Five-year follow-up of 165 Italian patients with undifferentiated connective tissue diseases

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Abstract

Objective

To study those conditions with a proven or hypothesised immunologic pathogenesis and denominated under a working definition of undifferentiated connective tissue diseases (UCTD).

Methods

A multicentre prospective study was organised involving 10 tertiary referral centers of internal medicine in Italy, with the aim of describing the natural history of UCTD and the prevalence of its different clinical and immunological manifestations.

Results

After a five-year follow-up period, data on 165 patients were available for analysis. UCTDs occur mainly in females in their fourth decade of life. Articular and mucocutaneous features and Raynaud's phenomenon represent the most common findings. Nevertheless, we also detected a relatively high incidence of permanent major organ damage. Regarding the immunologic parameters, we documented some conflicting results in the correlation between serologic abnormalities and clinical features. In 10 patients UCTD evolved to a major disease, generally systemic lupus erythematosus or Sjögren's syndrome.

Conclusion

A low rate of evolution to a defined autoimmune disease, the limited use of steroid or immunosuppressive therapy, and a favourable course in the majority of cases are the main characteristics of patients with UCTDs.

Key words
Autoimmune diseases, connective tissue diseases (CTDs), undifferentiated CTD, outcome.

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Introduction

The major systemic autoimmune diseases represent a broad group of clinical disorders presenting with both distinguishing and shared features, and characterised by tissue destruction caused by an ongoing immune response. Among these syndromes, it is possible to recognise two different clinical conditions. In the first group are included those diseases, such as the connective tissue diseases (CTDs), whose diagnoses are based on specific criteria (1-6). The second group comprises those patients who do not fulfill all the criteria required for a defined disease. These cases have been classified as undifferentiated CTD or early undifferentiated CTD by various authors (7-10).

We decided to study those clinical and serological conditions which are autoimmune in nature, but where it is not possible to reach a definite diagnosis. We use the global term undifferentiated connective tissue diseases (UCTD) to refer to these conditions. In this group we also included patients with few signs or symptoms of suspected autoimmune origin, in order to determine whether we are being confronted with a distinct disease or an immune system dysfunction devoid of any specific nosologic connotation. We set up a prospective study involving 10 clinical centres in Italy, our principal aim being to define the clinical and serological features in subjects with UCTDs at disease onset and after a five-year follow-up.

Patients and methods

Patients

For this study we recruited subjects, aged 14 - 70 years, being seen as in- or outpatients at 10 different clinics in the northern, central and southern parts of Italy. Each centre enrolled from 15 to 20 cases and studied them prospectively for a period of 5 years following a standardised protocol. We excluded from the study patients diagnosed as having a defined CTD, such as rheumatoid arthritis (RA) (1), systemic lupus erythematosus (SLE) (2), systemic sclerosis (SSc or scleroderma) (3), Sjögren's syndrome (SS) (4), polydermatomyositis (PM) (5) and mixed connective tissue disease (MCTD) (6), or other diseases such as antiphospholipid syndrome (11), mixed cryoglobulinemia (12), Behçet's disease (13), spondyloarthropathy (14) and vasculitis as defined by the ACR 1990 criteria (15).

The inclusion parameters, which must have been present for less than 24 months or at the study entry, were at least two of the clinical signs or one clinical and one serologic sign from those listed in Table I and defined according to the ARA glossary. For each patient a complete medical history, physical examination and laboratory tests were performed at study entry and subsequently at least twice a year. We also recorded data on permanent organ damage. We did not exclude from the study those patients re-

Table I. Clinical and laboratory criteria for the diagnosis of UCTD.

