

## Students as Co-Creators DRC Project Evaluation Guidelines

As you approach the end of your research project. You are expected to reflect on your findings, evaluate the research process, and think about the impact of your research. These guidelines have been created to help you write your final report.

Your report should consist of the following sections:

1. **Executive Summary**
2. **Background and Aims**
3. **Methods**
4. **Results**
5. **Discussion**
6. **Conclusion**
7. **Lessons Learned**
8. **Research Group Reflection**

Only **one report needs to be produced per team**. All members of the team should contribute towards writing this report.

### Section 1. Abstract (300-400 words)

The main aim of the project was to develop an easy to use health index test based on extracellular vesicles (EVs) and key microRNA EV-markers from plasma.

For this purpose, EVs were isolated from plasma from 23 recruited healthy volunteers. Profiles of EVs was assessed using Nanoparticle Tracking Analysis (NTA) using the NS300 Nanosight (Malvern) to quantify numbers and assess modal size of EVs in plasma. These markers were correlated with age, BMI and grip-strength (as indirect measurement of fitness) alongside other markers. Furthermore, a subset of 10 volunteers (5 males and 5 females) were assessed for the metabolic microRNA miR23b and this was correlated to EV profile.

A large part of the study included study design (questionnaire), recruitment of volunteers and selection of key microRNAs for metabolic function based on the literature.

This pilot study revealed a significant correlation of EV numbers with age, showing decreased EV numbers in plasma of older volunteers. Modal size changes were not significant. Furthermore, EV release could be associated with a selection of non-invasive markers. There was no significant difference in these parameters between males and females.

Next steps: A larger cohort of volunteers will allow for refinement of modelling and the development of a standard health EV-test from plasma, based on EV enumeration and EV microRNA profile.

*Tip: it might be a good idea to write this section last, although it needs to be at the beginning of your report.*

### Section 2. Background and Aims (200-300 words)

**Background:** Extracellular vesicles (EVs) are released from most cells as part of cellular communication, EVs carry cargo proteins and genetic material, including microRNAs. EVs can be isolated from most body fluids, including plasma, and can be used as biomarkers for assessment of health status.

**Overall Aim:** To develop an easy to use health index test based on extracellular vesicle (EV) profile and key microRNA EV-markers from human plasma.

**SUB-AIMS A-D:**

- a) To isolate EVs from plasma from a range of volunteers.
- b) To assess changes in numbers and size distribution profile of EVs and relate them to health markers that are easy to measure such as grip strength, BMI and age.
- c) To select a microRNA panel for key metabolic microRNAs that can be measured in the EV isolates.
- d) To model a standard optimal health status EV-test from plasma, based on EV enumeration and EV microRNA biomarker profile.

**Objectives:**

- a) The students designed a recruitment poster to attract healthy volunteers for the study. Plasma was isolated from 23 volunteers.
- b) The Nanosight NS300 system was used for nanoparticle tracing analysis (NTA) to count EVs isolated from volunteer's plasma. A key-element was to develop an effective fast-step EV isolation protocol, moving from a 3 hour 100,000 g ultracentrifugation protocol to a 2 step abbreviated protocol at 4,000 and 37,000 g, allowing for processing of 30 samples in 1h 20 min (compared to 8 samples previously in 3 hours).
- c) An extensive literature search for key inflammatory and metabolic markers lead to the conclusion to focus on miR23b and assess this marker in a selection of volunteers. Further miR markers have been selected to make up 3 key markers for the panel-test, but due to cost-effectiveness of this pilot project the focus was on this one key metabolic miR.
- d) EV numbers and modal sizes were found to be associated with some of the non-invasive tests such as BMI, age, grip-strength and blood pressure. Therefore this model can be refined including higher numbers of volunteers and include further microRNA profiling.

*How do you think this research will impact YOUR learning and teaching?*

-Understanding of how to design an effective recruitment poster and which questions are key to assessing health in volunteers for this type of study when they come to the laboratory.

-A major obstacle was the recruitment of participants and this project has given a very good insight into how hard and time consuming it is to carry out such an *in vivo* study which relies on volunteers and how important it is to have a team that works effectively together.

-Reliability of team members, co-ordination of tasks and standard approaches to methodologies are key factors.

-It is important to gain insight into how to optimise sample collection and processing to simplify the process for large-scale sample analysis.

-co-ordinating availability of academic staff and students, both with very busy and diverse time-tables is a major challenge.

**Section 3. Methods (150-300 words)**

For this section you need to think about exactly what you did it and be able to describe it clearly.

Think about:

What exactly did you do (surveys, interviews, library & archival research etc.)?

*We designed a health questionnaire for recruitment of healthy volunteers, this was based on similar questionnaires from previous studies.*

**How many participants did you use and who were they?**

23 volunteers all “healthy” in an age range of 18-65, mixed genders, with a BMI 18.6-29.7.

Blood was collected from a vein in the arm into EDTA coated cuvettes and plasma was isolated by centrifugation and frozen at -80°C until used for EV analysis.

For EV isolation, step-wise centrifugation was used, based on previously established protocols in Lang laboratory (Kosgodage et al., 2018 and 2019) and according to the recommendations of MISEV2018 (the minimal information for studies of extracellular vesicles 2018; Théry et al., 2018). The protocol was adjusted specifically in this pilot study for small scale samples as follows: plasma were diluted 1:4 in Dulbecco's PBS (DPBS which was ultrafiltered using a 0.22 µm filter) by adding 100 µl plasma to 400 µl DPBS per EV isolation. The diluted plasma were centrifuged at 4,000 g for 30 min at 4 °C for removal of cells and cell debris and thereafter the supernatant was collected and centrifuged at 37,500 g for 1 h at 4 °C for EV enrichment. The resulting EV pellet was then resuspended in 50 µl DPBS and stored frozen at -80°C until used. For nanoparticle tracking analysis (NTA), based on Brownian motion of particles in suspension (Soo et al., 2012), the EV samples were diluted 1/100 in DPBS (10 µl EVs added to 990 µl DPBS) and applied to the NanoSight NS300 system (Malvern, U.K.) using a syringe pump to ensure continuous flow of the sample. Videos were recorded for 4 x 60 sec, with approximately 30-60 particles per frame. The replicate histograms generated from the recordings were averaged for assessment of EV size distribution profiles of the plasma.

For microRNA analysis, RNA was extracted from EV preparations from 5 male and 5 female volunteers ranging in age and BMI, using Trizol (Sigma, U.K.). The purity and concentration of the isolated RNA were measured using the NanoDrop Spectrophotometer at 260 nm and 280 nm absorbance. For cDNA production, the qScript microRNA cDNA Synthesis Kit (Quantabio, U.K.) was used according to the manufacturer's instructions. The cDNA was used to assess the expression of miR23b, while U6-snRNA was used as reference RNAs for normalization of miR expression levels. The PerfeCTa SYBR Green SuperMix (Quantabio, U.K.) was used together with MystiCq microRNA qPCR primers for the miR23b (hsa-miR-23b-5p). All miR primers were obtained from Sigma (U.K.). Thermocycling conditions were used as follows: denaturation at 95 °C for 2 min, followed by 40 cycles of 95 °C for 2 sec, 60 °C for 15 sec, and extension at 72 °C for 15 sec. The  $2^{-\Delta\Delta CT}$  method (Livak and Schmittgen, 2001) was used for calculating relative miR expression levels and for normalisation.

**Did you take into account any ethical issues?**

All measurements and questionnaires were carried out according to approved Ethics.

**How did you analyse your data (eg. Software, statistics)?**

EV analysis was carried out using the NanoSight300 associated software to create histograms of EV distribution.

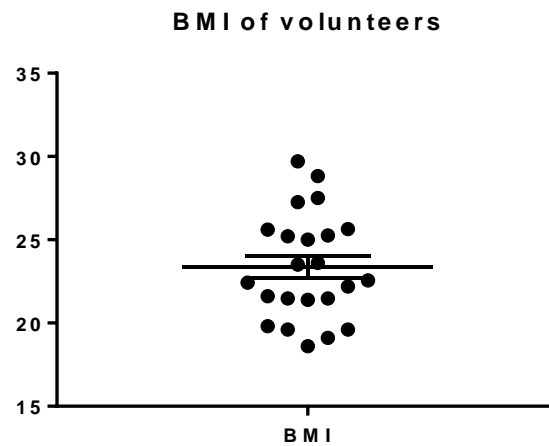
All graphs were created in either GraphPad Prims (version 7) or Excel. Statistical significance was at p < 0.05 following ANOVA.

