European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

Received 23 March 2024
Accepted 5 June 2024
Available online 5 June 2024
Version of Record published 24 June 2024

RESEARCH

Subacute thyroiditis in the SARS-CoV-2 era: a multicentre prospective study

Sara De Vincentis[1,2], Simona Loiacono[1], Eleonora Zanni[1], Roberta Sueri[1], Maria Laura Monzani[1,2], Daniele Santi[1,2], Ilaria Muller[3,4], Francesco Di Marco[5], Erica Crivicich[5], Mirco Armenti[5], Uberto Pagotto[6,7], Lorenzo Tucci[6,7], Carolina Cecchetti[6,7], Tommaso Trenti[8], Valentina Pecoraro[8], Giulia Canu[8], Manuela Simoni[1,2] and Giulia Brigante[1,2]

[1]Unit of Endocrinology, Department of Biomedical, Metabolic and Neural Sciences, University of Modena and Reggio Emilia, Modena, Italy
[2]Unit of Endocrinology, Department of Medical Specialties, Azienda Ospedaliero-Universitaria of Modena, Modena, Italy
[3]Department of Clinical Sciences and Community Health, University of Milan, Milan, Italy
[4]Department of Endocrinology, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[5]School of Specialisation in Endocrinology, University of Milan, Milan, Italy
[6]Department of Medical and Surgical Sciences, Alma Mater Studiorum - Bologna University, Bologna, Italy
[7]Department of Endocrinology and Diabetes Care and Prevention Unit, IRCCS Sant'Orsola-Malpighi Polyclinic, Bologna, Italy
[8]Department of Laboratory Medicine and Anatomy Pathology, Azienda USL Modena, Modena, Italy

Correspondence should be addressed to G Brigante: giulia.brigante@unimore.it

Abstract

Objective: Many cases of subacute thyroiditis (SAT) have been described related to SARS-CoV-2 infection, but no prospective data about follow-up are known. This prospective, longitudinal, 3-year, multicentre study aims to explore the clinical peculiarities and outcome of SAT in relation to SARS-CoV-2 infection, ascertained with antibody dosage.

Methods: All patients receiving SAT diagnosis from November 2020 to May 2022 were enrolled. Data on anamnesis, physical examination, blood tests (TSH, freeT4, freeT3, thyroglobulin, anti-thyroid antibodies, C-reactive protein, erythrocyte sedimentation rate, complete blood count), and thyroid ultrasound were collected. At baseline, the presence of IgG against the SARS-CoV-2 spike protein or nucleocapsid was investigated. Patients were evaluated after 1, 3, 6, and 12 months.

Results: Sixty-six subjects were enrolled. At baseline, 54 presented with pain, 36 (67%) for at least 15 days. Serum SARS-CoV-2 IgG measurements documented that 7 out of 52 subjects (13.5%) had infection before SAT diagnosis (COVID+). No significant differences between the COVID+ and COVID− groups were found at baseline, except for respiratory symptoms and fever, which were more common in COVID+ ($P = 0.039$ and $P = 0.021$, respectively). Among the 41 subjects who completed follow-up, COVID+ and COVID− did not differ for therapeutic approach to SAT or outcome, all having an improvement in neck pain, inflammation parameters, and ultrasound features.

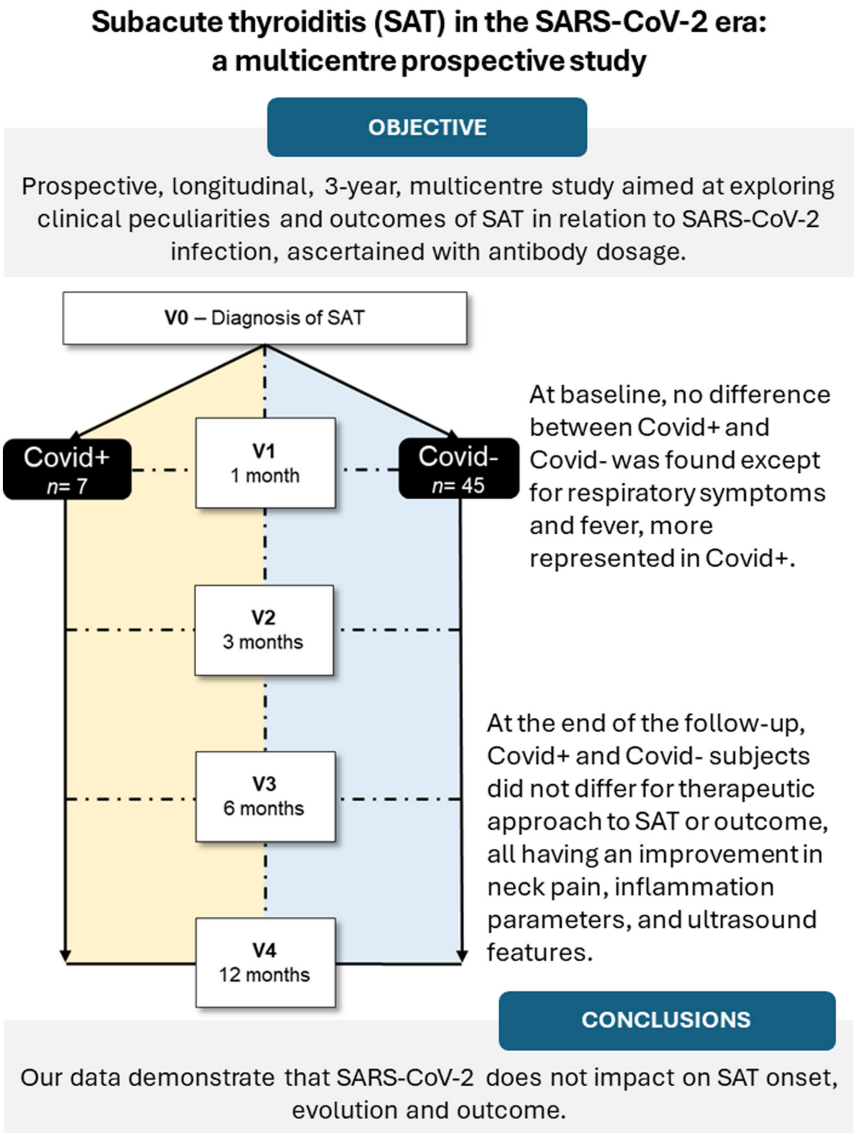
Conclusion: This is the first prospective study investigating any difference both at diagnosis and at follow-up between SAT presentation in patients with previous SARS-CoV-2 infection and those without. Our data demonstrate that SARS-CoV-2 does not impact on SAT onset, evolution, and outcome.

