Impact of modified short-term fasting and the combination with a fasting supportive diet during chemotherapy on the incidence and severity of chemotherapy-induced toxicities in cancer patients – a randomised controlled cross-over pilot study

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Abstract
Background This pilot trial aimed to investigate whether modified short-term fasting (mSTF) reduces the incidence of chemotherapy-induced toxicities and whether an initial ketogenic diet (KD) as fasting supportive diet reduces the fasting-related discomfort and increases the compliance.

Methods In this randomised controlled cross-over trial, gynaecologic cancer patients receiving chemotherapy with a minimum of 4 cycles were randomised to mSTF for 96 h during half of chemotherapy cycles and to consume a normocaloric diet during the other chemotherapy cycles. The caloric intake during mSTF was restricted to 25% of each patient's daily requirement. In addition, half of the patients should eat a 6-day normocaloric KD prior to each mSTF period to investigate the hunger-suppression effect of a KD. Chemotherapy-induced toxicities, fasting-related discomfort, body composition, quality of life, laboratory values, and compliance were assessed on each chemotherapy.

Results Thirty patients (30-74 years) completed the study. During mSTF frequency and severity scores of stomatitis [-0.16 ± 0.06; 95% CI -0.28 - (-0.03); P = 0.013], headaches [-1.80 ± 0.55; 95% CI -2.89 – (-0.71); P = 0.002], weakness [-1.99 ± 0.87; 95% CI -3.72 – (-0.26); P = 0.024] and the score of total toxicities were significantly reduced [-10.36 ± 4.44; 95% CI -19.22 – (-1.50); P = 0.023]. Additionally, significant fewer postponements of the chemotherapy were observed post-mSTF, reflecting an improved tolerance of chemotherapy [-0.80 ± 0.37; 95% CI -1.53 – (-0.06); P = 0.034]. A significant reduction of mean body weight by -0.79 ± 1.47 kg during mSTF could not be compensated and remained until the end of study ( P <0.005). On average, Insulin [-169.4 ± 44.1; 95% CI -257.1 – (-81.8); P <0.001] and Insulin-like growth factor 1 levels [-33.3 ± 5.4; 95% CI -44.1 – (-22.5); P <0.001] significantly decreased during fasting. The KD as fasting supportive diet could neither reduce fasting-related discomfort nor increase compliance of our fasting regime.

Conclusion MSTF is safe and feasible in gynaecologic cancer patients. The results indicate that mSTF during chemotherapy can reduce chemotherapy-induced toxicities and increase the tolerance of
chemotherapy. Larger clinical trials are required to recommend mSTF for cancer patients.

Background

Periods of intentional fasting are practiced worldwide, usually for traditional, cultural, or religious reasons. Fasting is defined as the partial or total cessation of food intake for a specific period. There is ample empirical and observational evidence that medically supervised modified fasting lasting one to three weeks is effective in the treatment of several chronic and acute diseases, eg, rheumatism, hypertension, and metabolic syndrome (1). Recent animal experiments revealed that short-term fasting (STF) prior to high-dose chemotherapy decreases chemotherapy-induced toxicities dramatically without affecting the therapeutic effect (2). Acute chemotherapy-induced toxicities could lead to premature discontinuation and dose reduction in chemotherapy, both are risk factors for an inferior therapeutic outcome. Adjuvant metabolic nutritional therapies during cancer treatment such as STF or a ketogenic diet (KD) are currently discussed and promoted in major public newspapers, telecasts, and online, making it a popular topic.

In 2012, Safdie et al. (3) demonstrated that fasting for 48 h sensitised murine, rat, and human glioma cells, but not primary mixed glia cells, to chemotherapy. In the same year, Lee et al. (4) demonstrated that treatment under fasting conditions sensitised 15 of 17 mammalian cancer cell lines to chemotherapeutic agents and was as effective as chemotherapeutic agents in delaying the progression of different tumours. In mouse models of neuroblastoma, fasting cycles plus chemotherapy drugs - but not either treatment alone - resulted in long-term cancer-free survival (3).

A recent article by Brandhorst et al. (2) described stress resistance in mice fed either an ad libitum standard diet or macronutrient-defined dietary restriction for three days or 60 h fasting prior to high-dose doxorubicin treatment. In contrast to the ad libitum-fed mice, the great majority of fasted (60 h) mice survived to day 25 after chemotherapy (16% vs. 89%). Furthermore, they reported that the mice in the STF group showed no visible signs of stress or pain, whereas the ad libitum-fed mice exhibited signs of toxicity including reduced mobility, ruffled hair, and hunched-back posture. Raffaghello et al. (5) investigated whether fasting for 48–60 h before etoposide treatment would enhance resistance in mice. Only one mouse out of 28 in the STF group died. By contrast, of the 37 mice treated with
etoposide alone, 20 died of toxicity.

Fasting induces wide-ranging changes in metabolic pathways and cellular processes, including decreases in circulating insulin-like growth factor-1 (IGF-1) and glucose. This affects different oncogenes including RAS and the AKT signalling pathway and leads to downregulation of proliferation and cell growth (6). Cell culture experiments showed that healthy cells are protected from treatment toxicity while tumour cells become more vulnerable to chemotherapy during short-term fasting. This phenomenon is described as differential stress resistance. Normal cells enter an alternate state characterised by reduced or a lack of cell division and resistance to multiple stresses, upregulation of DNA repair and induced autophagy. Tumour cells are unable to activate a protective response and growth pathways keep persistently overactivated. Hence, tumour cells are more sensitive to chemotherapy (4–7).

In a case series report published in 2012 (8), patients who fasted voluntarily during chemotherapy proved the feasibility and safety of various fasting regimens and reported significantly less chemotherapy-induced side effects, including asthenia, fatigue and gastrointestinal problems such as vomiting and diarrhoea. In the patients whose cancer progression could be followed, there was no evidence that fasting protected tumours or interfered with chemotherapeutic potency. De Groot et al. (9) published the first randomised controlled pilot trial evaluating feasibility and effects of STF on tolerance of adjuvant chemotherapy in HER2-negative breast cancer patients (N=13), demonstrating that a fasting period of 48 h during chemotherapy was well tolerated and reduced both bone marrow toxicity and chemotherapy-induced DNA damage in peripheral blood mononucleated cells, while no differences in non-haematological toxicities were found. Bauersfeld et al. (10) found that STF during chemotherapy was safe and well tolerated, and seemed to improve quality of life and fatigue in a pilot trial with 34 gynaecological cancer patients. In 2016 Dorff et al. (11) published the results of a dose-escalation fasting-study where 20 patients with advanced cancer receiving platinum-based combination chemotherapy fasted for 24, 48 and 72 h during chemotherapy administration. They found that fasting for 72 h was safe and feasible for cancer patients and that fasting-related toxicities were limited to ≤grade 2. Subjects who fasted for 48 hours or longer had reduced DNA damage in
leukocytes and IGF-1 levels decreased in all fasting cohorts. Michalsen et al. (12) published a non-randomised trial with 209 patients suffering from chronic pain, investigating the impact of STF on common side effects and well-being. There were no serious side effects, however a slight drop in mood and well-being on the first days in combination with feeling of hunger were reported. Typical complaints reported in the initial fasting period at a level that did not interfere with daily activities include hunger, tiredness, irritability, headache, and light-headedness (8,12,13). Fasting related discomforts occur especially on the fasting days 2 and 3, when metabolism is changes to physiological ketosis (13). Fasting leads to strong neuroendocrine adaptations that resemble the metabolic responses to a KD (14). A KD involves the intake of a high-fat, adequate-protein and very low- carbohydrate regimen (<30-40 g/day) and induces a metabolic condition called “physiological ketosis”, namely the state of elevated levels of ketone bodies in the blood (15). Previous studies revealed a hunger-suppression effect of ketosis, although the mechanisms of action of ketosis on appetite reduction are still not completely understood (16-18). We performed this pilot study to evaluate the influence of 96 h-fasting on chemotherapy-induced toxicities in patients with gynaecological cancer. Furthermore, quality of life, fatigue, fasting-related discomfort, compliance, nutritional status and laboratory values are assessed. For the first time it was investigated whether a KD as a fasting supportive diet is able to reduce the fasting-related discomfort during the first days and increase the compliance of our fasting regimen.

