Statistical analysis plan (SAP)
Behaviours and outcomes after liver transplant (BOLT): Diet
V1 27/11/2023

SAP revision history

Date	Version	Justification for SAP version
06/10/2023	Draft 1	Draft SAP sent draft to SG, KR, LO, AM, MA for initial review.
07/11/2023	Draft 2	Draft SAP revised following comments from SG, AM and KR, to include a more detailed section about descriptive analysis.
27/11/2023	Version 1	Approved by SG, AM, LO and KR.

Background

What is the problem being addressed?

Liver transplant cures liver disease and survival rates are good with 64% of liver transplant recipients (LTRs) alive at 10 years post-transplant (NHSBT, 2017). Cardiovascular disease (CVD) is a leading cause of death for liver transplant recipients (LTRs) causing 19% of non-hepatic deaths after LT (Watt et al., 2010). The presence of CVD risk factors has a negative impact on health and quality of life (Yang et al., 2014) . Hypertension, diabetes, dyslipidaemia and obesity are common conditions for LTRs; research has found 41-63% have high blood pressure, 21-45% have diabetes, 31-70% have high cholesterol and 67-87% are overweight or obese (Akarsu et al., 2013; Albeldawi et al., 2012; Anastácio et al., 2011; De Sena Ribeiro et al., 2014; Dehghani et al., 2007; Everhart et al., 1998; Pinto et al., 2016; Ribeiro et al., 2014; Richards et al., 2005; Richardson et al., 2001) .

CVD risk is modifiable through diet, however, post-transplant diet and the determinants of diet (and therefore the best way to support LTRs to achieve a healthy diet) are not well understood. In our systematic review of all published studies reporting nutritional intake post-transplant, we found that on average LTRs do not achieve energy and protein recommendations needed for recovery for the first month after transplant but after this energy intake increases and patients likely consume a diet high in energy for longer than required. Compared to international and national recommendations for general populations, we found that, on average, LTR generally consume a diet high in total fat and low in fibre, fruits and vegetables. Limited evidence from our systematic review suggests that time since transplant, gender and geographical location may be associated with nutritional intake. Only one study included in the review studied UK-based LTRs in Scotland. Most studies were based in countries where diet and its determinants are different to the UK, including Brazil, Mediterranean countries, eastern Europe and India, therefore further research in UK populations is required (Spillman et al., 2023).

When considering diet after transplant, attention should also be given to lifestyle prior to transplantation as unhealthy lifestyle behaviours are associated with some causes of liver failure. Alcohol-related liver disease (ARLD) and metabolic dysfunction-associated steatotic liver disease (MASLD) are the leading two causes of liver disease in UK patients waiting for transplant (Williams et al., 2014). MASLD is caused by an unhealthy lifestyle, including poor diet (Marchesini et al., 2016). It is hypothesised that unhealthy dietary behaviours replace alcohol addiction (Brunault et al., 2015). Additionally, before liver transplant, patients are advised to consume six energy and protein-dense meals or snacks per day, independent of liver disease aetiology (Plauth et al., 2019). Although appropriate in the peri-transplant stage, this may encourage unhealthy dietary behaviours long-term after liver transplant if patients are not supported with changing behaviours once recovered.

Rationale for the project

Measuring diet post liver transplant will help to identify if an intervention to promote healthier diet is needed. Quantifying dietary intake and determinants of diet will help to understand what an intervention should target, for example the elements of diet that need to be improved and potential modifiable factors influencing diet.

Research questions

What are the diets of patients after liver transplant?

What are the determinants of diet in LTRs?

Study population

English-speaking patients aged 18 years or over who had a liver transplant at Cambridge University Hospitals NHS Foundation Trust in the period 6 months to three years before the study start date were eligible for inclusion. Patients were excluded if they were not fluent in the English language, had an additional solid organ transplant other than liver +/- kidney (e.g. pancreas or bowel transplant) and were unable to provide informed consent.

Measurements

The items measured and method of measurement are shown in table 1 with more detail below. Measurements were taken at baseline (study recruitment) and follow-up (6 months after recruitment to study).

Table 1: Items measured and method of measurement

Item measured	Method of measurement
Dietary intake	Multiple 24-hour recalls using Intake24 – self-completed computerised dietary recall system based on multiple-pass 24-hour recall.