Keywords: thyroid; SARS-CoV-2; vaccine; pandemic; thyrotoxicosis; COVID-19

Published by Bioscientifica Ltd.
https://etj.bioscientifica.com

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

Graphical abstract



Introduction

Since the advent of the coronavirus disease (COVID-19) pandemic in early 2020 (1), a link between subacute thyroiditis (SAT) and severe acute respiratory syndrome coronavirus (SARS-CoV-2) infection has been observed. SAT is a thyroid inflammatory disease characterized by follicle damage that usually presents with anterior neck pain that may radiate to the ears and jaw (2). Other systemic and laboratory findings of SAT include low-grade fever, fatigue, elevated C-reactive protein (CRP), and erythrocyte sedimentation rate (ESR) (3). The SAT clinical course is divided into three phases: thyrotoxicosis (about 50% of cases) lasting 3–6 weeks, followed by hypothyroidism (about 30%), and restoration of normal thyroid function within 12 months (4). Notably, 5–15% of patients have persistent hypothyroidism after resolution of the symptomatic phase (4, 5). The thyroid ultrasonography (US) pattern is usually characterized by diffuse inhomogeneity, focal hypoechoic areas, and decreased or normal color flow at Doppler examination (3).

The SAT pathogenesis has not been fully elucidated yet (2, 4). The occurrence is believed to be a consequence of immunological and inflammatory mechanisms activated by viral infections or vaccinations. Beyond many viruses associated with SAT, namely coxsackie viruses, adenoviruses, and influenza, several cases of SAT related to COVID-19 (6, 7, 8, 9) and SARS-CoV-2 vaccine (10, 11, 12, 13) have been reported in the literature. Different possible theories have been elaborated to explain this thyroid involvement during the COVID-19

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

era. One of the most accredited hypotheses suggests that the thyroid gland is a potential target for viral damage because of the expression of the angiotensin-converting enzyme 2 (ACE2) on thyroid follicular cells, and its receptor being used by SARS-CoV-2 to invade the cells (14). Another possible mechanism is molecular mimicry, where antibodies developed against SARS-CoV-2 proteins could cross-react against thyroid antigens (15).

From a clinical standpoint, only a few studies explored the difference between COVID-19-related SAT and classical SAT. Some authors have observed that patients hospitalized for severe COVID-19 disease may develop an atypical form of SAT (e.g. absence of neck pain) compared to those previously described (15), while other studies, along with a recent systematic review, did not document any difference in the clinical presentation and outcomes between COVID-19-related SAT and classical SAT (16, 17, 18). Thus, whether SAT manifests differently from conventional SAT in relation to SARS-CoV-2 infection or its vaccine remains to be established. It should be noted that all the aforementioned studies have a retrospective design. The only prospective study found no clinical differences at diagnosis between classical SAT and COVID-19-related SAT (19). However, there are no data comparing the two groups during follow-up.

The aim of our prospective study was to describe the clinical characteristics of SAT cases at diagnosis and during a 12-month follow-up, comparing patients with previous exposure to SARS-CoV-2 to those without, in a multicentre cohort from Northern Italy, one of the regions most affected by the pandemic. In late 2020, the SARS-CoV-2 vaccination campaign started in Italy (20), therefore its possible link with SAT was also considered.

Methods

Study design

A multicentre, longitudinal, prospective study was conducted, enrolling all patients diagnosed with SAT at the participating centers between November 2020 and May 2022. The following Italian centers participated: Endocrinology Unit of Azienda Ospedaliero-Universitaria of Modena (Coordinating center); Endocrinology Unit of IRCCS Ca' Granda Ospedale Maggiore Policlinico of Milano; Endocrinology and Diabetes Prevention and Care Unit of the IRCCS Azienda Ospedaliero-Universitaria Policlinico of Bologna. These units were involved through a call launched by the coordinating center to the young Italian members of the Club EnGioI (Endocrinologia Giovane in Italia) of the Italian Society of Endocrinology (SIE).

Five visits were planned: at diagnosis (V0) and after 1, 3, 6, and 12 months (V1, V2, V3, and V4, respectively). At each visit, subjects were evaluated with anamnesis, physical examination, thyroid US, and blood tests. Patients were treated according to the clinical presentation and the current guidelines (2). Nonsteroidal anti-inflammatory drugs (NSAIDs) were preferred for patients with mild symptoms and mild laboratory findings, while steroid therapy was preferred for those with severe symptoms and/or those who did not respond to NSAIDs within 1–2 weeks. Beta-blockers were prescribed as symptomatic treatment in cases of tachycardia. During the follow-up phase, the therapeutic approach and any changes to it were recorded.

Finally, the treatment responsiveness and outcomes of transient hypothyroidism, permanent hypothyroidism, or recurrence during the follow-up period were all documented.

Ethics

The Institutional Review Board of Modena (protocol no. 1104/2020), Bologna (161/2021/Oss/AOUBo), and Milano (967_2020bis, CE Milano Area 2) approved the study. All enrolled subjects signed written informed consent. The research was conducted in accordance with the World Medical Association Declaration of Helsinki. The ClinicalTrials.gov registration number for the study is NCT06391515.