*Tip: If you engaged with research participants, it would be useful to attach a copy of the Participant Information Sheet and questionnaire you used, as an Appendix at the end of the report.*

Please see questionnaire attached in Appendix.

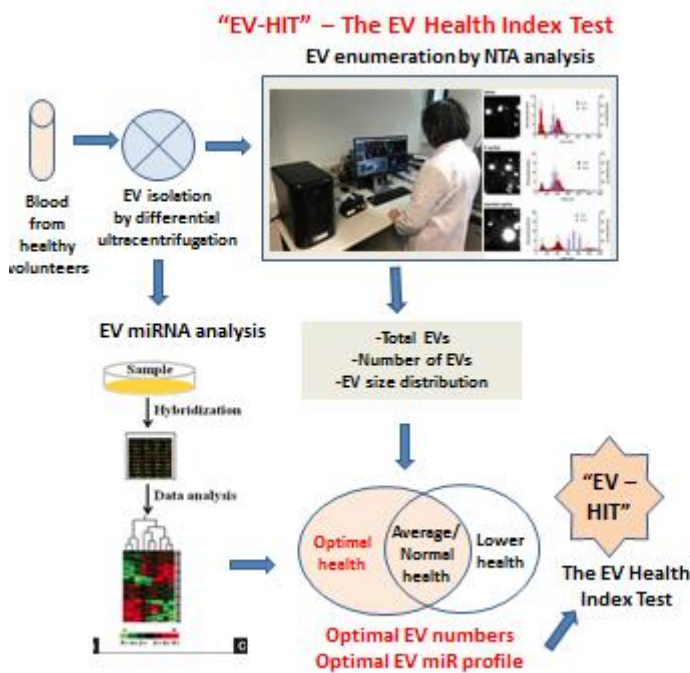
**Section 4. Results**

**4.1. Design of the Recruitment poster:** The recruitment poster was designed by the student team and recruitment was advertised at UoW as well as in a range of gyms. A range of 23 volunteers, with a BMI of 18.6-29.7 was recruited (Fig. 1B).



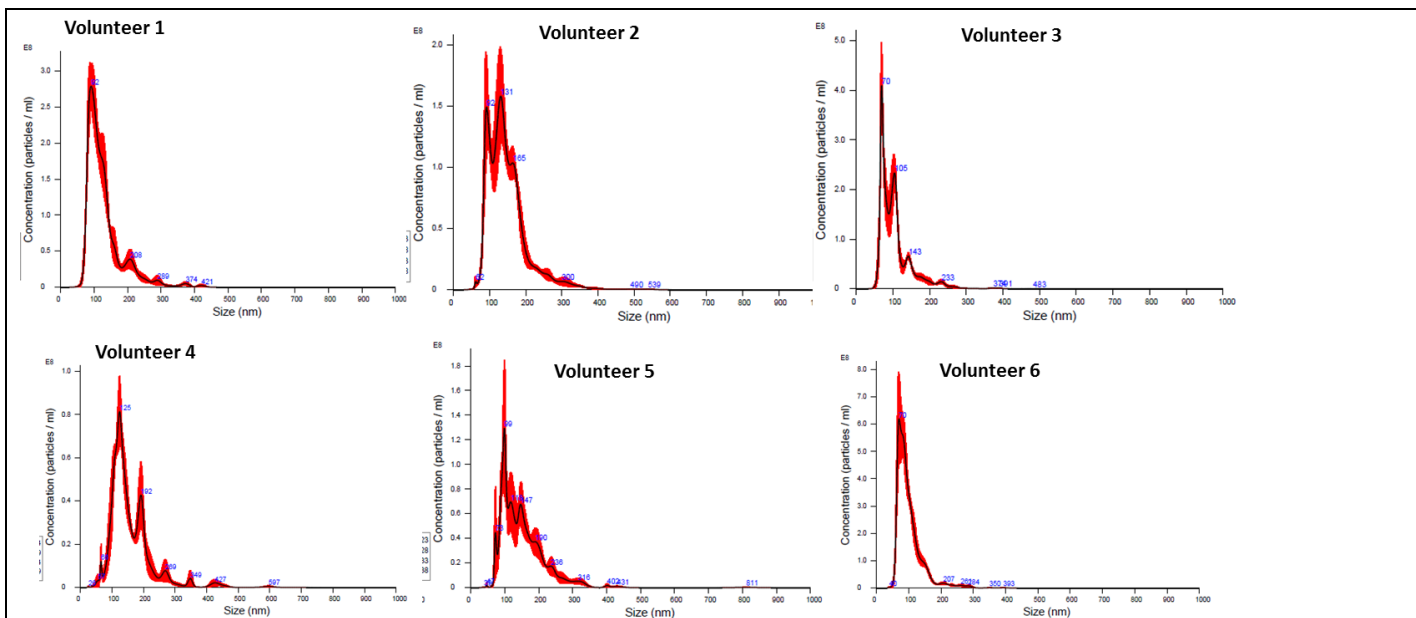
**Figure 1:** A. Recruitment poster for EV-HIT. B. BMI range of volunteers recruited.

**4.2. EV profiling:** EVs were isolated from plasma using step wise centrifugation and thereafter analysed by NTA. EVs were furthermore assessed for the metabolic miR23b. The flow of sample processing is summarised in Figure 2. below:



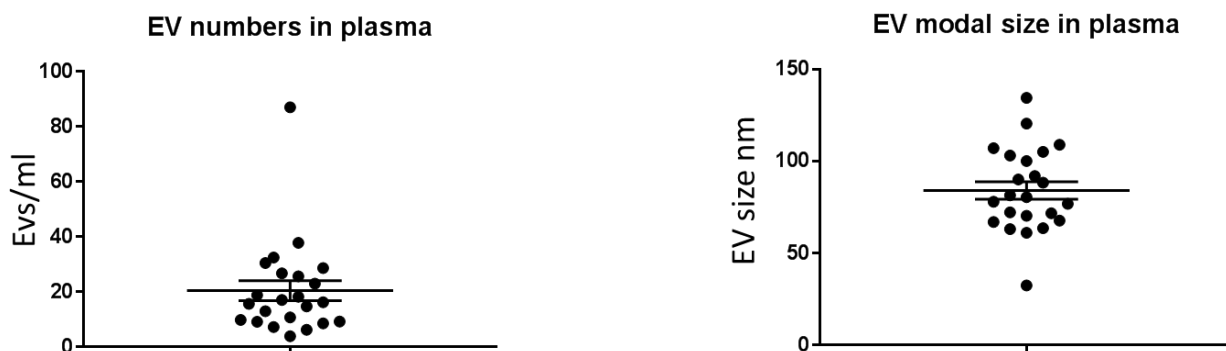
**Figure 2:** A summary of plasma processing for EV analysis and miR analysis in EVs.

**4.3. EV profiles in healthy volunteers:** NTA analysis was carried out for EVs from plasma from 23 volunteers in the age range of 18-65 years and with a range of BMI 18-29. A clear difference in EV profiles could be observed between individuals as shown in 6 examples in Fig. 2. While plasma EV profiles fell within the typical size range of 50 – 300 nm, in some volunteers a broad poly-dispersed size distribution was observed, including larger EVs upto 600 nm, while some plasma showed a very narrow monodispersed peak around 100 nm (Fig. 3).