Plasma carotenoids and vitamins	Carotenoids measured from blood sample: lutein, α-cryptoxanthin, β-cryptoxanthin, lycopene, α-carotene and β-carotene. Vitamins measured from blood sample: retinol, α-tocopherol and γ-tocopherol.
Eating behaviour traits	Adult eating behaviour questionnaire and restraint items from the three-factor eating questionnaire.
Coping	Brief-COPE
Depression	Patient Health Questionnaire (PHQ-8)
Anxiety	Generalised Anxiety Disorder Questionnaire (GAD-7)
Stress	Perceived Stress Scale Questionnaire (PSS)
Age	Questionnaire
Sex	Questionnaire
Ethnicity	Questionnaire
Marital status	Questionnaire
Age left full time education	Questionnaire
Employment status	Questionnaire
Index of multiple deprivation	Derived from postcode
Smoking	Questionnaire
Self-reported health status	EQ-5D-5L and SF-8 questionnaire
Aetiology of liver disease	Electronic health record
Liver transplant reason	Electronic health record
Hepatocellular carcinoma (HCC)	Electronic health record
Time since transplant	Electronic health record
Total number of transplants	Electronic health record
Baseline liver function tests	Electronic health record
Medication	Electronic health record
Co-morbidities	Electronic health record
Weight and height	Electronic health record
BMI	Calculated from weight and height
Physical activity	Accelerometer worn for 7 days

Subjective dietary assessment was used to assess overall diet quality. A nutritional biomarker can provide a method with less error than subjective dietary assessment to examine associations between diet and determinants, therefore plasma carotenoids were also measured (Woodside et al., 2017). Multiple 24-hour dietary recalls were used as the subjective method. A total of four 24-hour recalls at each data collection time point was used to measure individual usual intake. The 24-hour recalls included 3 weekdays (Monday-Friday) and 1 weekend day (Saturday or Sunday). Multiple recalls are more reliable than single 24-hour recall and average intakes better represent habitual intakes (Cade et al., 2017). Validity of energy intakes reported using Intake24 has been assessed against concurrent measurement of total energy expenditure using doubly-labelled water, an objective method of measuring energy intake. In this study, estimation of energy intake using Intake24 was found to be comparable with 24hr dietary recall and estimated food diaries (Foster et al., 2019). Plasma carotenoids were measured as an objective marker of fruit and vegetable intake at the Cambridge Biomedical Research Centre (BRC) Nutritional Biomarker Lab (NBL) using standard operating procedures (SOPs).

The adult eating behaviour questionnaire (AEBQ) was used to measure food approach and avoidance appetite traits. This has been shown to be valid and reliable

(Hunot et al., 2016; Mallan et al., 2017). The AEBQ does not measure food restraint, therefore restraint items from the three-factor eating questionnaire (TFEQ) R21 was used (Cappelleri et al., 2009). Food restraint may be an important determinant of eating behaviour for this population.

The Brief-COPE questionnaire was used to measure coping strategies (Carver, 1997). Qualitative research suggests effective coping strategies are required before diet and activity behaviours can be addressed and therefore coping strategies may influence these behaviours (Hickman et al., 2019). This questionnaire has been validated and used with liver transplant patients (Amoyal et al., 2016; Ángeles Pérez-San-Gregorio et al., 2017).

Depression was measured using the Patient health questionnaire (PHQ-8) (Kroenke et al., 2009). Anxiety was measured using the Generalised Anxiety Disorder questionnaire (GAD-7) (Spitzer et al., 2006). PHQ-8 and GAD-7 assess symptoms occurring in the past two weeks. Perceived stress was measured using the Perceived Stress Scale (PSS) (Cohen et al., 1983). PSS assesses stress over the past month. These questionnaires are all validated self-report tools and widely used in clinical populations.

SF-8™ and the EQ-5D-5L (Herdman et al., 2011) are questionnaires which measure functional status and health utility, respectively. Both functional status and health utility are important information to understand for this population. The EQ-5D-5L measures five dimensions of health (mobility, self-care, usual activities, pain/discomfort and anxiety/depression) with five response levels (no problems, slight problems, moderate problems, severe problems, unable to/extreme problems). It also includes a visual analogue scale of overall health ranging from 0 (worst imaginable health) to 100 (best imaginable health) (Herdman et al., 2011). The SF-8™ is a short version of the SF-36™ QOL questionnaire and was chosen to minimise the number of questions asked and reduce participant burden. The SF-8™ consists of eight questions with Likert scale rating that vary by question. Each question measures one domain: general health perception (GH), physical functioning (PF), role limitations due to physical health problems (role physical, RP), bodily pain (BP), energy/fatigue (vitality, VT), social functioning (SF), role limitation due to emotional problems (role emotional, RE), and psychological distress and well-being (mental health, MH).