Subjects

Subjects with a clinical diagnosis of SAT were enrolled. The diagnosis was made by experienced endocrinologists according to guidelines (2), based on the presence of thyroid tenderness and anterior neck pain, often radiating to the ears, jaw, or throat, spreading from one side to the other, low-grade fever, pharyngitis symptoms, fatigue, thyroid slightly enlarged, firm and painful to palpation, ESR, or CRP elevation.

Inclusion criteria: clinical diagnosis of SAT, age ≥ 18 years, willingness to sign an informed consent. Exclusion criteria: ongoing pregnancy, alcohol abuse.

Division into COVID+ and COVID− groups

At V0, a blood sample was collected and centrifuged for serological analysis. Subsequently, sera were stored at −20°C until the end of the enrollment phase, when all samples were centralized at the coordinating center for SARS-CoV-2 IgG measurements. Both IgG against the spike protein (anti-S IgG) and against nucleocapsid (anti-N IgG) were tested. Anti-N IgG increases only after natural infection since the nucleocapsid protein is not contained in the vaccines, whereas anti-S IgG can increase due to either vaccination or infection (21). Thus, we subdivided patients according to their serological status: (i) group COVID+ included those patients who had both positive anti-S and anti-N IgG demonstrating contact with SARS-CoV-2 before the

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

diagnosis of SAT; (ii) group COVID− consisted of patients with only anti-S IgG positivity (due to vaccination) or negative anti-N/anti-S IgG.

In order to compare the possible impact of SARS-CoV-2 infection or vaccine on SAT, we performed a further analysis by subgrouping patients as follows: (i) positive anti-N and positive and anti-S (previous infection); (ii) negative anti-N and positive anti-S (previous vaccine); (iii) negative anti-N and anti-S (no previous exposure to COVID-19, neither infection nor vaccine).

Specific IgG antibodies to SARS-CoV-2 were detected for all serum samples using the Abbott Alinity platform, using a chemiluminescent microparticle immunoassay (CLIA) technology. Patient samples were incubated with SARS-CoV-2 antigen-coated paramagnetic microparticles that react to form antigen–antibody complexes. Then, acridinium-labeled anti-human IgG conjugates were added to form antigen–antibody anti-human IgG antibody complexes. The resulting chemiluminescent reaction was measured in relative light units (RLUs). A direct relationship exists between the amount of SARS-CoV-2 IgG Abs in the sample and the RLUs detected, reflecting the calculated index. We considered a positive cut-off for anti-N IgG 1.7 S/C and for anti-S IgG 7.1 BAU/mL. The procedures and the interpretation of the results were performed according to the manufacturers' instructions.

Procedures

Data collection at V0 included subjects' demographic data, history of thyroid disease, history of SAT, history of familial thyroid disease, previous and/or current anterior neck pain (localization, migrating, duration in days, evaluation on a scale from 0 to 10), previous and/or current medical treatments (drug, dosage, start and stop date), previous and/or current symptoms suggestive of SARS-CoV-2 infection (respiratory symptoms or body temperature above 37.5°C in the previous 6 months), previous known contacts with positives, and results of any swabs or serological tests for SARS-CoV-2 previously carried out. In Italy, the anti-SARS-CoV-2 vaccination campaign began on December 30th, 2020, therefore after the start of the study. Since then, the case report form has been updated with entries regarding vaccination (date and type of vaccine).

At each visit (from V0 to V4), data about typical symptoms of SAT such as neck pain, neck swelling, tenderness, tremors, fever, weight loss, sweating, fatigue, and signs such as goiter and tachycardia were also evaluated. Subjects underwent a physical examination, with specific reference to body weight and height, blood pressure and heart rate, and body temperature. BMI was calculated as body weight (kilograms) divided by the square of height (square meters). Body surface area (BSA) was calculated according to Mosteller's equation as follows: BSA (m^2) = square root of (height (cm) × weight (kg)/3600) (22).

Biochemical tests were performed at each center to assess the SAT course and to adjust medical management, measuring thyroid stimulating hormone (TSH), free thyroxine (fT4), free triiodothyronine (fT3), thyroglobulin (Tg), thyroglobulin antibodies (TgAb), anti-thyroid peroxidase antibodies (TPOAb), thyrotropin receptor antibodies (TRAb), ESR, high-sensitivity CRP, and complete blood count (CBC). All assays were performed in high-volume laboratories, and the units were made uniform in the laboratory data.

US examination was performed by endocrinologists with at least 5 years of experience in US neck examination using a linear probe. SAT-related findings such as inhomogeneity, hypoechoic areas, and increased gland volume were examined. Some subjects had new or known nodules, but none were suspicious. Data relating to nodules will not be shown here as they are considered not relevant to the SAT study. Thyroid gland vascular flow was evaluated by Doppler sonography.

Statistical analysis

Proportions and rates were calculated for categorical data, while continuous data were reported as median and interquartile range (IQR). Clinical, biochemical, and US parameters were compared between COVID+ and COVID− subgroups. The non-parametric Mann–Whitney *U* test or Kruskal–Wallis test were used for comparisons of continuous variables since they were not normally distributed according to the Kolmogorov–Smirnov test. Categorical variables were compared by Pearson's Chi-squared test.

Statistical analyses were performed using the SPSS software for Windows (version 28.0; SPSS Inc., USA). For all comparisons, $P < 0.05$ was considered statistically significant.

Results

In total, 66 patients receiving a SAT diagnosis during the study period were enrolled. Of them, eight were excluded from subsequent analyses because they had an onset of symptoms more than 60 days before diagnosis. We arbitrarily decided on this threshold to exclude patients with symptom onset times very distant from those of all other enrolled subjects, as symptoms and clinical data of these patients would alter the picture of the SAT trend.

The clinical characteristics of the remaining 58 patients at V0 (corresponding to the SAT diagnosis and not to the onset of symptoms) are summarized in Table 1. Only four patients did not report previous or concomitant neck pain, but they all presented with inhomogeneous thyroid echostructure as per thyroiditis, negative anti-thyroid Ab, thyrotoxicosis, and elevated ESR and/or CRP. Among the 54 subjects who presented with pain, 36 (67%) had complained of pain for more than 15 days.

