96 delirious and non-delirious patients, but showed a perioperative decrease of BChE which was  
97 potentially caused by cardiopulmonary bypass (17). However, due to high variability in the etiology  
98 and progress of clinical conditions, it was difficult to determine whether the changes in the enzyme  
99 activity correlated with the emergence of disease or was affected by concomitant factors such as  
100 cardiopulmonary bypass.

101 This is the first study to investigate the roles of AChE and BChE as inflammatory markers in cardiac  
102 surgical patients under standardized perioperative conditions without using cardiopulmonary bypass.  
103 Our aim of the present study is to evaluate if there is a predictive association of perioperative  
104 determination of AChE and BChE activity and the occurrence of postoperative complications after  
105 TAVI.

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3 106 Material and Methods:  
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5 107 *Study design and patient population:*  
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7 108 This work is a secondary analysis of the prospective observational multi-center CESARO study,  
8 109 powered for the detection of postoperative delirium. The CESARO study was initiated at Charité –  
9 110 Universitätsmedizin Berlin, Department of Anesthesiology and Operative Intensive Care Medicine  
10 111 (Clinicaltrials.gov ID: NCT01964274) and approved by the local independent Charité Ethics Committee,  
11 112 Charité – Universitätsmedizin Berlin, Germany (ref.: EA1/220/13) on 14 August 2013. After further  
12 113 approval of the local ethics board of the University of Regensburg a total of 48 patients were included  
13 114 into the study between March 2014, and June 2016 at University Hospital of Regensburg. Written  
14 115 informed consent was obtained from each patient.

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17 116 Inclusions criteria: minimum age of 18 years, admission to intensive care unit (ICU) following elective  
18 117 TAVI in general anesthesia.

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20 118 Exclusion criteria: missing consent, patients with a known pseudocholinesterase deficiency, patients  
21 119 with language, visual or hearing impairments.

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25 121 *Data*  
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27 122 Data were acquired from anesthetic charts (Medlinq V.1.3, Hamburg, Germany), the patient document  
28 123 system used in the ICU (Metavision, iMDsoft, Tel Aviv, Israel) and medical reports from the electronic  
29 124 hospital information system (SAP, Walldorf, Germany) from the preoperative, intraoperative and  
30 125 postoperative periods until the patients were discharged from the hospital.

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34 127 *Preoperative variables:*  
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36 128 Preoperative data included demographic data, such as age, sex, height, weight, regular use of alcohol  
37 129 and nicotine, American Society of Anesthesiologists (ASA) class, logistic EuroSCORE (European System  
38 130 for Cardiac Operative Risk Evaluation), New York Heart Association (NYHA) class and left ventricular  
39 131 ejection fraction (EF). The patients' previous medical history was examined for conditions such as  
40 132 chronic kidney disease, cerebrovascular events, including stroke and transient ischemic attacks,  
41 133 myocardial infarction, chronic obstructive pulmonary disease, diabetes mellitus and pre-existing  
42 134 cardiac arrhythmias. Every patient was screened for preoperative delirium, using the nursing delirium  
43 135 screening scale (NU-DESC). Preoperative assessment of AChE, BChE, CRP, leukocytes, haemoglobin and  
44 136 creatinine were performed (table 1).

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49 138 *Intraoperative variables:*  
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51 139 Key elements of intraoperative data included the selected access type, anesthetic procedure,  
52 140 transfusion of erythrocyte concentrates and extubation rate as well as the procedure duration.

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56 142 *Postoperative variables:*  
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58 143 Postoperative data included the patient's stay in the ICU and the stay in hospital in general. Next to  
59 144 the sampling of laboratory markers, every patient was screened for delirium with NU-DESC for the first  
60 145 3 days after surgery. Patients were daily assessed for pain, using the numeric rating scale (NRS score:

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3 146 0 = no pain – 10 = maximum pain). Furthermore, any complication in recovery time was noticed.  
4 147 Mortality reasons are divided into cardiac, acute kidney injury, cardiovascular events and infections.

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8 149 Variables:

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10 150 *Delirium:*

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12 151 Delirium screening was conducted perioperatively using a validated screening tool (NU-DESC) (18). NU-  
13 152 DESC assesses five dimensions: orientation, behaviour, communication, illusion/hallucination and  
14 153 psychomotor retardation. The symptoms are rated on a three-point scale, whereas a score of two or  
15 154 more cumulative points indicated delirium. Delirium assessment was performed one day prior to the  
16 155 operation, on admission to ICU and daily up to the third postoperative day. Patients with Richmond  
17 156 Agitation Sedation Scale (RASS)  $\leq -2$  were excluded for the current testing.

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21 158 *Laboratory parameters:*

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23 159 Blood samples were taken from every patient at following time points: one day before operation  
24 160 (screening), shortly before anesthetic induction, on admission to ICU, one day after surgery and two  
25 161 days after surgery (figure 1). The measurements included the determination of AChE and BChE. Both  
26 162 were measured in 10  $\mu$ l whole blood, using *ChE check mobile*, a validated point-of-care testing device  
27 163 (*ChE check mobile*<sup>®</sup>, Securetec Detektions-Systeme AG, Neubiberg, Germany; In-Vitro-Diagnostics  
28 164 Guideline 98/79/EG; DIN EN ISO 18113-2 and -3) by following the manufacturer's instructions. Also,  
29 165 blood count, C-reactive protein (CRP), creatinine were measured at each time point. Creatine kinase  
30 166 (CK) and heart enzymes (CK-MB) were measured on the first postoperative day in the normal  
31 167 laboratory control. Brain natriuretic Peptide (NT-proBNP) was measured at the screening day. To deal  
32 168 with missing values, we included three defined measurements (time points) into the analysis.

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38 170 *Postoperative complications:*

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40 171 Since delirium, pneumonia, heart rhythm disturbances and acute renal failure are the most frequently  
41 172 reported complications after TAVI (4), we have screened all patients until the discharge of the hospital.  
42 173 Infection was defined as an increase in CRP, fever and diagnosed infection-focus (pneumonia, urinary  
43 174 tract infection, other infections). Delirium was diagnosed by using NU-DESC. Postoperative heart  
44 175 rhythm disturbances occurred by AV-block and atrial fibrillation. Patients were divided into two  
45 176 groups: those who did not develop any postoperative complications (non-complication group) and  
46 177 patients who showed one of these complications within 3 days after TAVI (complication group).

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51 179 *Operation procedure:*

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53 180 All patients were admitted and evaluated at least one day before the operation. TAVIs were performed  
54 181 by the cardiac team (cardiac surgeon, cardiologist, and cardiac anesthetist) in a hybrid operating  
55 182 theatre, according to the German guidelines for TAVI procedures. All procedures were performed with  
56 183 the patients placed under general anesthesia. In all patients, monitoring consisted of pulsoximetry, 5-  
57 184 channel electrocardiogram, invasive blood pressure, central venous pressure, urinary output and  
58 185 bladder temperature. The maintenance of normothermia was accomplished by a heating blanket  
59 186 placed beneath the patient. The patients received right ventricular pacemakers for rapid ventricular  
60 187 pacing during balloon aortic valvuloplasty and valve-expansion. Pre-oxygenation was performed with



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3 188 pure oxygen using a facemask. Anesthesia was induced with etomidate (Etomidat-Lipuro®, B. Braun  
4 189 Melsungen AG, Melsungen, Germany), remifentanil (Ultiva®, GlaxoSmithKline GmbH & Co. KG,  
5 190 Munich, Germany) and rocuronium (Rocuronium Inresa®, Inresa Arzneimittel GmbH, Freiburg,  
6 191 Germany) and maintained with sevoflurane (Sevorane®, AbbVie Deutschland GmbH & Co.KG,  
7 192 Wiesbaden, Germany). Piritramide and metamizole were used as additional pain medication. PONV  
8 193 prophylaxis was used intraoperatively, depending on the patient's risk. Cardiovascular drugs (e.g.  
9 194 norepinephrine, and dobutamine) were administered, as needed. A prophylactic antibiotic (1.5 g,  
10 195 Cefuroxim Hikma®, Hikma Pharma GmbH, Gräfelfing, Germany) was administered to each patient. In  
11 196 the operating theatre, the patient was connected to a defibrillator, and a TEE probe was introduced.  
12 197 After preparing the access points and anticoagulation with heparin (Ratiopharm GmbH, Ulm, Germany;  
13 198 mean given dose  $5293 \pm 2643$  IU), the native valve was opened under rapid ventricular pacing, and the  
14 199 prosthesis was implanted. The position and function of the prosthesis was verified with TEE. Extubation  
15 200 of the patient was the goal at the end of each procedure. After surgery, patients were monitored for  
16 201 at least 12 hours in the ICU. Following this period, patient care continued either in the ICU or in the  
17 202 general ward. There was no use of heart lung machines.

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#### 23 204 *Patient and public involvement*

24 205 Patients were not involved in the study.

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#### 26 207 *Statistics:*

27 208 The data were electronically gathered and stored by using Excel (Excel 2013, Microsoft Corporation,  
28 209 Redmond, Washington, USA). Data analysis was performed by using SPSS (Version 22.0; SPSS Inc.,  
29 210 Chicago, IL, USA). Frequency distributions and percentage rates were used for the categorical variables.  
30 211 Data are presented as mean with standard deviation and with Cohen's  $r$  effect size. Shapiro-Wilk test  
31 212 was used to verify Gaussian distribution of the study groups. Statistical significance between the  
32 213 patient groups was tested using t-test, Mann-Whitney U test, analysis of variance and chi-quadrat.  
33 214 Friedman analysis of ranks was performed to compare the cholinesterase activity change over time in  
34 215 each group. A multivariate logistic regression analysis was performed to investigate the association  
35 216 between cholinesterase activity and postoperative complications. A  $p$ -value  $< 0.05$  indicated statistical  
36 217 significance.