Accelerometers were used as an objective measure of PA. A validated waterproof 3 axis wrist accelerometer (GeneActiv) was worn continuously for 7 days at baseline and follow-up as an objective measure of physical activity (Esliger et al., 2011; Pavey et al., 2016).

Analysis

Descriptive analysis – frequency, central tendency and variability

For continuous data, parametric data will be summarised as mean and standard deviation (SD) and non-parametric data as medians and interquartile range (IQR). Categorical data will be reported as counts and percentages.

We will explore if there are different proportions of missing values by socioeconomic status (SES) by looking a differences in averages for continuous data and proportions for categorical data by tertiles of the index of multiple deprivation (IMD) obtained from postcode data. We will explore if there are differences between baseline and follow-up.

We will summarise the following data at baseline and follow-up (where available).

- Demographic, clinical, anthropometry, cardiovascular disease risk factors (table 2)
- Self-reported health status
- Behaviour determinants
- Diet
- Physical activity
- Mental health

Table 2: Demographic, clinical, anthropometry and cardiovascular risk data

Variable	Summary statistic
Measured and dry weight and participant reported pre-morbid (pre liver disease) weight (kg)	Average and variance
Estimated dry and patient reported pre-morbid body mass index (BMI) (kg/m2) –	Average and variance and by World Health Organisation Category
Age	Average and variance
Sex	Proportion male and female
Ethnicity	Proportion White, Multi-ethnic, Asian, Black, other
Employment status	Proportion working full time, part-time, keeping house, retired, waiting to start a new job, unemployed, temporarily sick, permanently sick or disabled and other
Marital status	Proportion single, married or living as married, widowed, separated, divorced
Age completed full time education	Proportion ≤ 16 years
Index of Multiple Deprivation	Tertiles of deprivation
Smoking	Proportion current smoker, past smoker, never smoked Pack years
Aetiology of liver disease	Proportion with: Alcohol-related liver disease Metabolic dysfunction-associated steatotic liver disease Hepatitis C Hepatitis B PSC PBC Autoimmune hepatitis Polycystic liver disease Other

Acute or chronic liver failure	Proportion with acute liver failure Proportion with chronic liver disease
Hepatocellular carcinoma (HCC)	Proportion with HCC
Time since most recent liver transplant at baseline	Average and variance
Total number of liver transplants	Proportion having had 1, 2, 3 and 4 transplants
Liver function tests at baseline	Proportion with high Alanine Transaminase (>40 U/L), Alkaline Phosphatase (>130 U/L) and total bilirubin (>20 U/L), and low albumin (<35 g/L).
Medication	Proportion taking the following medication: Antihypertensives, oral glucose lowering, insulin, cholesterol lowering, tacrolimus, cyclosporin, mycophenolate mofetil, sirolimus and steroids.
Co-morbidities	Proportion with diabetes, hypertension, dyslipidaemia, ischaemic heart disease. • Diabetes is defined as a diabetes diagnosis, HbA_{1c}: ≥48 mmol/mol and/or diabetes medication • Hypertension is defined as hypertension diagnosis, BP ≥140/90mmHg or BP medication • Dyslipidaemia is defined as dyslipidaemia diagnosis, TC:HDL ratio: ≥6 mmol/L, triglycerides: ≥2.3 mmol/L (non-fasting), or cholesterol-lowering medication.

Diet

We will describe average blood carotenoid and vitamin levels, total energy intake, grams and percent energy macronutrients, grams of fibre, food groups (table 3), relative Mediterranean Diet Score (rMDS) (Buckland et al., 2010) (table 4), and Eatwell Guide score (Public Health England, 2016) (table 5).

Table 3: Food groups as described by the National Diet and Nutrition Survey

Cereals and cereal products
Pasta, rice, pizza and other miscellaneous cereals
Sandwiches
White bread
Wholemeal bread
Brown, granary and wheatgerm bread
High fibre breakfast cereals
Other breakfast cereals
Biscuits
Buns, cakes, pastries and fruit pies
Puddings
Milk and milk products
Whole milk (3.8% fat)
Semi skimmed milk (1.8% fat)
Other milk and cream
Cheese
Cheddar cheese
Other cheese
Yoghurt, fromage frais and other dairy desserts