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

Forty-one subjects completed the 12-month follow-up, while 17 subjects dropped out, mainly after the symptoms resolved, skipping the following visits (Fig. 1). Of the subjects who completed follow-up, 10% were COVID positive, compared to 18% of subjects who dropped out before the end of the study ($P > 0.05$). They reported less pain at diagnosis (33% vs 74%, $P = 0.03$) and had less thyrotoxicosis (47% vs 81%, $P = 0.01$).

Serum samples at diagnosis were collected from 52 out of 58 subjects. Serum SARS-CoV-2 IgG measurement was performed, documenting that seven subjects (13.5%) had both positive anti-N and anti-S IgG as per direct contact with the virus before the diagnosis of SAT; they were included in the COVID+ group. Only two of these subjects had previously tested positive on a swab test.

Comparison between COVID+ and COVID− groups

At diagnosis, most subjects were already receiving analgesic or symptomatic drugs, without any difference between COVID+ and COVID− groups: 17 were treated with NSAIDs, 3 with beta-blockers, and 14 with steroids (Table 2). Two subjects were taking levothyroxine due to previous hypothyroidism; on the contrary, methimazole (range: 5–20 mg daily) was started in three thyrotoxic patients before referral to our centers (V0) and promptly stopped at SAT diagnosis. Obviously, these drugs impacted the presentation of the disease at the time of diagnosis. However, this bias equally affected both the COVID+ and COVID− groups, the comparison of which is the primary objective of this study (Table 2).

At diagnosis, the COVID+ group reported respiratory symptoms or fever more frequently than the COVID− ($P = 0.039$ and 0.021, respectively), despite significantly lower ESR values ($P = 0.021$). No other differences emerged at diagnosis between the two groups (Table 2). Globally, COVID+ subjects had mild symptoms that did not require hospitalization or antiviral treatment. Fifty-seven percent had respiratory symptoms and 71% had temperature >37.5°C in the previous 6 months.

A total of 19 out of 58 patients (32.8%) of the entire cohort had already received the first dose of COVID-19 vaccine before the SAT diagnosis at V0; the mean ± S.D. time between the vaccine administration and V0 was 161 ± 123 days. Of these 19 subjects, 17 tested COVID− and two tested COVID+. We then compared anti-S negative (no previous exposure to COVID, either infection or vaccine) to anti-S positive + anti-N negative (previous vaccine) and to anti-S positive + anti-N positive (previous infection) subjects. The only significant difference was again ESR lower in anti-S positive + anti-N positive subjects compared to the other groups (Supplementary Tables 1 and 2, see section on supplementary materials given at the end of this article).

Subjects were followed up for 12 months. NSAIDs were prescribed in 43% of COVID+ and 34% of COVID−

Table 1 Characteristics of patients at the time of diagnosis (V0) expressed as *n* (%) or median (IQR).

Characteristics	Normal range	Values
n		58
Sex		
Males		11 (19%)
Females		47 (81.0%)
Age (years)	n.a.	50.0 (42.3–56.4)
BMI (kg/m^2)	18.5–25.0	23.8 (21.2–25.3)
SBP (mm Hg)	n.a.	115 (110–130)
DBP (mm Hg)	n.a.	75 (70–80)
HR (bpm)	n.a.	80 (69–89)
Body temperature (°C)	n.a.	36.5 (36–36.9)
Thyroid volume-to-BSA ratio	n.a.	8.3 (5.7–10.7)
ESR (mm)	2–37	46 (27–73)
CRP (mg/dL)	0–0.7	1.6 (0.5–4.8)
TSH (μUI/mL)[a]	0.35–4.94	0.18 (0.01–2.21)
fT3 (pg/mL)[a]	2.5–3.9	3.6 (3.1–5.1)
fT4 (pg/mL)[a]	6–12	13.2 (10.2–23.1)
Tg (ng/mL)	1–48	14.7 (5.3–48.8)
Positive TPOAb, *n* (%)	<9 mIU/L	6 (12%)
Positive TgAb, *n* (%)	<4 mIU/L	9 (19%)
Positive TRAb, *n* (%)	<0.4 U/L	7 (16%)

[a]Seven patients were excluded from this analysis due to previous/current methimazole treatment.
BMI, body mass index; BSA, body surface area; CRP, C-reactive protein; DBP, diastolic blood pressure; ESR, erythrocyte sedimentation rate; fT3, free triiodothyronine; fT4, free thyroxine; HR, heart rate; IQR, interquartile range; SBP, systolic blood pressure; TSH, thyroid-stimulating hormone; Tg, thyroglobulin; TPOAb, thyroid peroxidase antibodies; TgAb, thyroglobulin antibodies; TRAb, anti-TSH receptor antibodies.

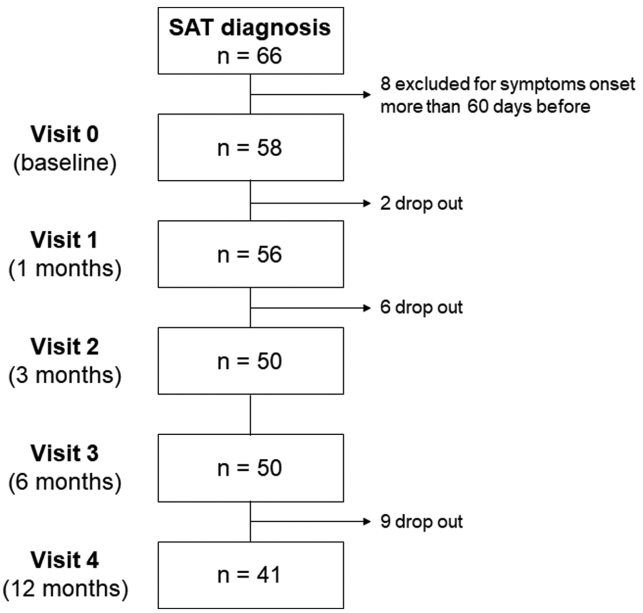


Figure 1
Flow chart of the study.