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3 219 Results:

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5 220 *Baseline data*

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7 221 A total of 48 patients were included, and 43 patients completed the assessment battery. The mean age  
8 222 was 79.5 +/- 5.71 years and the mean BMI was 27.93 +/- 5.36. There were almost equally men and  
9 223 women (22 (51.2 %) vs. 21 (48.8 %), table 1). All patients received elective TAVI in general anesthesia.  
10 224 TF access was selected for 32 (74 %), with TA chosen for 11 (26 %) patients. There was no use of heart-  
11 225 lung-machines. The demographic data and pre-existing conditions are shown in table 1. 32 (74.4 %)  
12 226 had an ASA-class of three, 11 (25.6 %) of four. Except of four, every patient was extubated immediately  
13 227 after operation and brought to ICU. One high risk patient was still intubated when brought to ICU and  
14 228 died two days after operation by multiorgan failure. Another patient was extubated on the first  
15 229 postoperative day. Two patients were extubated a few hours after brought to ICU. Patients were  
16 230 discharged to a normal ward after one day and left the hospital after 13.28 +/- 6.2 days.

19 231

20 232 *Postoperative complications*

21 233 24 patients (55.8 %) had postoperative complications as defined above. One multimorbid and high-risk  
22 234 patient died due to multiorgan-failure at ICU two days after surgery.

23 235 Of 43 patients, 12 developed postoperative delirium (27.9 %). Most patients developed their delirium  
24 236 on the first day after surgery.

25 237 Of 43 patients, 2 developed pneumonia. However, in 3 patients with raised infection markers and  
26 238 suspected infection no focus was found. All of them received antibiotics.

27 239 There were 7 patients with postoperative indication for pacemaker (16.3%). Overall 12 patients  
28 240 developed heart rhythm disturbances (27.9%). Some of the patients developed more than one  
29 241 complication, e.g. delirium or infection.

30 242

31 243 *Comparison between complication and non-complication group*

32 244 Preoperative variables

33 245 Preoperative assessment showed no significant differences regarding demographic data and  
34 246 laboratory routine markers like haemoglobin (p = 0.917), leukocytes (p = 0.383), CRP (p = 0.716), NT-  
35 247 proBNP (p = 0.563) and creatinine (p = 0.089). Preoperative BChE levels were significantly lower in  
36 248 patients who developed postoperative complications (D 1 complication group 2589.2 ± 556.4 vs. D 1  
37 249 non-complication group 3295.7 ± 628.0 Cohen's r = 0.514, p < 0.001, table 2). Preoperative AChE  
38 250 enzyme activity in contrast did not show any difference between complication and non-complication  
39 251 group. There was also no difference regarding alcohol (p = 0.226) or nicotine (p = 0.807) consumption.  
40 252 Men showed a higher incidence of postoperative complications (p = 0.095).

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42 254 Postoperative variables

43 255 All patients were postoperatively admitted to the ICU extubated and hemodynamic supported by  
44 256 catecholamines. Two patients did not meet the extubation criteria in the operation room and were  
45 257 extubated a few hours later at ICU. One high risk patient died at ICU due to multiorgan-failure. One  
46 258 patient was extubated on the first postoperative day.

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3 259 Complication group showed an early, sustained and statistically significant decrease in BChE activity  
4 260 from the preoperative to the first postoperative measurement (D 0:  $2784.0 \pm 534.9$  vs POD 0:  $2379.6$   
5 261  $\pm 525.1$ ,  $p < 0.001$ , figure 2). In contrast in patients without postoperative complications we observed  
6 262 a delayed decrease in BChE activity from the preoperative to postoperative period (D 0:  $3072.6 \pm 652.1$   
7 263 vs POD 2:  $2713.5 \pm 510.6$ ,  $p < 0.001$ , figure 2). In all time points a significantly lower BChE activity was  
8 264 observed in patients with complications compared to patients without postoperative complication  
9 265 (figure 2).

12 266 Both groups showed a moderate decrease in AChE activity after preoperative screening measurement  
13 267 (figure 3). From anesthesia induction to the second postoperative measurement we observed no  
14 268 significant changes in AChE activity over time in both groups. There were no significant differences in  
15 269 AChE activity between patients with and without complication in any time point (figure 3).

17 270 Further analysis showed high effect sizes for the perioperative measurements of BChE. In contrast,  
18 271 effect sizes for AChE were much lower, which affirms the results above (table 2).

21 272 Patients, who developed postoperative complications had a significantly longer stay in hospital in  
22 273 general (complication-group:  $15.2 \pm 6.3$  vs. non-complication-group:  $11.1 \pm 5.5$  days, Cohen's  $r = 0.325$ ,  
23 274  $p = 0.033$ ). There was no difference regarding the stay on ICU (complication group vs. non-complication  
24 275 group Cohen's  $r = 0.132$ ,  $p = 0.379$ ). Patients with postoperative delirium showed highest NU-DESC  
25 276 score on the first postoperative day (delirium:  $3.3 \pm 2.6$  vs. non-delirium:  $0.27 \pm 0.79$ ). The preoperative  
26 277 score of NU-DESC was  $0.42 \pm 0.67$  within patients, who developed postoperative delirium. Routine  
27 278 laboratory markers like haemoglobin, leukocytes, CRP, CK, CK-MB and creatinine did not show any  
28 279 difference (complication group vs. non-complication group  $p = n. s.$ , table 2).

31 280 Furthermore, there was no difference in EuroSCORE regarding on complication (complication group  
32 281 vs. non-complication group Cohen's  $r = 0.034$ ,  $p = 0.824$ , table 1)).

34 282

### 36 283 TA vs. TF

38 284 Patients, who underwent TA approach declared postoperative higher pain levels measured by NRS ( $p$   
39 285  $< 0.001$ ). They also showed higher CRP levels on POD 2 ( $88.8 \pm 44.5$  vs.  $161.6 \pm 70.2$ , Cohen's  $r = 0.574$ ,  
40 286  $p < 0.001$ ) and higher levels of CK ( $110.8 \pm 134.5$  vs.  $398.7 \pm 139.0$ , Cohen's  $r = 0.728$ ,  $p < 0.001$ ) and  
41 287 CK-MB ( $8.3 \pm 11.8$  vs.  $29.8 \pm 14.7$ , Cohen's  $r = 0.650$ ,  $p < 0.001$ ) on the first postoperative day. There  
42 288 were no further differences between patients with TF and TA approach, especially regarding on  
43 289 complications or BChE and AChE enzyme levels.

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293 Discussion:

294 TAVI has become the therapeutic standard for medical treatment in elderly patients with severe aortic  
295 valve stenosis and increased operative risks. The primary objective of the present investigation was to  
296 evaluate the roles of AChE and BChE as predictive markers for prevalent complications in cardiosurgical  
297 patients after TAVI.

298 Previous studies assumed an interaction of the immune and cholinergic system (19) and identified  
299 AChE and BChE as useful biomarkers for early detection of patients with emerging inflammation (16).  
300 Rapid changes in cholinesterase activity have been reported in patients after acute trauma, infections,  
301 delirium and critical illness (10–14). Both enzymes may serve as indicators of systemic inflammation  
302 and have a remarkable predictive value for mortality in critically ill patients. Zivkovic et al. showed that  
303 bedside-measurement of BChE activity predicts patient morbidity and length of ICU stay following  
304 major traumatic injury (20). Another study with patients undergoing venoarterial extracorporeal  
305 membrane oxygenation therapy after cardiac surgery revealed BChE as a strong predictor of all-cause  
306 and cardiovascular mortality (10).

307 In our present study patients with postoperative complications after TAVI had significant lower  
308 preoperative levels of BChE compared to the non-complication group. Effect sizes were particularly  
309 high for BChE measurements in this homogeneous patient group, which may rule out a great variance  
310 of enzyme activities. In combination with common preoperative evaluation procedures, BChE activity  
311 may serve as a useful predictive indicator to identify high-risk patients. Future studies are needed to  
312 clarify clinical implications.

313 Due to high variability in the onset, aetiology and progress of clinical conditions among patients,  
314 determining whether changes in the enzyme activity are correlated with the emergence of disease or  
315 are affected by concomitant factors is difficult. John et al. tested the hypothesis that AChE and BChE  
316 have an impact on patients after cardiac surgery with postoperative delirium. They showed that AChE  
317 increased and BChE decreased within the first 3 days after surgery but did not discern between patients  
318 with and without delirium. The authors supposed that the perioperative change of AChE and BChE  
319 activity might possibly be explained by an interaction of AChE and BChE and the use of a  
320 cardiopulmonary bypass (17). In our present study we evaluated the role of AChE and BChE activity in  
321 cardiosurgical patients after TAVI, as a standardized operative procedure without cardiopulmonary  
322 bypass. We could show that complication group showed a significantly perioperative decrease of BChE  
323 within the first 3 days after TAVI, despite the fact that there was no use of heart-lung machines in our  
324 patients. Furthermore, there was no use of blood products in the present study, so we can rule out a  
325 possible interaction of AChE and BChE with blood products as well. While in the CESARO study a wide  
326 spectrum of operative disciplines has been analysed and the perioperative enzyme activities showed  
327 small effect sizes, we can show high effect sizes for BChE in this secondary analysis of a homogeneous  
328 patient group with standardized operative procedure.

329 BChE activity could be regarded as an inflammatory parameter in this context. In literature, lower  
330 levels of BChE activity have already been described during inflammatory processes, stress and  
331 malnutrition. Therefore, lower levels of BChE activity in complication group might reflect perioperative  
332 inflammation, which is known to promote complications like delirium or infections. Conventional  
333 markers like CRP and leucocytes did not differ in both groups.