Ice cream
Eggs and egg dishes
Fat spreads Butter
Meat and meat products Bacon and ham Beef, veal and dishes Pork and dishes Coated chicken and turkey Chicken, turkey and dishes Burgers and kebabs Sausages Meat pies and pastries Other meat, meat products and dishes
Fish and fish dishes White fish coated or fried including fish fingers Other white fish, shellfish or fish dishes and canned tuna Oily fish
Vegetables and potatoes Salad and other raw vegetables Vegetables (not raw) including vegetable dishes Chips, fried and roast potatoes and potato products Other potatoes, potato salads and dishes
Savoury snacks
Nuts and seeds
Fruit
Sugar, preserves and confectionery Sugars, including table sugar, preserves and sweet spreads Sugar confectionery Chocolate confectionery
Non-alcoholic beverages Fruit juice Soft drinks, not low calorie Tea, coffee and water
Alcoholic beverages Wine Beer, lager, cider and perry
Miscellaneous Soup, manufactured/retail and homemade Savoury sauces, pickles, gravies and condiments

Table 4: Relative Mediterranean Diet Score (rMDS) scoring system (Buckland et al., 2010)

	Points

Food groups	Tertile 1	Tertile 2	Tertile 3
Fruit (including nuts, smoothies and fruit juice)	0	1	2
Vegetables (excluding potatoes)	0	1	2
Legumes	0	1	2
Fish	0	1	2
Cereals (including all food groups in the NDNS cereal category which includes biscuits, cakes, pizza and pastry)	0	1	2
Total meat	2	1	0
Dairy products	2	1	0
Olive oil*	0 = Used in 0 recalls (non-consumers)	1 = Used in up to half of recalls	2 = Used in half of recalls or more
Alcohol	0 = above or below 5-25g/day for women & 10-50g/d for men		2 = 5-25g/day for women & 10-50g/d for men

Intakes are adjusted to grams/1000kcal/day. *The original rMDS scored olive oil based on grams of olive oil intake, however Intake24 does not collect these data, therefore the olive oil score has been adapted based on the data available.

Table 5: Eatwell Guide score scoring system (Public Health England, 2016)

Food/nutrient	Cut off
Free sugars	≤5% total energy
Total fat	≤35% total energy
Saturated fat	≤11% total energy
Salt, not adding salt	≤6g/day
Carbohydrate	≥50% energy
Protein	14.5-15.5% energy
Fibre	≥30g (AOAC)
Fruit and vegetables*	≥400g/day (equivalent to ≥5 portions per day)
Oily fish	≥20g/day (equivalent to ≥1 portion per week)
Non-oily fish	≥20g/day (equivalent to ≥1 portion per week)
Red and processed meat	≤70g per day

*30g of dried fruit, max 150ml fruit juice or smoothie and max 80g beans considered as one portion
Score 0 if not meeting recommendation and 1 if meeting recommendation, maximum score is 11.

Physical activity

Describe the time spent in light activity (Euclidean Norm Minus One (ENMO 30) and above) and moderate-vigorous physical activity (MVPA) (ENMO 125 and above) from accelerometry data.

Self-reported health status

Describe the self-reported health status scores from EQ-5D-5L and SF-8.

The SF-8 has eight subscales with a 5-6 level Likert scale response. In the original scoring, the eight subscales were scored from 0 (worst health) to 100 (best heath). More recently, norm-based scoring (NBS) has been recommended, where the mean score in the US general population is set to 50 and the standard deviation (SD) is set to 10. The rescaling from the old (0-100) to the new (mean=50, SD=10) scoring is done by a linear transformation (Ware et al., 2001).

The summary health components, Physical Heath Component (PCS) and Mental Health Component (MCS), are derived from the eight subscales above. Both summary health components summarise information from all eight subscales but with different weights. These weights were derived from a principal components analysis. For PCS, highest weights are given to the physical subscales while some mental subscales are given negative weights. For MCS, highest weights are given to the mental subscales while some physical subscales are given negative weights (Ware et al., 2001).

We will describe the frequency of each level for the eight SF-8 subscales, and the overall, PCS and MCS scores.

To derive a health state from the EQ-5D-5L, each of the five dimensions is divided into five levels of perceived problems:

- Level 1: indicating no problem
- Level 2: indicating slight problems
- Level 3: indicating moderate problems
- Level 4: indicating severe problems
- Level 5: indicating unable to/extreme problems

A unique health state is defined by combining one level from each of the five dimensions. A total of 3125 possible health states is defined in this way. Each state is referred to by a 5-digit code. State 12345 indicates no problems with mobility, slight problems with washing or dressing, moderate problems with doing usual activities, severe pain or discomfort and extreme anxiety or depression, while state 11111 indicates no problems on any of the five dimensions and 55555 indicates extreme problems with all five dimensions. The VAS is scored 0-100 (EuroQol, 2019).