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

Table 2 Comparison between the characteristics of COVID+ and COVID− at baseline (V0); data are expressed as *n* (%) or median (IQR).

	COVID+	COVID−	*P*
n	7	45	
Females	6 (85.7%)	36 (80%)	0.72
Age (years)	52.1 (43.5–59.7)	49.5 (41.0–56.0)	0.78
BMI (kg/m^2)	23.8 (20.5–24.5)	24.0 (21.1–25.5)	0.73
Previous thyroid disease	2 (28.6%)	13 (28.9%)	0.99
Physical examination			
SBP (mm Hg)	110 (102–114)	120 (110–130)	0.09
DBP (mm Hg)	70 (66–81)	80 (70–80)	0.35
HR (bpm)	82 (72–92)	80 (68–94)	0.85
Respiratory symptoms	4 (57.1%)	9 (20.5%)	0.04
Temperature > 37.5°C	5 (71.4%)	12 (27.3%)	0.02
Temperature (°C)	36.0 (36.0–36.8)	36.5 (36.1–37.0)	0.25
Increased thyroid consistency	4 (66.7%)	20 (55.3%)	0.48
Pain at palpation	3 (50.0%)	22 (55.0%)	0.82
Pain before diagnosis (>15 days)	5 (71.4%)	31 (68.9%)	0.69
Current pain at V0	3 (42.9%)	30 (66.7%)	0.22
Biochemical parameters			
ESR (mm)	29 (17–41)	52 (30–75)	0.02
CRP (mg/dL)	0.5 (0.2–3.3)	1.7 (0.5–4.2)	0.15
TSH (μUI/mL)[a]	0.30 (0.02–1.46)	0.11 (0.01–1.88)	0.53
fT3 (pg/mL)[a]	3.10 (2.61–4.10)	3.89 (3.15–5.20)	0.12
fT4 (pg/mL)[a]	12.4 (11.0–20.4)	13.6 (10.3–24.4)	0.62
Tg (ng/mL)	37.0 (1.7–92.1)	15.0 (8.8–68.2)	0.81
Thyrotoxicosis[b]	4 (57.1%)	33 (78.6%)	0.22
Positive TPOAb	1/7 (14.3%)	5/39 (12.8%)	0.92
Positive TgAb	0/2 (0%)	6/28 (21.4%)	0.46
Positive TRAb	2/5 (40.0%)	5/34 (14.7%)	0.17
US characteristics			
Thyroid volume-to-BSA ratio	8.1 (6.9–10.7)	8.3 (5.7–10.8)	0.77
Increased thyroid volume	3 (60.0%)	21 (51.2%)	0.71
Thyroid inhomogeneity	7 (100%)	39 (95.3%)	0.55
Previous medical treatment (before V0)			
Beta-blockers	1 (14.3%)	3 (15.0%)	0.44
NSAIDs	3 (42.9%)	14 (31.1%)	0.44
Steroids	5 (71.4%)	9 (20.0%)	0.08
Levothyroxine	1 (14.3%)	1 (2.2%)	0.39
Methimazole	1 (14.3%)	2 (4.4%)	0.55

[a]Seven patients (two COVID+ and five COVID−) were excluded from this analysis due to previous/current methimazole treatment; [b]Two COVID+ and five COVID− patients receiving previous/current methimazole treatment were included.
BMI, body mass index; BSA, body surface area; CRP, C-reactive protein; DBP, diastolic blood pressure; ESR, erythrocyte sedimentation rate; fT3, free triiodothyronine; fT4, free thyroxine; HR, heart rate; IQR, interquartile range; NSAIDs, non-steroidal anti-inflammatory drugs; SBP, systolic blood pressure; Tg, thyroglobulin; TSH, thyroid-stimulating hormone; TPOAb, thyroid peroxidase antibodies; TgAb, thyroglobulin antibodies; TRAb, anti-TSH receptor antibodies; V0, baseline.

(P = 0.441), and steroids in 86% of COVID+ and 60% of COVID− (P = 0.145), with a mean steroid duration of 44 and 47 days, respectively (Table 3); prednisone was the most used steroid (27/33), followed by methylprednisolone (1/33) and betamethasone (1/33), whereas for four patients, we had no details about steroid therapy. Beta-blockers were used in 14% of the COVID+ group and 15% of the COVID− group (P = 0.961) (Table 3). Overall, no significant difference was observed in comparing the therapeutic approach (steroids, NSAIDs, beta-blockers) adopted for COVID+ and COVID− (Table 3). Both COVID+ and COVID− patients showed improvement in neck pain and US parameters over visits (Fig. 2). Data about laboratory measurements, including both thyroid function and inflammation markers, improved over visits without significant differences between the two groups (COVID+ and COVID−). No differences in the proportions of patients with transient hypothyroidism at V1–V2–V3 (P = 0.653), normal thyroid function at V4 (P = 0.612), and hypothyroidism requiring LT4 therapy at V4 (P = 0.713) were observed between the groups (Table 3). Similarly, no difference

European Thyroid Journal (2024) **13** e240083
https://doi.org/10.1530/ETJ-24-0083

Table 3 Comparison between COVID+ (*n* = 7) and COVID− (*n* = 45) in medical management and outcome parameters; data are expressed as *n* (%) or median (IQR).