334 Delirium is a complex symptom which is very common in operative and non-operative disciplines in  
335 the course of hospital stay. The incidence is especially high among patients undergoing heart surgery  
336 (21). The incidence in this patients population has been described to be from 30 up to 80 % (22). The

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3 337 incidence of delirium after TAVI is reported as 29 % in literature (6). Delirium occurred significantly  
4 338 more frequently following TA procedures (23). In the present study 26,7 % of the patients were  
5 339 diagnosed with delirium overall. There was no difference depending on TA or TF approach.  
6 340 Perioperative measurement of AChE and BChE did not discern between patients with and without  
7 341 delirium, which is in accordance with the findings by John et al.

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10 342 The present study highlights the validity of BChE measurements for early detection of high-risk patients  
11 343 after TAVI. Surprisingly, the BChE assessment proved more effective than the EuroSCORE in  
12 344 discriminating between the patient groups making it a valuable biomarker for the early detection of  
13 345 high-risk patients. EuroSCORE is a well-established clinical assay for the patient mortality analysis-(24)  
14 346 and requires documenting multiple and diverse datasets. The datasets are in most cases readily  
15 347 available; however, in some cases, a particular set of data might not be accessible, delaying or making  
16 348 the scoring impossible. By using a POCT system for a single BChE measurement, the results of an  
17 349 equally efficient outcome assessment tool are readily available at the bedside and may complete  
18 350 conventional assessments. Further studies with a greater patient population siare needed to  
19 351 investigate the clinical implications.

22 352 Prompt assessment of the systemic immune response with an immediate, rapid and affordable  
23 353 bedside measurement of the BChE activity might improve risk evaluation and help optimize  
24 354 postoperative management and therapy of patients after TAVI. Predicting the length of the hospital  
25 355 stay might play an important role in staff and resources management for these patients.

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### 31 358 Limitations

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35 360 Limitations of the present study might be the short duration of 3 days' measurement. Blood was taken  
36 361 from each patient; in case the analysis could not be performed immediately (during anesthesia  
37 362 induction), the sample was cooled down in a refrigerator. Maybe values of AChE and BChE changed in  
38 363 combination with lower temperature. Furthermore, it was only one measurement performed with  
39 364 each sample, so no control values could be achieved.

42 365 The study protocol required daily cholinesterase activity measurements in the postoperative period,  
43 366 without specifying time or requesting multiple daily measurements. Therefore, circadian fluctuations  
44 367 in enzyme activities could not be considered.

46 368 The biggest limitation of the present study is the low number of included patients. Further studies with  
47 369 larger patient groups and with focus on the underlying mechanisms of the different complications  
48 370 would be needed to validate our findings and the clinical implications.

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Characteristic	Total sample (n = 43)
<b>Age (years) [M (SD)]</b>	79.47 (5.7)
<b>Sex [n (%)]</b>	
Male	22 (51.2)
Female	21 (48.8)
<b>BMI [M (SD)]</b>	27.93 (5.4)
<b>ASA – PS [n (%)]</b>	
3	32 (74.4)
4	11 (25.6)
<b>Operative procedure [n (%)]</b>	
Transapicale TAVI	11 (25.6)
Transfemorale TAVI	32 (74.4)
<b>Relevant comorbidities [n (%)]</b>	
Hypertension	37 (86)
Diabetes	16 (37.2)
Congestive heart failure	36 (83.7)
Congestive kidney failure	20 (46.5)
Coronary heart disease	31 (72.1)
Cardiac arrhythmias	25 (58.1)
Stroke	8 (18.6)
Nicotine	19 (44.2)
Alcohol	8 (18.6)
Hypothyreosis	10 (23.3)
Hypercholesterinemia	14 (32.6)
<b>EuroSCORE [n (%)]</b>	
low	15 (34.9)
middle	18 (41.9)
high	10 (23.3)

373 Table 1 Description of baseline data; all data are presented as n (number) and (%). ASA, American  
 374 Society of Anaesthesiologists Classification; BMI, body mass index

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Variables	Complication	N	M	SD	p-value	Cohens' r
EuroSCORE	Yes	24	21.8	15.4	0.824	0.034
	no	19	22.8	13.5		
Weight [kg]	Yes	24	78.3	15.5	0.504	0.102
	no	19	74.9	18.0		
BMI [kg/m <sup>2</sup> ]	Yes	24	28.1	4.7	0.860	0.037
	no	19	27.7	6.2		
Age [years]	Yes	24	79.9	5.3	0.556	0.086
	no	19	78.9	6.3		
NT-proBNP [pg/ml]	Yes	24	6244.8	6773.1	0.563	0.099
	no	19	4806.6	7809.4		
Hemoglobin D 0 [g/dl]	Yes	24	12.1	1.9	0.917	0.029
	no	19	12.2	1.4		
Hemoglobin POD 0 [g/dl]	Yes	24	10.7	1.5	0.565	0.068
	no	19	10.9	1.4		
Hemoglobin POD 1 [g/dl]	Yes	24	10.1	1.4	0.986	0.000
	no	19	10.1	1.1		
Hemoglobin POD 2 [g/dl]	Yes	24	10.1	1.1	0.673	0.087
	No	19	10.3	1.2		
Hemoglobin POD 3 [g/dl]	Yes	24	9.6	1.0	0.272	0.173
	No	19	10.0	1.3		
Creatinine D 0 [mg/dl]	Yes	24	1.4	0.7	0.089	0.247
	no	19	1.1	0.4		
Creatinine POD 1 [mg/dl]	Yes	24	1.2	0.4	0.347	0.124
	No	19	1.1	0.4		
Creatinine POD 2 [mg/dl]	Yes	24	1.5	0.8	0.188	0.223
	no	19	1.2	0.4		
Creatinine POD 3 [mg/dl]	Yes	24	1.5	1.0	0.240	0.244
	No	19	1.1	0.4		
Leukocytes D 0 [/nl]	Yes	24	7.8	2.1	0.383	0.131
	no	19	7.3	1.6		
Leucocytes POD 1 [/nl]	Yes	24	9.6	4.3	0.496	0.113
	No	19	8.8	2.1		
Leukocytes POD 2 [/nl]	Yes	24	9.9	2.7	0.616	0.081
	No	19	9.4	3.5		
Leukocytes POD 3 [/nl]	Yes	24	8.8	3.1	0.079	0.336
	No	19	7.0	1.5		
CRP D 0 [mg/l]	Yes	24	16.3	17.8	0.716	0.094
	no	19	19.8	18.2		
CRP POD 1 [mg/l]	Yes	24	31.8	21.1	0.177	0.236
	No	19	22.0	18.9		
CRP POD 2 [mg/l]	Yes	24	116.3	52.9	0.516	0.114
	No	19	101.3	78.2		
CRP POD 3 [mg/l]	Yes	24	115.3	68,5	0.113	0.284
	No	19	72.7	76,0		
BChE D 0 [U/l]	Yes	24	2784.0	534,9	0.118	0.238
	no	19	3072.6	652,1		
BChE D 1 [U/l]	Yes	24	2589.2	556,4	<0.001	0.514
	no	19	3295.7	628,0		
BChE POD 0 [U/l]	Yes	24	2379.6	525.1.	<0.001	0.469
	No	19	2972.5	599.2		
BChE	Yes	24	2300.3	561.0	<0.001	0.504

POD 1 [U/l]	No	19	2936.2	523.1		
BChE	Yes	24	2166.7	537.0	<b>0.002</b>	0.462
POD 2 [U/l]	No	19	2713.5	510.6		
AChE	Yes	24	45.0	8.1	0.446	0.118
D 0 [U/gHb]	no	19	43.3	6.0		
AChE	Yes	24	42.0	10.0	0.263	0.172
D 1 [U/gHb]	no	19	39.2	4.7		
AChE	Yes	24	42.9	10.0	0.051	0.295
POD 0 [U/gHb]	No	19	37.6	6.4		
AChE	Yes	24	41.5	9.2	0.196	0.198
POD 1 [U/gHb]	No	19	38.4	5.2		
AChE	Yes	24	41.2	8.2	0.058	0.294
POD 2 [U/gHb]	No	19	36.5	6.8		
CK	Yes	24	189.6	186.8	0.953	0.009
POD 1 [U/l]	No	19	186.1	190.5		
CK-MB	Yes	24	15.5	18.2	0.946	0.012
POD 1 [ng/ml]	No	19	15.9	14.3		
CK-Index	Yes	24	7.2	3.6	0.314	0.174
POD 1	No	19	8.5	3.8		
Stay in hospital [days]	Yes	24	15.2	6.3	<b>0.033</b>	0.325
	no	19	11.1	5.5		

385 Table 2 Perioperative laboratory markers; all data are presented as number (n) or as mean  $\pm$  SD

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5 388 Figure 1 Timeline of measurements of BChE and AChE: blood samples were taken one day preoperative  
6 389 (D 0), shortly before anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery  
7 390 (POD 1) and two days after surgery (POD 2). If the measurements could not be conducted immediately,  
8 391 the samples have been cooled in a refrigerator and the measurement was performed up to 2 hours  
9 392 later.

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13 394 Figure 2 Time trajectories of BChE activities in TAVI-patients (n = 43). Pre-operative (D0), shortly before  
14 395 anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery (POD 1) and two days  
15 396 after surgery (POD 2) measurements in patients with (dashed) and without (solid) complication. Data  
16 397 are presented as median  $\pm$  standard deviation. \* Difference between groups; # Difference within  
17 398 groups.

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22 400 Figure 3 Time trajectories of AChE activities in TAVI-patients (n = 43). Pre-operative (D0), shortly before  
23 401 anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery (POD 1) and two days  
24 402 after surgery (POD 2) measurements in patients with (dashed) and without (solid) complication. Data  
25 403 are presented as median  $\pm$  standard deviation.