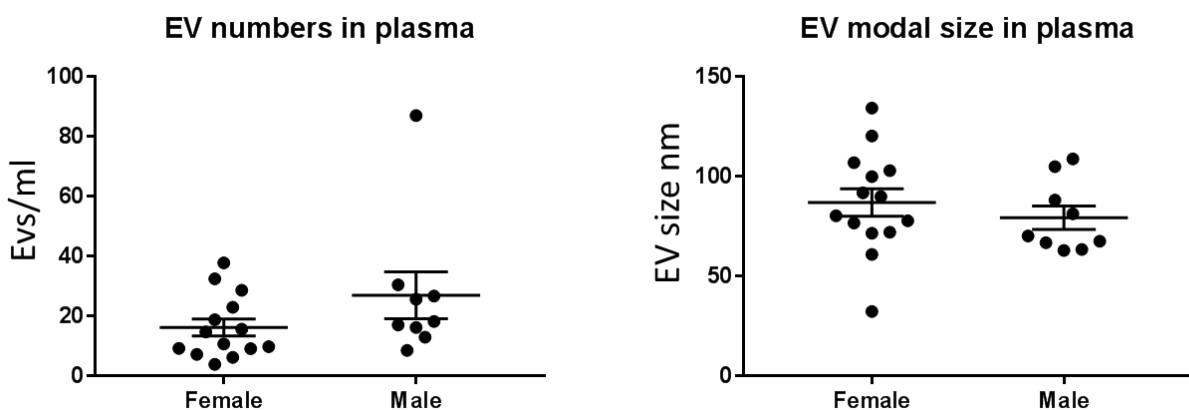


**Figure 3:** Examples of NTA EV profile analysis showing EV size distribution profiles from plasma of 6 volunteers.

**4.4 Total EV numbers in plasma across the volunteer range:** Distribution of EV total count profile was compared between all volunteers to assess distribution of EV size profiles (Figure 4).



**Figure 4:** EV numbers and modal size in plasma across the volunteer range. EV count and modal size of EVs released as by NTA analysis from plasma of the 23 volunteers.



**Figure 5:** EV numbers and modal size in plasma compared between genders. EV count and modal size of EVs released as by NTA analysis from plasma of the 23 volunteers; note outliers.

**4.5 Correlation of EV numbers and EV modal size with BMI, grip strength, fat mass:** Association between EV numbers and EV modal size to some of the parameters included in the questionnaire. A strong association was found with age, while a direct correlation to other parameters was not as significant. Note outliers.

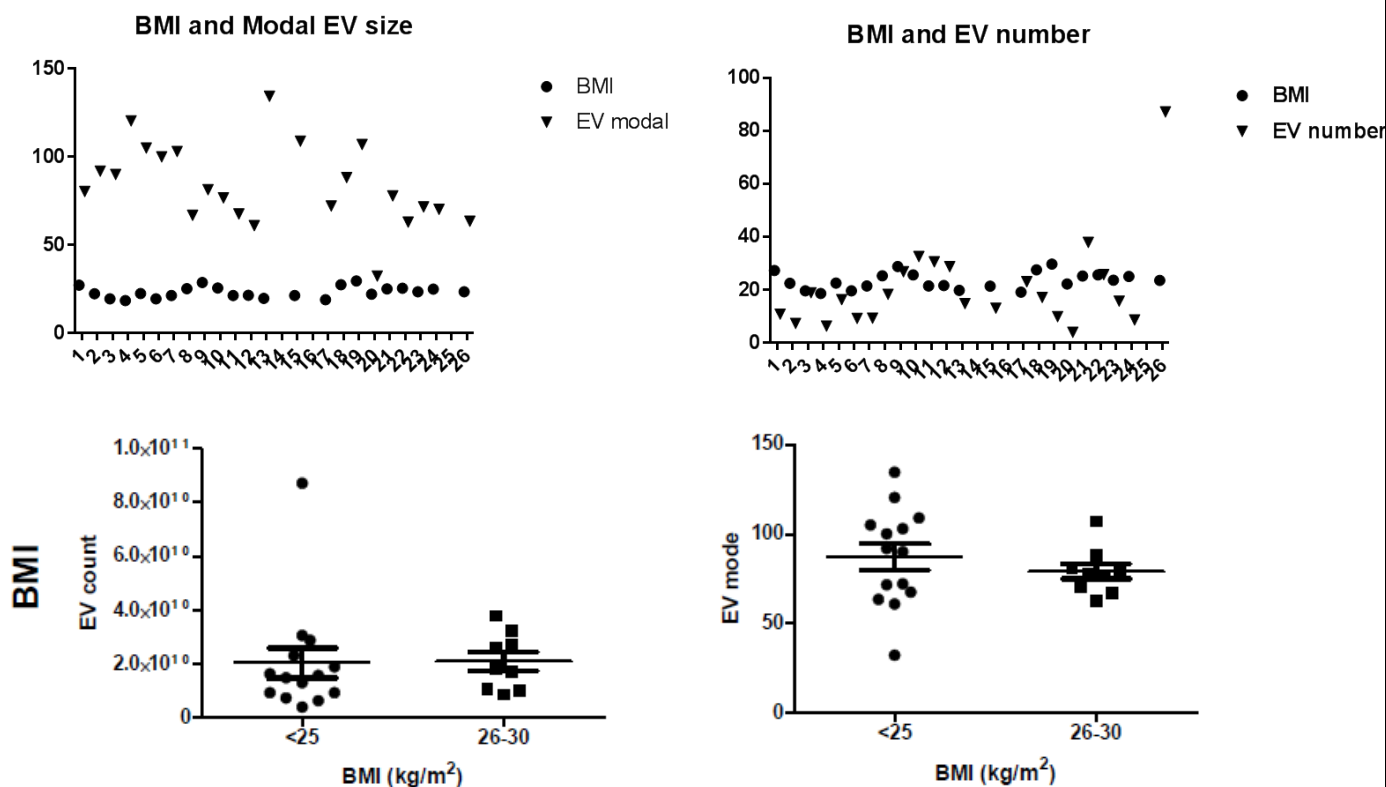


Figure 6: BMI correlation with EV numbers and modal size in volunteer plasma.

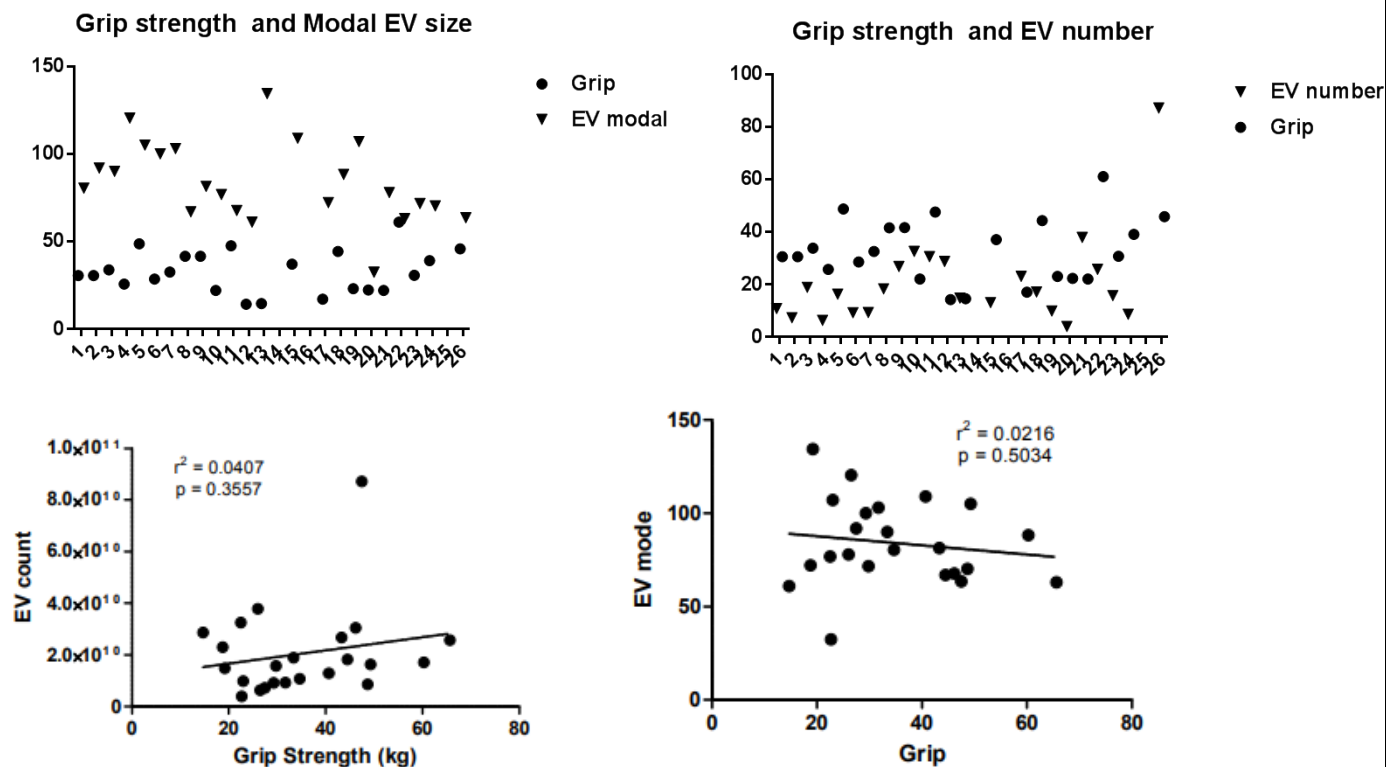
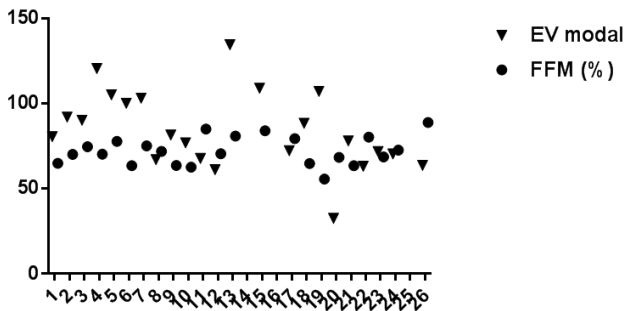


