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ORIGINAL ARTICLE

How Effective is Frailty and Comprehensive Geriatric Assessment to Predict the Long-Term Mortality After General Surgery?

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~ ABSTRACT Com

Introduction: This study investigated the effect of preoperative comprehensive geriatric assessment(CGA) and frailty assessment on long-term mortality.

Methods: This study which evaluated a total of 81 older patients underwent the CGA prior to general surgery. Katz ADL, the Lawton Brody IADL, the Mini-Nutrition Assessment test (MNAsf), the Mini-Mental State Examination (MMSE), and Yesavage Geriatric Depression Scale (GDS) were performed. Fried criteria were utilized for the assessment of frailty. The Physiological and Operative Severity Scores for the Enumeration of Mortality and Morbidity (POSSUM) score, and the Charlson Comorbidity Index (CCI) were used for operative risk assessment. The patients were screened for 3-year mortality.

Results: The median age of the patients was 71 years (range, 65-84 years). 58.02% of the patients were female and 24.69% were in the frail group. The mortality rate of the frail group was significantly higher than those of the pre-frail and robust groups (p: 0.030). The Cox regression analyses revealed that MMSE (p: 0.020), Physiological Severity Score (PSS) (p: 0.034), BUPA score (p: 0.030) and educational background (p: 0.031) were independently correlated with mortality in Model 1, while MNA (p: 0.003), PSS score (p: 0.080) and educational background (p: 0.002) were correlated with mortality in Model 2. ADL, MMSE, CDT, MNA-SF, Fried score, length of hospital stay, PSS score, and BUPA score were the best predictors of mortality (AUC values: 0.61, 0.74, 0.72, 0.73, 0.69, 0.74, 0.64, and 0.66respectively).

Conclusion: The results of the study demonstrated that CGA components and frailty predicted long-term mortality in general surgery patients.

Keywords: comprehensive geriatric assessment, frailty, long-term mortality, general surgery.

INTRODUCTION

Frailty is a geriatric syndrome characterized by a physiological decline in multiple systems and increased vulnerability to stress factors and adverse clinical outcomes. Although the use of frailty as a medical syndrome and as a measure of decreased physiological reserve is well known, there is no gold standard definition of frailty universally used in the clinic. A compilation by Lin et al. evaluated 23 studies. These studies show that 21 different scales were used to measure frailty. There is strong evidence of an association between frailty and increased 30-90-day and 1-year mortality, postoperative complications, and prolonged length of hospital stay [1]. Given the significance of general surgery as a common and main therapeutic intervention in older patients along with its risk of complications and other adverse clinical outcomes, it is critical to develop reliable risk stratification tools that will appropriately guide clinicians and patients in medical decision-making. A critical first step for achieving this goal is to determine whether frailty, a measure of physiological reserve and vulnerability in older patients, is predictive of adverse clinical outcomes after general surgery [2].

Improved perioperative care and medical advances make older adults eligible for surgery. Routine determination of preoperative requirements does not provide the information required to predict outcomes and optimal and specific treatment. Moreover, older patients are still underrepresented in clinical trials, and the results of studies evaluating young or only fit older patients cannot be directly predicted for all older patients. Comprehensive geriatric assessment is used for accurate and versatile evaluation of older patients. Besides others, it allows for the initial assessment of the patient's condition, identification of previously unknown health problems, diagnosis of vulnerability, and assessment of the likelihood of complications. Frailty, not chronological age, is the most important preoperative risk factor for poor surgical outcomes in the older population [3]. As a result of studies showing that frailty predicts postoperative complications, its effect on mortality aroused curiosity. Comprehensive geriatric assessment (CGA) allows clinicians to accurately assess the preoperative health status of older patients and focuses not only on somatic domains but also on functional, nutritional, and psychosocial

domains. It helps uncover impairments that are not documented by routine medical evaluation [1]. Due to advances in surgical protocols and the use of less invasive techniques and surgical procedures, there is an increasing need for new interventions to prevent postoperative complications, especially in older patients [4].

There is growing interest in using the time to surgery to prevent postoperative complications and reduce mortality rates in patients undergoing abdominal surgery. "Prehabilitation" aims to optimize patients' basic health and functional capacity to alleviate the impact of a stressor before surgery. As all these factors are independently associated with the number of postoperative complications, multimodal prehabilitation programs have been developed to simultaneously optimize multiple domains [5]. A study of patients undergoing elective major abdominal surgery showed a reduction in postoperative complications in prehabilitated patients [6]. In addition, a meta-analysis of data from 15 prehabilitation studies showed a significant reduction in overall and pulmonary morbidity in patients undergoing elective major abdominal surgery [7].