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3 405 **Contributorship statement**  
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6 407 DB, YZ, AM, AH and BM were responsible for study design, statistical analyses and drafting of the  
7  
8 408 manuscript. WP, AB, BM and BG performed the experiments and drafted the manuscript. BM and AM  
9  
10 409 were responsible for statistical analysis. All authors read and approved the final manuscript.

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15 412 **Competing interests**

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18 414 The authors declare no conflict of interest.

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27  
28 420 The authors have not declared a specific grant for this research from any funding agency in the public,  
29  
30 421 commercial or not-for-profit sectors.

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35 424 **Data sharing statement**

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38 426 No additional data are available.

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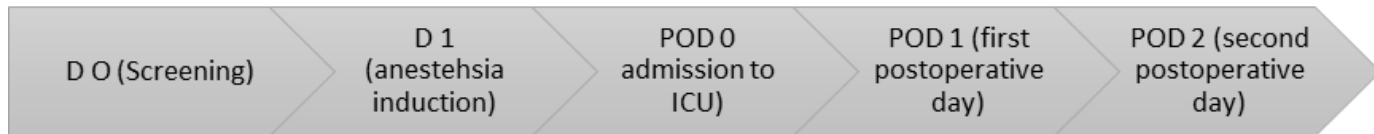


Figure 1

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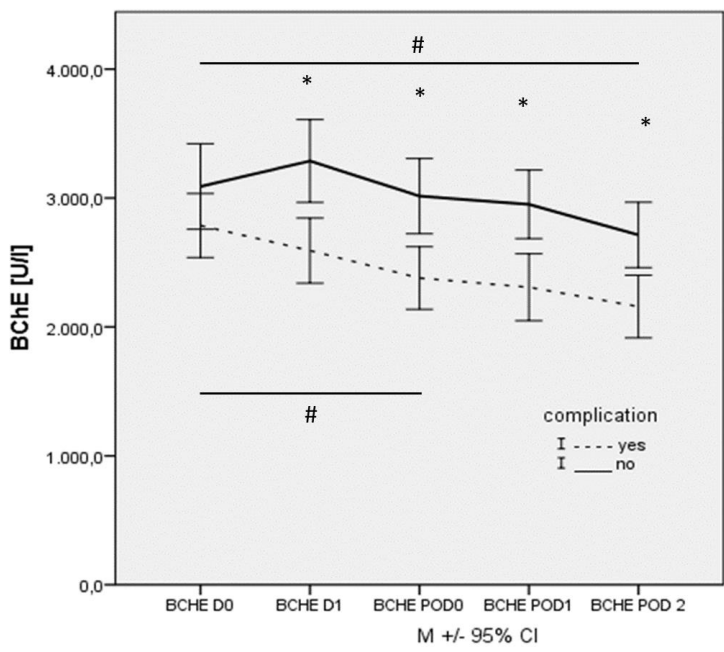


Figure 2

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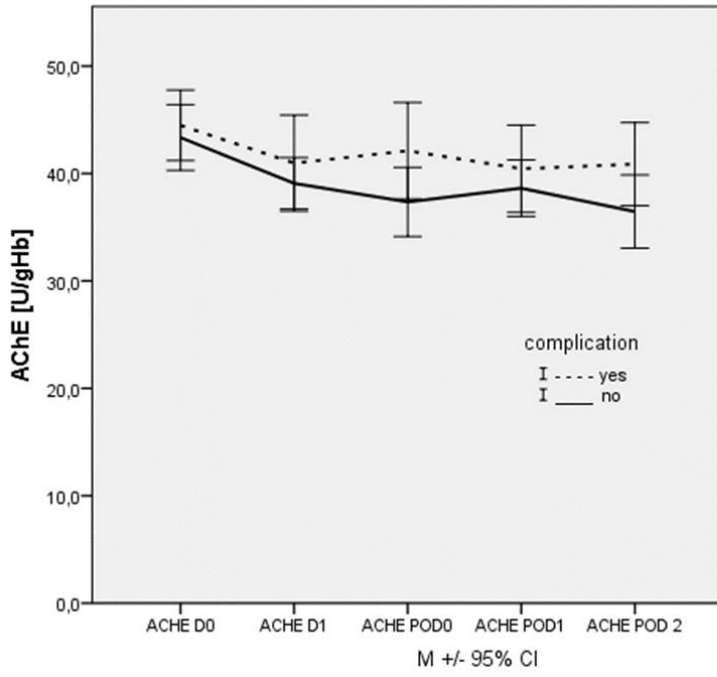


Figure 3

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## STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
<b>Title and abstract</b>	1	(a) Indicate the study's design with a commonly used term in the title or the abstract [line 34-36] (b) Provide in the abstract an informative and balanced summary of what was done and what was found [line 30-55]
<b>Introduction</b>		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported [line 73-100]
Objectives	3	State specific objectives, including any prespecified hypotheses [line 73-105]
<b>Methods</b>		
Study design	4	Present key elements of study design early in the paper [line 108-119]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection [line 122-125]
Participants	6	(a) <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up [line 116-119] <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants (b) <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable [line 127-177]
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group [line 122-125]
Bias	9	Describe any efforts to address potential sources of bias [line 159-168]
Study size	10	Explain how the study size was arrived at [line 108-115]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why [line 127-177]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding [line 208-217] (b) Describe any methods used to examine subgroups and interactions [not included] (c) Explain how missing data were addressed [line 166-168] (d) <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses [not included]

Continued on next page



<b>Results</b>		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed [line 221-230] (b) Give reasons for non-participation at each stage [line 221-230] (c) Consider use of a flow diagram → removed (as suggested by reviewer)
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders [line 221-230] (b) Indicate number of participants with missing data for each variable of interest [line 233-241] (c) <i>Cohort study</i> —Summarise follow-up time (eg, average and total amount) [line 226-230]
Outcome data	15*	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time [line 226-230] <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included [line 232-289] (b) Report category boundaries when continuous variables were categorized [line 232-289] (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period [line 232-289]
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses [not applicable]
<b>Discussion</b>		
Key results	18	Summarise key results with reference to study objectives [line 294-351]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias [line 356-373]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence [line 294-351]
Generalisability	21	Discuss the generalisability (external validity) of the study results [line 294-351]
<b>Other information</b>		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based [line 461-463]

\*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

**Note:** An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at [www.strobe-statement.org](http://www.strobe-statement.org).

# BMJ Open

## Butyrylcholinesterase as a perioperative complication marker in patients after transcatheter aortic valve implantation: a prospective observational study

Journal:	<i>BMJ Open</i>
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<b>Primary Subject Heading</b>:	Intensive care
Secondary Subject Heading:	Anaesthesia, Cardiovascular medicine
Keywords:	Adult intensive & critical care < ANAESTHETICS, Anaesthesia in cardiology < ANAESTHETICS, Adult cardiology < CARDIOLOGY, Valvular heart disease < CARDIOLOGY

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3 **1 Butyrylcholinesterase as a perioperative complication marker in patients after**  
4 **2 transcatheter aortic valve implantation: a prospective observational study**  
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11 6 Bernhard Michels<sup>1</sup>, Andreas Holzamer<sup>2</sup>, Bernhard Graf<sup>3</sup>, Andre Bredthauer<sup>3</sup>, Walter Petermichl<sup>3</sup>, Anika  
12 7 Müller<sup>4</sup>, York Zausig<sup>5</sup>, Diane Bitzinger<sup>3</sup>  
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**Abstract**

*Objectives:* Transcatheter aortic valve implantation (TAVI) is performed in elderly patients with severe aortic valve stenosis and increased operative risks. We tested the hypothesis that acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) have a predictive value for prevalent complications after TAVI and could serve as indicators of systemic inflammation in the early postoperative period.

*Design:* Prospective observational study.

*Setting:* This study is a secondary analysis of multi-center CESARO-study.

*Participants:* 48 TAVI patients were included, 43 obtained the complete assessment.

*Primary and secondary outcome measures:* Patients clinical parameters, demographic data, peripheral AChE and BChE-activities and routine blood markers were assessed throughout the perioperative period using bedside point-of-care measurements for AChE and BChE. Postoperative complications screening was conducted up to the 3<sup>rd</sup> postoperative day and included infections, delirium and heart-rhythm disturbances. After assessment the patients were divided into complication and non-complication group.

*Results:* Of 43 patients, 24 developed postsurgical complications (55.8%). Preoperative assessment showed no significant differences regarding demographic data and laboratory markers, but preoperative BChE-levels were significantly lower in patients who developed postoperative complications (complication group  $2589.2 \pm 556.4$  vs. non-complication group  $3295.7 \pm 628.0$ , Cohen's  $r = 0.514$ ,  $p < 0.001$ ). In complication group we observed an early, sustained reduction in BChE-activity from preoperative to postoperative period. In complication group BChE-levels were significantly lower at each time point compared to non-complication group. AChE-activity showed no significant difference between both groups. Complication group also had longer stay in hospital overall.

*Conclusion:* BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative complications after TAVI. By using point-of-care measurements the levels of BChE are fast available and can lead to an early targeted therapy. Predicting the length of the hospital stay might play an important role in staff and resources management for these patients.