Figure 7: Grip strength correlation with EV numbers and modal size in volunteer plasma.

Free Fat Mass (%) and Modal EV size



Free Fat Mass (%) and EV number

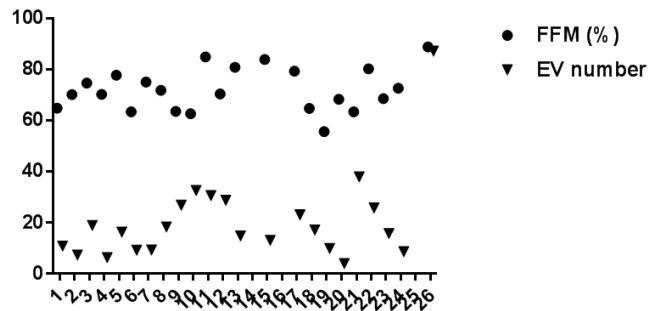
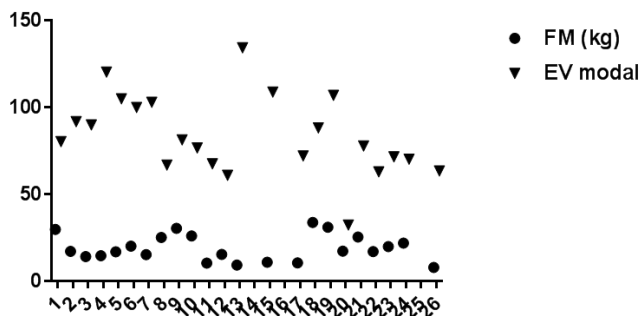


Figure 8: Free fat mass (%) correlation with EV numbers and modal size in volunteer plasma.

Fat Mass (kg) and Modal EV size



Fat Mass (kg) and EV number

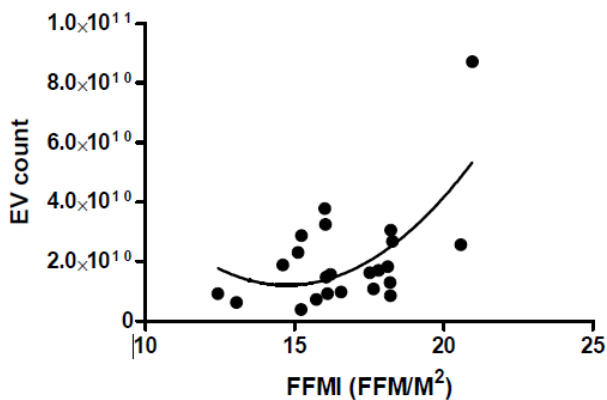
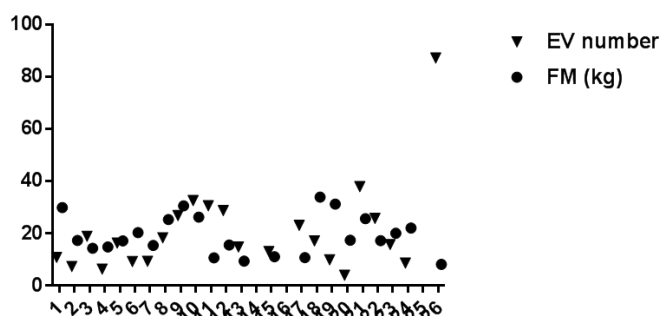


Figure 9: Fat mass (kg) and free fat mass (FFM) correlation with EV numbers and modal size in volunteer plasma.

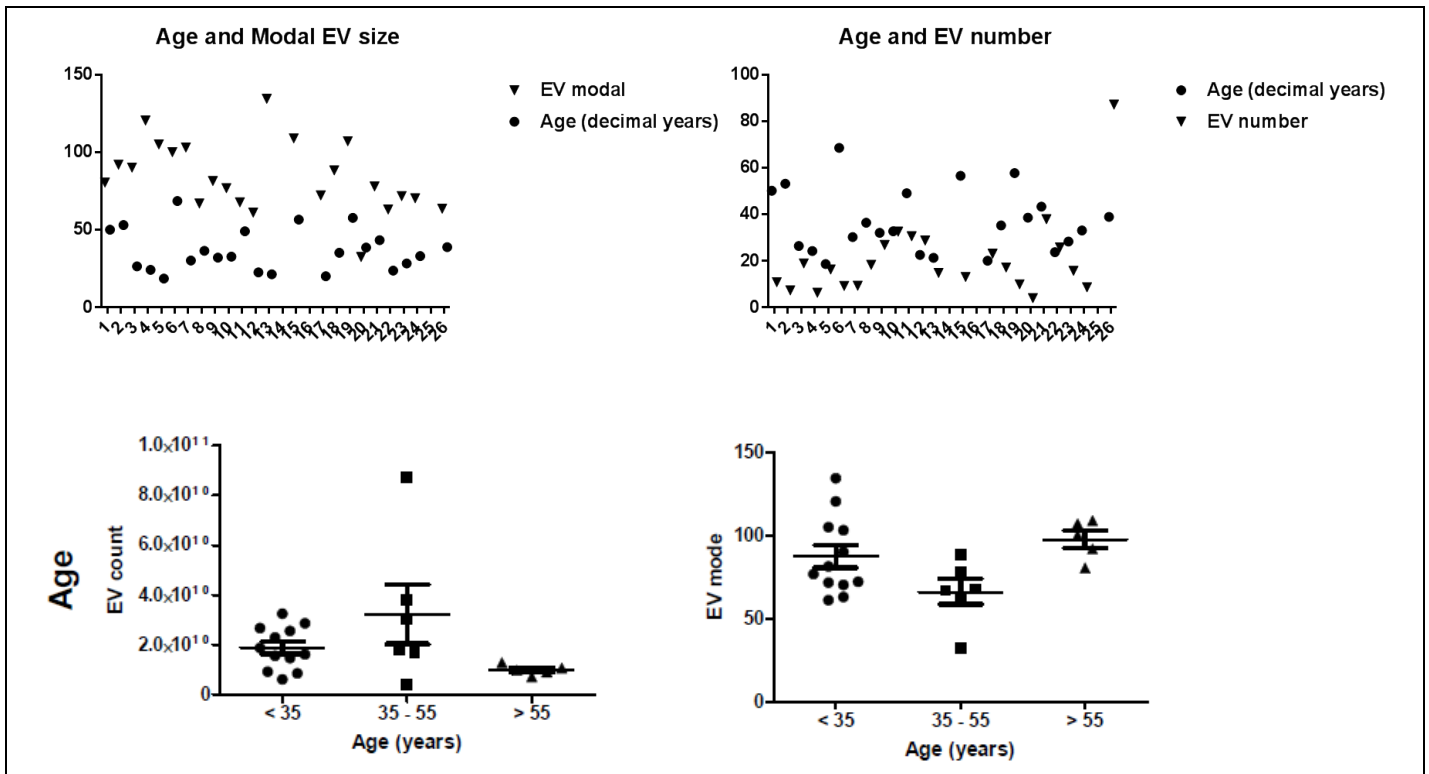


Figure 10: Age correlation with EV numbers and modal size in volunteer plasma.

