KPI	Activity Type	Target Value
n. of new longitudinal data collections designed and launched	Project realization	3 at M12

The key performance indicator (KPI) expected at month 12, for Spoke 3, concerns the design and launch of at least 3 new longitudinal data collections. Here are listed the data collections launched within Spoke 3 during the first year of the Age-it project.

1. **IDENTIFICATION AND QUANTIFICATION OF MULTIDIMENSIONAL FRAILTY IN ELDERLY PATIENTS IN DIFFERENT CARE SETTINGS (fra-SET)**

   A state of increased "vulnerability", resulting from aging-associated decline in reserve and function across multiple physiologic systems, may lead to "multidimensional frailty", considered a dynamic state with a lost in one or more domains (physical, mental, nutritional, and social) which increases the risk of adverse outcomes.
   Moreover, the main problem is the identification and quantization of this condition. According to this definition, the most used tool for multidimensional frailty recognition is the "Rockwood deficit–accumulation frailty index" (FI), expressed as the ratio between the number of deficits found in a patient and the total number of deficits investigated. Several studies have shown that FI is more predictive of adverse clinical outcomes than other tools in both hospital and community settings. Recently, we have developed and validated an Italian modified version of FI, the Italian Frailty index (IFi) but, despite the numerous advantages and reliability, IFi shows a prolonged administration time (≈30 min.) (Abete P et al., 2017). To overcome this limitation, we have recently created and validated a rapid tool for the evaluation of multidimensional frailty called "Fr-AGILE" that has been proved to be reliable with a diagnostic power comparable to that of the IFi with a high predictive value on adverse clinical outcomes, and more importantly, with an administration time of less than 5 min. Fr-AGILE was built by selecting among the 40 items of Italian version of Frailty Index the 10 ones most predictive of mortality. The total score ranged from 0 to 10, with higher score indicate more severe frailty. The Fr-AGILE scores were divided into degrees: absent (0), light (1–3), moderate (4–7) and severe frailty (8–10). However, Fr-AGILE has been validated in non-institutionalized out-patients (Liguori I et al., 2020).

Fr-AGILE items are the following ones:

n.	AGILE's Items	Score	FRAILTY domains
1	Feel everything is an effort	yes=1; no=0	**PHYSICAL**
2	Help up/down stairs	yes=1; no=0	
3	Muscular grip strength (1)	yes=1; no=0	
4	Temporal orientation at MMSE (2)	yes=1; no=0	**MENTAL**
5	Re-enactment deficit at MMSE (3)	yes=1; no=0	
6	Feel depressed	yes=1; no=0	
7	Weight loss over 4.5 kg in the last year	yes=1; no=0	**NUTRITIONAL**
8	Help in eating	yes=1; no=0	
9	Concrete help from family members	yes=1; no=0	**SOCIO-ECONOMIC**
10	Financial help from family members	yes=1; no=0	

1) less than 27 kg in men, less than 17 kg in women at dynamometers
2) referred exact date (day / month / year)
3) the words are referred to the beginning of the questionnaire and made to repeat at the end of the test

Thus, we aimed to validate Fr-AGILE as a new tool for the assessment of multidimensional frailty in various hospital care settings. In-hospital patients aged ≥ 65 years consecutively admitted from January 2024 to December 2024, in relation to the different levels of intensity of care, will be enrolled. The patients will be consecutively enrolled in the more important hospitals of Campania region, Italy (n.5 low-medium and n. 4 high intensity of care units). The study protocol will evaluate the degree of frailty of each patient through the Edmonton Frail Scale (a validated method) (Rolfson DB et al., 2006) and Fr-AGILE (a method to be validated). The tools will be administered in the clinical stability phase (pre-discharge) and a series of intra-hospital events will be evaluated. Mortality, disability (a reduction in the ADL score ≥1) and new hospitalizations at 6, 12, and 24 months of follow-up, via a telephone interview. To evaluate the sample size, Lin's Coefficient of Correlation of Concordance (CCC), a method used to compare a new measurement method (Fr-AGILE) with the standard method (Edmonton Frail Scale) will be used. (Lin HM et al. , 2015) The sample size will be ≈800 with a study power of 0.90 (one-sided z test with alpha=0.05) with a CCC value between 0.65-0.75.

The study complied with the Declaration of Helsinki principles and the study protocol was approved by the Ethics Committee Campania 3 of Campania Region, Italy (reg. n.100/2023 of October 4, 2023).

The implementation of this project aims to provide clinicians with an effective and easily applicable tool for the diagnosis and estimation of the degree of frailty of in-hospital elderly patient.