We will describe the frequency of each level of the five EQ-5D-5L dimensions. We will also report the average and variance of the VAS.

Eating behaviour traits

Describe the adult eating behaviour questionnaire (AEBQ) scores and restraint item scores from the three-factor eating questionnaire (TFEQ-21). The AEBQ consists of 35 questions that measure enjoyment of food, emotional overeating, emotional undereating, food fussiness, food responsiveness, hunger, slowness in eating and satiety responsiveness. Questions are measured using a five level Likert scale,

scored 1-5. Reversed questions are scored in the opposite direction. We have used seven questions from the TFEQ-21 to measure restraint. Five questions are measured using a four-level Likert scale and one question uses a 1-8 VAS. All questions are scored 1-4 (Cappelleri et al., 2009). We will describe the average and variance of scores for each domain.

Brief COPE questionnaire

Describe the Brief COPE scores. The Brief COPE is a 28-item self-report measure developed from the full version COPE inventory (60 items) to assess coping behaviours. Responses are measured using a four-level Likert scale. The Brief COPE assesses 14 coping behaviours (measured using 2 items), categorised across three coping styles:

Problem-focused coping style, including active coping, planning, positive reframing, using instrumental support. High scores are positive, indicative of psychological strength, grit, a practical approach to problem solving.

Emotion-focused coping, including using emotional support, acceptance, humour, venting, religion, and self-blame. A high score indicates coping strategies that are aiming to regulate emotions associated with the stressful situation.

Avoidance coping, including self-distraction, denial, substance use, behavioural disengagement. High scores are negative and indicative of maladaptive coping.

https://novopsych.com.au/assessments/formulation/brief-cope/

We will describe average scores for these three coping styles and for the 14 coping behaviours:

- Active coping, items 2 & 7 (Problem-Focused)
- Use of informational support, items 10 & 23 (Problem-Focussed)
- Positive reframing, items 12 & 17 (Problem-Focused)
- Planning, items 14 & 25 (Problem-Focused)
- Emotional support, items 5 & 15 (Emotion-Focused)
- Venting, items 9 & 21 (Emotion-Focused)
- Humor, items 18 & 28 (Emotion-Focused)
- Acceptance, items 20 & 24 (Emotion-Focused)
- Religion, items 22 & 27 (Emotion-Focused)
- Self-blame, items 13 & 26 (Emotion-Focused)
- Self-distraction, items 1 & 19 (Avoidant)
- Denial, items 3 & 8 (Avoidant)
- Substance use, items 4 & 11 (Avoidant)
- Behavioral disengagement, items 6 & 16 (Avoidant)

Mental health

Describe the depression Patient Health Questionnaire (PHQ-8) score. PHQ-8 assesses symptoms occurring in the past two weeks. Each item is rated as not present (0), several days (1), more than half the days (2), and nearly every day (3), giving a total depression symptom score of 0-24. A score of ≥10 indicates depression. Describe the Generalised Anxiety Disorder questionnaire (GAD-7) score. GAD-7 assesses symptoms occurring in the past two weeks. Each item is rated as not present (0), several days (1), more than half the days (2), and nearly every day (3), giving a total score of 0-21. A score of ≥10 indicates anxiety. Describe the Perceived Stress Scale (PSS) score. PSS is a ten-item questionnaire to assess stress levels over the past month. Each item is rated as never (0); almost Never (1); sometimes (2); fairly often (3), and very often (4), giving a total score of 0-40. PSS scores are obtained by reversing responses (e.g., 0 = 4, 1 = 3, 2 = 2, 3 = 1 & 4 = 0) to the four positively stated items (items 4, 5, 7, & 8) and then summing across all 10 items. There are no cut-offs for PSS and higher scores indicate higher stress. We will describe average and variance for all scores and frequencies for PHQ-8 and GAD-7 <10 and ≥10.

Descriptive analysis – Associations between diet and other variables

We will describe associations between diet (rMDS, Eatwell Diet score and carotenoids) and the baseline variables shown in table 6. For categorical variables, for example sex, we will use the t-test if data are parametric and Mann-Whitney test if data are non-parametric to assess differences in average diet scores and carotenoids between categories. For continuous data, for example mental health scores, we will use Pearson's correlation for parametric data and Spearman's correlation for non-parametric data. For polytomous data, for example tertiles of IMD, we will use one-way ANOVA for parametric data and the Kruskal-Wallis test for non-parametric data.

Univariable analysis will be used to explore differences for outcomes (rMDS and total carotenoids) between baseline and follow-up using the paired t-test for parametric and Wilcoxon test for non-parametric data.