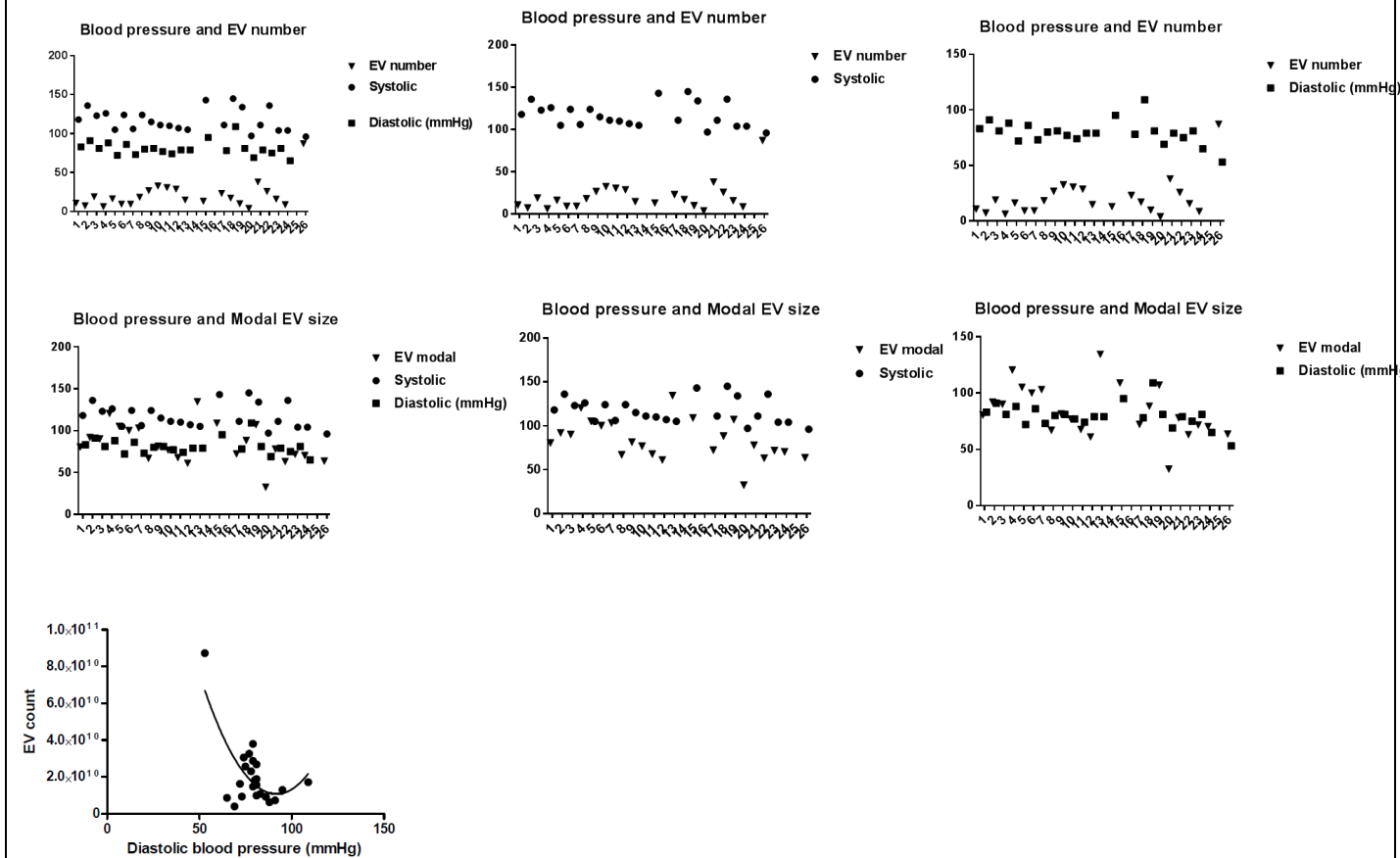


Figure 11: Blood pressure correlation with EV numbers and modal size in volunteer plasma. Systolic and diastolic blood pressure is shown in correlation with EV number and EV modal size.



#### 4.6 Assessment of the metabolic microRNA 23b in volunteer plasma

EVs isolated from the individual plasma were used to extract RNA which then was used for analysis of microRNA 23b, a key metabolic related miR. A selected 10 (5 males and 5 females) samples, with a range in BMI and age were selected.

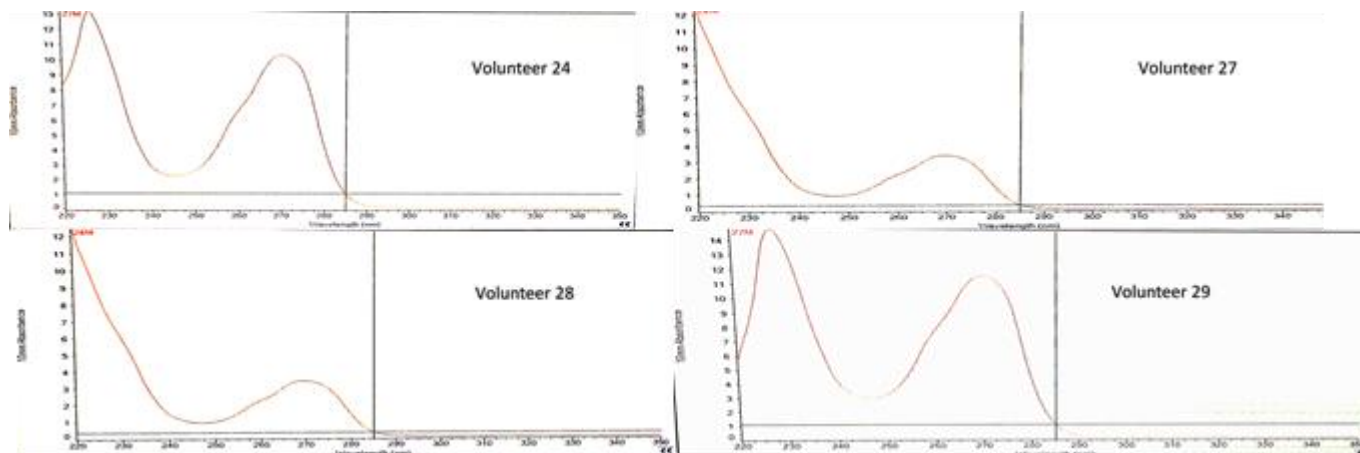
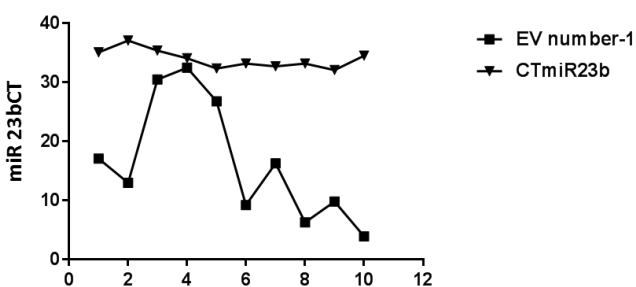