*Trial registration:* NCT01964274

**Key words:** cardiac surgery, TAVI, inflammation, delirium, butyrylcholinesterase, acetylcholinesterase

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3 59 Strengths and limitations of this study  
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5 60 This study is a secondary analysis of the prospective observational multi-center CESARO-study.  
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9 62 BChE could be a useful perioperative biomarker to identify patients with a higher risk for postoperative  
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72 Introduction:

73 Recently, transcatheter aortic valve implantation (TAVI) has become the therapeutic standard for  
74 medical treatment in elderly, multimorbid patients with severe aortic valve stenosis and increased  
75 operative risks (1, 2). TAVI involves the implantation of a prosthetic valve, which is introduced with a  
76 catheter through transfemoral (TF), transapical (TA) or direct transaortic access. Usually, the TF  
77 approach is preferred because thoracotomy and penetration of the myocardium are not needed. The  
78 TA approach is common, if severe atherosclerotic disease does not allow retrograde insertion of the  
79 catheter. In patients with severe aortic stenosis, who could not undergo a surgical replacement of the  
80 aortic valve, TAVI significantly reduced the rates of death at any cause, compared to standard therapy  
81 (3). However, previous studies have shown that pneumonia, acute renal failure, indication for a  
82 permanent pacemaker and delirium were the most frequently recorded complications after TAVI (4).  
83 Covello et. al. reported a pneumonia rate of 7-8 % after TAVI (5). The incidence of delirium after TAVI  
84 is reported as 29 % in literature (6).

85 Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) are a focus of current research. Recent  
86 studies have shown that AChE and BChE serve as diagnostic markers of low-grade systemic  
87 inflammation (7–9). Rapid changes in cholinesterase activity have also been reported in patients after  
88 acute trauma, infections, burns and critical illness (10–14). Both enzymes may serve as indicators of  
89 systemic inflammation and may have a predictive value for mortality in critically ill patients. Zivkovic  
90 et. al. showed that reduced serum activity of BChE indicates severe systemic inflammation in critical ill  
91 patients (13). Furthermore, a recent study showed, that a sustained reduction in serum cholinesterase  
92 enzyme activity predicts patient outcome following sepsis (15).

93 Other studies postulate low preoperative plasma cholinesterase activity as a risk marker of  
94 postoperative delirium in elderly patients (16). A recently published study on cholinesterase activity in  
95 cardiac surgical patients showed no postoperative differences in cholinesterase activities between  
96 delirious and non-delirious patients, but showed a perioperative decrease of BChE which was  
97 potentially caused by cardiopulmonary bypass (17). However, due to high variability in the etiology  
98 and progress of clinical conditions, it was difficult to determine whether the changes in the enzyme  
99 activity correlated with the emergence of disease or was affected by concomitant factors such as  
100 cardiopulmonary bypass.

101 This is the first study to investigate the roles of AChE and BChE as inflammatory markers in cardiac  
102 surgical patients under standardized perioperative conditions without using cardiopulmonary bypass.  
103 Our aim of the present study is to evaluate if there is a predictive association of perioperative  
104 determination of AChE and BChE activity and the occurrence of postoperative complications after  
105 TAVI.

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3 106 Material and Methods:

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5 107 *Ethics approval statement and patient population:*

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7 108 This work is a secondary analysis of the prospective observational multi-center CESARO study,  
8 109 powered for the detection of postoperative delirium. The CESARO study was initiated at Charité –  
9 110 Universitätsmedizin Berlin, Department of Anesthesiology and Operative Intensive Care Medicine  
10 111 (Clinicaltrials.gov ID: NCT01964274) and approved by the local independent Charité Ethics Committee,  
11 112 Charité – Universitätsmedizin Berlin, Germany (ref.: EA1/220/13) on 14 August 2013. After further  
12 113 approval of the local ethics board of the University of Regensburg a total of 48 patients were included  
13 114 into the study between March 2014, and June 2016 at University Hospital of Regensburg. Written  
14 115 informed consent was obtained from each patient.

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17 116 Inclusions criteria: minimum age of 18 years, admission to intensive care unit (ICU) following elective  
18 117 TAVI in general anesthesia.

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20 118 Exclusion criteria: missing consent, patients with a known pseudocholinesterase deficiency, patients  
21 119 with language, visual or hearing impairments.

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25 121 *Data*

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27 122 Data were acquired from anesthetic charts (Medlinq V.1.3, Hamburg, Germany), the patient document  
28 123 system used in the ICU (Metavision, iMDsoft, Tel Aviv, Israel) and medical reports from the electronic  
29 124 hospital information system (SAP, Walldorf, Germany) from the preoperative, intraoperative and  
30 125 postoperative periods until the patients were discharged from the hospital.

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34 127 *Preoperative variables:*

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36 128 Preoperative data included demographic data, such as age, sex, height, weight, regular use of alcohol  
37 129 and nicotine, American Society of Anesthesiologists (ASA) class, logistic EuroSCORE (European System  
38 130 for Cardiac Operative Risk Evaluation), New York Heart Association (NYHA) class and left ventricular  
39 131 ejection fraction (EF). The patients' previous medical history was examined for conditions such as  
40 132 chronic kidney disease, cerebrovascular events, including stroke and transient ischemic attacks,  
41 133 myocardial infarction, chronic obstructive pulmonary disease, diabetes mellitus and pre-existing  
42 134 cardiac arrhythmias. Furthermore, we evaluated the preoperative anticholinergic burden using the  
43 135 anticholinergic drug scale (18). This scale ranges from zero (no anticholinergic activity) to three  
44 136 (highest anticholinergic activity). Each long-term drug was screened for its anticholinergic activity and  
45 137 for each patient the number of points was assessed. Every patient was screened for preoperative  
46 138 delirium, using the nursing delirium screening scale (NU-DESC). Preoperative assessment of AChE,  
47 139 BChE, CRP, leukocytes, haemoglobin and creatinine were performed (table 1).

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53 141 *Intraoperative variables:*

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55 142 Key elements of intraoperative data included the selected access type, anesthetic procedure,  
56 143 transfusion of erythrocyte concentrates and extubation rate as well as the procedure duration.

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60 145 *Postoperative variables:*



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3 146 Postoperative data included the patient's stay in the ICU and the stay in hospital in general. Next to  
4 147 the sampling of laboratory markers, every patient was screened for delirium with NU-DESC for the first  
5 148 3 days after surgery. Patients were daily assessed for pain, using the numeric rating scale (NRS score:  
6 149 0 = no pain – 10 = maximum pain). Furthermore, any complication in recovery time was noticed.  
7 150 Mortality reasons are divided into cardiac, acute kidney injury, cardiovascular events and infections.  
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11 152 Variables:

12 153 *Delirium:*

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15 154 Delirium screening was conducted perioperatively using a validated screening tool (NU-DESC) (19). NU-  
16 155 DESC assesses five dimensions: orientation, behaviour, communication, illusion/hallucination and  
17 156 psychomotor retardation. The symptoms are rated on a three-point scale, whereas a score of two or  
18 157 more cumulative points indicated delirium. Delirium assessment was performed one day prior to the  
19 158 operation, on admission to ICU and daily up to the third postoperative day. Patients with Richmond  
20 159 Agitation Sedation Scale (RASS)  $\leq -2$  were excluded for the current testing.  
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24 161 *Laboratory parameters:*

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27 162 Blood samples were taken from every patient at following time points: one day before operation  
28 163 (screening), shortly before anesthetic induction, on admission to ICU, one day after surgery and two  
29 164 days after surgery (figure 1). The measurements included the determination of AChE and BChE. Both  
30 165 were measured in 10  $\mu$ l whole blood, using *ChE check mobile*, a validated point-of-care testing device  
31 166 (*ChE check mobile*<sup>®</sup>, Securetec Detektions-Systeme AG, Neubiberg, Germany; In-Vitro-Diagnostics  
32 167 Guideline 98/79/EG; DIN EN ISO 18113-2 and -3) by following the manufacturer's instructions. Also,  
33 168 blood count, C-reactive protein (CRP), creatinine were measured at each time point. Creatine kinase  
34 169 (CK) and heart enzymes (CK-MB) were measured on the first postoperative day in the normal  
35 170 laboratory control. Brain natriuretic Peptide (NT-proBNP) was measured at the screening day. To deal  
36 171 with missing values, we included three defined measurements (time points) into the analysis.  
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41 173 *Postoperative complications:*

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43 174 Since delirium, pneumonia, heart rhythm disturbances and acute renal failure are the most frequently  
44 175 reported complications after TAVI (4), we have screened all patients until the discharge of the hospital.  
45 176 Infection was defined as an increase in CRP, fever and diagnosed infection-focus (pneumonia, urinary  
46 177 tract infection, other infections). Delirium was diagnosed by using NU-DESC. Postoperative heart  
47 178 rhythm disturbances occurred by AV-block and atrial fibrillation. Patients were divided into two  
48 179 groups: those who did not develop any postoperative complications (non-complication group) and  
49 180 patients who showed one of these complications within 3 days after TAVI (complication group).  
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54 182 *Operation procedure:*

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56 183 All patients were admitted and evaluated at least one day before the operation. TAVIs were performed  
57 184 by the cardiac team (cardiac surgeon, cardiologist, and cardiac anesthetist) in a hybrid operating  
58 185 theatre, according to the German guidelines for TAVI procedures. All procedures were performed with  
59 186 the patients placed under general anesthesia. In all patients, monitoring consisted of pulse oximetry, 5-  
60 187 channel electrocardiogram, invasive blood pressure, central venous pressure, urinary output and

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3 188 bladder temperature. The maintenance of normothermia was accomplished by a heating blanket  
4 189 placed beneath the patient. The patients received right ventricular pacemakers for rapid ventricular  
5 190 pacing during balloon aortic valvuloplasty and valve-expansion. Pre-oxygenation was performed with  
6 191 pure oxygen using a facemask. Anesthesia was induced with etomidate (Etomidat-Lipuro®, B. Braun  
7 192 Melsungen AG, Melsungen, Germany), remifentanyl (Ultiva®, GlaxoSmithKline GmbH & Co. KG,  
8 193 Munich, Germany) and rocuronium (Rocuronium Inresa®, Inresa Arzneimittel GmbH, Freiburg,  
9 194 Germany) and maintained with sevoflurane (Sevorane®, AbbVie Deutschland GmbH & Co.KG,  
10 195 Wiesbaden, Germany). Piritramide and metamizole were used as additional pain medication. PONV  
11 196 prophylaxis was used intraoperatively, depending on the patient's risk. Cardiovascular drugs (e.g.  
12 197 norepinephrine, and dobutamine) were administered, as needed. A prophylactic antibiotic (1.5 g,  
13 198 Cefuroxim Hikma®, Hikma Pharma GmbH, Gräfelfing, Germany) was administered to each patient. In  
14 199 the operating theatre, the patient was connected to a defibrillator, and a TEE probe was introduced.  
15 200 After preparing the access points and anticoagulation with heparin (Ratiopharm GmbH, Ulm, Germany;  
16 201 mean given dose  $5293 \pm 2643$  IU), the native valve was opened under rapid ventricular pacing, and the  
17 202 prosthesis was implanted. The position and function of the prosthesis was verified with TEE. Extubation  
18 203 of the patient was the goal at the end of each procedure. After surgery, patients were monitored for  
19 204 at least 12 hours in the ICU. Following this period, patient care continued either in the ICU or in the  
20 205 general ward. There was no use of heart lung machines.