The surgical procedure itself is the next important elementthataffectsthefinalresult.Somemeasurable intraoperative factors may predict postoperative complications. Some scoring systems have been designed to help quantify the risk of postoperative complications. However, these tests are extremely complex and require a large number of data items, which limits their use in daily life [8]. Given the high rate of postoperative complications, there is a need for tools to accurately and reliably identify risks in an older population who are vulnerable to adverse surgery-related sequelae. This study aimed to examine the effect of preoperative CGA and frailty assessment on long-term mortality and to demonstrate its relationship with other operative risk scorings used for preoperative assessment.

MATERIALS AND METHODS

Study population

The study included a total of 81 patients over the age of 65 who presented to the geriatric

outpatient clinic between January 2017 and August 2018 and were scheduled for elective general surgery (Cholecystectomy, bile duct excision, liver segment tumor resection, modified radical mastectomy, esophagectomy, gastrectomy, cholecystoenterostomy, Whipple, right hemicolectomy, rectosigmoid mass resection, colectomy, hemicolectomy, small bowel resection+enterostomy, adrenalectomy, incisional hernia repair, parathyroidectomy). The 3-year mortality follow-up of the general surgery patients of our previous study was performed [3]. The exclusion criteria included patients who did not want to participate in the study and who had communication problems so much that CGA could not be performed, and emergency surgery, day surgery, surgery under local anesthesia, and palliative surgery. Age, gender, body mass index (BMI), smoking and alcohol use, co-living individuals, educational background, comorbidities, incontinence, falls, and the number of medications were recorded. The type of surgery and 30-day complications were obtained from the records. Complications were defined as any event occurring within 30 days of surgery that required treatment not routinely administered in the postoperative period.

Comprehensive geriatric assessment

A comprehensive geriatric assessment was preoperatively performed by a geriatrician. Functional status assessment, a component of the CGA, was performed using the Katz Activities of Daily Living (ADL) and Lawton Instrumental Activities of Daily Living (IADL) scales [9-11]. The Folstein Mini-Mental State Examination (MMSE) and the Clock Drawing Test (CDT) were used for cognitive status assessment [12-14]. Generally, the MMSE score higher than 24 points is thought to indicate good cognitive performance. The CDT score ranges from 0 to 6 points and the point <4 in CDT was accepted to show low cognitive performance. Mood assessment, another component of the CGA, was performed using the short form of the Yesavage Geriatric Depression Scale (GDS) [15, 16]. A score of five or more is considered clinically significant for depression. The Mini Nutritional Assessment-Short Form (MNA-SF) was used for the nutritional status assessment where the total score of ≤ 11 was described as the risk of malnutrition [17, 18].

The frailty assessment of the patients was carried out using Fried criteria. In the phenotype model described by Fried et al., frailty is characterized by 5 clinical features: unintentional weight loss, exhaustion, weakness, slow walking speed, and low physical activity. According to these criteria, patients with a score of 3 or more are evaluated as 'frail', 1 or 2 points as 'pre-frail', and 0 points as 'robust' [19, 20].

Preoperative risk assessment with operative scores

Operative risk assessment was performed preoperatively with the Physiological and Operative Severity Score for the Enumeration of Mortality and Morbidity (POSSUM), the American Society of Anesthesiologists (ASA), and the British United Provident Association (BUPA) [21-23]. POSSUM, which is recommended to predict postoperative morbidity and mortality, consists of components of the Physiological Severity Score (PSS) and the Operative Severity Score (OSS). While PSS is based on 12 factors including preoperative measurements such as laboratory results, age, and cardiac status, OSS is calculated using 6 intraoperative factors such as operative severity, number of procedures, total blood loss, and peritoneal contamination. The ASA classification, a well-known scoring system, was developed to offer clinicians a simple classification of a patient's physiological status that can help predict operative risk with a score from 1 to 5. The British United Provident Association evaluates the operative severity. An increase in postoperative risk is shown with an increase in surgical scores. The Charlson Comorbidity Index (CCI), which includes 19 comorbidity parameters, was used for the risk assessment of medical comorbidity burden [24].