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2. **FRASNET study** of the “Vita Salute San Raffele University”. The protocol is currently being evaluated at the Clinical Trial Center of our hospital and we are awaiting approval from the Ethics Committee.
   The study will recruit healthy volunteers from a previous cohort of 2016-2017 (1250 patients) and neurological patients suffering from mild cognitive impairment and dementia followed at the Neurological Clinic of the San Raffaele Hospital (approximately 100 patients).
   The new study involves multidimensional geriatric assessments that will include anthropometric and body composition assessments using bioimpedance measurement. The frailty of the patients will then be assessed by calculating the Frailty Index and Fried's phenotype. Moreover, the degree of sarcopenia will be evaluated. Participants will also be subjected to a venous sampling which will be biobanked. Thanks to previous biobanked samples in 2016 and 2017, we could establish any predictive biomarkers of frailty and sarcopenia. In the task 5.3 we are going to explore the role of damage-associated molecular patterns (DAMPs) to stratify aging trajectories. DAMPs are released by damaged/dying cells, and are key mediators of the sterile inflammation typical of inflammaging and hold potential as novel biomarkers of frailty and sarcopenia. In particular, we are going to study HMGB1 as a novel biomarker of age-related sarcopenia and its association with the development of disability and other negative health outcomes in a large cohort of geriatric patients.
   HMGB1 is implicated in muscle waste in cancer patients, but it is still unexplored in age-related sarcopenia.
   We plan to study DAMPs as serum biomarkers, in association with functional and imaging-related correlates to stratify clinical phenotypes and predict

sarcopenia, disability and frailty in a large cohort of geriatric patients. HMGB1 will be dosed in a sample of 100 patients (including both neurologic patients and FRASNET participants).

3. **Technology-assisted Cognitive-Motor Dual Task Rehabilitation in a Population of Older Adults with Multimorbidity: A Prospective Cohort Study**
   Version 1.0, 4th/Dec/2023
   Acronim: AROMA -Assistenza Robotica per la Riabilitazione degli Anziani con Multimorbidità.

   Rationale and objectives of the study

   Cognitive-motor dual-tasking (CMDT) is the simultaneous processing and execution of a motor (e.g., walking, maintaining balance, or exercising) and cognitive (e.g., focusing attention, making decisions, or memorizing) activity.
   Older adults, especially those who suffer from multiple chronic, with systemic inflammation or a high burden of vascular damage, have particular difficulty in carrying out dual-tasks. Initial evidence suggests that dual-task training can exert a beneficial effect in old age and improve motor and functional performance both in the context of normal physiological aging and in the case of chronic and neurodegenerative diseases of the older adults.
   The objective of this protocol is to propose a prospective observational cohort study with the aim of comparing older patients aged ≥ 70 years with multimorbidity who have experienced an acute motor disability (e.g. femur fracture, stroke) and who have been treated with dual-task rehabilitation assisted by robotic technology versus conventional rehabilitation training not assisted by robotic technology with respect to the overall level of functional recovery achieved and the level of recovery of motor and cognitive abilities, in an intensive rehabilitation hospital setting.

   Methods

   This is a prospective cohort study. The study setting is the Geriatric Rehabilitation Unit of the A. Gemelli University Hospital, Catholic University (UCSC) in Rome. Participants will be enrolled in sequential order among the inpatients of the Geriatric Rehabilitation Unit. To date, rehabilitation with robotic technology is offered to a limited number of patients due to the limited availability of staff trained and experienced in providing these treatments. An enrolment period of 24 months is expected. From the date of enrollment of each patient, the follow-up period will coincide with the duration of hospitalization. Participants will be selected based on the type of exposure (dual task robotic rehabilitation versus conventional rehabilitation). Participants will be assessed at the time of enrollment (T0 - baseline assessment) and at the time of discharge from the Unit (T1 - follow-up