25 206

#### 27 207 *Patient and public involvement*

28 208 Patients were not involved in the study.

30 209

#### 31 210 *Statistics:*

32 211 The data were electronically gathered and stored by using Excel (Excel 2013, Microsoft Corporation,  
33 212 Redmond, Washington, USA). Data analysis was performed by using SPSS (Version 22.0; SPSS Inc.,  
34 213 Chicago, IL, USA). Frequency distributions and percentage rates were used for the categorical variables.  
35 214 Data are presented as mean with standard deviation and with Cohen's r effect size. Shapiro-Wilk test  
36 215 was used to verify Gaussian distribution of the study groups. Statistical significance between the  
37 216 patient groups was tested using t-test, Mann-Whitney U test, analysis of variance and chi-quadrat.  
38 217 Bonferroni correction was done in case of repeat-measurements to rule out alpha error accumulation.  
39 218 Friedman analysis of ranks was performed to compare the cholinesterase activity change over time in  
40 219 each group. A multivariate logistic regression analysis was performed to investigate the association  
41 220 between cholinesterase activity and postoperative complications. A p-value < 0.05 indicated statistical  
42 221 significance.

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3 223 Results:

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5 224 *Baseline data*

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7 225 A total of 48 patients were included, and 43 patients completed the assessment battery. The mean age  
8 226 was 79.5 +/- 5.71 years and the mean BMI was 27.93 +/- 5.36. There were almost equally men and  
9 227 women (22 (51.2 %) vs. 21 (48.8 %), table 1). All patients received elective TAVI in general anesthesia.  
10 228 TF access was selected for 32 (74 %), with TA chosen for 11 (26 %) patients. There was no use of heart-  
11 229 lung-machines. The demographic data and pre-existing conditions are shown in table 1. 32 (74.4 %)  
12 230 had an ASA-class of three, 11 (25.6 %) of four. Except of four, every patient was extubated immediately  
13 231 after operation and brought to ICU. One high risk patient was still intubated when brought to ICU and  
14 232 died two days after operation by multiorgan failure. Another patient was extubated on the first  
15 233 postoperative day. Two patients were extubated a few hours after brought to ICU. Patients were  
16 234 discharged to a normal ward after one day and left the hospital after 13.28 +/- 6.2 days.

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21 236 *Postoperative complications*

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23 237 24 patients (55.8 %) had postoperative complications as defined above. One multimorbid and high-risk  
24 238 patient died due to multiorgan-failure at ICU two days after surgery.

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26 239 Of 43 patients, 12 developed postoperative delirium (27.9 %). Most patients developed their delirium  
27 240 on the first day after surgery.

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29 241 Of 43 patients, 2 developed pneumonia. However, in 3 patients with raised infection markers and  
30 242 suspected infection no focus was found. All of them received antibiotics.

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32 243 There were 7 patients with postoperative indication for pacemaker (16.3%). Overall 12 patients  
33 244 developed heart rhythm disturbances (27.9%). Some of the patients developed more than one  
34 245 complication, e.g. delirium or infection.

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39 247 *Comparison between complication and non-complication group*

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41 248 Preoperative variables

42 249 Preoperative assessment showed no statistically significant differences regarding demographic data  
43 250 and laboratory routine markers like haemoglobin (p = 0.917), leukocytes (p = 0.383), CRP (p = 0.716),  
44 251 NT-proBNP (p = 0.563) and creatinine (p = 0.089). Preoperative BChE levels were significantly lower in  
45 252 patients who developed postoperative complications (D 1 complication group 2589.2 ± 556.4 vs. D 1  
46 253 non-complication group 3295.7 ± 628.0 Cohen's r = 0.514, p < 0.001, table 2). Preoperative AChE  
47 254 enzyme activity in contrast did not show any statistically significant difference between complication  
48 255 and non-complication group. There was no statistically significant difference regarding pre-existing  
49 256 anticholinergic medication (p = 0.153). There was also no statistically significant difference regarding  
50 257 alcohol (p = 0.226) or nicotine (p = 0.807) consumption. Men or women did not show a significantly  
51 258 higher incidence of postoperative complications (p = 0.095). There was no statistically significant  
52 259 difference between complication and non-complication group regarding the anticholinergic burden (p  
53 260 = 0.229).

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59 262 Postoperative variables

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3 263 All patients were postoperatively admitted to the ICU extubated and hemodynamic supported by  
4 264 catecholamines. Two patients did not meet the extubation criteria in the operation room and were  
5 265 extubated a few hours later at ICU. One high risk patient died at ICU due to multiorgan-failure. One  
6 266 patient was extubated on the first postoperative day.

7  
8 267 Complication group showed an early, sustained and statistically significant decrease in BChE activity  
9 268 from the preoperative to the first postoperative measurement (D 0:  $2784.0 \pm 534.9$  vs POD 0:  $2379.6$   
10 269  $\pm 525.1$ ,  $p < 0.001$ , figure 2). In contrast in patients without postoperative complications we observed  
11 270 a delayed decrease in BChE activity from the preoperative to postoperative period (D 0:  $3072.6 \pm 652.1$   
12 271 vs POD 2:  $2713.5 \pm 510.6$ ,  $p < 0.001$ , figure 2). In all time points a significantly lower BChE activity was  
13 272 observed in patients with complications compared to patients without postoperative complication  
14 273 (figure 2).

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17 274 Further analysis involving partial correlation and regression analysis showed, that there was no  
18 275 influence of pre-operative anticholinergic medication on BChE results ( $p = n. s.$ ).

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21 276 Both groups showed a moderate decrease in AChE activity after preoperative screening measurement  
22 277 (figure 3). From anesthesia induction to the second postoperative measurement we observed no  
23 278 significant changes in AChE activity over time in both groups. There were no significant differences in  
24 279 AChE activity between patients with and without complication in any time point (figure 3).

25  
26 280 Further analysis showed large effect sizes for the perioperative measurements of BChE. In contrast,  
27 281 effect sizes for AChE were much lower, which affirms the results above (table 2).

28  
29 282 Patients, who developed postoperative complications had a significantly longer stay in hospital in  
30 283 general (complication-group:  $15.2 \pm 6.3$  vs. non-complication-group:  $11.1 \pm 5.5$  days, Cohen's  $r = 0.325$ ,  
31 284  $p = 0.033$ ). There was no statistically significant difference regarding the stay on ICU (complication  
32 285 group vs. non-complication group Cohen's  $r = 0.132$ ,  $p = 0.379$ ). Patients with postoperative delirium  
33 286 showed highest NU-DESC score on the first postoperative day (delirium:  $3.3 \pm 2.6$  vs. non-delirium:  
34 287  $0.27 \pm 0.79$ ). The preoperative score of NU-DESC was  $0.42 \pm 0.67$  within patients, who developed  
35 288 postoperative delirium. Routine laboratory markers like haemoglobin, leukocytes, CRP, CK, CK-MB and  
36 289 creatinine did not show any statistically significant difference (complication group vs. non-  
37 290 complication group  $p = n. s.$ , table 2). he

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44 293 Furthermore, there was no statistically significant difference in EuroSCORE regarding on complication  
45 294 (complication group vs. non-complication group Cohen's  $r = 0.034$ ,  $p = 0.824$ , table 1).

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#### 47 296 TA vs. TF

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50 297 Patients, who underwent TA approach declared postoperative higher pain levels measured by NRS ( $p$   
51 298  $< 0.001$ ). They also showed higher CRP levels on POD 2 ( $88.8 \pm 44.5$  vs.  $161.6 \pm 70.2$ , Cohen's  $r = 0.574$ ,  
52 299  $p < 0.001$ ) and higher levels of CK ( $110.8 \pm 134.5$  vs.  $398.7 \pm 139.0$ , Cohen's  $r = 0.728$ ,  $p < 0.001$ ) and  
53 300 CK-MB ( $8.3 \pm 11.8$  vs.  $29.8 \pm 14.7$ , Cohen's  $r = 0.650$ ,  $p < 0.001$ ) on the first postoperative day. There  
54 301 were no further statistically significant differences between patients with TF and TA approach,  
55 302 especially regarding on complications or BChE and AChE enzyme levels.

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3 305 Discussion:  
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5 306 TAVI has become the therapeutic standard for medical treatment in elderly patients with severe aortic  
6 307 valve stenosis and increased operative risks. The primary objective of the present investigation was to  
7 308 evaluate the roles of AChE and BChE as predictive markers for prevalent complications in cardiosurgical  
8 309 patients after TAVI.