Delirium assessment

Delirium was assessed by а geriatrician preoperatively and on postoperative days 3 and 7. The 4AT test was used for delirium assessment [25]. 4AT is a delirium assessment tool that includes alertness, AMT4 (age, date of birth, place, current year), attention, acute change, or fluctuating course questions. A score of 4 or above is considered 'possible delirium +/- cognitive impairment', 1-3 points as 'possible cognitive impairment', and 0 points as 'delirium or severe cognitive impairment unlikely'.

Mortality evaluation

Postoperative length of stay (LOS) was defined as the number of days from surgery to discharge. The 3-year mortality of the patients was recorded by searching the death notification system.

Ethics

The study protocol was evaluated and approved by the Local Ethics Committee. Informed consent was obtained from each patient before participating in the study.

Statistical analyses

SPSS version 22.0 was used for statistical analyses. Descriptive statistical results were presented as frequencies and percentages for categorical variables. Numerical parameters for the normal distribution were analyzed by histogram and Kolmogorov-Smirnov tests. Normally distributed continuous parameters were presented as mean ± SD, while skewed parameters were presented as median (minimum-maximum). Comparison of categorical variables was carried out with Chisquare or Fisher exact tests as appropriate. Normally distributed continuous variables were evaluated with Student's t-test, while skewed variables were evaluated with the Mann-Whitney U test. A p-value less than 0.05 was considered statistically significant. Parameters with a significant difference in univariate analyses or a p-value less than 0.20 were included in Binary Logistic Regression analysis to determine parameters independently correlated with mortality.

Kaplan-Meier survival estimates were calculated. A ROC (Receiver Operating Characteristics) curve analysis was used to determine the ability of relevant factors to predict mortality. In the case of observation of a significant threshold value, the area under the curve (AUC) values, sensitivity, specificity, and positive and negative predictive values were presented.

RESULTS

A total of 81 patients were included in the study. The median age of the patients was 71 years (range, 65-84 years) and 58% of the patients were female. The median time from initial admission to the determination of the survival status of the patients was 53.0 (range, 0.1-69.5) months. The rates of frail, pre-frail, and robust patients were 24.69%, 44.44%, and 30.86%, respectively. The categorization of the patients into two groups as survivors and nonsurvivors revealed a significantly higher frequency of frailty in non-survivors than in survivors (p: 0.022). The CGA components of Katz ADL (p: 0.034), CDT (p<0.001), MMSE (p<0.001), and MNA-SF (p<0.001) scores were significantly lower in the non-survivor group. The operative risk scores of PSS (0.035) and BUPA (p:0.015) were higher in the non-survivor group. The analysis of comorbidities showed more frequent mortality in Parkinson's disease (p: 0.018) and dementia patients (p: 0.018). General characteristics, CGA test scores, and operative risk scores by groups are presented in Table 1.

According to Fried Criteria, the mortality rate was significantly higher in the frail group than in the pre-frail and robust groups (84%, 61.14%, and 45.02% respectively, p:0.030) (Figure 1).

Models were created in Cox regression analyses. MMSE (OR: 0.932, 95% CI: 0.879-0.989, p: 0.020), PSS (OR: 1.130, 95% CI: 1.009-1.265, p: 0.034), BUPA score (OR: 1.463, 95 %CI: 1.038-2.062, p: 0.030) and educational background (OR:0.332, 95%CI:0.123-0.902, p:0.031) were independently correlated with mortality in Model 1, whereas MNA (OR: 0.809, 95% CI: 0.704-0.930, p: 0.003), PSS score (OR: 1.106, 95% CI: 0.988-1.237, p: 0.080) and educational background (OR: 0.241, 95% CI: 0.096-0.606, p: 0.002) were correlated with mortality in Model 2. Independently correlated factors of mortality are presented in Table 2.

Katz ADL, MMSE, CDT, MNA-SF, Fried score, LOS, PSS score, BUPA score, and 4AT score were the best predictors of mortality (AUC values: 0.61, 0.74, 0.72, 0.73, 0.69, 0.74, 0.64, 0.66 and 0.58, respectively). The cut-off, AUC, sensitivity, specificity, and positive and negative predictive values of the best predictive factors of mortality are presented in Table 3.