Methods

Study population

Adult women with first diagnosis or first recurrence of histologically confirmed gynaecologic cancers in all stages receiving neoadjuvant or adjuvant chemotherapy with a minimum of 4 cycles of the same chemotherapy protocol at a 3- to 4-week interval administrated within 24 h were recruited. Exclusion criteria included present malnutrition (Nutritional Risk Screening >3, weight loss >5% in the last 3 months, body mass index <18.5 kg/m²), eating disorders, diabetes mellitus undergoing drug therapy, gout, severe cardiovascular diseases, pregnancy or lactation, parenteral nutrition, administration of steroids or IGF-1-receptor blockers. Patients were enrolled between March 2017 and December 2017
at the Department of Gynecology and Gynecologic Oncology of the University Medical Center Freiburg by employees of the Department of Nutritional Medicine and Dietetics. All study participants gave their written informed consent. The study protocol was reviewed and approved by the Ethics Committee of Albert-Ludwig University Freiburg (313/16). The study was registered at germanctr.de as DRKS00011610.

**Study design and intervention**

Figure 1 Randomised crossover study design to test the effect of modified short-term fasting (mSTF) during 2 or 3 cycles of chemotherapy (CTX) depending on CTX regimen on CTX-induced toxicities compared to a normocaloric diet (NC) during 2 or 3 cycles of CTX as control and to test the effect of a normocaloric ketogenic diet (KD) followed by mSTF on fasting related discomfort compared to mSTF alone.

This pilot study had a randomised, interventional, open-label cross-over design with a dietary intervention. The primary objective of this trial was to evaluate the influence of modified short-term fasting (mSTF) on chemotherapy-induced toxicities in gynaecologic cancer patients on the basis of the probability of adverse events grade III and higher. The dietary intervention consisted of an mSTF or a 6-day normocaloric KD prior to each mSTF. Patients were randomly assigned to one of the four study groups: A, B, C, D (Figure 1). All study groups included 2-3 periods of mSTF or mSTF combined with KD and normocaloric diet (NC), respectively. Patients in study groups A and C started with mSTF or KD prior to mSTF for the first half of chemotherapy cycles, whereas the groups B and D began with NC. For statistical analysis we mainly compared cycles of NC versus cycles of mSTF. Furthermore, we compared cycles of mSTF versus cycles of mSTF combined with KD. All patients entered the preparation period after randomisation, which consisted of individually teaching units and the development of an individual diet plan for the 4 day- mSTF period by a skilled nutritional scientist. One day before each dietary intervention period, patients received a reminder by phone or by email. In periods between the dietary interventions close to the chemotherapy cycles, patients were on their usual NC. During mSTF, the level of caloric restriction was limited to 25% of each patient’s daily calorie
requirement, consisting of the resting energy expenditure multiplied by a factor that corresponds to the individual overall physical activity level. The resting energy expenditure was estimated with the improved Harris-Benedict equation (19). The energy content of the mSTF for most patients was between 400-600 kcal/day. The mSTF was an individually adapted, very low-calorie diet whose macronutrients revealed to a ketogenic composition (75% fat, 15% protein and 10% carbohydrates) aiming to support the metabolic changes of fasting and to minimise the burden of water-only fasting. The patients were instructed to drink a daily minimum of 2.5 L of any calorie-free liquids, including water, herbal tea, and diet drinks without stimulants such as caffeine and alcohol. The fasting period took place on 5 consecutive days lasting a total of just 4 days (96h). It started 3 days prior to chemotherapy in the evening at 6 pm. The fasting period ended one day after chemotherapy at 6 pm, approximately 24 h after the end of drug administration to extend beyond the half-life of most toxic drugs (20).

While patients of the study groups A and B received only a mSTF, patients of the study groups C and D additionally obtained a 6-day normocaloric KD prior each mSTF period. Both fasting and normocaloric KD lead to similar metabolic changes (21) including a state of ketosis which can suppress hunger (16–18). Accordingly, fasting metabolism was induced via a KD prior each 4-day mSTF period. The patients of the study groups C and D received advice and recipes to follow a KD ad libitum according to individual food preferences. The patients were encouraged to limit their carbohydrate intake to a maximum of 20-40 g/day and to derive at least 75% of total consumed energy from fat. The macronutrient content during KD was the same as during the mSTF period except for the energy content.

**Outcome assessment**

The outcome measures were determined at baseline, at each chemotherapy cycle and as follow-up 3 weeks after the 4th or 6th cycle depending on the chemotherapy regimen. For measuring the main endpoint we assessed the percentage of patients with grade III or higher chemotherapy-induced toxicities. Toxicities were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE v4.0) (22). This comprehensive toxicity grading scale is well
established and used in clinical practice. Toxicities were reported by grade (level of severity) on a scale of I to V, whereby grade V (death) is inappropriate for some adverse events. Further outcome measures included chemotherapy-induced toxicities grade I/II (NCI CTCAE v4.0Table 2), nutritional status, body composition via bioelectrical impedance analysis (BIA, Body Impedance Analyser Nutriguard-MS™, Data Input GmbH, Germany), and validated questionnaires issued by the European Organisation for Research and Treatment of Cancer (EORTC) and the Functional Assessment of Chronic Illness Therapy (FACIT) focussing on quality of life (EORTC QLQ-C30 (23)), chemotherapy-induced polyneuropathy (EORTC QLQ-CIPN20 (24)) and fatigue (FACIT-Fatigue v4.0 (25)). In addition to the chemotherapy-induced toxicities (NCI CTCAE v4.0), a self-reporting questionnaire was developed especially for this trial to evaluate the chemotherapy-induced side effects in the week following chemotherapy. Daily, the patients rated their toxicities on a scale of 0 (none) to 3 (severe).

Venous blood sample were drawn before each chemotherapy administration and analysed at the Institute for Clinical Chemistry and Laboratory Medicine, University Medical Centre Freiburg. The effect of fasting was determined by recording hematologic parameters (differential blood count), inflammatory response [C-Reaktive Protein (CRP)], metabolic parameters [insulin, IGF-1] and endocrine parameters [thyroid-stimulating hormone (TSH), free triiodothyronine (fT3), free thyroxine (fT4)].

Safety measures were based on recommendations published 2013 by the Expert Panel Update of the 2002 Consensus Guidelines for Fasting Therapy (26). They included laboratory values of electrolytes (sodium, potassium, calcium, magnesium), renal (creatinine, urea nitrogen, uric acid) and liver function (bilirubin, alanine aminotransferase [ALT], aspartate aminotransferase [AST], alkaline phosphatase [ALP], and gamma-glutamyltransferase [GGT]). In order to document compliance with the mSTF regimen and to verify that the control group was not fasting, the patients were obligated to record their daily food intake and to monitor their ketosis state by documenting daily measures of urinary ketones via strips for self-testing (Ketostix®, Bayer AG, Switzerland) during each fasting cycle. Blood ketone levels were also checked at each chemotherapy via a testing device called FreeStyle Precision Neo Blood Glucose and Ketone Monitoring System with blood β-ketone test strips.
(Abbott GmbH & Co. KG, Germany). Semi-quantitative food records were analysed by professional software (PRODI® expert v6.5, Nuri-Science GmbH, Germany). At the follow-up visit, a specially developed questionnaire was conducted to evaluate patient’s subjective feeling and the feasibility of this pilot study.

**Sample size calculation and statistics**

The determination of the sample size was based on the primary outcome probability of adverse events grade III or higher and the primary objective to demonstrate a difference between the two main treatment groups (NC vs. mSTF). We assume a probability for grade 3 or higher adverse events of 0.60 (data source: GOIM 9902 study (27)) with NC and of 0.30 with fasting, i.e. a difference in proportions of 0.30. We further assume that the proportion of patients with differing results between NC and mSTF is 0.40 (discordant pairs). A sample size of 30 pairs would have 80% power to detect a difference in proportions of 0.30 when the proportion of discordant pairs is expected to be 0.40 and the method of analysis is a McNemar’s test of equality of paired proportions with a 0.05 two-sided significance level. Taking into account the possibility of dropouts, we would have needed 40 patients to ensure sufficient power.

All variables were tested for normal distribution (Kolmogorow-Smirnow test). Normally distributed variables are presented as mean ± standard deviation and compared using two sample t-test for differences between intervention groups or paired t-tests for differences between baseline (T₀) and follow-up (FU). Not normally distributed variables are shown as median (minimum – maximum). The Mann-Whitney test was used for independent groups, and the Wilcoxon rank-sum test for paired groups.