10 310 Previous studies assumed an interaction of the immune and cholinergic system (20) and identified  
11 311 AChE and BChE as useful biomarkers for early detection of patients with emerging inflammation (16).  
12 312 Rapid changes in cholinesterase activity have been reported in patients after acute trauma, infections,  
13 313 delirium and critical illness (10–14). Both enzymes may serve as indicators of systemic inflammation  
14 314 and may have a predictive value for mortality in critically ill patients. Zivkovic et al. showed that  
15 315 bedside-measurement of BChE activity predicts patient morbidity and length of ICU stay following  
16 316 major traumatic injury (21). Another study with patients undergoing venoarterial extracorporeal  
17 317 membrane oxygenation therapy after cardiac surgery revealed BChE as a strong predictor of all-cause  
18 318 and cardiovascular mortality (10).

19 319 In our present study patients with postoperative complications after TAVI had significant lower  
20 320 preoperative levels of BChE compared to the non-complication group. Effect sizes were particularly  
21 321 large for BChE measurements in this homogeneous patient group. In combination with common  
22 322 preoperative evaluation procedures, BChE activity may serve as a useful predictive indicator to identify  
23 323 high-risk patients. Future studies are needed to clarify clinical implications.

24 324 Due to high variability in the onset, aetiology and progress of clinical conditions among patients,  
25 325 determining whether changes in the enzyme activity are correlated with the emergence of disease or  
26 326 are affected by concomitant factors is difficult. John et al. tested the hypothesis that AChE and BChE  
27 327 have an impact on patients after cardiac surgery with postoperative delirium. They showed that AChE  
28 328 increased and BChE decreased within the first 3 days after surgery but did not discern between patients  
29 329 with and without delirium. The authors supposed that the perioperative change of AChE and BChE  
30 330 activity might possibly be explained by an interaction of AChE and BChE and the use of a  
31 331 cardiopulmonary bypass (17). In our present study we evaluated the role of AChE and BChE activity in  
32 332 cardiosurgical patients after TAVI, as a standardized operative procedure without cardiopulmonary  
33 333 bypass. We could show that complication group showed a significantly perioperative decrease of BChE  
34 334 within the first 3 days after TAVI, despite the fact that there was no use of heart-lung machines in our  
35 335 patients. Furthermore, there was no use of blood products in the present study, so we can rule out a  
36 336 possible interaction of AChE and BChE with blood products as well. While in the CESARO study a wide  
37 337 spectrum of operative disciplines has been analysed and the perioperative enzyme activities showed  
38 338 small effect sizes, we can show large effect sizes for BChE in this secondary analysis of a homogeneous  
39 339 patient group with standardized operative procedure.

40 340 BChE activity could be regarded as an inflammatory parameter in this context. In literature, lower  
41 341 levels of BChE activity have already been described during inflammatory processes, stress and  
42 342 malnutrition (9, 11, 22). Therefore, lower levels of BChE activity in complication group might reflect  
43 343 perioperative inflammation, which is known to promote complications like delirium or infections (20,  
44 344 23). Conventional markers like CRP and leucocytes did not differ in both groups.

45 345 Delirium is a complex symptom which is very common in operative and non-operative disciplines in  
46 346 the course of hospital stay. The incidence is especially high among patients undergoing heart surgery  
47 347 (24). The incidence in this patients population has been described to be from 30 up to 80 % (25). The  
48 348 incidence of delirium after TAVI is reported as 29 % in literature (6). Delirium occurred significantly  
49 349 more frequently following TA procedures (26). In the present study 26,7 % of the patients were  
50 350 diagnosed with delirium overall. There was no difference depending on TA or TF approach.

351 Perioperative measurement of AChE and BChE did not discern between patients with and without  
352 delirium, which is in accordance with the findings by John et al.

353 The present study highlights the validity of BChE measurements for early detection of high-risk patients  
354 after TAVI. Surprisingly, the BChE assessment proved more effective than the EuroSCORE in  
355 discriminating between the patient groups making it a valuable biomarker for the early detection of  
356 high-risk patients. EuroSCORE is a well-established clinical assay for the patient mortality analysis (27)  
357 and requires documenting multiple and diverse datasets. The datasets are in most cases readily  
358 available; however, in some cases, a particular set of data might not be accessible, delaying or making  
359 the scoring impossible. By using a POCT system for a single BChE measurement, the results of an  
360 equally efficient outcome assessment tool are readily available at the bedside and may complete  
361 conventional assessments. Further studies with a greater patient population are needed to investigate  
362 the clinical implications.

363 Prompt assessment of the systemic immune response with an immediate, rapid and affordable  
364 bedside measurement of the BChE activity might improve risk evaluation and help optimize  
365 postoperative management and therapy of patients after TAVI. Predicting the length of the hospital  
366 stay might play an important role in staff and resources management for these patients.

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#### 369 Limitations

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371 Limitations of the present study might be the short duration of 3 days' measurement. Blood was taken  
372 from each patient; in case the analysis could not be performed immediately (during anesthesia  
373 induction), the sample was cooled down in a refrigerator. Maybe values of AChE and BChE changed in  
374 combination with lower temperature. Furthermore, it was only one measurement performed with  
375 each sample, so no control values could be achieved.

376 The study protocol required daily cholinesterase activity measurements in the postoperative period,  
377 without specifying time or requesting multiple daily measurements. Therefore, circadian fluctuations  
378 in enzyme activities could not be considered.

379 The biggest limitation of the present study is the low number of included patients and the related  
380 statistical power. Further studies with larger patient groups and with focus on the underlying  
381 mechanisms of the different complications would be needed to validate our findings and the clinical  
382 implications. A larger, possibly multicenter study would be needed to evaluate more postoperative  
383 complications and the roles of BChE and AChE in particular complications.

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Characteristic	Total sample (n = 43)
<b>Age (years) [M (SD)]</b>	79.47 (5.7)
<b>Sex [n (%)]</b>	
Male	22 (51.2)
Female	21 (48.8)
<b>BMI [M (SD)]</b>	27.93 (5.4)
<b>ASA – PS [n (%)]</b>	
3	32 (74.4)
4	11 (25.6)
<b>Operative procedure [n (%)]</b>	
Transapicale TAVI	11 (25.6)
Transfemorale TAVI	32 (74.4)
<b>Relevant comorbidities [n (%)]</b>	
Hypertension	37 (86)
Diabetes	16 (37.2)
Congestive heart failure	36 (83.7)
Congestive kidney failure	20 (46.5)
Coronary heart disease	31 (72.1)
Cardiac arrhythmias	25 (58.1)
Stroke	8 (18.6)
Nicotine	19 (44.2)
Alcohol	8 (18.6)
Hypothyreosis	10 (23.3)
Hypercholesterinemia	14 (32.6)
<b>EuroSCORE [n (%)]</b>	
low	15 (34.9)
middle	18 (41.9)
high	10 (23.3)
<b>Pre-operative anticholinergic drugs</b>	16 (37.2)

Table 1 Description of baseline data; all data are presented as n (number) and (%). ASA, American Society of Anaesthesiologists Classification; BMI, body mass index

Variables	Complication	N	M	SD	p-value	Cohens' r
EuroSCORE	Yes	24	21.8	15.4	0.824	0.034
	no	19	22.8	13.5		
Weight [kg]	Yes	24	78.3	15.5	0.504	0.102
	no	19	74.9	18.0		
BMI [kg/m <sup>2</sup> ]	Yes	24	28.1	4.7	0.860	0.037
	no	19	27.7	6.2		
Age [years]	Yes	24	79.9	5.3	0.556	0.086
	no	19	78.9	6.3		
NT-proBNP [pg/ml]	Yes	24	6244.8	6773.1	0.563	0.099
	no	19	4806.6	7809.4		
Hemoglobin D 0 [g/dl]	Yes	24	12.1	1.9	0.917	0.029
	no	19	12.2	1.4		
Hemoglobin POD 0 [g/dl]	Yes	24	10.7	1.5	0.565	0.068
	no	19	10.9	1.4		
Hemoglobin POD 1 [g/dl]	Yes	24	10.1	1.4	0.986	0.000
	no	19	10.1	1.1		
Hemoglobin POD 2 [g/dl]	Yes	24	10.1	1.1	0.673	0.087
	No	19	10.3	1.2		
Hemoglobin POD 3 [g/dl]	Yes	24	9.6	1.0	0.272	0.173
	No	19	10.0	1.3		
Creatinine D 0 [mg/dl]	Yes	24	1.4	0.7	0.089	0.247
	no	19	1.1	0.4		
Creatinine POD 1 [mg/dl]	Yes	24	1.2	0.4	0.347	0.124
	No	19	1.1	0.4		
Creatinine POD 2 [mg/dl]	Yes	24	1.5	0.8	0.188	0.223
	no	19	1.2	0.4		
Creatinine POD 3 [mg/dl]	Yes	24	1.5	1.0	0.240	0.244
	No	19	1.1	0.4		
Leukocytes D 0 [/nl]	Yes	24	7.8	2.1	0.383	0.131
	no	19	7.3	1.6		
Leucocytes POD 1 [/nl]	Yes	24	9.6	4.3	0.496	0.113
	No	19	8.8	2.1		
Leukocytes POD 2 [/nl]	Yes	24	9.9	2.7	0.616	0.081
	No	19	9.4	3.5		
Leukocytes POD 3 [/nl]	Yes	24	8.8	3.1	0.079	0.336
	No	19	7.0	1.5		
CRP D 0 [mg/l]	Yes	24	16.3	17.8	0.716	0.094
	no	19	19.8	18.2		
CRP POD 1 [mg/l]	Yes	24	31.8	21.1	0.177	0.236
	No	19	22.0	18.9		
CRP POD 2 [mg/l]	Yes	24	116.3	52.9	0.516	0.114
	No	19	101.3	78.2		
CRP POD 3 [mg/l]	Yes	24	115.3	68.5	0.113	0.284
	No	19	72.7	76.0		
BChE D 0 [U/l]	Yes	24	2784.0	534.9	0.118	0.238
	no	19	3072.6	652.1		
BChE D 1 [U/l]	Yes	24	2589.2	556.4	<b>&lt;0.001</b>	0.514
	no	19	3295.7	628.0		
BChE POD 0 [U/l]	Yes	24	2379.6	525.1	<b>&lt;0.001</b>	0.469
	No	19	2972.5	599.2		
BChE	Yes	24	2300.3	561.0	<b>&lt;0.001</b>	0.504