	COVID+	COVID−	*P* value
Medical treatment during study period			
Beta blockers	1 (14.3%)	6 (15.0%)	0.96
NSAIDs	3 (42.9%)	14 (34.1%)	0.44
NSADs duration (days)	8 (2–8)	5 (2–13)	0.79
Steroids	6 (85.7%)	27 (60.0%)	0.14
Steroids duration (days)	29 (19–74)	39 (21–62)	0.70
Cumulative steroids dosage (mg)	284 (175–1075)	543 (306–905)	0.49
Levothyroxine	1 (14.3%)	2 (4.4%)	0.37
Thyroid functionality evolution			
Transient hypothyroidism at V1, V2, or V3	2/5 (40.0%)	14/46 (30.4%)	0.65
Normal thyroid function at V4	3/3 (100%)	29/36 (80.6%)	0.61
Hypothyroidism at V4 (requiring LT4 therapy)	0/3 (0%)	3/36 (8.3%)	0.71

IQR, interquartile range; NSAIDs, non-steroidal anti-inflammatory drugs; V1, visit at 1 month; V2, visit at 3 months; V3, visit at 6 months; V4, visit at 12 months.

was observed at V4 in the percentage of subjects who restored normal thyroid function (100% in COVID+ and 81% in COVID−) or developed permanent hypothyroidism requiring L-T4 therapy (0% in COVID+ and 8.3% in COVID−) (Table 3). The percentages of dysthyroidism and euthyroidism at each visit are graphed in Fig. 3.

Finally, only one patient had a recurrence of SAT during follow-up at V4.

Discussion

This is the first prospective study investigating any difference in diagnosis or follow-up among SAT cases in patients with serologically ascertained exposure to SARS-CoV-2 and in those without previous contact with SARS-CoV-2.

Respiratory symptoms and fever were more prevalent in the COVID+ group, surprisingly accompanied by lower ESR values, the only significant difference in diagnosis. Apparently, the difference in ESR levels was not associated with anti-inflammatory treatments received before SAT diagnosis since no significant difference was observed between COVID+ and COVID−. However, a *P*-value that bordered on statistical significance was found for corticosteroid use, more frequent in the COVID+ group than in the COVID− (71% vs 20%). Hence, it is reasonable to suppose that corticosteroids may have at least partly impacted ESR values recorded at baseline.

Otherwise, the two groups did not differ at baseline in thyroid function, painful symptoms, or thyroid US appearance.

Our data confirmed that the diagnosis of SAT is often delayed (23), with a rather long latency from

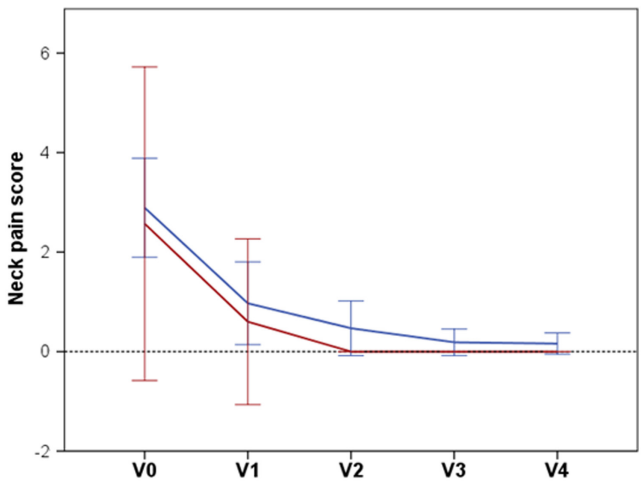


Figure 2

Temporal trend along visits of neck pain score in COVID− (blue line) and COVID+ (red line). Whiskers correspond to the 95% confidence interval. V0, baseline; V1, visit at 1 month; V2, visit at 3 months; V3, visit at 6 months; V4, visit at 12 months.

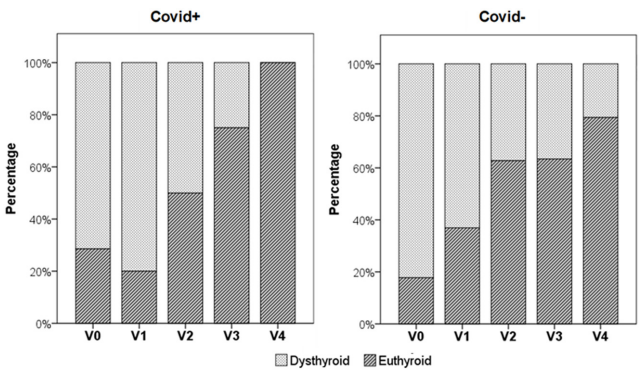


Figure 3

Percentages of dysthyroid and euthyroid subjects at each visit in the COVID− and COVID+ groups. V0, baseline; V1, visit at 1 month; V2, visit at 3 months; V3, visit at 6 months; V4, visit at 12 months.

symptom onset. This diagnostic delay was even more exacerbated in pandemic times, when people were afraid of going to hospitals that admitted SARS-CoV-2-infected patients. Although we excluded subjects with symptom onset more than 60 days before SAT diagnosis, 70% of the remaining patients presented with neck pain for 15 days before, starting symptomatic drugs in most cases. This certainly reduced the number of subjects with pain at diagnosis, which was instead present at the onset of symptoms in almost all those enrolled. Only four subjects did not present pain, but had other clinical, serological, and US characteristics that led to the diagnosis of SAT. Even if COVID+ patients had lower rate of pain at diagnosis, this difference was not statistically significant from COVID− patients and what was expected in classical SAT. However, it has been observed that patients with severe COVID-19 disease presented frequent episodes of mild painless thyroiditis (24), with quick and spontaneous normalization of thyroid dysfunction shortly after the end of SARS-CoV-2 infection, but long-term persistence of hypoechoic areas at thyroid ultrasound scans (25). The different characteristics of thyroiditis in this group of patients are likely due to the severe form of COVID-19 disease and the coexistence of non-thyroidal illness syndrome.

Considering thyroid function at diagnosis, the rate of thyrotoxicosis in COVID+ was 57%, without difference from COVID− and in accordance with the expected rate in classical SAT (4). During monitoring for the following 12 months, there was a rise in TSH values and a decline in fT4 in all subjects, with no differences between COVID+ and COVID−. About 30–40% of subjects experienced transient hypothyroidism during follow-up, without differences between groups. At the end of the follow-up, all COVID+ cases reached a state of spontaneous euthyroidism, without needing levothyroxine replacement therapy. About 8% of the COVID− cases required levothyroxine, as expected (4, 5). These results would suggest an excellent *restitutio ad integrum* of the thyroid gland after SARS-CoV-2-related SAT, but the small sample size at the end of the follow-up in the COVID+ group makes the data not generalizable.