Variables measured at several points in time (chemotherapy-induced toxicities, body composition, validated questionnaires, safety and compliance parameters) were analysed as outcomes with a linear mixed model appropriate for the cross-over design, where treatment, period and sequence were treated as fixed effects and patients are considered as random effects. Results are presented as parameter estimates (PE), standard errors (SD) and 95% confidence intervals. The PE represents the difference for each variable between cycles of NC and those of mSTF. Chemotherapy cycles were
modeled as continuous variable. Statistical significance was set at \( P < 0.05 \). The data were analysed using IBM SPSS Statistics (Version 24, IBM, New York, USA).

Results

Patient characteristics

From March 2017 until December 2017, 121 women with gynaecological cancers were screened for eligibility, 51 were randomised to one of the four study arms (Figure 2). Over the course of the study period, 21 patients dropped out due to various reasons, including adjustments of the chemotherapy protocol \((n = 3)\), deterioration of the general condition, independent of fasting \((n = 4)\), fasting-related discomfort \((n = 2)\), postponements of chemotherapy \((n = 2)\) or without giving reasons \((n = 10)\).

Overall, 30 patients completed the intervention study. Seven patients (group A) started with mSTF during the first half of chemotherapy cycles, one patient began with a 6-day normocaloric KD followed by mSTF (group C). Twenty-two patients started with NC during the first half of chemotherapy cycles and changed to one of the fasting interventions afterwards (group B: \(n = 9\); group D: \(n = 13\)). Unexpectedly, the randomisation could not be implemented as originally planned due to the strict predefined daily routine in the outpatient chemotherapy ward. Consequently, the sizes of the groups were unequal and the patients’ characteristics differed significantly between groups A-D. Baseline characteristics of the whole study population are summarized in Table 1. The median age of the population was 54 years and ranged from 30 – 74 years. Mean body mass index (BMI) was 26.0 ± 4.6 kg/m^2. The majority of patients \((n = 22, 73.3\%)\) were diagnosed with breast cancer and treated with a neoadjuvant chemotherapy regimen of Epirubicin/Cyclophosphamid. Further treated tumor entities included endometrial cancer \((n = 2)\), ovarian cancer \((n = 2)\), and cervical cancer \((n = 4)\). According to the nutritional risk screening, twenty-one patients (70%) had a low risk (NRS = 1), only one patient had a risk for malnutrition (NRS = 3). In total, patients fasted during 56 of 118 chemotherapy cycles. Daily energy requirement was estimated at 1933 ± 178 kcal per day of NC or KD and 484 ± 44 kcal per fasting days.

Table 1 Baseline characteristics of the study cohort
| Randomisation          | MW ± SD |
|------------------------|---------|
| Group A                | 7 (23.3)|
| Group B                | 9 (30.0)|
| Group C                | 1 (3.3) |
| Group D                | 13 (43.3)|

| Demography          |         |
|---------------------|---------|
| Age (years)         | 54 ± 11 |
| Height (m)          | 1.65 ± 0.07 |
| BMI (kg/m²)         | 26.0 ± 4.6 |

| Diagnosis           |         |
|---------------------|---------|
| Adjuvant            | 26 (86.7)|
| Metastatic or local advanced | 4 (13.3)|

| Tumour entity          |         |
|------------------------|---------|
| Breast Cancer          | 22 (73.3)|
| Endometrial Cancer     | 2 (6.7)  |
| Ovarian Cancer         | 2 (6.7)  |
| Cervical Cancer        | 4 (13.3) |

| T-categories |         |
|--------------|---------|
| T1           | 10 (33.3)|
| T2           | 14 (46.7)|
| T3           | 3 (10.0) |
| T4           | 3 (10.0) |

| Nodal status       |         |
|--------------------|---------|
| N0                  | 20 (66.7)|
| N1                  | 8 (26.7) |
| N2                  | 1 (3.3)  |
| N3                  | 1 (3.3)  |

| Metastasis |         |
|------------|---------|
| M0         | 27 (90.0)|
| M1         | 3 (10.0) |

| Chemotherapy regimens |         |
|-----------------------|---------|
| Paclitaxel/Carboplatin| 7 (23.3)|
| Epirubicin/Cyclophosphamid | 22 (73.3)|
| Docetaxel/Cyclophosphamid | 1 (3.3)|

| Treatment           |         |
|---------------------|---------|
| Adjuvant chemotherapy| 6 (20)  |
| Neoadjuvant chemotherapy | 22 (73.3)|
| Palliative chemotherapy | 3 (10.0)|

| Nutritional status (NRS 2002) |         |
|-------------------------------|---------|
| 1                             | 21 (70.0)|
| 2                             | 8 (26.7) |
| 3                             | 1 (3.3)  |

| Energy requirement |         |
|--------------------|---------|
| Basal metabolic rate (kcal) | 1391 ± 125 |
| Physical Activity Level | 1.39 ± 0.06 |
| Active metabolic rate (kcal) | 1933 ± 178 |
| 25% of the active metabolic rate (kcal) | 484 ± 44 |

**Compliance and diet compositions**

Physiological blood ketosis (blood ketone level ≥ 0.6 mmol/l) was detected for 71.4% (n = 40) of the 56 chemotherapy cycles. State of ketosis was significantly different between mSTF and NC (mSTF 1.27 ± 1.18 mmol/l, NC 0.21 ± 1.98 mmol/l; P < 0.001). In addition to blood ketosis on the day of chemotherapy administration, patients measured urinary ketosis for daily self-testing during periods of fasting interventions. The mean urinary ketosis on fasting days confirmed the metabolic shift to ketosis and fat oxidation (1.47 ± 1.06 mmol/l). Neither the frequency nor the intensity of blood ketosis on fasting days was significantly different for patients in study groups C and D with a 6-day normocaloric KD prior to mSTF compared to patients in study groups A and B with only mSTF. The mean daily caloric intake was 1631 ± 566 kcal during NC. There were no significant differences in daily energy intake between NC and KD before mSTF. However, macronutrient composition of the KD
changed significantly in comparison to NC (P < 0.001) with higher fat (KD 64%; NC 39%) and protein (KD 20%; NC 17%) as well as lower carbohydrate consumption (KD 16%; NC 44%), respectively. The mean daily caloric intake on fasting days was 493 ± 157 kcal and was in line with the estimated energy requirements. While mSTF, the intake of the macronutrient composition was similar to the KD with 58% energy intake from fat, 23% from carbohydrates and 19% from proteins. There were no significant differences in macronutrient composition on fasting days between study arms C and D (KD prior mSTF) and study arms A and B (only mSTF).

**Toxicity**

We found that patients experienced significant less frequent grade I/II stomatitis during cycles of mSTF compared to cycles of NC [-0.16 ± 0.06; 95% CI -0.28 - (-0.03); P = 0.013, Table 2]. Regarding our primary endpoint, percentage of patients with grade III or higher chemotherapy-induced toxicities, only one single case of CTCAE grade III nausea was documented during a cycle of mSTF. Therefore the study failed to meet the primary endpoint. Most frequent toxicities grade I/II were Fatigue (41.6%), nausea (33.6%) and stomatitis (15.9%).

**Table 2 Chemotherapy related toxicities according to CTCAE (Grade I/II)**

|                 | NC (n = 58) | mSTF (n = 55) | PE ± SD* | 95% CI       | P-Va |
|-----------------|-------------|---------------|----------|--------------|------|
| Infection       | 2 (3.4%)    | 4 (7.3%)      | 0.07 ± 0.04 | 0.01 – 0.15  | 0.09 |
| Fatigue         | 22 (37.9%)  | 25 (45.5%)    | -0.01 ± 0.09 | -0.20 – 0.17 | 0.88 |
| Insomnia        | 2 (3.4%)    | 4 (7.3%)      | 0.03 ± 0.04 | -0.05 – 0.11 | 0.50 |
| Headachesa      | 2 (5.7%)    | 0             | -0.03 ± 0.06 | -0.14 – 0.09 | 0.64 |
| Dizzinessa      | 3 (8.6%)    | 2 (7.1%)      | -0.10 ± 0.07 | -0.25 – 0.05 | 0.18 |
| Depression      | 2 (3.4%)    | 2 (3.6%)      | -0.02 ± 0.04 | -0.10 – 0.06 | 0.56 |
| Nausea          | 22 (37.9%)  | 16 (29.1%)    | -0.01 ± 0.10 | -0.21 – 0.18 | 0.89 |
| Vomiting        | 0           | 2 (3.6%)      | 0.02 ± 0.03  | -0.03 – 0.08 | 0.41 |
| Diarrhea        | 2 (3.4%)    | 2 (3.6%)      | -0.01 ± 0.04 | -0.09 – 0.07 | 0.87 |
| Obstipationb    | 5 (8.8%)    | 4 (7.3%)      | -0.02 ± 0.06 | -0.13 – 0.09 | 0.68 |
| Stomach painsa  | 1 (2.9%)    | 0             | -0.02 ± 0.04 | -0.10 – 0.07 | 0.69 |
| Reduced appetitea | 7 (20.0%)   | 2 (7.1%)      | -0.16 ± 0.11 | -0.39 – 0.06 | 0.15 |
| Hungerc         | 2 (5.6%)    | 0             | -0.03 ± 0.06 | -0.15 – 0.09 | 0.60 |
| Stomatitis      | 15 (25.9%)  | 3 (5.5%)      | -0.16 ± 0.06 | -0.28 – (-0.03) | 0.01 |
| Esophagitis     | 2 (3.4%)    | 2 (3.6%)      | 0.01 ± 0.04  | -0.08 – 0.09 | 0.89 |
| Neuroses        | 8 (13.8%)   | 10 (18.2%)    | 0.07 ± 0.07  | -0.06 – 0.20 | 0.28 |
| Arthralgia      | 1 (1.7%)    | 2 (3.6%)      | 0.02 ± 0.03  | -0.05 – 0.09 | 0.61 |
| Pain            | 11 (19 %)   | 3 (5.5%)      | -0.11 ± 0.06 | -0.23 – 0.01 | 0.07 |
| Dyspnea         | 0           | 2 (3.6%)      | 0.03 ± 0.02  | -0.01 – 0.07 | 0.09 |
| Oedemas         | 0           | 1 (1.8%)      | 0.0 ± 0.02   | -0.04 – 0.04 | 0.84 |