assessment). The hospitalization time will coincide with the duration of follow-up for each participant.
Inclusion criteria: age ≥ 70 years; recent motor disability; multimorbidity, defined as the presence of two or more coexisting chronic diseases in the same individual; signature of informed consent.
Exclusion criteria: severe neurosensorial deficits (severe vision or hearing loss); diagnosis of dementia.
The main outcome of the study is the level of functional recovery measured by the Barthel index (BI) Italian version. The BI consists of the analysis of 10 activities of daily living (ADL): nutrition, ability to bathe or shower, grooming or ability to take care of external appearance, ability to dress, control of intestinal transit, bladder and urination control, toilet use (sitting down and getting up, cleaning and dressing), transfers (from sitting on the bed to the chair and vice versa), mobility, stairs. The total score ranges from 0 (complete functional dependence) to 100 (complete functional autonomy).
Secondary outcomes include assessment of balance, gait, walking, manual dexterity, global cognitive function, executive functions, attention, praxis. These measures will be conducted using tools and scales routinely adopted in the clinical practice of the Geriatric Rehabilitation Unit.
We plan to recruit at least 180 participants (90 per type of exposure) over a two-year period. The expected dropout rate for various reasons during the follow-up is approximately 10%.
Differences in the changes in mean score on the BI (T1-T0, gain score) between the two groups will be tested using the analysis of covariance (ANCOVA). Additional ANCOVA models will be produced to measure secondary outcomes. Stratified analyzes by etiological type of motor damage (orthopedic or neurological) will be conducted. The statistical significance of the estimated associations and effect sizes will be expressed by a p-value equal to or less than 0.05. All analyses will be conducted using R - 4.3.2 software.

Current stage

The present protocol has been approved by the UCSC Ethical Committee on December 21st, 2023. The kick-off meeting is scheduled on January 30th,2024. Participants' screening and enrolment is expected to start February 1st, 2024.

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4. *OPA Study: Relationship between neurological, metabolic, cognitive and functional indicators in aging*

P.I. Prof Giuseppe Sergi

STUDY PROTOCOL

Background: Inflammaging, a chronic low-grade inflammation associated with the aging process, represents a significant risk factor for morbidity and mortality in older adults. Elevated levels of inflammatory biomarkers, such as C-reactive protein and interleukin-6, are associated with several aspects of aging, including changes in body composition, immune senescence, and neuronal health. Inflammaging, involving microglia, astrocytes and neurons, could play a key role in neurodegenerative diseases such as Parkinson's and Alzheimer's diseases. Additionally, inflammatory cytokines released by immune cells in skeletal muscle may contribute to sarcopenia, the loss of muscle mass and strength, which often coexists with obesity in older adults (sarcopenic obesity). Studies in human and animal models suggest that sex hormones influence the innate immune system, with gender differences in the phenomena of immunosenescence and cellular senescence. However, the exact cause of inflammaging and its potential contribution to adverse health outcomes in older adults are largely unknown.

Objectives of the study and significance: This project will allow us to understand any common pathophysiological pathways in the field of inflammaging between (1) neurodegeneration, (2) obesity and malnutrition, (3) sarcopenia and osteosarcopenia to identify profiles at high risk of disability in

short and long term, which could benefit from targeted and personalized multidimensional interventions. This aim will be pursued through the following main actions: (1) use digital tools and physical performance tests to investigate the prevalence of sarcopenia in patients suffering from obesity, Parkinson's disease and mild cognitive impairment (MCI) and collect non-invasive physiological and functional variables of the themselves; (2) use biohumoral data to investigate the presence of inflammaging in the enrolled patients; (3) identify associations between these measures and markers derived from genetic, clinical, neuropsychological, neurophysiological, neuropathological, and neuroimaging data; and (4) use these associations to define possible aging promoting strategies also in the field of "gender medicine". The innovative potential of the project is to bring added value in the field of aging, delving into the pathophysiological mechanisms underlying the multimorbid older patient, with particular attention to the prevention and treatment of geriatric pathologies that could lead to frailty and disability.

Materials and methods: prospective cross-sectional study. Approximately 200 subjects will be enrolled (50 patients suffering from obesity, 50 from Parkinson's disease, 50 from Alzheimer's disease and 50 healthy subjects), aged between 65 and 90 years, who will be subjected to the following assessments:

a) Collection of immediate and remote pathological, physiological and pharmacological anamnesis, as well as comorbidities and indicators of severity of the disease. The degree of comorbidity will be assessed using the Cumulative Illness Rating Scale (CIRS).

b) Anthropometric measurements. Weight and height with body mass index (BMI) calculation; waist circumference (cm); calf circumference (cm).

c) Functional evaluation. Based on the assessment of autonomy in activities of daily living (Activities of Daily Living - ADL) and in instrumental activities of daily living (Instrumental Activities Daily Living - IADL). In addition, for the assessment of functionality in Parkinson's disease, the Parkinson's disease - Cognitive Functional Rating Scale (PD-CFRS) will be administered, which allows the contribution of cognitive deficits to interfering with functional autonomy, rather than motor problems, to be assessed.