Although the study protocol did not provide predefined therapeutic schemes, the subjects were treated with NSAIDs, steroids, and beta-blockers without differences between the two groups. Thanks to these treatments, a gradual improvement in symptoms, thyroid US features, and inflammation markers was achieved in both COVID+ and COVID−.

The analysis of SAT cases occurring after the administration of the COVID-19 vaccine was outside the focus of the present study since vaccines were not yet available at the time of study conceptualization. However, with the introduction of vaccines in Italy, we started to collect data about the time interval between vaccine administration and the onset of SAT symptoms. We observed that up to one-third of our cohort had already received the first dose of the vaccine before V0, with a median time distance of 161 days. This period was much longer than all intervals reported in the literature, both for single SAT cases appearing 1–21 days (median 7 days) after COVID-19 vaccination (11, 12, 13), and for prospective studies reporting a median of 4 (1–12) weeks (19) and 45 (7–90) days (26). Due to these limitations, it is not possible to draw further conclusions regarding cases related to vaccines in our cohort.

The present study has the strength of being prospective, multicentre, having defined previous contact with the virus with antibody dosage measured centrally, and monitoring patients for a follow-up of up to 12 months.

However, results are limited by the fact that the prospective study inevitably started from the diagnosis of SAT and not from the onset of symptoms. This is secondary to the well-known diagnostic delay of SAT, initially mistaken for a sore throat, and aggravated by poor access to treatment during pandemic times. For the same reason, the serum sample on which the anti-SARS-CoV-2 immunoglobulins were assayed was collected at diagnosis, not at onset. Some patients classified as COVID+ may have had contact with the virus between onset and diagnosis of SAT. Therefore, the percentage of SAT likely to be secondary to SARS-CoV-2 infection could drop further. Furthermore, it is fair to point out that our case series only includes outpatients who may present different characteristics compared to patients hospitalized for more severe COVID-19. Moreover, the study design did not include a common therapeutic regimen, so each patient was treated differently according to clinical practice. Finally, the generalizability of our results is limited by the small sample size, especially of the COVID+ group. However, having centralized the measurement of anti-SARS-CoV-2 antibodies at the end of enrollment, we could not know *a priori* how many subjects had previous contact with the virus. Moreover, the dropout rate was higher than expected, maybe because of the pandemic, which kept patients away from doctors as soon as the symptoms resolved for fear of contagion.

In conclusion, our data demonstrate that SAT developed in patients with serologically documented previous exposure to SARS-CoV-2 presents with similar clinical and biochemical characteristics both at diagnosis and during the following 12 months in comparison to patients without previous contact with SARS-CoV-2.

Supplementary materials

This is linked to the online version of the paper at https://doi.org/10.1530/ETJ-24-0083.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the study reported.

Funding

This work was supported by the 'ESE COVID-19 Research Grant' from the European Society of Endocrinology.

Author contribution statement

SDV, MLM, DS, and GB conceived the study; SDVA, SL, EZ, RS, MLM, DS, IM, FDM, EC, MA, LT, CC, and GB enrolled and followed the patients; MS, UP, and IM supervised the enrollment of patients; GB coordinated the centers; TT, VP, GC performed the laboratory analyses; SDV, SL, EZ, FDM, EC, MA, LT, CC, and GB collected the data; SDV, DS, LT performed the statistical analyses and analyzed the results; SDV, GB, VP wrote the paper; all the authors approved the final version of the manuscript.

References

1 Dong E, Du H & Gardner L. An interactive web-based dashboard to track COVID-19 in real time. *Lancet. Infectious Diseases* 2020 **20** 533–534. (https://doi.org/10.1016/S1473-3099(20)30120-1)

2 Ross DS, Burch HB, Cooper DS, Greenlee MC, Laurberg P, Maia AL, Rivkees SA, Samuels M, Sosa JA, Stan MN, *et al.* 2016 American Thyroid Association guidelines for diagnosis and management of hyperthyroidism and other causes of thyrotoxicosis. *Thyroid* 2016 **26** 1343–1421. (https://doi.org/10.1089/thy.2016.0229)

3 Frates MC, Marqusee E, Benson CB & Alexander EK. Subacute granulomatous (de Quervain) thyroiditis: grayscale and color Doppler sonographic characteristics. *Journal of Ultrasound in Medicine* 2013 **32** 505–511. (https://doi.org/10.7863/jum.2013.32.3.505)

4 Fatourechi V, Aniszewski JP, Fatourechi GZ, Atkinson EJ & Jacobsen SJ. Clinical features and outcome of subacute thyroiditis in an incidence cohort: Olmsted County, Minnesota, study. *Journal of Clinical Endocrinology and Metabolism* 2003 **88** 2100–2105. (https://doi.org/10.1210/jc.2002-021799)

5 Benbassat CA, Olchovsky D, Tsvetov G & Shimon I. Subacute thyroiditis: clinical characteristics and treatment outcome in fifty-six consecutive patients diagnosed between 1999 and 2005. *Journal of Endocrinological Investigation* 2007 **30** 631–635. (https://doi.org/10.1007/BF03347442)

6 Abobaker A, Raba AA & Alzwi A. Extrapulmonary and atypical clinical presentations of COVID-19. *Journal of Medical Virology* 2020 **92** 2458–2464. (https://doi.org/10.1002/jmv.26157)

7 Ippolito S, Dentali F & Tanda ML. SARS-CoV-2: a potential trigger for subacute thyroiditis? Insights from a case report. *Journal of Endocrinological Investigation* 2020 **43** 1171–1172. (https://doi.org/10.1007/s40618-020-01312-7)

8 Brancatella A, Ricci D, Viola N, Sgrò D, Santini F & Latrofa F. Subacute thyroiditis after Sars-COV-2 infection. *Journal of Clinical Endocrinology and Metabolism* 2020 **105**. (https://doi.org/10.1210/clinem/dgaa276)

