**Project protocol**

**1. Title**

Systemic Sclerosis and Cancer: A Multicenter Study

**2. Background**

Systemic sclerosis (SSc) is a rare and complex disease characterized by fibrosis of the skin and internal organs, vascular abnormalities, and dysregulation of the immune system.1 The clinical manifestations of systemic sclerosis vary widely, ranging from mild forms with skin involvement to severe cases with multi-organ complications, including pulmonary, cardiac, and renal involvement, accounting for chronic morbidity and high mortality.1

The most common causes of death in SSc are related to pulmonary and cardiovascular complications, though malignancy also plays a significant role in the increased mortality observed in this patient population, accounting for 11% of all-cause deaths and 31% of non-SSc related causes.2

In recent years, growing evidence suggests that SSc may be associated with an increased risk of cancer, particularly certain types such as breast, lung and hematologic malignancies,3-5 The exact mechanisms underlying this association are still under investigation, but it is hypothesized that chronic inflammation, immune dysregulation, and the presence of autoantibodies may play a role in tumorigenesis, and the concept of systemic sclerosis as a paraneoplastic syndrome is now arising.6,7 Emerging data suggest that distinct subsets of SSc patients, identified by the presence of specific autoantibodies and clinical features, may have an increased or decreased risk of cancer around SSc onset or during the disease course.3,7,8

Enhancing knowledge and raising awareness about cancer in SSc is important for early diagnosis and for improving the prognosis of these patients. Nonetheless, this is an evolving field with much yet to be explored and validated. Indeed, some studies reveal novel associations, such as an increased cancer risk in SSA/Ro-positive patients,9-11 while others do not support the previously observed link with anti-RNA polymerase III antibodies,12 and some present contradictory findings regarding anti-RNP antibodies.11,12 However, most studies are highly heterogeneous regarding study design, inclusion criteria and “cancer-related SSc” and “SSc disease onset” definitions.

Further research is needed to clarify these associations and uncover pathogenic links. Identifying high-risk subgroups and validating biomarkers predictive of cancer risk in SSc may allow for the development of targeted surveillance strategies and more personalized approaches to patient management.

In this context, our study aims to contribute to the growing body of evidence by characterizing the most frequent malignancies in a large nationwide SSc cohort and exploring associations between cancer and clinical or immunological features. We hope this work may support the identification of SSc subgroups with increased oncologic risk and provide a foundation for developing national recommendations for cancer screening in SSc. In the absence of standardized guidelines or uniform practices in Portugal, these findings may inform future strategies for early cancer detection and individualized follow-up, ultimately improving patient outcomes in clinical practice.

**3. Objectives**

Primary objectives:

* To assess the proportion of patients who develop incident cancer following systemic sclerosis diagnosis;
* To identify clinical features and immunological markers associated with cancer in SSc patients.

Secondary objectives:

* To identify most frequent cancer types in SSc patients;
* To determine the association of specific cancers with SSc subtypes, if sample size allows;
* To estimate the incidence rate of cancer in SSc, compared to general Portuguese population.

Exploratory objective:

* To determine whether clinical features and immunological markers are associated with cancer diagnosis before SSc disease onset.

**4. Methods**

**4.1. Study design**

This is a retrospective study based on nationwide SSc patients registered in the Rheumatic Diseases Portuguese Registry (Reuma.pt), through which data are collected prospectively, with more than 1700 patients currently registered. Missing data will be retrospectively collected through the review of electronic clinical reports.

**4.2. Study population/inclusion criteria**

1. Age ≥18 years at diagnosis.
2. Registered diagnosis of SSc defined by 2013 ACR/EULAR classification criteria, or having definite Raynaud’s, abnormal nailfold capillaries and a scleroderma-specific autoantibody.

**4.3. Operational Definitions and Methodological Notes**

SSc onset is defined as the development of the first Raynaud or non-Raynaud’s symptom, whichever occurred first.

Cancer diagnoses will be identified primarily through the Reuma.pt registry. For each case, validation will be performed through the review of electronic medical records from the participating centers. The minimum evidence required to confirm a cancer diagnosis will include one of the following: (1) a pathology report confirming malignancy; (2) a radiology report highly suggestive of cancer in combination with oncology referral and treatment initiation; or (3) a clear diagnosis documented by the attending physician in the clinical notes, accompanied by treatment records or follow-up in an oncology setting. Patient self-reported diagnoses will not be considered valid unless corroborated by medical documentation.

For the exploratory analysis, we will identify patients with a confirmed cancer diagnosis dated prior to the defined onset of SSc. Among these, clinical and immunological data collected at SSc diagnosis will be compared to those of patients with no cancer or with cancer diagnosed after SSc onset. Although all biomarker data will have been collected after SSc diagnosis, we aim to assess whether particular immunological profiles are more frequently observed in patients whose cancer preceded SSc onset, suggesting a possible paraneoplastic context. We acknowledge that autoantibody and biomarker data will have been collected post-SSc diagnosis; therefore, any associations identified cannot establish temporality or causality, but may suggest relevant clinical patterns worth further prospective investigation.

Although this is a retrospective study based on pre-existing data, we conducted an approximate estimation of the number of events (cancer cases) to ensure the feasibility of the planned analyses. Reuma.pt currently includes over 1,700 patients with systemic sclerosis. Based on previous literature, the reported prevalence of cancer in SSc cohorts ranges from 7% to 15%.9-12 Assuming a conservative estimate of 10% cancer prevalence, we expect approximately 170 cancer cases in our cohort. This number of events is expected to provide adequate power for multivariable logistic regression analyses, particularly when assessing associations with major clinical and immunological features.