*PE ± SD represents the difference for CTCAE points between cycles of NC and those of mSTF. All side effects were scored according to CTCAE v.4.0. Each side effect was scored once per patient during
each chemotherapy cycle. \(^a\) \(n = 63; \(^b\) \(n = 112; \(^c\) \(n = 64.

In addition to the incidence of side effects documented with the CTCAE by physicians, patients reported their chemotherapy-induced side effects in the week following chemotherapy (Table 3). Besides a significant reduction of frequency and severity score of self-reported headaches \([-1.80 \pm 0.55; 95\% \text{ CI} -2.89 \text{–} (-0.71); P = 0.002\]}, the frequency and severity score of self-reported feeling weak decreased significantly during cycles of mSTF compared to those of NC \([-1.99 \pm 0.87; 95\% \text{ CI} -3.72 \text{–} (-0.26); P = 0.024\]. Furthermore, the frequency and severity score of total self-reported toxicities were significantly reduced at cycles with mSTF compared to cycles of NC \([-10.36 \pm 4.44; 95\% \text{ CI} -19.22 \text{–} (-1.50); P = 0.023\]. There were no significant differences in the incidence and severity of adverse events between mSTF alone or in combination with a prior KD, neither in the documented CTCAE nor in the self-reported chemotherapy-induced toxicities.

Table 3 Self-reported chemotherapy-induced toxicities one week after chemotherapy

|                  | NC (n = 53) | mSTF (n = 51) | PE ± SD* | 95% CI     | P-\(V_δ\) |
|------------------|------------|---------------|----------|------------|-----------|
| Reduced Appetite | 4.51 ± 5.01| 2.75 ± 4.75   | -1.01 ± 0.71 | -2.43 \text{–} 0.42 | 0.16      |
| Hunger           | 5.36 ± 5.48| 4.98 ± 5.23   | -0.14 ± 0.60 | -1.35 \text{–} 1.06 | 0.81      |
| Nausea           | 3.72 ± 4.34| 4.24 ± 4.91   | -0.13 ± 0.57 | -1.26 \text{–} 0.99 | 0.81      |
| Vomiting         | 0.21 ± 0.84| 0.45 ± 1.14   | 0.32 ± 0.19  | 0.07 \text{–} 0.70  | 0.10      |
| Stomach pains    | 1.66 ± 3.25| 2.25 ± 3.69   | 0.79 ± 0.63  | -0.47 \text{–} 2.04 | 0.21      |
| Diarrhea         | 1.00 ± 2.88| 0.78 ± 2.34   | -0.41 ± 0.58 | -1.57 \text{–} 0.76 | 0.48      |
| Obstipation      | 3.17 ± 4.42| 3.06 ± 3.83   | -0.73 ± 0.84 | -2.41 \text{–} 0.94 | 0.38      |
| Fever            | 0.11 ± 0.51| 0.14 ± 0.98   | 0.13 ± 0.18  | -0.23 \text{–} 0.48 | 0.48      |
| Headaches        | 2.74 ± 4.30| 1.18 ± 2.06   | -1.80 ± 0.55 | -2.89 \text{–} (-0.71) | 0.00      |
| Insomnia         | 5.21 ± 5.35| 4.18 ± 4.97   | -1.18 ± 0.77 | -2.73 \text{–} 0.36 | 0.13      |
| Fatigue          | 7.96 ± 5.52| 6.63 ± 5.66   | -1.45 ± 0.85 | -3.15 \text{–} 0.26 | 0.09      |
| Dizziness        | 2.23 ± 9.19| 2.84 ± 4.15   | -0.13 ± 0.59 | -1.31 \text{–} 1.05 | 0.83      |
| Weakness         | 7.11 ± 5.40| 5.78 ± 5.26   | -1.99 ± 0.87 | -3.72 \text{–} (-0.26) | 0.02      |
| Exhaustion       | 6.74 ± 5.46| 6.20 ± 5.67   | -1.23 ± 0.85 | -2.92 \text{–} 0.46  | 0.15      |
| Total toxicities | 56.36 ± 32.14| 47.52 ± 33.21| -10.36 ± 4.44| -19.22 \text{–} (-1.50) | 0.02      |

* PE ± SD represents the difference for each toxicity score points between cycles of NC and those of mSTF. Per day, patients awarded each toxicity with 0 (none) to 3 (severe) points. Consequently, each toxicity received 0–21 points. The scores of all toxicities in the following seven days after chemotherapy are summarised in total toxicities (0-294 points).
Despite the fact that mSTF was safe, patients reported low grade fasting-related side effects. Overall, the most common reported fasting-related side effects included hunger (n = 8), dizziness (n = 5), weakness (n = 4), and headaches (n = 4).

Unfortunately, chemotherapy-induced toxicities frequently lead to postponements of the chemotherapy. This circumstance is an additional and extremely burdensome stress factor for patients. We compared the number of days of postponements at cycles of mSTF with those of NC. Patients had significant fewer chemotherapy postponements at cycles of mSTF [-0.80 ± 0.37; 95% CI -1.53 - (-0.06); P = 0.034], reflecting an improved tolerance of chemotherapy.

**Weight and body composition**

Comparing cycles of mSTF with cycles of NC, we observed a significant loss of mean BIA fat mass [-0.63 ± 0.23; 95% CI -1.09 - (-0.17); P = 0.008], leading to a significant reduction of body weight during mSTF [-0.84 ± 0.26; 95% CI -1.35 - (-0.33); P = 0.002]. Apart from BIA fat mass, body composition remained on average constant. In contrast to the baseline measures, losses of body weight and fat mass during mSTF were not counterbalanced and remained significantly reduced at the end of the study (P < 0.005). Mean body weight and mean fat mass were 71.4 ± 12.3 kg and 23.0 ± 8.8 kg at the beginning and 69.8 ± 11.6 kg and 21.4 ± 8.4 kg at the end of the intervention, respectively. When comparing body composition in the total study population at baseline with that at follow-up mean BIA body cell mass and mean BIA phase angle demonstrated a significant reduction from 23.2 ± 3.2 kg to 22.1 ± 3.0 kg (P = 0.007) and from 5.27 ± 0.74 kg to 4.8 ± 0.67 kg (P = 0.001), whereas the mean BIA extracellular cell mass showed a significant increase from 25.2 ± 3.3 kg to 26.8 ± 3.2 kg (P = 0.001), respectively. However, there were no significant differences in the measured anthropometric parameters comparing mSTF alone or in combination with a prior KD.