Figure 12: Examples of Nanodrop analysis showing EV-RNA concentration profiles from plasma of 4 volunteer: two females (volunteer 28 and 29) and two males (volunteer 24 and 27)

miR23b expression related to EV numbers



miR23b expression related to EV modal size

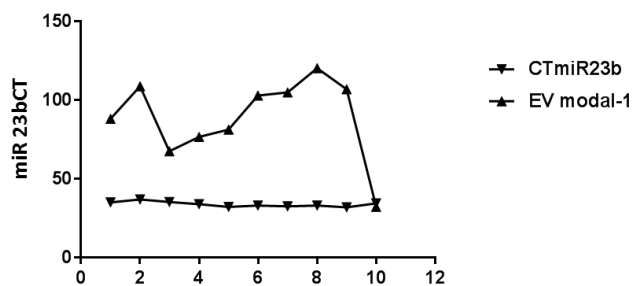
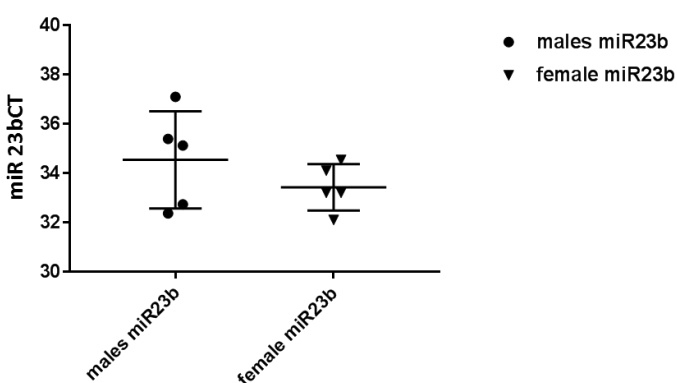


Figure 13: miR23b expression related to EV numbers and EV modal size.

miR23b expression between genders



miR23b expression according to age

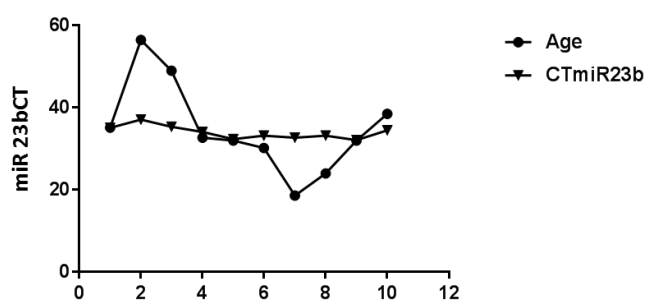
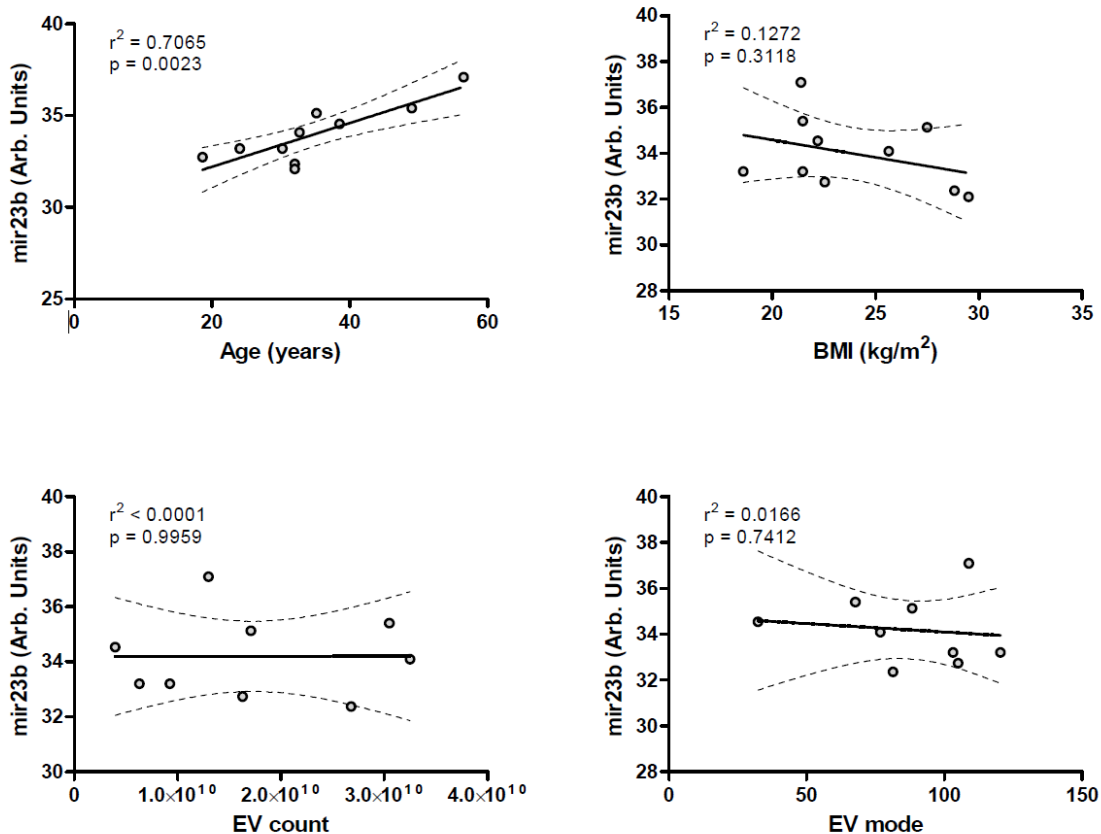
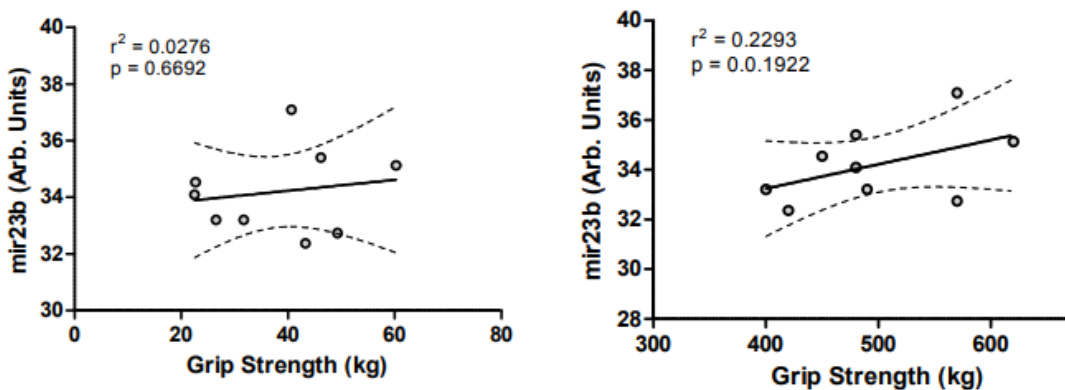


Figure 14: miR23b expression between genders and relating to age.



**Figure 15: miR23b expression correlated with age, BMI, EV count and EV mode**



**Figure 16: miR23b expression correlation with grip strength.**

*Tip: When writing this section, look at your results, graphs and tables and ask yourself – what does this data mean?*

### Section 5. Discussion (300-600 words)

Overall our findings show that EV profiles can be linked to some non-invasive health parameters and may therefore be developed as a novel tool to assess health. Furthermore, refinement of the test, including more key microRNAs for metabolic health and inflammation status can be incorporated into such a health index.

Our findings from this pilot study indeed do support the initial idea of development of a health index test based on EV profiling.

A clear limitation of this pilot study is the number of volunteers, with 23 overall, although this did indeed give some significant data relating to age. Resources also only allowed for 10 volunteers to be analysed for miR. For refinement of the test, this cohort of volunteers will need to be increased and the current index can serve as a basic model that data can be added to. With increased numbers of participants, the modelling of the health index test can be refined.

*Tip: When writing this section think about your aims. Did you achieve your aims? Were your results as expected? Are you findings consistent with other work in the literature?*

### **Section 6. Conclusions and Future Work (200-300 words)**

*This section should include: key messages/findings as well as suggestions for how your work could be continued in the future.*

- EV profiles vary between individuals
- EV numbers associate with age groups
- EVs are enriched in microRNAs and can be used to assess miR differences between individuals
- An EV-health index test can be developed based on EV profiling

### **Section 7. Lessons Learned (200-600 words)**

What did you learn from working in this partnership? Where there any novel techniques you employed?