**4.4. Covariates to be extracted from Reuma.pt**

* Classification criteria (ACR/EULAR 2013)
* Demographic, clinical and immunological features:
  + Age at symptom onset (years)\*
  + Sex (male; female)\*
  + Ethnicity (caucasian; black; asian; other)
  + Date of SSc symptom onset (month/year)
  + Date of SSc diagnosis (month/year)
  + SSc subtype (diffuse cutaneous; limited cutaneous; sine scleroderma; overlap; preclinical)\*
  + Baseline (diagnosis) Rodnan score (0-51)
  + Cumulative clinical features:
    - Raynaud phenomenon (yes; no)
    - Telangiectasis (yes; no)
    - Digital ulcers (yes; no)
    - Calcinosis (yes; no)
    - Tendon friction rubs (yes; no)
    - Arthralgias / arthritis (yes; no)
    - Myositis (yes; no)
    - Gastrointestinal involvement (and subtype – esophageal, gastric or intestinal) (yes; no)
    - Pulmonary arterial hypertension (yes; no)
    - Interstitial lung disease (yes; no)
    - Renal involvement (yes; no)
  + Autoantibodies (ANA, anti-SSA, anti-SSB and SSc-specific autoantibodies)\*
* Capillaroscopy pattern at SSc diagnosis (early; active; late)
* Cancer diagnosis (yes; no)
* Cancer primary site (organ)\*
* Date of cancer diagnosis (month/year)
* SSc-related treatments before cancer diagnosis [calcium channel blockers, phosphodiesterase inhibitors, prostacyclin analogues, endothelin receptor antagonists, corticosteroids, immunosuppressives, conventional and biologic immunomodulators, antifibrotics] (yes; no [for each drug])\*
* Cancer-related modifiable risk factors
  + Obesity (yes; no)\*
  + Tobacco exposure (current smoker; ex-smoker; non-smoker)\*
  + Alcohol consumption (current consumption; former consumption; no consumption)\*
* Date of first registered visit (month/year)
* Date of last registered visit (month/year)

Variables marked with an asterisk (\*) are considered core variables for statistical analysis purposes.

**4.5. Statistical analysis**

Data will be analyzed using IBM-SPSS Statistics 28.

To describe the demographic, clinical and immunological features, statistics will be presented as absolute and relative frequencies for categorical variables, as mean ± standard deviation for continuous variables with normal distribution, and as median (interquartile range) for continuous variables non-normally distributed.

A comparative analysis of clinical, immunological and demographic variables will be performed between SSc patients with and without cancer diagnosis and, if sample size allows, between SSc subtypes. For dichotomic variables chi-squared test or Fisher’s exact test will be used, and for continuous variables with normal distribution we will use student’s T-test, Mann-Whitney Test or Anova, whereas Kruskal-Wallis test will be used for non-normally distributed continuous variables.

Logistic regression will be used to determine the associations of different variables with cancer. Variables with *p* values <0.05 in the univariable analysis and those deemed to be of clinical significance to the outcome will be included in the multivariable logistic regression analysis.

Standardized incidence ratios (SIR) with exact Poisson 95% confidence intervals (CIs) will be calculated as ratios of cancer incidence in patients with SSc (observed cases) to that in the general Portuguese population (expected cases), weighted according to age and sex. Cancer incidence data for Portugal will be obtained from the National Oncologic Registry (2020 version).13

Statistical significance will be considered with a *p* value < 0.05.

**5. Timeline**

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| Study design and elaboration of project protocol | December 2024 – January 2025 |
| Submission of research protocol to Reuma.pt and to local Hospital Health Committee | February 2025 |
| Invitation of all national centres to participate in the project | July 2025 |
| Extraction of data from Reuma.pt | September 2025 |
| Filling of missing data by participating centres collaborators | September - October 2025 |
| Extraction of updated data from Reuma.pt and compilation | November 2025 |
| Data analysis | November - December 2025 |
| Final report, abstract submissions and publication | January-June 2026 |

**6. Expected results, strengths and limitations**

We aim to identify associations between cancer-related SSc and its clinical and immunological features, not only to validate previously described links but also to uncover novel ones. Although recent studies have provided valuable insights into the association between cancer and SSc,9-12 our study offers several distinct contributions. Firstly, it is based on a large, nationwide, multicenter cohort from Reuma.pt, ensuring comprehensive and representative data from routine clinical practice in Portugal. Secondly, by calculating standardized incidence ratios (SIRs) using data from the National Oncologic Registry, our study provides a unique comparison with the general Portuguese population, which has not been previously reported. Additionally, we will explore a wide range of clinical, serological, and therapeutic factors, allowing for a multifaceted analysis of cancer risk in SSc. Importantly, the study aims to generate clinically applicable knowledge to inform future screening strategies and optimize personalized care pathways for SSc patients in the Portuguese context.

Study limitations are the ones expected in a retrospective study, namely missing data.

**7. Ethical considerations**

The study will be conducted according to the principles of the Declaration of Helsinki. It will be submitted for evaluation and approval to the Ethics Committee of Hospital Garcia de Orta. All patients have signed the Reuma.pt informed consent.

**8. Research team**

Rodrigo Rei, Tomás Stein Novais, Ana Catarina Duarte, Ana Cordeiro, Maria José Santos

**9. Co-authoring**

Clinicians who actively collaborate in the project will be co-authors according to the principles of the International Committee of Medical Journal Editors (ICMJE). The number of authors per participating centre will be proportional to the number of valid cases included in the analysis. Centers contributing 1–50 patients may designate one co-author; those contributing 51–150 patients may designate two co-authors; and those contributing more than 150 patients may designate up to three co-authors.

**10. Funding and conflicts of interest**

There are no conflicts of interest. This project received no funding.

**11. References**

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