**Blood parameters**

Table 4 Blood parameters of chemotherapy cycles with short-term fasting or normocaloric diet
|                  | NC (MW ± SD n = 61) | mSTF (MW ± SD n = 56) | PE ± SD  | 95% CI       |
|------------------|---------------------|-----------------------|----------|--------------|
| **Blood count**  |                     |                       |          |              |
| Leucocytes       | 10³/µl              | 6.67 ± 2.23           | 6.09 ± 2.25 | -0.25 ± 0.30 | -0.84 - 0.35 |
| Thromobies       | 10³/µl              | 288.2 ± 76.6          | 306.6 ± 76.9 | 4.85 ± 10.76 | -16.56 - 26.25 |
| Erythrocytes     | 10³/µl              | 3.91 ± 0.47           | 3.95 ± 0.37 | 0.11 ± 0.06  | -0.01 - 0.23  |
| Haemoglobin      | g/dl                | 11.7 ± 1.4            | 11.6 ± 1.1 | 0.18 ± 0.18  | -0.19 - 0.54  |
| Haematocrit      | %                   | 33.5 ± 3.9            | 33.1 ± 2.9 | 0.31 ± 0.52  | -0.72 - 1.33  |
| MCH               | pg                  | 29.9 ± 1.5            | 29.5 ± 1.6 | -0.38 ± 0.13 | -0.63 - (-0.13) |
| MCV               | 10⁹/µl              | 85.7 ± 4.0            | 84.1 ± 3.7 | -1.68 ± 0.33 | -2.33 - (-1.02) |
| MCHC              | g/dl                | 34.9 ± 0.9            | 35.1 ± 0.9 | 0.23 ± 0.14  | -0.05 - 0.51  |
| Neutrophils       | 10³/µl              | 4.70 ± 2.22           | 4.29 ± 2.24 | -0.17 ± 0.30 | -0.76 - 0.43  |
| Lymphocytes       | %                   | 21.0 ± 10.7           | 19.7 ± 12.4 | -0.89 ± 1.32 | -3.50 - 1.73  |
| Monocytes         | %                   | 8.22 ± 4.70           | 10.1 ± 5.3 | 0.90 ± 0.72  | -0.52 - 2.33  |
| Eosinophils       | %                   | 0.71 ± 1.29           | 0.51 ± 1.18 | -0.05 ± 0.19 | -0.42 - 0.33  |
| Basophils         | %                   | 0.68 ± 0.76           | 0.74 ± 0.59 | -0.02 ± 0.10 | -0.18 - 0.22  |
| **Electrolytes**  |                     |                       |          |              |
| Potassium         | mmol/l              | 4.29 ± 0.42           | 4.28 ± 0.32 | -0.01 ± 0.07 | -0.15 - 0.12  |
| Sodium            | mmol/l              | 139.2 ± 2.2           | 137.9 ± 3.3 | -1.17 ± 0.42 | -2.0 - (-0.33) |
| Calcium           | mmol/l              | 2.33 ± 0.14           | 2.34 ± 0.15 | 0.02 ± 0.03  | -0.04 - 0.07  |
| Magnesium         | mmol/l              | 0.80 ± 0.07           | 0.78 ± 0.07 | -0.00 ± 0.01 | -0.02 - 0.02  |
| **Liver and kidney parameters** | | | | | |
| Urea              | mg/dl               | 26.9 ± 10.0           | 25.3 ± 10.6 | -1.087 - 1.34 | -4.54 - 0.79  |
| Creatinine        | mg/dl               | 0.71 ± 0.16           | 0.72 ± 0.19 | 0.02 ± 0.01  | -0.01 - 0.05  |
| Uric acid         | mg/dl               | 4.44 ± 1.33           | 5.98 ± 1.79 | 1.35 ± 0.17  | 1.01 - 1.68   |
| Bilirubin         | mg/dl               | 0.33 ± 0.25           | 0.29 ± 0.11 | 0.01 ± 0.03  | -0.05 - 0.07  |
| GOT               | U/l                 | 24.2 ± 8.7            | 25.0 ± 7.2 | 1.38 ± 1.48  | -1.56 - 4.32  |
| GPT               | U/l                 | 22.4 ± 11.8           | 23.5 ± 12.2 | -0.10 ± 1.46 | -3.01 - 2.81  |
| AP                | g/dl                | 71.4 ± 22.0           | 73.6 ± 25.0 | -1.96 ± 2.29 | -6.52 - 2.58  |
| Albumin           | g/dl                | 4.31 ± 0.35           | 4.29 ± 0.32 | 0.02 ± 0.06  | -0.09 - 0.13  |
| CRP               | mg/l                | 5.57 ± 4.93           | 8.1 ± 13.5 | 0.87 ± 1.95  | -3.0 - 4.73   |
| **Hormones**      |                     |                       |          |              |
| TSH               | µU/ml               | 1.05 ± 0.74           | 1.03 ± 0.73 | -0.1 ± 0.07  | -0.24 - 0.05  |
| ft3               | pmol/l              | 4.21 ± 0.81           | 3.75 ± 0.76 | -0.47 ± 0.09 | -0.64 - (-0.30) |
| ft4               | pmol/l              | 15.5 ± 2.7            | 16.4 ± 2.9 | 0.82 ± 0.37  | 0.09 - 1.55   |
| Insulin           | pmol/l              | 252.7 ± 372.8         | 140.1 ± 252.4 | -169.4 ± 44.1 | -257.1 - (-81.8) |
| IGF-1             | ng/ml               | 127.7 ± 55.7          | 97.8 ± 46.4 | -33.3 ± 5.4  | -44.1 - -(-22.5) |

Abbreviations: AP alkaline phosphatase, CRP C-reactive protein, GOT glutamic-oxaloacetic transaminase, GPT glutamic-pyruvic transaminase, IGF-1 insulin-like growth factor 1, MCH mean corpuscular haemoglobin, MCHC mean corpuscular haemoglobin concentration, MCV mean corpuscular volume, ft3 free triiodothyronine, ft4 free thyroxine, TSH thyroid stimulating hormone

All haematological, inflammatory, metabolic and endocrine parameters were compared between cycles of mSTF and those of NC (Table 4). Significant differences of haematological parameters were found in the mean corpuscular cell volume [MCV; -1.68 ± 0.33; 95% CI -2.33 -(-1.02); P <0.001] and mean corpuscular haemoglobin [MCH; -0.38 ± 0.13; 95% CI -0.63 -(-0.13); P = 0.004]. Both parameters were significantly lowered during cycles of mSTF compared to cycles of NC (Table 4).

However, erythrocytes were on average slightly increased at mSTF [0.11 ± 0.06; 95% CI -0.01 - 0.23;
For leucocytes, thrombocytes and neutrophils, no significant differences in counts were observed between cycles of mSTF and cycles of NC.

Although the mean level of sodium in the blood was significantly decreased during cycles of mSTF compared with those of NC [-1.17 ± 0.42; 95% CI -2.0 –(-0.33); P = 0.007], mean sodium levels remained within the reference range. All other electrolytes remained unchanged throughout the intervention. Monitoring the renal function, significantly elevated mean uric acid levels, exceeding the reference value, were measured during mSTF [1.35 ± 0.17; 95% CI 1.01 – 1.68; P <0.001]. No significant changes were observed for urea and creatinine values as well as liver parameters and CRP comparing cycles of mSTF and cycles of NC.

The mean thyroid levels fT3 and fT4 showed significant changes within the reference range at cycles of mSTF compared to those of NC, whereas mean TSH remained unaffected. In contrast to the significantly reduced mean fT3 levels [-0.47 ± 0.09; 95% CI -0.64 –(-0.30); P <0.001], mean fT4 rose significantly at cycles of mSTF compared to cycles of NC [0.82 ± 0.37; 95% CI 0.09 – 1.55; P = 0.028].

Both, mean Insulin [-169.4 ± 44.1; 95% CI -257.1 – (-81.8); P <0.001] and mean IGF-1 level [-33.3 ± 5.4; 95% CI -44.1 – (-22.5); P <0.001] decreased significantly during cycles of mSTF compared to cycles of NC. Until the follow-up visit, all blood parameters that had altered due to fasting had returned to normal levels. Comparing mSTF alone with mSTF in combination with a prior KD, there were no significant differences for any blood parameter.

**Questionnaires of QoL, CIPN and Fatigue**

No significant changes of QoL, CIPN and fatigue were found neither comparing cycles of mSTF with those of NC, nor comparing mSTF alone with mSTF in combination with a prior KD. The severity of fatigue remained unchanged at follow-up compared to baseline.

In general, fasting was well tolerated by the patients. At follow-up, twenty-three patients estimated the effect of mSTF on general health as “very good” or “good”. Only one patient declared mSTF as “difficult”, while 11 outlined mSTF as “rather difficult”. MSTF was evaluated as “rather easy” or “easy” by 9 patients, respectively. In total, more than half the patients would fast again during chemotherapy (n = 24).
Discussion
The primary objective of this randomised clinical trial was to evaluate the effects of a 4-day mSTF with a ketogenic macronutrient composition on chemotherapy-induced toxicities CTCAE grade III and higher in patients with gynaecological cancers. Secondary objectives included the assessment of toxicities CTCAE I/II, self-reported toxicities, body composition, blood parameters and subjective well-being (QoL, CIPN and fatigue). In addition, this was the first clinical trial to explore the effects of a 6-day normocaloric KD prior mSTF as a fasting supportive diet on fasting-related discomfort and compliance.