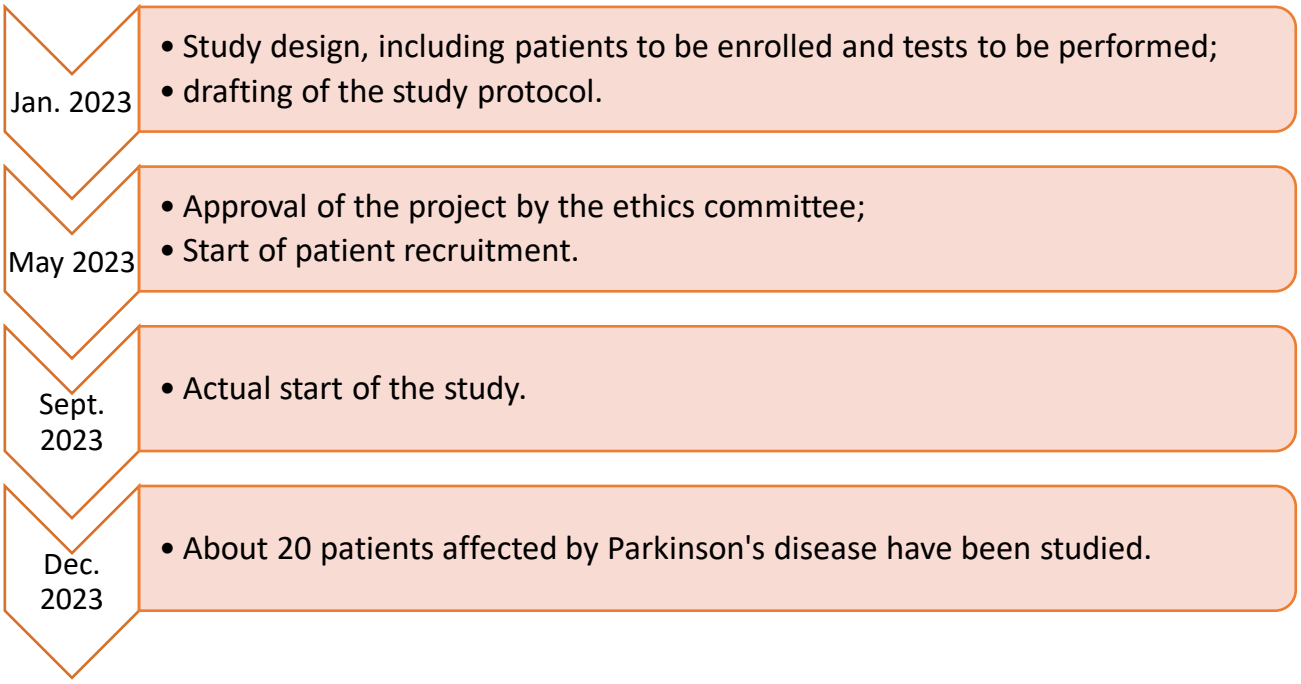
d) Nutritional assessment using Mini-Nutritional Assessment (MNA) tests.

e) Evaluation of physical performance: Short Physical Performance Battery (SPPB), 6-minute walking test (6-MWT), and measurement of muscle strength using a dynamometer. In the sample of Parkinson's patients, motor severity will be assessed with part 3 of the Movement Disorder Society Unified Parkinson's Disease Rating Scale (MDS-UPDRS).

f) Cognitive assessment. Based on the use of the Mini Mental State Examination (MMSE), the Montreal Cognitive Assessment (MoCA), the Cognitive Reserve Index (CRI-q) and the Geriatric Depression Scale (GDS). Finally, the Beck Depression Inventory (BDI-II, the STAI Y1 and Y2 for the assessment of anxiety and the Apathy scale for the evaluation of apathy will be performed.
g) Instrumental investigations: Lumbar and femoral bone densitometry; Brain MRI to investigate the diagnosis of cognitive impairment/dementia.
h) Genetic and biohumoral investigations. All patients will be characterized on a clinical metabolic level including superoxide dismutase, uric acid, cholesterol, blood glucose, cystatin C, HDL-C, LDL-C, Hs-CRP, erythrocyte sedimentation rate. The main inflammatory cytokines involved in the process of chronic inflammation (leptin, adiponectin, IL-6, IL-17, INF-gamma, TNFα, sestrins) will be measured by blood sampling. EVPs from adipose tissue will also be isolated from the blood sample and an analysis of microRNA expression will be carried out.

6 months after the first evaluation, patients will be contacted by telephone to record information relating to new hospitalizations, new falls and to verify whether or not there have been significant functional and neurobehavioral changes.

FLOW-CHART PROGRESS STATUS OF THE PROTOCOL



APPROVAL OF THE ETHICS COMMITTEE (PROVINCE OF PADUA): Approved on 18 May 2023 (Code 5757/AO/23). The amendment for telephone follow-ups of patients 6 months after the first evaluation is being submitted.