Co-ordination of academics and 4 undergraduate students, with very diverse time tables, is a challenge but could be overcome with careful planning. Flexibility and good time management and careful planning is needed when working with volunteers.

Novel techniques developed in this project were the adjustment of previous EV isolation protocols to use for small scale samples in an abbreviated protocol that still yielded similar EV profiles to what is normally seen from human plasma. This is a great improvement to assess multiple samples in a shorter time frame.

*Tip: Think about how you are going to get your research across to your stakeholders. Be realistic about this and consider whose help you may need in the process.*

### **Section 8. Group Reflection (200-300 words)**

Combining the different expertise of the participating academics was a great advantage for bringing together skills and knowledge to create a model for a health index test based on EV analysis, microRNAs and physiological parameters.

Bringing together a range of skill sets from students and academics, as well as experimental and analytical knowledge, made this a successful research collaborations.

Recruitment of participants and co-ordination of the academic and student teams, all with busy time tables and multiple commitments was a challenge, but could be overcome.

From an academic's perspective, working within a group that every member has different expertise was the greatest strength. We learnt from each other and analysed our data in a wider context. Our student members were highly motivated and hard working.

### **Submission Instructions:**

Please attach the cover sheet to the front of your report and email the report to [studentpartnership@westminster.ac.uk](mailto:studentpartnership@westminster.ac.uk) by the 24<sup>th</sup> July 2019.



## **EV-HIT PARTICIPANT CONSENT FORM**

### ***Developing a new health blood test***

You are being invited to take part in a research study that aims to develop a health index from certain blood biomarkers associated with health. These biomarkers include extracellular vesicles, released by cells into blood plasma and key microRNA EV-markers. Extra cellular vesicles are membrane-bound structures, varying in size (30-1000nm) that are associated with several disease-processes as they carry RNAs, proteins, microRNAs and other signalling molecules which influence cellular communication and behaviour.

Currently, there is a lot of research interest in the role of these extracellular vesicles and microRNAs in diagnostics, as therapeutic targets for disease and for their role in cellular communication. The purpose of this study is to isolate chosen biomarkers, further analyse and create a panel of critical health biomarkers to be used as a health test: in order to do this, in a group of younger non-smoker people (18 – 35 years of age), body composition will be measured and a small sample of blood will be drawn. In order to find an optimal number of vesicles for a healthy individual, isolated extracellular vesicle will be assessed by the number and used as a starting point to compare and contrast changes in number and presence based on age and health within the participants.

This research is taking place at the University and financial support is provided by Student Co-Creators funding at the University.

The study will involve you:

- 1) Having a small sample of blood drawn (approx. 6mL)
  
- 2) Having body composition tests including blood pressure and strength measured
  
- 3) Attending the University of Westminster on specified times and dates for approximately 30 minutes

If desired, a copy of the experimental results can be provided for your information.

## Please Note:

- Your participation in this research is entirely voluntary
- You have the right to withdraw at any time without giving a reason.
- Wherever practicable, withdrawal from the research will not affect any treatment and/or services that you receive.
- You have the right to ask for your data to be withdrawn as long as this is practical, and for personal information to be destroyed.
- The possibility exists that abnormal results regarding your health or well-being will be uncovered by this research. Whilst not clinically qualified, we will inform you if we expect this to be the case, provide you with a copy of this information, and ask that you independently seek further medical guidance.
- You do not have to answer particular questions either on questionnaires or in interviews if you do not wish to do so.
- Your responses will normally be made anonymous, unless indicated above to the contrary, and will be kept confidential unless you provide explicit consent to do otherwise, for example, the use of your image from photographs and/or video recordings.  
*[NOTE: it may not be possible to maintain confidentiality in certain circumstances, e.g. where issues of child safety have been identified. You should seek clarification from the researcher and/or their supervisor if you are concerned about this].*
- No individuals should be identifiable from any collated data, written report of the research, or any publications arising from it.
- All computer data files will be encrypted and password protected. The researcher will keep files in a secure place and will comply with the requirements of the Data Protection Act.
- All hard copy documents, e.g. consent forms, completed questionnaires, etc. will be kept securely and in a locked cupboard, wherever possible on University premises. Documents may be scanned and stored electronically. This may be done to enable secure transmission of data to the university's secure computer systems.
- Please notify the researcher immediately if any adverse symptoms arise during or after the research.
- If you wish, it will be possible to receive information on the results of the research.  
*Please indicate on the consent form if you would like to receive this information.*
- The head academic researchers can be contacted during and after participation by email: Dr S. Lange ([S.Lange@my.westminster.ac.uk](mailto:S.Lange@my.westminster.ac.uk)), Dr P. U. Onganer ([P.onganer@my.westminster.ac.uk](mailto:P.onganer@my.westminster.ac.uk)), Dr B. Elliott ([B.Elliott@my.westminster.ac.uk](mailto:B.Elliott@my.westminster.ac.uk)) and Prof. J. Bell ([J.Bell@my.westminster.ac.uk](mailto:J.Bell@my.westminster.ac.uk))

**Appendix B: EV-HIT Consent Form**



**EV-HIT CONSENT FORM**

**Title Study: Developing a new health blood test**

I have been given the Participation Information Sheet and/or had its contents explained to me. Yes  No

I have had an opportunity to ask any questions and I am satisfied with the answers given. Yes  No

I understand I have a right to withdraw from the research at any time and I do not have to provide a reason. Yes  No

I understand that if I withdraw from the research any data included in the results will be removed if that is practicable (I understand that once anonymised data has been collated into other datasets it may not be possible to remove that data). Yes  No

I would like to receive information relating to the results of this study. Yes  No

I wish to receive a copy of this Consent form. Yes  No

I confirm I am willing to be a participant in the above research study. Yes  No

I note the data collected may be retained in an archive and I am happy for my data to be reused as part of future research activities. I note my data will be fully anonymised (if applicable). Yes  No

**Participant's Name:** \_\_\_\_\_

**Signature:** \_\_\_\_\_ **Date:** \_\_\_\_\_

This consent form will be stored separately from any data you provide so that your responses remain anonymous.

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I confirm I have provided a copy of the Participant Information Sheet approved by the Research Ethics Committee to the participant and fully explained its contents. I have given the participant an opportunity to ask questions, which have been answered.

**Researcher's Name:** \_\_\_\_\_

**Signature:** \_\_\_\_\_ **Date:** \_\_\_\_\_

## **Appendix C: EV-HIT Medical Questionnaire**



### **MEDICAL QUESTIONNAIRE** **STRICTLY CONFIDENTIAL**

**Participant code:** \_\_\_\_\_

**Title of study:** Developing a new health blood test

**Mark if you have suffered from any of the following:**

- |   |   |
|---|---|
| <input type="checkbox"/> Severe anxiety or depression | <input type="checkbox"/> Eating or Mental Disorder        |
| <input type="checkbox"/> Hay fever or any allergies   | <input type="checkbox"/> Epilepsy                         |
| <input type="checkbox"/> Liver disorder               | <input type="checkbox"/> Any blood disorder               |
| <input type="checkbox"/> Bladder disorder             | <input type="checkbox"/> Genito-Urinary Complaints        |
| <input type="checkbox"/> Any recurrent infections     | <input type="checkbox"/> Asthma                           |
| <input type="checkbox"/> Rheumatic Fever (Rheumatism) | <input type="checkbox"/> Alcohol or drug related problems |
| <input type="checkbox"/> Any mental impairment        | <input type="checkbox"/> Thyroid or gland problems        |
| <input type="checkbox"/> Stomach or Bowel Complaint   | <input type="checkbox"/> High or Low Blood pressure       |
| <input type="checkbox"/> Diabetes                     | <input type="checkbox"/> Heart condition/Angina           |
| <input type="checkbox"/> Infection of Kidneys         | <input type="checkbox"/> Fainting or Migraine             |
| <input type="checkbox"/> Tumours or other masses      | <input type="checkbox"/> Autoimmune disease               |