Unexpectedly, only a very low incidence of CTCAE grade ≥III was observed during the study period and thus, our primary endpoint was not evaluable. However, we found that mSTF improved tolerability of chemotherapy due to less chemotherapy-induced toxicities as less stomatitis, fewer headaches, less weakness and overall toxicities. Concomitant with previous studies, these outcomes confirm that mSTF during chemotherapy is safe and well tolerated (8,11). We found different results between CTCAE and self-reported toxicities, what may be explained by the different time points of the queries. Coolbrandt et al. (28) demonstrated that delayed self-assessments of chemotherapy-induced toxicities from retrospective queries, as performed in the CTCAE, provide a distorted and weaker overview of the actual symptoms and symptom severity experienced by patients compared to immediate self-report. Hence, we assume that the prospective self-reported toxicities reflect the actual side effects better than the retrospective CTCAE. Considering previous human fasting studies during chemotherapy, it must be noted that there are several differences between the study populations, designs, types, and durations of fasting interventions. It is thus not surprising that findings about chemotherapy-induced toxicities varied between different trials. While Safdie et al. (8) and Dorff et al. (11) detected less gastrointestinal toxicities, including less nausea, vomiting and diarrhea after STF, we observed no differences in these symptoms. This can probably be explained by the fact that in general patients had very few cases of gastrointestinal toxicities during cycles of mSTF as well as during those of NC, due to an individually adapted preventive concomitant medication against nausea and vomiting in each patient. Significant reductions in fatigue after STF
were recognised by Safdie et al. (8) and Bauersfeld et al. (10). However, in line with Safdie et al. (8), patients in this trial reported significant less weakness in the week after mSTF, indicating that mSTF could promote the regeneration after chemotherapy. Unexpectedly, patients experienced significant fewer headaches during mSTF, even though fasting and irregular meals are some of the frequent triggers for migraine (29). However, there is some evidence that ketones reduce the frequency and severity of migraine and headaches presumably by an improved mitochondrial function and less oxidative stress (30–32). It is well known that many serious chemotherapy-induced side effects compromise the continuation of the therapy, leading to postponements or interruptions of the chemotherapy. Overall, in this trial, the reduced toxicities due to mSTF probably resulted in a faster physical regeneration after chemotherapy and demonstrable fewer postponements of the chemotherapy.

Previous animal studies investigating STF during chemotherapy indicated that STF leads to fewer damages and faster regeneration in bone marrow cells and resulting cells, especially leukocytes (33). Safdie et al. (8) confirmed these observations in a first case series report, indicating that especially leukocytes, thrombocytes and neutrophils showed faster regeneration due to STF during chemotherapy. De Groot et al. (9) observed significant increases of erythrocytes and thrombocytes, and Dorff et al. (11) found less neutropenia and thrombocytopenia and concluded that STF protects against depletion and DNA damage of leukocytes. Contrary to the previous studies, we found no significant changes in any of the above mentioned blood parameters, which can probably be attributed to the small study population. However, we observed significant reductions of MCH and MCV during mSTF. These blood parameters are considered as independent prediction parameters of survival in various cancers prior to surgery (34). In case of breast cancer patients, higher MCH levels suggest adverse effects on disease-free survival (35). Even though the reductions of MCH and MCV can be seen as positive results, the clinical relevance of these findings remain unclear, particularly in view of the small sample size of this pilot trial.

We measured significant increased ketone body concentrations during mSTF, which proved that patients were compliant in fasting. This finding was consistent with the result of Dorff et al. (11).
While both, de Groot et al. (9) and Dorff et al. (11) observed only a slight trend for reduced insulin levels in cycles of STF, we detected a significant insulin reduction. Similarly to de Groot et al. (9), we found a significant reduction of IGF-1 during mSTF. This reduction of IGF-1 seems to be one of the key mediators of differential stress resistance in response to nutrient restriction (36). Previous experimental data indicate that mainly low levels of IGF-1 induce differential stress resistance, leading to a protection of healthy cells against toxicity of chemotherapy, mediated by the downregulation of cell growth and proliferation, while tumour cells are unable to activate a protective response (4,5,37). Since we observed both reductions of IGF-1 and chemotherapy-induced toxicities, our results support the hypothesis that reduction of IGF-1 promotes differential stress resistance. In future trials blood glucose should additionally be measured to get a better understanding of the fluctuations of ketones, insulin and IGF-1. The observed increase of the endocrine parameter fT4 was in line with the results of the human fasting intervention trial of de Groot et al. (9), and our previous clinical trial, investigating the effect of a normocaloric six-week KD on biochemical parameters in healthy adults (38). While de Groot et al. (9) revealed no change of fT3, we found a significant reduction of fT3 during KD in our previous trial in healthy subjects (38). We observed comparable changes in thyroid hormones in a clinical trial with a normocaloric KD as well as in this trial with mSTF. This can probably be attributed to the state of physiological ketosis, which appears in both, mSTF and KD due to carbohydrate restriction. The change of the fasting safety parameters sodium and uric acid during mSTF has previously been demonstrated in fasting studies with healthy adults (13,39). Fasting is known to provoke dehydrating effects, including enhanced excretion of minerals, especially sodium. Enhanced natriuresis during the first fasting days comes along with the excretion of ketones via urine, which is always accompanied by cations, namely initially by sodium, followed by ammonium (40,41). The increased concentration of uric acid during fasting is due to the reduced renal uric acid clearance by impairment of tubular uric acid secretion because ketone bodies compete with uric acid for common tubular secretion (41,42).

Consistent with previous fasting studies in healthy adults, we observed a mild weight loss (<5%) during mSTF, which remained until the end of our examination (13,39). In contrast, previous animal
and human trials investigating fasting during chemotherapy showed that animals as well humans regained their usual weight after an initial weight loss (3-5,8,10,11,43). In this respect, it should particularly be noted that the documented weight loss in our trial can primarily be attributed to a loss of fat mass, whereas lean body mass remained stable. Compared to water-only fasting, the mSTF allowed a slight caloric intake per each fasting day to minimise the protein catabolism, including the loss of lean body mass (44). Given that loss of lean body mass is associated with an impaired nutritional status and a poor general condition, it should be avoided by all means (45,46). Although weight loss was finally harmless in our trial, fasting accompanying chemotherapy may only be implemented if a risk of malnutrition can be excluded.

Indeed, this was the first human fasting intervention study, which assessed body composition during fasting using BIA measurements. Thereby, significant reductions of body cell mass and phase angle were observed over the course of trial. In general, phase angle is an independent prognostic factor for impaired functional and nutritional status and overall survival (47,48). Reductions of the phase angle during cancer treatment can frequently be explained by chemotherapy-induced damages, because cytostatic drugs reduce the total cell amount and membrane integrity (47,49). Notably, phase angle was already below the reference values for healthy women stratified by age and BMI at the beginning of our study (50). Especially for oncological patients, an adjusted reference value, more precisely, the fifth percentile of the phase angle reference value is commonly used (48,51). Nevertheless, the phase angle was slightly below the adjusted reference value at the end of our trial. To date, no published clinical trial investigated the impact of fasting on the phase angle. Moreover, it is still unclear whether fasting has an impact on body composition, especially on the phase angle.

Similarly to Bauersfeld et al. (10), we collected data on QoL, fatigue and additionally CIPN. While Bauersfeld et al. (10) observed significant improvements in fatigue and QoL due to STF during chemotherapy, our results were not able to confirm these findings. However, it should be noted, that the informative value of the three above mentioned questionnaires is limited due to an unfavourable point in time for data collection on the day of chemotherapy, reflecting QoL, CIPN and fatigue in the week before chemotherapy. Thus, concerning the impact of fasting on QoL, CIPN, and fatigue in the
week following chemotherapy, no valid conclusions can be drawn from our results.

Since both mSTF and KD lead to similar metabolic changes, including a state of ketosis which can suppress hunger (16-18), we hypothesized that a KD as a fasting supportive diet prior to mSTF should improve the compliance of the fasting regimen and reduce the initial fasting-related discomfort. However, no beneficial effects could be found for a 6-day normocaloric KD prior mSTF compared to mSTF alone, based on compliance, fasting-related discomfort, chemotherapy-induced toxicities and other outcome parameters in this trial. On the contrary, patients performing a KD prior to mSTF considered the feasibility of this intervention as more difficult compared to patients with mSTF alone. Therefore, a KD prior mSTF seemed to be an additional burden to patients undergoing chemotherapy, without benefiting from any advantage.