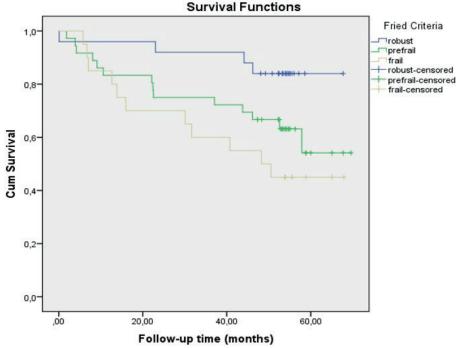
DISCUSSION

This study evaluated the correlation between the CGA components and frailty and postoperative long-term mortality. The results of the study demonstrated an independent correlation between the CGA components, namely MMSE, MNA, frailty, and long-term mortality. The evaluation of operative

	Total (n=81)	Survivor (n=52)	Non-survivor (n=29)	р
Age, year, median (min-max)	71 (65-84)	72 (65-84)	70 (65-84)	0.250
Female, n (%)	47 (58.02)	33 (63.46)	14 (48.28)	0.184
Education, n (%)				0.006
Primary school and lower	47 (58.02)	24 (46.15)	23 (79.31)	
Secondary school and higher	32 (39.51)	26 (50)	6 (20.69)	
BMI, kg/m², median (min-max)	27.24 (18.82-43.14)	28.23 (20.15-43.0)	26.67 (18.82-43.14)	0.197
Smoking, n (%)				0.187
No	55 (67.90)	39 (75)	16 (55.17)	
Yes	16 (19.75)	8 (15.38)	8 (27.59)	
Ex-smoker	10 (12.35)	5 (9.62)	5 (17.24)	
Presence of weight loss, n (%)	41 (50.62)	21 (40.38)	20 (68.97)	0.014
Hypertension, n (%)	49 (60.49)	33 (63.46)	16 (55.17)	0.464
Diabetes Mellitus, n (%)	22 (27.16)	15 (28.85)	7 (24.14)	0.648
Coronary artery disease, n (%)	19 (23.46)	10 (19.23)	9 (31.03)	0.229
Congestive heart failure, n (%)	2 (2.47)	0 (0)	2 (6.90)	0.055
Chronic obstructive pulmonary disease, n (%)	7 (8.64)	4 (7.69)	3 (10.34)	0.684
Malignancy, n (%)	45 (55.56)	26 (50)	19 (65.52)	0.178
Dementia, n (%)	3 (3.75)	0 (0)	3 (10.34)	0.018
Depression, n (%)	8 (9.88)	7 (13.46)	1 (3.45)	0.148
Parkinson's disease, n (%)	3 (3.70)	0 (0)	3 (10.34)	0.018
History of falls, n (%)	19 (23.46)	11 (21.15)	8 (27.59)	0.512
Urinary incontinence, n (%)	21 (25.92)	12 (23.08)	9 (31.04)	0.433
Number of medications, n, median (min-max)	3 (0-10)	3 (0-10)	3 (0-9)	0.8500
Katz ADL, median (min-max)	6 (2-6)	6 (3-6)	6 (2-6)	0.034
Lawton IADL, median (min-max)	8 (0-8)	8 (0-8)	8 (1-8)	0.084
CDT, median (min-max)	6 (0-6)	6 (0-6)	3 (0-6)	<0.001
MMSE, median (min-max)	28 (2-30)	29 (20-30)	26 (2-30)	<0.001
MNA-SF, median (min-max)	12 (4-14)	12 (7-14)	10 (4-14)	<0.001
Yesavage GDS, median (min-max)	2 (0-11)	2 (0-8)	3 (0-11)	0.071
Handgrip, kg, mean	23.19±7.17	23.14±6.98	23.27±7.61	0.617
Length of stay, median (min-max)	8 (2-63)	6 (2-63)	11 (3-39)	<0.001
Postoperative morbidity	24 (29.63)	12 (23.08)	12 (41.38)	0.084
Presence of delirium, n (%)	2 (2.47)	1 (1.92)	1 (3.45)	1.000
Fried score, median (min-max)	2 (0-4)	1 (0-4)	2 (0-4)	0.003
Fried group, n (%)				0.022
Robust	25 (30.86)	21 (40.38)	4 (13.79)	
Pre-frail	36 (44.44)	22 (42.31)	14 (48.28)	
Frail	20 (24.69)	9 (17.31)	11 (37.93)	
PSS, median (min-max)	19 (13-29)	18 (13-28)	20 (15-29)	0.035
OSS, median (min-max)	10 (6-31)	9 (6-31)	13 (6-31)	0.127
ASA, median (min-max)	2 (1-3)	2 (1-3)	2 (1-2)	0.882
BUPA, median (min-max)	3 (2-5)	3 (2-5)	3 (2-5)	0.015**
4AT, median (min-max)	0 (0-3)	0 (0-3)	0 (0-3)	0.013**
CCI, median (min-max)	5 (2-11)	5 (2-11)	5 (2-9)	0.533
Follow-up time, months, mean	53.03 (0.1-69.5)	54.24 (47.3-69.5)	22.43 (0.1-57.8)	<0.001