Table 6: Variables for descriptive analysis exploring associations with diet

Type of variable	Variable	Categorical (categories) or continuous	Exposure or effect modifier/confounder
Demographics	Age	continuous	Effect modifier/confounder
	Sex	Categorical: male/female	Effect modifier/confounder
	Marital status	Categorical: married or living as married/other	Effect modifier/confounder
	Age left full time education	Categorical: ≤16/>16 years	Effect modifier/confounder
	Employment status	Categorical: working (full-time or part-time)/other	Effect modifier/confounder
	Index of Multiple Deprivation	Categorical: tertiles of deprivation	Effect modifier/confounder

Liver disease information	Aetiology of liver disease	Categorical: ARLD or MASLD/other	Effect modifier/confounder
	Liver transplant reason	Categorical: acute/chronic	Effect modifier/confounder
	Time since transplant	Continuous and Categorical: <12 month, ≥12 months	Effect modifier/confounder
	Hepatocellular carcimoma present	Categorical: Yes/No	Effect modifier/confounder
	Immunosuppression use	Categorical: steroids/no steroids Categorical: tacrolimus/no tacrolimus Categorical: ciclosporin/no ciclosporin	Effect modifier/confounder
	Abnormal liver function tests (LFT)	Categorical: Any LFT results above normal/all results within normal range	Effect modifier/confounder
Other health behaviours	Smoking status	Categorical: Current smoker/not current smoker Continuous:pack years	Effect modifier/confounder
	Physical activity	Categorical: Time spent in light activity/time spend below light activity and time spent in MVPA/time spent below MVPA. Quartiles of ENMO. Continuous: ENMO	Effect modifier/confounder
Health status	Self-reported health status	Continuous: SF-8 scores	Effect modifier/confounder
	Diabetes	Categorical: diabetic/not diabetic	Effect modifier/confounder
	Hypertension	Categorical: with hypertension/without hypertension	Effect modifier/confounder
	Dyslipidaemia	Categorical: with dyslipidaemia/without dyslipidaemia	Effect modifier/confounder
Eating behaviours	Eating behaviour traits	Continuous: AEBQ score and restrain item score	Exposure
Mental health	Coping	Continuous: Brief-COPE score for problem-focused coping, emotion-focused coping, and avoidant coping	Exposure
	Depression	Continuous: PHQ-8 score Categorical: depressed and not depressed	Exposure
	Anxiety	Continuous: GAD-7 score	Exposure

		Categorical: Anxiety and no anxiety	
	Stress	Continuous: PSS score	Exposure

Inferential analysis – potential determinants of diet

The primary outcomes are shown in table 7.

Table 7: Primary outcomes and methods of measurement

Outcome	Method of measurement
Relative Mediterranean Diet Score (rMDS)	Multiple 24-hour recalls using Intake24 – self-completed computerised dietary recall system based on multiple-pass 24-hour recall.
Total plasma carotenoids	Carotenoids measured from blood sample: lutein, α-cryptoxanthin, β-cryptoxanthin, lycopene, α-carotene and β-carotene. Vitamins measured from blood sample: retinol, α-tocopherol and γ-tocopherol.

The average intake from all diet recalls and average total plasma carotenoids will be used to calculate the primary outcomes.

Simple linear regression will be used to investigate relationships between exposures/effect modifiers/confounders (see table 6) and outcomes in a univariable analysis.

Multiple linear regression will be used to investigate relationships between exposures and outcomes with adjustment for confounders/effect modifiers. We will add the potential confounders/effect modifiers to the model by category (demographics, liver disease information, other health behaviours and health status) in a stepwise way until we achieve a final maximally adjusted model.

From research in general populations and LTRs we know that the factors we have defined as confounders and effect modifiers are associated with dietary intake, however the best multiple regression model for the study sample cannot be defined *a priori*. To build the best possible model we will explore simple regression, the interactions between independent variables to avoid multicollinearity and residual plots, review changes in adjusted R-square and Mallow's cp as independent variables are added/removed from the model and use theoretical knowledge to include important variables to avoid omitted variable bias. Due to the small sample size, the number of independent variables in the multiple regression models will be limited.

Subgroup analysis

Participants with ARLD/MASLD liver disease diagnosis.