9 Asfuroglu Kalkan E & Ates I. A case of subacute thyroiditis associated with Covid-19 infection. *Journal of Endocrinological Investigation* 2020 **43** 1173–1174. (https://doi.org/10.1007/s40618-020-01316-3)

10 Yorulmaz G & Sahin Tekin M. SARS-CoV-2 vaccine-associated subacute thyroiditis. *Journal of Endocrinological Investigation* 2022 **45** 1341–1347. (https://doi.org/10.1007/s40618-022-01767-w)

11 İremli BG, Şendur SN & Ünlütürk U. Three cases of subacute thyroiditis following SARS-CoV-2 vaccine: postvaccination Asia syndrome. *Journal of Clinical Endocrinology and Metabolism* 2021 **106** 2600–2605. (https://doi.org/10.1210/clinem/dgab373)

12 Tomic AZ, Zafirovic SS, Gluvic ZM, Samardzic VS, Macvanin MT, Radunovic ML & Isenovic ER. Subacute thyroiditis following COVID-19 vaccination: case presentation. *Antiviral Therapy* 2023 **28** 13596535231208831. (https://doi.org/10.1177/13596535231208831)

13 Saygılı ES & Karakilic E. Subacute thyroiditis after inactive SARS-CoV-2 vaccine. *BMJ Case Reports* 2021 **14**. (https://doi.org/10.1136/bcr-2021-244711)

14 Rotondi M, Coperchini F, Ricci G, Denegri M, Croce L, Ngnitejeu ST, Villani L, Magri F, Latrofa F & Chiovato L. Detection of SARS-COV-2 receptor ACE-2 mRNA in thyroid cells: a clue for COVID-19-related subacute thyroiditis. *Journal of Endocrinological Investigation* 2021 **44** 1085–1090. (https://doi.org/10.1007/s40618-020-01436-w)

15 Vojdani A, Vojdani E, Melgar AL & Redd J. Reaction of SARS-CoV-2 antibodies with other pathogens, vaccines, and food antigens. *Frontiers in Immunology* 2022 **13** 1003094. (https://doi.org/10.3389/fimmu.2022.1003094)

16 Batman A, Yazıcı D, Dikbaş O, Ağbaht K, Saygılı ES, Demirci İ, Bursa N, Ayas G, Anıl C, Cesur M, *et al.* Subacute THYROiditis related to SARS-CoV-2 vaccine and Covid-19 (THYROVAC study): a multicenter nationwide study. *Journal of Clinical Endocrinology and Metabolism* 2023 **108** e1013–e1026. (https://doi.org/10.1210/clinem/dgad235)

17 Meftah E, Rahmati R, Zari Meidani F, Khodadadi S, Chitzan-Zadeh K, Esfahanian F & Afshar S. Subacute thyroiditis following COVID-19: a systematic review. *Frontiers in Endocrinology* 2023 **14** 1126637. (https://doi.org/10.3389/fendo.2023.1126637)

18 Angelopoulos N, Askitis DP, Androulakis I, Valvis N, Paparodis R, Petkova V, Boniakos A, Zianni D, Perogamvros I, Toulis K, *et al.* Clinical aspects in subacute thyroiditis: a real-life study on 226 cases in Greece amid the COVID-19 pandemic. *Journal of Clinical Medicine* 2023 **12**. (https://doi.org/10.3390/jcm12227171)

19 Bahçecioğlu AB, Karahan ZC, Aydoğan BI, Kalkan IA, Azap A & Erdoğan MF. Subacute thyroiditis during the COVID-19 pandemic: a prospective study. *Journal of Endocrinological Investigation* 2022 **45** 865–874. (https://doi.org/10.1007/s40618-021-01718-x)

20 Dai L & Gao GF. Viral targets for vaccines against COVID-19. *Nature Reviews. Immunology* 2021 **21** 73–82. (https://doi.org/10.1038/s41577-020-00480-0)

21 Assis R, Jain A, Nakajima R, Jasinskas A, Khan S, Palma A, Parker DM, Chau A, Specimen Collection Group, Obiero JM, *et al.* Distinct SARS-CoV-2 antibody reactivity patterns elicited by natural infection and mRNA vaccination. *NPJ Vaccines* 2021 **6** 132. (https://doi.org/10.1038/s41541-021-00396-3)

22 Mosteller RD. Simplified calculation of body-surface area. *New England Journal of Medicine* 1987 **317** 1098. (https://doi.org/10.1056/NEJM198710223171717)

23 Bostan H, Sencar ME, Calapkulu M, Hepsen S, Duger H, Ozturk Unsal I, Ozbek M & Cakal E. Two important issues in subacute thyroiditis management: delayed diagnosis and inappropriate use of antibiotics. *European Thyroid Journal* 2021 **10** 323–329. (https://doi.org/10.1159/000513745)

24 Muller I, Cannavaro D, Dazzi D, Covelli D, Mantovani G, Muscatello A, Ferrante E, Orsi E, Resi V, Longari V, *et al.* SARS-CoV-2-related atypical thyroiditis. *Lancet. Diabetes and Endocrinology* 2020 **8** 739–741. (https://doi.org/10.1016/S2213-8587(20)30266-7)

25 Muller I, Daturi A, Varallo M, Re TE, Dazzi D, Maioli S, Crivicich E, Di Marco F, Longari V, Dazzi B, *et al.* Long-term outcome of thyroid abnormalities in patients with severe Covid-19. *European Thyroid Journal* 2023 **12**. (https://doi.org/10.1530/ETJ-22-0200)

26 Topaloğlu Ö, Tekin S, Topaloğlu SN & Bayraktaroglu T. Differences in clinical aspects between subacute thyroiditis associated with COVID-19 vaccines and classical subacute thyroiditis. *Hormone and Metabolic Research* 2022 **54** 380–388. (https://doi.org/10.1055/a-1840-4374)