*BMI: Body Mass Index, ADL: Activities of Daily Living, IADL: Instrumental Activities of Daily Living, CDT: Clock Drawing Test score, MMSE: Minimental State Examination, MNA-SF: Mini-Nutritional Assessment-Short Form, GDS: Geriatric Depression Scale, PSS: Physiological Severity Score, OSS: Operative Severity Score, ASA: American Society of Anesthesiologists, 4AT: Assessment Test for Delirium, BUPA: British United Provident Association, CCI: Charlson Comorbidity Index

** 95% CI value for BUPA is 0.023-0.208 and for 4 AT is 0.017-0.331.



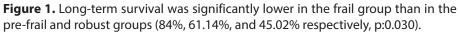


Table 2. Independent	y correlated factors	of mortality
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	OR	CI	р			
Model 1						
MMSE score	0.932	0.879-0.989	0.020			
PSS score	1.130	1.009-1.265	0.034			
BUPA score	1.463	1.038-2.062	0.030			
Educational background	0.332	0.123-0.902	0.031			
Model 2						
MNA-SF score	0.809	0.704-0.930	0.003			
PSS score	1.106	0.988-1.237	0.080			
Educational background	0.241	0.096-0.606	0.002			

*MMSE: Mini-mental State Examination, PSS Physiological Severity Score, BUPA: British United Provident Association, MNA-SF: Mini-Nutritional Assessment-Short Form

Model 1: In this model, the parameters with a significant correlation between survivors and non-survivors in the univariate analyses (ADL, MMSE, length of stay, PSS score, BUPA score, 4AT score, educational background, Fried score) were included in the multivariate logistic regression analysis. The backward stepwise method was used. The last step (step 5) was presented in the table. The Omnibus test in this model yielded a p-value <0.001 and the chi-square test yielded 24.975. In this model, the -2 log-likelihood was 214.333.

Model 2: In this model, the parameters with a significant correlation between survivors and non-survivors in the univariate analyses (ADL, MMSE, length of stay, PSS score, BUPA score, 4AT score, educational background, MNA-SF) were included in the multivariate logistic regression analysis. The backward stepwise method was used. The last step (step 6) was presented in the table. The Omnibus test in this model yielded a p-value <0.001 and the chi-square test yielded 25.185. In this model, the -2 log-likelihood was 213.352.

risk scoring for long-term mortality together with CGA comprised the superiority of this study over other studies. Cut-off values of factors associated with long-term mortality were determined by ROC analysis.

Operative risk assessment for older adults has historically focused on age and pre-existing medical comorbidities [26]. There are many operative risk classification tools used by clinicians. However, the estimation accuracy of these tools is highly variable among different patients in different populations, different surgical indications, procedures, and age groups. One possible explanation for the limitations of these risk stratification strategies in assessment may be their failure to catch the physiological compromise typical of older adults. Therefore, the ability to better quantify the physiological reserve of older patients in preoperative risk assessment may be key to recovery [2].

	Cut-off	AUC (95% CI)	Sensitivity %	Specificity %	PPV %	NPV %	P-Value
Katz ADL	≤5	0.61 (0.49-0.72)	41.38	78.85	52.24	70.77	0.044
MMSE	≤28	0.74 (0.63-0.84)	79.31	60	53.53	83.38	<0.001
CDT	≤5	0.72 (0.61-0.82)	72.41	68	56.84	81.02	<0.001
MNA-SF	≤11	0.73 (0.62-0.83)	75.86	65.38	55.02	82.91	<0.001
Fried score	>1	0.69 (0.58-0.79)	75.86	59.62	51.23	81.64	0.001
Length of stay (days)	>5	0.74 (0.64-0.84)	89.66	46.15	48.17	88.93	<0.001
PSS	>17	0.64 (0.53-0.75)	86.21	36.54	43.18	82.64	0.025
BUPA	>2	0.66 (0.54-0.75)	79.31	48.08	46.04	80.64	0.010
4AT	>0	0.58 (0.46-0.69)	17.24	98.08	83.33	68.07	0.040

Table 3. Results of ROC curve analyses for the best predictors of all-cause mortality