POD 1 [U/l]	No	19	2936.2	523.1		
BChE	Yes	24	2166.7	537.0	<b>0.002</b>	0.462
POD 2 [U/l]	No	19	2713.5	510.6		
AChE	Yes	24	45.0	8.1	0.446	0.118
D 0 [U/gHb]	no	19	43.3	6.0		
AChE	Yes	24	42.0	10.0	0.263	0.172
D 1 [U/gHb]	no	19	39.2	4.7		
AChE	Yes	24	42.9	10.0	0.051	0.295
POD 0 [U/gHb]	No	19	37.6	6.4		
AChE	Yes	24	41.5	9.2	0.196	0.198
POD 1 [U/gHb]	No	19	38.4	5.2		
AChE	Yes	24	41.2	8.2	0.058	0.294
POD 2 [U/gHb]	No	19	36.5	6.8		
CK	Yes	24	189.6	186.8	0.953	0.009
POD 1 [U/l]	No	19	186.1	190.5		
CK-MB	Yes	24	15.5	18.2	0.946	0.012
POD 1 [ng/ml]	No	19	15.9	14.3		
CK-Index	Yes	24	7.2	3.6	0.314	0.174
POD 1	No	19	8.5	3.8		
Stay in hospital [days]	Yes	24	15.2	6.3	<b>0.033</b>	0.325
	No	19	11.1	5.5		
Anticholinergic burden	Yes	24	0.82	1.191	0.229	0.189
	No	19	0.42	0.838		

400 Table 2 Perioperative laboratory markers; all data are presented as number (n) or as mean  $\pm$  SD

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5 403 Figure 1 Timeline of measurements of BChE and AChE: blood samples were taken one day preoperative  
6 404 (D 0), shortly before anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery  
7 405 (POD 1) and two days after surgery (POD 2). If the measurements could not be conducted immediately,  
8 406 the samples have been cooled in a refrigerator and the measurement was performed up to 2 hours  
9 407 later.

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13 409 Figure 2 Time trajectories of BChE activities in TAVI-patients (n = 43). Pre-operative (D0), shortly before  
14 410 anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery (POD 1) and two days  
15 411 after surgery (POD 2) measurements in patients with (dashed) and without (solid) complication. Data  
16 412 are presented as median  $\pm$  standard deviation. \* Difference between groups; # Difference within  
17 413 groups.

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22 415 Figure 3 Time trajectories of AChE activities in TAVI-patients (n = 43). Pre-operative (D0), shortly before  
23 416 anesthetic induction (D 1), on admission to ICU (POD 0), one day after surgery (POD 1) and two days  
24 417 after surgery (POD 2) measurements in patients with (dashed) and without (solid) complication. Data  
25 418 are presented as median  $\pm$  standard deviation.

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3 420 **Contributorship statement**  
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5 421  
6 422 DB, YZ, AM, AH and BM were responsible for study design, statistical analyses and drafting of the  
7  
8 423 manuscript. WP, AB, BM and BG performed the experiments and drafted the manuscript. BM and AM  
9  
10 424 were responsible for statistical analysis. All authors read and approved the final manuscript.  
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15 427 **Competing interests**

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18 429 The authors declare no conflict of interest.  
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23 433 **Funding**

24 434

25 435 The authors have not declared a specific grant for this research from any funding agency in the public,  
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27 436 commercial or not-for-profit sectors.  
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32 440 **Data sharing statement**

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34 442 No additional data are available.  
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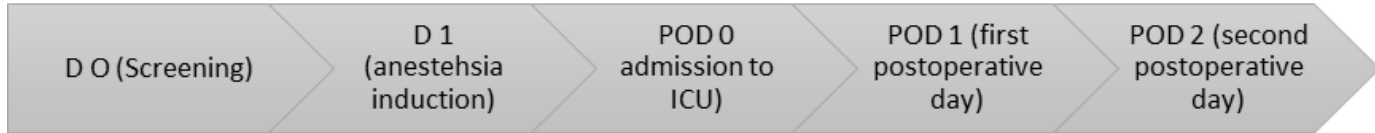


Figure 1

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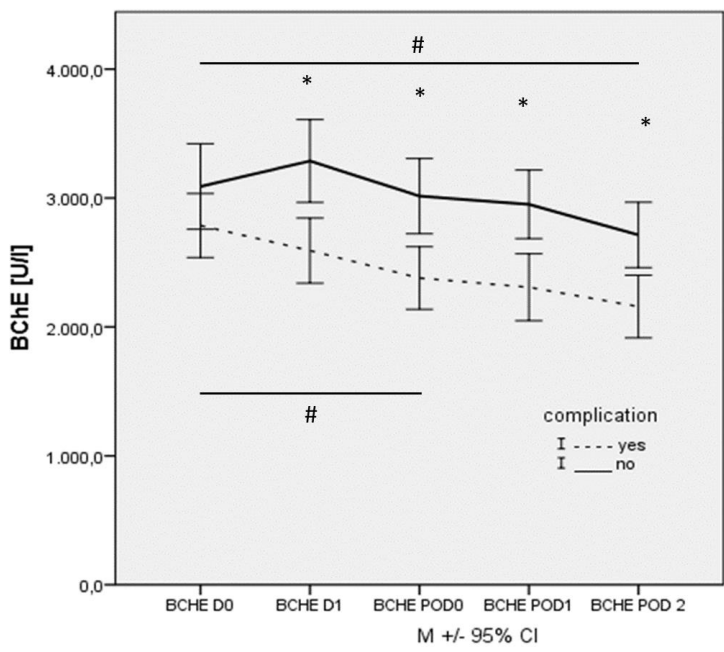


Figure 2

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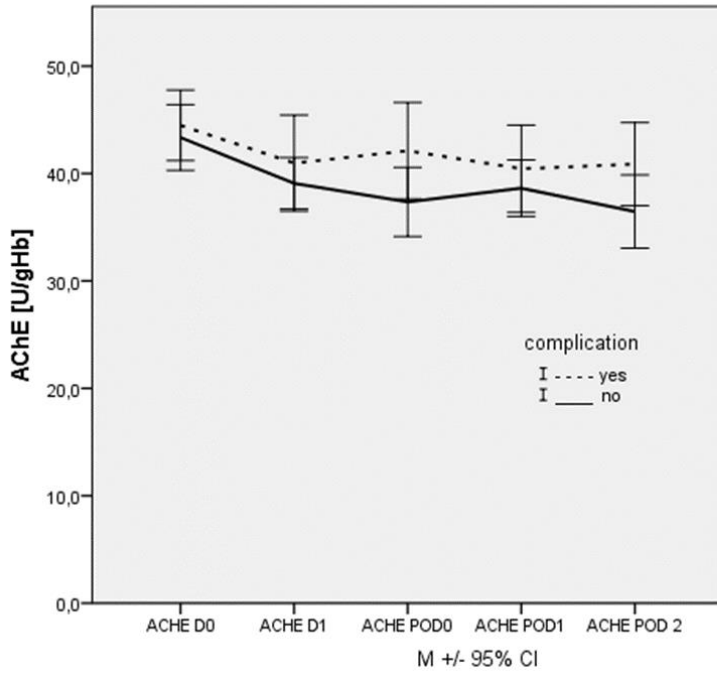


Figure 3

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## STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
<b>Title and abstract</b>	1	(a) Indicate the study's design with a commonly used term in the title or the abstract [line 34-36, page 2] (b) Provide in the abstract an informative and balanced summary of what was done and what was found [line 30-55, page 2]
<b>Introduction</b>		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported [line 73-105, page 4]
Objectives	3	State specific objectives, including any prespecified hypotheses [line 73-105, page 4]
<b>Methods</b>		
Study design	4	Present key elements of study design early in the paper [line 108-119, page 5]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection [line 122-125, page 5]
Participants	6	(a) <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up [line 116-119, page 5] <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants (b) <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable [line 127-180, page 5-6]
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group [line 122-125, page 5]
Bias	9	Describe any efforts to address potential sources of bias [line 108-208, page 5-7]
Study size	10	Explain how the study size was arrived at [line 108-115, page 5]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why [line 127-180, page 5-6]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding [line 211-221, page 7] (b) Describe any methods used to examine subgroups and interactions [line 211-221, page 7] (c) Explain how missing data were addressed [line 170-171, page 6] (d) <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed [line 106-180, page 5-6] <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of

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sampling strategy

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(e) Describe any sensitivity analyses [211-221, page 7]

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60**Results**

Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed [line 225-234, page 8] (b) Give reasons for non-participation at each stage [line 225-234, page 8] (c) Consider use of a flow diagram → removed (as suggested by reviewer)
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders [line 225-234, page 8 and 12] (b) Indicate number of participants with missing data for each variable of interest [225-234, page 8] (c) <i>Cohort study</i> —Summarise follow-up time (eg, average and total amount) [line 230-234, page 8]
Outcome data	15*	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time [line 237-245, page 8] <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included [line 236-302, page 8-9] (b) Report category boundaries when continuous variables were categorized [line 236-302, page 8-9] (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period [line 236-302, page 8-9]
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses [211-221, page 7]

**Discussion**

Key results	18	Summarise key results with reference to study objectives [line 305-366, page 10-11]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias [line 371-383, page 11]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence [line 305-366, page 10-11]
Generalisability	21	Discuss the generalisability (external validity) of the study results [line 305-366, page 10-11]

**Other information**

Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based [line 433-436, page 16]
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\*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

**Note:** An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at [www.strobe-statement.org](http://www.strobe-statement.org).