### **STATEMENT OF PERSONAL HEALTH**

**Your statement of personal health?**

- Excellent
- Good
- Fair/poor

**Have you had any specialist or hospital investigation, X-Ray or E.C.G.?** \_\_\_\_\_

**Is any investigation pending?**

If so please specify \_\_\_\_\_

**Have you suffered an injury?**



If so state when and how \_\_\_\_\_

**Are you at present on any form of treatment, medication or medical advice?**

If so please specify \_\_\_\_\_

**Are you, at present, taking any dietary supplements or over the counter pain killers?**

If so, please specify \_\_\_\_\_

**Do you feel in good health?** \_\_\_\_\_

**Do you drink alcohol? If so how many units per week?** \_\_\_\_\_

**Are you a smoker? If so, please give details** \_\_\_\_\_

**Approximately, how many hours of sleep do you get per night?** \_\_\_\_\_

**Do you have any condition not mentioned in here?** \_\_\_\_\_

**How often do you eat healthy food?** \_\_\_\_\_

**How often do you indulge in junk food?** \_\_\_\_\_

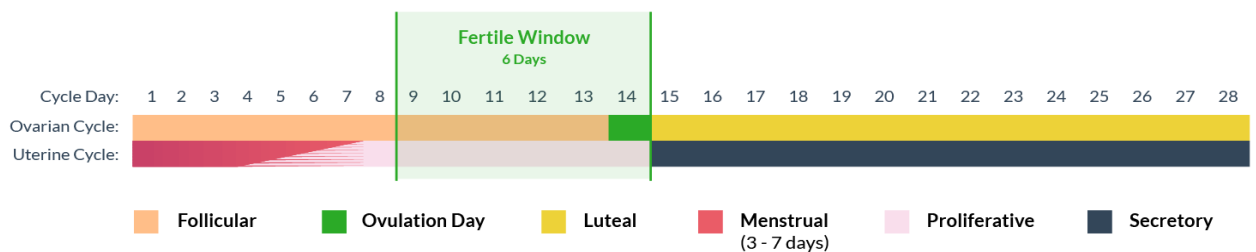
## FEMALES ONLY

Do you consider yourself to be pre-, mid-, or post-menopausal? Yes / No

Do you use a prescription oral or implantable contraception method?  
[e.g. the oral contraceptive pill] Yes / No

Do you consider your menstrual cycle to be regular?  
[e.g. occurs in a predictable 28-day cycle] Yes / No

In the following diagram where [approximately] do you think you are right now?



---

## FAMILY HISTORY

Check if there is any history in your family of

- |  |  |  |   |
|--|--|--|---|
| <input type="checkbox"/> Diabetes                | <input type="checkbox"/> Blood disorders | <input type="checkbox"/> Obesity             | <input type="checkbox"/> Allergy            |
| <input type="checkbox"/> Stroke                  | <input type="checkbox"/> Jaundice        | <input type="checkbox"/> Gout                | <input type="checkbox"/> Autoimmune disease |
| <input type="checkbox"/> Heart Disease           | <input type="checkbox"/> Alcoholism      | <input type="checkbox"/> Asthma              | <input type="checkbox"/> Cancer             |
| <input type="checkbox"/> High/Low Blood Pressure | <input type="checkbox"/> Tuberculosis    | <input type="checkbox"/> Psychiatric Illness | <input type="checkbox"/> High Blood Fats    |

Other (please specify) \_\_\_\_\_

Researcher: \_\_\_\_\_ Date \_\_\_\_\_

## **Appendix D: International Physical Activity Questionnaire**



### **INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE**

The main aim of the project is to put the health-associated biomarkers found in humans on centre stage and in the future to show how specific health-associated biomarker disturbances are linked to host processes.

In order to do so, it will be essential to assess your health status through a very simple physical activity questionnaire based on what kinds of physical activities people do as part of their everyday lives. Your answers and your data will be of great value for developing an optimum health index.

The below questions are about the time you spent being physically active in the last 7 days. They include questions about activities you do at work, as part of your house and yard work, to get from place to place, and in your spare time for recreation, exercise or sport.

**Please answer each question even if you do not consider yourself to be an active person.**

**THANK YOU FOR PARTICIPATING.**

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**In answering the following questions,**

◆ **vigorous** physical activities refer to activities that take hard physical effort and make you breathe much harder than normal.

◆ **moderate** activities refer to activities that take moderate physical effort and make you breathe somewhat harder than normal.

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**1a.** During the last 7 days, on how many days did you do physical activities like heavy lifting, digging, bicycling?

vigorous  
aerobics, or fast

Think about only those physical activities that you did for at  
minutes at a time.

least 10

\_\_\_\_\_ **days per week**                      or                      **None**

**1b.** If so, how much time in total did you usually spend on one of those days doing vigorous physical activities?

\_\_\_\_\_ **hours** \_\_\_\_\_ **minutes**

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**2a.** Again, think only about those physical activities that you did for at least 10 minutes at a time. During the last 7 days, on how many days did you do moderate physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis? Do not include walking.

\_\_\_\_\_ **days per week**                      or                      **None**

**2b.** If so, how much time in total did you usually spend on one of those days doing moderate physical activities?

\_\_\_\_\_ **hours** \_\_\_\_\_ **minutes**

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**3a.** During the last 7 days, on how many days did you walk for at least 10 minutes at a time? This includes walking at work and at home, walking to travel from place to place, and any other walking that you did solely for recreation, sport, exercise or leisure.

\_\_\_\_\_ **days per week**                      or                      **None**

**3b.** How much time in total did you usually spend walking on one of those days?

\_\_\_\_\_ **hours** \_\_\_\_\_ **minutes**

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The last question is about the time you spent sitting on weekdays while at work, at home, while doing course work and during leisure time. This includes time spent sitting at a desk, visiting friends, reading traveling on a bus or sitting or lying down to watch television.

**4.** During the last 7 days, how much time in total did you usually spend sitting on a week day? \_\_\_\_\_ **hours** \_\_\_\_\_ **minutes**

## Appendix E: EV-HIT Data Collection Sheet



### Data Collection Sheet

Participant Code: \_\_\_\_\_ Researcher: \_\_\_\_\_

Age: \_\_\_\_\_ (decimal) Year of birth: \_\_\_\_\_ Sex: \_\_\_\_\_

Date: \_\_\_\_\_

**1. Consent form**

**2. Medical History**

**3. Blood taken**

WBC: \_\_\_\_\_

MCV: \_\_\_\_\_

RBC: \_\_\_\_\_

MCH: \_\_\_\_\_

HGB: \_\_\_\_\_

MCHC: \_\_\_\_\_

HCT: \_\_\_\_\_

PLT: \_\_\_\_\_

**4. Blood pressure**

Systolic: \_\_\_\_\_ mm Hg

Diastolic \_\_\_\_\_ mm Hg

**5. Body Composition**

Weight: \_\_\_\_\_ (kg)

Height: \_\_\_\_\_ (m)

BMI: \_\_\_\_\_

Waist: \_\_\_\_\_ (m)

Fat Mass: \_\_\_\_\_ (kg)

FF Mass: \_\_\_\_\_

SkM  $M$  \_\_\_\_\_ (kg)

Visceral fat: \_\_\_\_\_ (L)

**6. Physical activity status**

Grip Strength (kg): \_\_\_\_\_ Hand grip #: \_\_\_\_\_

Leg Strength (kg): \_\_\_\_\_ Chain link #: \_\_\_\_\_

Pre		

Pre		

Astrand predicted  $VO_2$  max: \_\_\_\_\_ ( $mL.kg.min^{-1}$ )

Watts =  $6 \times RPM \times (KG + 1) = ? / 6.12$

Max HR ( $208 - 0.7 \times age$ ): \_\_\_\_\_ 85% MHR: \_\_\_\_\_ Wt used: \_\_\_\_\_

Min	Heart Rate
5	
6	
Average	

Notes:

**A STUDENTS AS CO-CREATORS PROJECT**

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