References

Akarsu, M., Bakir, Y., Karademir, S., Unek, T., Bacakoglu, A., & Astarcioglu, I. (2013). Prevalence and risk factors for obesity after liver transplantation: a single-center experience. *Hepatitis Monthly*, *13*(8), 3. https://doi.org/10.5812/HEPATMON.7569

Albeldawi, M., Aggarwal, A., Madhwal, S., Cywinski, J., Lopez, R., Eghtesad, B., & Zein, N. N. (2012). Cumulative risk of cardiovascular events after orthotopic liver transplantation. *Liver Transplantation : Official Publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society*, *18*(3), 370–375. https://doi.org/10.1002/LT.22468

Anastácio, L. R., Ferreira, L. G., De Sena Ribeiro, H., Lima, A. S., Garcia Vilela, E., & Toulson Davisson Correia, M. I. (2011). Body composition and overweight of liver transplant recipients. *Transplantation*, *92*(8), 947–951. https://doi.org/10.1097/TP.0B013E31822E0BEE

Brunault, P., Salamé, E., Jaafari, N., Courtois, R., Réveillère, C., Silvain, C., Benyamina, A., Blecha, L., Belin, D., & Ballon, N. (2015). Why do liver transplant patients so often become obese? The addiction transfer hypothesis. *Medical Hypotheses*, *85*(1), 68–75. https://doi.org/10.1016/J.MEHY.2015.03.026

Buckland, G., Agudo, A., Luján, L., Jakszyn, P., Bas Bueno-de-Mesquita, H., Palli, D., Boeing, H., Carneiro, F., Krogh, V., Sacerdote, C., Tumino, R., Panico, S., Nesi, G., Manjer, J., Regnér, S., Johansson, I., Stenling, R., Sanchez, M. J., Dorronsoro, M., … González, C. A. (2010). Adherence to a Mediterranean diet and risk of gastric adenocarcinoma within the European Prospective Investigation into Cancer and Nutrition (EPIC) cohort study. *The American Journal of Clinical Nutrition*, *91*(2), 381–390. https://doi.org/10.3945/AJCN.2009.28209

Cappelleri, J. C., Bushmakin, A. G., Gerber, R. A., Leidy, N. K., Sexton, C. C., Lowe, M. R., & Karlsson, J. (2009). Psychometric analysis of the Three-Factor Eating Questionnaire-R21: results from a large diverse sample of obese and non-obese participants. *International Journal of Obesity (2005)*, *33*(6), 611–620. https://doi.org/10.1038/IJO.2009.74

De Sena Ribeiro, H., Anastácio, L. R., Ferreira, L. G., Lagares, É. B., Lima, A. S., & Correia, M. I. T. D. (2014). Prevalence and factors associated with dyslipidemia after liver transplantation. *Revista Da Associacao Medica Brasileira (1992)*, *60*(4), 365–372. https://doi.org/10.1590/1806-9282.60.04.016

Dehghani, S. M., Taghavi, S. A. R., Eshraghian, A., Gholami, S., Imanieh, M. H., Bordbar, M. R., & Malek-Hosseini, S. A. (2007). Hyperlipidemia in Iranian liver transplant recipients: prevalence and risk factors. *Journal of Gastroenterology*, *42*(9), 769–774. https://doi.org/10.1007/S00535-007-2092-2

Esliger, D. W., Rowlands, A. V., Hurst, T. L., Catt, M., Murray, P., & Eston, R. G. (2011). Validation of the GENEA Accelerometer. *Medicine and Science in Sports and Exercise*, *43*(6), 1085–1093. https://doi.org/10.1249/MSS.0B013E31820513BE

EuroQol. (2019). *EQ-5D-5L User Guide*. https://euroqol.org/publications/user-guides/

Everhart, J. E., Lombardero, M., Lake, J. R., Wiesner, R. H., Zetterman, R. K., & Hoofnagle, J. H. (1998). Weight change and obesity after liver transplantation: incidence and risk factors. *Liver Transplantation and Surgery : Official Publication of the American Association for the Study of*

Liver Diseases and the International Liver Transplantation Society, *4*(4), 285–296. https://doi.org/10.1002/LT.500040402

Herdman, M., Gudex, C., Lloyd, A., Janssen, M., Kind, P., Parkin, D., Bonsel, G., & Badia, X. (2011). Development and preliminary testing of the new five-level version of EQ-5D (EQ-5D-5L). *Quality of Life Research : An International Journal of Quality of Life Aspects of Treatment, Care and Rehabilitation*, *20*(10), 1727–1736. https://doi.org/10.1007/S11136-011-9903-X

Marchesini, G., Day, C. P., Dufour, J. F., Canbay, A., Nobili, V., Ratziu, V., Tilg, H., Roden, M., Gastaldelli, A., Yki-Jarvinen, H., Schick, F., Vettor, R., Fruhbeck, G., & Mathus-Vliegen, L. (2016). EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. *Journal of Hepatology*, *64*(6), 1388–1402. https://doi.org/10.1016/J.JHEP.2015.11.004

NHSBT. (2017). *Organ Donation and Transplantation Activity Report 2016/2017*. http://www.odt.nhs.uk.