*ADL: Activities of Daily Living, MMSE: Mini-mental State Examination, CDT: Clock Drawing Test Score, MNA-SF: Mini-Nutritional Assessment-Short Form, PSS: Physiological Severity Score, BUPA: British United Provident Association, 4AT: Assessment Test for Delirium

The preoperative frailty assessment of the patients who underwent elective surgery in the present study showed significantly higher 3-year mortality in the prefrail and frail groups. Another study reported frailty as the best predictor of 30-day and 12-month mortality in older patients with cancer undergoing elective abdominal surgery for curative purposes [27]. Hall D et al. demonstrated the clinical benefit of the screening tool applied in the preoperative decision process of 9153 patients undergoing various surgical procedures. A significant reduction in mortality was observed at 30, 180, and 365 days after surgery by clinicians conducting a detailed preoperative assessment of patients and modifying their perioperative plans [28]. Another study evaluating three different frailty screening methods in patients undergoing elective abdominal surgery found frailty to be associated with 90-day mortality. The comparison of non-frail patients with frail patients using the FRAIL scale, Frailty index, and Clinical Frailty Scale revealed significantly higher 90-day mortality in frail patients [29]. The superiority of the present study over these studies is the longer follow-up period and assessment with operative risk scoring systems. The benefit of frailty assessment must go beyond its role in preoperative risk stratification. Identification of frailty in the geriatric patient scheduled for surgery should trigger the initiation of a series of interventions that can reduce morbidity and increase postoperative functional recovery [30].

Another study evaluating older cancer patients undergoing high-risk abdominal surgery found ADL, CDT, and frailty as valid predictors of 12-month mortality [31]. Numerous studies have confirmed that the functional domain is of great importance in predicting postoperative outcomes. In the present study, the failure of ADL and IADL to predict long-term mortality may be due to the generally good functional status of the included patients (43.2 patients had an ASA score of 1, and 55.5% had an ASA score of 2). The results of the present study showed an independent correlation between MMSE and mortality. Similarly, the study of Schmidt et al. evaluating 131 older surgical patients reported that a decrease in the MMSE score predicted 1-year mortality [32]. The univariate Cox regression analysis of another study revealed that cognitive impairment, which was measured using MMSE scores adjusted for age and education, increased the mortality risk [33]. The present study demonstrated an association between MNA in Model 2 and long-term mortality in the models created for factors independently correlated with mortality. In this study, it was shown that CGA performed before surgery will affect postoperative results. Interventions for patients who are in the risk group with MNA before surgery may cause a decrease in postoperative mortality. There is no nutritional assessment and general surgery longterm mortality study in the literature including MNA. Therefore, the result of this present study adds new information to the literature.

Ellis et al. defined CGA as a multidimensional diagnostic and therapeutic process that focuses on identifying the medical, functional, mental, and social abilities and limitations of a frail older person to ensure that problems are appropriately identified, measured, and managed [34]. Comprehensive geriatric assessment allows for appropriate preoperative examination, identification of age-related vulnerability domains that may be overlooked in routine clinical evaluation, and their preoperative modification. At the same time, it fully

supports the joint decision-making process with the patient and their relatives before the operation. The aim of treatment for older patients is not only to prolong life but more importantly, to restore the preoperative functional level of the patient in the postoperative period.

The limitations of this study include a study sample that may not be characteristic of the general population and a small number of patients. Due to the small number of patients in the study, the mortality-related causes of the subgroups could not be studied by classifying them according to the types of surgery. Many studies have evaluated only CGA or surgical risk assessment scores alone and many studies have examined the association with short-term mortality. The strength of the study is that it is the first study examining the well-known CGA and operative risk assessments along with a long follow-up period (3 years).

In conclusion, the results of the study showed that the CGA components and frailty predicted long-term mortality in general surgery patients. Although further research is needed, CGA and frailty assessment offer geriatric patient-focused perioperative and postoperative management in older adults undergoing surgery, determination of patient-centered clinical care pathways, and use of interdisciplinary care models as a comprehensive intervention.

Author contribution

Study conception and design: RTD, ABD, and BBD; data collection: RTD, ABD, HC, CB, GSA, CS and AK; analysis and interpretation of results: RTD, MCK and BBD; draft manuscript preparation: RTD, BBD, MH and MC. All authors reviewed the results and approved the final version of the manuscript.

Ethical approval

The study was approved by the Hacettepe University (Protocol no. GO 22/547 / 31.05.2022).

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Conflict of interest

The authors declare that there is no conflict of interest.

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