Our study had some limitations. The most obvious limitation was the small sample size within the scope of the pilot trial, which limits the power of the study and precludes firm statistical conclusions. Nonetheless that cross-over models are limited by carry-over effects, we chose this design for our pilot trial because of the low influence of confounding variables in control and intervention groups and the high statistically efficiency in small sample sizes. Another limitation was due to the randomisation. In the strictly predefined daily routine in the outpatient chemotherapy ward it was not possible to conduct the randomisation as planned. Consequently, patients were not randomly but selectively assigned to the four study groups. Additionally, we neglected the documentation of the concomitant medication during chemotherapy in our pilot trial. Finally, there is a possibility, that patients subjectively overestimated the positive effects of fasting due to the knowledge of the allocated interventions and a general positive attitude of fasting cures in Germany. This limitation can only be resolved by a double-blinded randomised study design, which is not possible with a natural nutrition.

Conclusion
In summary, our results reveal that mSTF is safe and feasible for gynaecologic cancer patients, however high motivation for this intervention is required. Moreover, mSTF during chemotherapy has the potential to reduce chemotherapy-induced toxicities and increase the tolerance of chemotherapy. Different blood parameters and metabolic parameters, particularly IGF-1 and insulin, were positively
influenced by mSTF. A 6-day normocaloric KD prior mSTF had no beneficial effects neither on chemotherapy-induced toxicities nor on fasting-related discomfort and compliance. Clearly, there is a need for larger randomised controlled trials, focusing on confirmation of the efficacy of mSTF accompanying chemotherapy as innovative, supportive approach.

List Of Abbreviations
ALP          alkaline phosphatase
ALT          alanine aminotransferase
AST          aspartate aminotransferase
BIA          bioelectrical impedance analysis
BMI          body mass index
CRP          C-Reaktive Protein
CIPN         Chemotherapy-induced Polyneuropathy
EORTC        European Organisation for Research and Treatment of Cancer
FACIT        Functional Assessment of Chronic Illness Therapy
fT3          free triiodothyronine
fT4          free thyroxine
FU           follow-up
GGT          gamma-glutamyltransferase
IGF-1        insulin-like growth factor-1
KD           ketogenic diet
NC           normocaloric diet
NCI CTCAE    National Cancer Institute Common Terminology Criteria for Adverse Events
mSTF         modified short-term fasting
PE           parameter estimate
QoL          quality of life
T₀           baseline
TSH          thyroid-stimulating hormone
Declarations

Ethics approval and consent to participate

The study protocol was approved by the Ethics Commission of Albert-Ludwig University Freiburg (313/16). All subjects provided written informed consent prior to participation.

Consent for publication

Not Applicable.

Availability of data and material

The datasets generated and analysed during the study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors’ contributions

PU and HB contributed to the conceptualisation and design of this study. SZ, AR and RS performed patient recruitment and data collection. BR contributed to the patient security and patient recruitment. SZ, AR, JE and GI performed statistical analysis and interpretation of the data analysis. SZ and AR wrote the manuscript. All authors edited and made critical revisions to the article. All authors have read and approved the final manuscript.

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References

1. Michalsen A, Li C. Fasting Therapy for Treating and Preventing Disease - Current State of Evidence. Forsch Komplementärmedizin Res Complement Med. 2013;20(6):444-53.

2. Brandhorst S, Wei M, Hwang S, Morgan TE, Longo VD. Short-term calorie and protein restriction provide partial protection from chemotoxicity but do not delay glioma progression. Exp Gerontol. 2013;48(10):1120-8.

3. Safdie F, Brandhorst S, Wei M, Wang W, Lee C, Hwang S, et al. Fasting Enhances the Response of Glioma to Chemo- and Radiotherapy. PLoS ONE. 2012;7(9):e44603.

4. Lee C, Raffaghello L, Brandhorst S, Safdie FM, Bianchi G, Martin-Montalvo A, et al. Fasting Cycles Retard Growth of Tumors and Sensitize a Range of Cancer Cell Types to Chemotherapy. Sci Transl Med. 2012;4(124):124ra27.

5. Raffaghello L, Lee C, Safdie FM, Wei M, Madia F, Bianchi G, et al. Starvation-dependent differential stress resistance protects normal but not cancer cells against high-dose chemotherapy. Proc Natl Acad Sci. 2008;105(24):8215-20.

6. Longo VD, Mattson MP. Fasting: Molecular Mechanisms and Clinical Applications. Cell Metab. 2014;19(2):181–92.

7. Laviano A, Rossi Fanelli F. Toxicity in chemotherapy--when less is more. N Engl J Med. 2012 Jun 14;366(24):2319–20.

8. Safdie FM, Dorff T, Quinn D, Fontana L, Wei M, Lee C, et al. Fasting and cancer treatment in humans: A case series report. Aging. 2009 Dec 31;1(12):988-1007.

9. de Groot S, Vreeswijk MP, Welters MJ, Gravesteijn G, Boei JJ, Jochems A, et al. The effects of short-term fasting on tolerance to (neo) adjuvant chemotherapy in HER2-negative breast cancer patients: a randomized pilot study. BMC Cancer. 2015;15(1).
10. Bauersfeld SP, Kessler CS, Wischnewsky M, Jaensch A, Steckhan N, Stange R, et al. The effects of short-term fasting on quality of life and tolerance to chemotherapy in patients with breast and ovarian cancer: a randomized cross-over pilot study. BMC Cancer. 2018;18(1):476.

11. Dorff TB, Groshen S, Garcia A, Shah M, Tsao-Wei D, Pham H, et al. Safety and feasibility of fasting in combination with platinum-based chemotherapy. BMC Cancer. 2016;16(1).

12. Michalsen A, Weidenhammer W, Melchart D, Langhorst J, Saha J, Dobos G. Kurzzeitiges therapeutisches Fasten in der Behandlung von chronischen Schmerz- und Erschöpfungssyndromen – Verträglichkeit und Nebenwirkungen mit und ohne begleitende Mineralstoffergänzung. Complement Med Res. 2002;9(4):221–7.

13. Michalsen A, Hoffmann B, Moebus S, Bäcker M, Langhorst J, Dobos GJ. Incorporation of fasting therapy in an integrative medicine ward: evaluation of outcome, safety, and effects on lifestyle adherence in a large prospective cohort study. J Altern Complement Med N Y N. 2005 Aug;11(4):601–7.

14. Brandhorst S, Choi IY, Wei M, Cheng CW, Sedrakyan S, Navarrete G, et al. A Periodic Diet that Mimics Fasting Promotes Multi-System Regeneration, Enhanced Cognitive Performance, and Healthspan. Cell Metab. 2015 Jul 7;22(1):86–99.

15. Veech RL. The therapeutic implications of ketone bodies: the effects of ketone bodies in pathological conditions: ketosis, ketogenic diet, redox states, insulin resistance, and mitochondrial metabolism. Prostaglandins Leukot Essent Fatty Acids. 2004 Mar;70(3):309–19.

16. Johnston CS, Tjoon SL, Swan PD, White A, Hutchins H, Sears B. Ketogenic low-carbohydrate diets have no metabolic advantage over nonketogenic low-carbohydrate diets. Am J Clin Nutr. 2006;83(5):1055–61.
17. Johnstone AM, Horgan GW, Murison SD, Bremner DM, Lobley GE. Effects of a high-protein ketogenic diet on hunger, appetite, and weight loss in obese men feeding ad libitum. Am J Clin Nutr. 2008;87(1):44–55.

18. Sumithran P, Prendergast LA, Delbridge E, Purcell K, Shulkes A, Kriketos A, et al. Ketosis and appetite-mediating nutrients and hormones after weight loss. Eur J Clin Nutr. 2013;67(7):759–64.

19. Mifflin MD, St Jeor ST, Hill LA, Scott BJ, Daugherty SA, Koh YO. A new predictive equation for resting energy expenditure in healthy individuals. Am J Clin Nutr. 1990;51(2):241–7.

20. Calsteren KV, Verbesselt R, Ottevanger N, Halaska M, Heyns L, Bree RV, et al. Pharmacokinetics of chemotherapeutic agents in pregnancy: a preclinical and clinical study. Acta Obstet Gynecol Scand. 2010;89(10):1338–45.