Pavey, T. G., Gomersall, S. R., Clark, B. K., & Brown, W. J. (2016). The validity of the GENEActiv wrist-worn accelerometer for measuring adult sedentary time in free living. *Journal of Science and Medicine in Sport*, *19*(5), 395–399. https://doi.org/10.1016/j.jsams.2015.04.007

Pinto, A. S., Chedid, M. F., Guerra, L. T., Cabeleira, D. D., & Kruel, C. D. P. (2016). DIETARY MANAGEMENT FOR DYSLIPIDEMIA IN LIVER TRANSPLANT RECIPIENTS. *Arquivos Brasileiros de Cirurgia Digestiva : ABCD = Brazilian Archives of Digestive Surgery*, *29*(4), 246–251. https://doi.org/10.1590/0102-6720201600040008

Plauth, M., Bernal, W., Dasarathy, S., Merli, M., Plank, L. D., Schütz, T., & Bischoff, S. C. (2019). ESPEN guideline on clinical nutrition in liver disease. *Clinical Nutrition*, *38*(2), 485–521. https://doi.org/10.1016/J.CLNU.2018.12.022

Public Health England. (2016). *The Eatwell Guide*. https://www.gov.uk/government/publications/the-eatwell-guide

Ribeiro, H. S., Anastácio, L. R., Ferreira, L. G., Lima, A. S., & Correia, M. I. T. D. (2014). Energy expenditure and balance among long term liver recipients. *Clinical Nutrition (Edinburgh, Scotland)*, *33*(6), 1147–1152. https://doi.org/10.1016/J.CLNU.2013.12.015

Richards, J., Gunson, B., Johnson, J., & Neuberger, J. (2005). Weight gain and obesity after liver transplantation. *Transplant International : Official Journal of the European Society for Organ Transplantation*, *18*(4), 461–466. https://doi.org/10.1111/J.1432-2277.2004.00067.X

Richardson, R. A., Garden, O. J., & Davidson, H. I. (2001). Reduction in energy expenditure after liver transplantation. *Nutrition (Burbank, Los Angeles County, Calif.)*, *17*(7–8), 585–589. https://doi.org/10.1016/S0899-9007(01)00571-8

Spillman, L. N., Madden, A. M., Richardson, H., Imamura, F., Jones, D., Nash, M., Lim, H. K., Hellawell, H. N., Rennie, K. L., Oude Griep, L. M., Allison, M., & Griffin, S. J. (2023). Nutritional Intake after Liver Transplant: Systematic Review and Meta-Analysis. *Nutrients*, *15*(11). https://doi.org/10.3390/NU15112487/S1

Ware, J., Kosinski, M., Dewey, J., & Gandek, B. (2001). *How to Score and Interpret Single-Item Health Status Measures: A Manual for Users of the SF-8 Health Survey.* . Quality Metric Incorporated.

Watt, K. D. S., Pedersen, R. A., Kremers, W. K., Heimbach, J. K., & Charlton, M. R. (2010). Evolution of Causes and Risk Factors for Mortality Post-Liver Transplant: Results of the NIDDK Long-Term

Follow-Up Study. *American Journal of Transplantation*, *10*, 1420–1427. https://doi.org/10.1111/j.1600-6143.2010.03126.x

Williams, R., Aspinall, R., Bellis, M., Camps-Walsh, G., Cramp, M., Dhawan, A., Ferguson, J., Forton, D., Foster, G., Gilmore, S. I., Hickman, M., Hudson, M., Kelly, D., Langford, A., Lombard, M., Longworth, L., Martin, N., Moriarty, K., Newsome, P., … Smith, T. (2014). Addressing liver disease in the UK: a blueprint for attaining excellence in health care and reducing premature mortality from lifestyle issues of excess consumption of alcohol, obesity, and viral hepatitis. *Lancet (London, England)*, *384*(9958), 1953–1997. https://doi.org/10.1016/S0140-6736(14)61838-9

Yang, L. S., Shan, L. L., Saxena, A., & Morris, D. L. (2014). Liver transplantation: a systematic review of long-term quality of life. *Liver International*, *34*(9), 1298–1313. https://doi.org/10.1111/LIV.12553