21. Paoli A, Rubini A, Volek JS, Grimaldi KA. Beyond weight loss: a review of the therapeutic uses of very-low-carbohydrate (ketogenic) diets. Eur J Clin Nutr. 2013;67(8):789–96.

22. National Institutes of Health, National Cancer Institute, editors. Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 [Internet]. 2009. Available from: http://evs.nci.nih.gov/ftp1/CTCAE

23. Aaronson NK, Ahmedzai S, Bergman B, Bullinger M, Cull A, Duez NJ, et al. The European Organization for Research and Treatment of Cancer QLQ-C30: A Quality-of-Life Instrument for Use in International Clinical Trials in Oncology. JNCI J Natl Cancer Inst. 1993;85(5):365–76.

24. Postma TJ, Aaronson NK, Heimans JJ, Muller MJ, Hildebrand JG, Delattre JY, et al. The development of an EORTC quality of life questionnaire to assess chemotherapy-induced peripheral neuropathy: The QLQ-CIPN20. Eur J Cancer. 2005;41(8):1135–9.
25. Yellen SB, Cella DF, Webster K, Blendowski C, Kaplan E. Measuring fatigue and other anemia-related symptoms with the Functional Assessment of Cancer Therapy (FACT) measurement system. J Pain Symptom Manage. 1997 Feb 1;13(2):63–74.

26. Wilhelmi de Toledo F, Buchinger A, Burggrabe H, Hölz G, Kuhn C, Lischka E, et al. Fasting Therapy - an Expert Panel Update of the 2002 Consensus Guidelines. Forsch Komplementärmedizin Res Complement Med. 2013;20(6):434–43.

27. Vici P, Brandi M, Giotta F, Foggi P, Schittulli F, Di Lauro L, et al. A multicenter phase III prospective randomized trial of high-dose epirubicin in combination with cyclophosphamide (EC) versus docetaxel followed by EC in node-positive breast cancer. GOIM (Gruppo Oncologico Italia Meridionale) 9902 study. Ann Oncol. 2012;23(5):1121–9.

28. Coolbrandt A, Heede KV den, Vanhove E, Bom AD, Milisen K, Wildiers H. Immediate versus delayed self-reporting of symptoms and side effects during chemotherapy: Does timing matter? Eur J Oncol Nurs. 2011;15(2):130–6.

29. Dalkara T, Kiliç K. How does fasting trigger migraine? A hypothesis. Curr Pain Headache Rep. 2013;17(10):368.

30. Di Lorenzo C, Pinto A, Ienca R, Coppola G, Sirianni G, Di Lorenzo G, et al. A Randomized Double-Blind, Cross-Over Trial of very Low-Calorie Diet in Overweight Migraine Patients: A Possible Role for Ketones? Nutrients. 2019;11(8):1742.

31. Di Lorenzo C, Coppola G, Sirianni G, Di Lorenzo G, Bracaglia M, Di Lenola D, et al. Migraine improvement during short lasting ketogenesis: a proof-of-concept study. Eur J Neurol. 2015;22(1):170–7.

32. Gross EC, Klement RJ, Schoenen J, D’Agostino DP, Fischer D. Potential Protective Mechanisms of Ketone Bodies in Migraine Prevention. Nutrients. 2019;11(4).

33. Cheng C-W, Adams GB, Perin L, Wei M, Zhou X, Lam BS, et al. Prolonged Fasting
Reduces IGF-1/PKA to Promote Hematopoietic-Stem-Cell-Based Regeneration and Reverse Immunosuppression. Cell Stem Cell. 2014;14(6):810–23.

34. Jomrich G, Hollenstein M, John M, Ristl R, Paireder M, Kristo I, et al. High Mean Corpuscular Volume Predicts Poor Outcome for Patients With Gastroesophageal Adenocarcinoma. Ann Surg Oncol. 2019;26(4):976–85.

35. Zhang P, Zong Y, Liu M, Tai Y, Cao Y, Hu C. Prediction of outcome in breast cancer patients using test parameters from complete blood count. Mol Clin Oncol. 2016;4(6):918–24.

36. de Groot S, Pijl H, van der Hoeven JJM, Kroep JR. Effects of short-term fasting on cancer treatment. J Exp Clin Cancer Res CR. 2019;38.

37. Lee C, Safdie FM, Raffaghello L, Wei M, Madia F, Parrella E, et al. Reduced Levels of IGF-I Mediate Differential Protection of Normal and Cancer Cells in Response to Fasting and Improve Chemotherapeutic Index. Cancer Res. 2010;70(4):1564–72.

38. Urbain P, Strom L, Morawski L, Wehrle A, Deibert P, Bertz H. Impact of a 6-week non-energy-restricted ketogenic diet on physical fitness, body composition and biochemical parameters in healthy adults. Nutr Metab. 2017;14(1).

39. Toledo FW de, Grundler F, Bergouignan A, Drinda S, Michalsen A. Safety, health improvement and well-being during a 4 to 21-day fasting period in an observational study including 1422 subjects. PLOS ONE. 2019;14(1):e0209353.

40. Sigler MH. The mechanism of the natriuresis of fasting. J Clin Invest. 1975;55(2):377–87.

41. Kerndt PR, Naughton JL, Driscoll CE, Loxterkamp DA. Fasting: The History, Pathophysiology and Complications. West J Med. 1982;137(5):379–99.

42. Mojto V, Gvozdjakova A, Kucharska J, Rausova Z, Vancova O, Valuch J. Effects of complete water fasting and regeneration diet on kidney function, oxidative stress
and antioxidants. Bratisl Med J. 2018;119(02):107-11.

43. Huisman SA, Bijman-Lagcher W, IJzermans JN, Smits R, Bruin RW de. Fasting protects against the side effects of irinotecan but preserves its anti-tumor effect in Apc15lox mutant mice. Cell Cycle. 2015;14(14):2333–9.

44. Owen OE, Smalley KJ, D’Alessio DA, Mozzoli MA, Dawson EK. Protein, fat, and carbohydrate requirements during starvation: anaplerosis and cataplerosis. Am J Clin Nutr. 1998;68(1):12–34.

45. Deutz NEP, Ashurst I, Ballesteros MD, Bear DE, Cruz-Jentoft AJ, Genton L, et al. The Underappreciated Role of Low Muscle Mass in the Management of Malnutrition. J Am Med Dir Assoc. 2019;20(1):22–7.

46. Pierik VD, Meskers CGM, Van Ancum JM, Numans ST, Verlaan S, Scheerman K, et al. High risk of malnutrition is associated with low muscle mass in older hospitalized patients - a prospective cohort study. BMC Geriatr. 2017;17(1):118.

47. Gupta D, Lammersfeld CA, Vashi PG, King J, Dahlk SL, Grutsch JF, et al. Bioelectrical impedance phase angle as a prognostic indicator in breast cancer. BMC Cancer. 2008;8(1).

48. Norman K, Stobäus N, Zocher D, Bosy-Westphal A, Szramek A, Scheufele R, et al. Cutoff percentiles of bioelectrical phase angle predict functionality, quality of life, and mortality in patients with cancer. Am J Clin Nutr. 2010;92(3):612–9.

49. Aslantürk ÖS. In Vitro Cytotoxicity and Cell Viability Assays: Principles, Advantages, and Disadvantages. Genotoxicity - Predict Risk Our Actual World. 2017;

50. Bosy-Westphal A, Danielzik S, Dörhöfer R-P, Later W, Wiese S, Müller MJ. Phase Angle From Bioelectrical Impedance Analysis: Population Reference Values by Age, Sex, and Body Mass Index. J Parenter Enter Nutr. 2006;30(4):309-16.

51. Urbain P, Birlinger J, Ihorst G, Biesalski H-K, Finke J, Bertz H. Body mass index and
bioelectrical impedance phase angle as potentially modifiable nutritional markers are independent risk factors for outcome in allogeneic hematopoietic cell transplantation. Ann Hematol. 2013;92(1):111-9.

Supplementary Information

Additional file 1: CONSORT extension for pilot and feasibility trials checklist

Figures

Randomised crossover study design to test the effect of modified short-term fasting (mSTF) during 2 or 3 cycles of chemotherapy (CTX) depending on CTX regimen on CTX-induced toxicities compared to a normocaloric diet (NC) during 2 or 3 cycles of CTX as control and to test the effect of a normocaloric ketogenic diet (KD) followed by mSTF on fasting realated discomfort compared to mSTF alone.
Figure 2

Flow diagram of the study participants from eligibility criteria screening to study completion

Supplementary Files

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