Myositis *
Arthralgias - arthritis
Raynaud's phenomenon in the hands and feet
Recurrent deep and superficial vein thrombosis
Pleuritis and/or pericarditis and/or peritonitis
Myocarditis - endocarditis
Oral erosion or aphtas
Skin erythema ("butterfly", discoid, heliotrope rash distribution)
Photosensitivity
Sclerodactyly
Superficial and palpable purpura
Erythema nodosum
Teleangiectasia
Subcutaneous calcifications
Alopecia
Livedo reticularis
Xerostomia
Xerophtalmia
Recurrent enlargement of the parotid gland
Dismotility of the oesophagus
Mononeuropathy
Psychosis and seizures
TIA, stroke
Interstitial lung disease
Anaemia, leucopenia, lymphopenia, thrombocytopenia
Recurrent abortion
Antinuclear antibodies
Anti-ds DNA antibodies
Anti-ENA antibodies
Rheumatoid factors
Anti-neutrophil cytoplasmic autoantibodies (ANCA)
Anti-cardiolipin antibodies, LAC, VDRL false positivity

* not fulfilling the established criteria (5).

ceiving the following treatments: colchicin, antimalarials, non-steroidal anti-inflammatory drugs (NSAIDs), and low dose steroids (< 10 mg/die of prednisone or equivalent).
The aims of the study and the procedures involved were explained to the patients and their informed consent was obtained.

Procedures
Raynaud's phenomenon (RP) was determined using nailfold capillary microscopy. More specifically targeted tests such as chest radiogram, carbon monoxide (CO) diffusion capacity, an oesophageal motility study and electromyogram (EMG) were performed as necessary. All patients complaining of xerostomia and xerophthalmia underwent a thorough work-up based on the procedures of Vitali *et al*. (4, 16).

Immunologic parameters
Immunologic parameters were analysed in all the patients. They had to be present on two separate occasions at least two months apart and validated by the specific reference centre (see below). The data presented in the paper were furnished by the respective reference centres and were documented as follows:

- antinuclear antibodies (ANA) by the immunofluorescent method. ANA titres values = 1: 160 were considered as positive;
- anti-dsDNA antibodies by the RIA-Farr technique (normal values: < 4.2 U/ml) (reference centre: Brescia);
- anti-ENA antibodies by counterimmunoelectrophoresis (17) and Western blot analysis to detect the differing patterns of RNP and SSA/Ro (18) (reference centre: Brescia);
- rheumatoid factor (RF) by the Waaler Rose test (positive if a serum dilution = 1: 8) and/or by latex agglutination (positive if 1: 20) and/or by laser nephelometry (positive if levels > 40 IU/ml);
- the third and fourth complement fractions (C3 and C4) by immunodiffusion plates or nephelometry;
- anticardiolipin antibodies (aCL) by a solid phase enzyme-linked immunosorbent assay (ELISA) using the technique described by Harris (19) (reference centre: I Clinica Medica, Rome);
- antineutrophil cytoplasmic autoantibodies (ANCA) by an indirect immunofluorescence assay according to the recommendations of the First International ANCA Workshop (20) using 1: 20 diluted sera on human ethanol-fixed neutrophils (reference centre: Florence).

Results

Baseline characteristics
One hundred and eighty-three subjects were enrolled in the study. At the end of the 5-year follow-up period, data on 165 patients were available for subsequent analysis (11% of the cases were lost during follow-up). There were 12 males and 153 females; the mean age at inclusion was 41.5 ± 13.9 years (range 16-70, median 41).

Clinical and serological features at the onset based on the medical history
The major target organs were the vascular system (mainly RP) and joints. The skin and mucocutaneous systems were also frequently involved. About 20% of the patients showed haematological abnormalities as the first manifestation of their disease. No lung, kidney, gut, central or peripheral nervous system involvement was recorded. Few specific laboratory abnormalities were present, i.e. ANA and RF in 8% and 5% of the patients, respectively.

Clinical features at inclusion
At inclusion, 61% and 77% of patients showed 4 or 5 clinical and/or serological features, respectively. Sixty-two patients out of the 165 complained of arthralgias (37%) and 37 of (22%) arthritis. RP was detected in 83 patients (50%). Eighty-seven patients (52%) showed mucocutaneous involvement, mainly characterised by photosensitivity, butterfly rash, sclerodactyly and alopecia, whereas discoid rash, purpura, aphthous ulcers and tissue calcifications were less frequently represented. Sicca syndrome was present in 37 patients (22%), in most cases in the form of associated xerostomia and xerophthalmia.
Cardiovascular system involvement was found in only 6% of the patients, the principal target being the pericardium with 7 cases of pericarditis. Five patients presented a history of arterial/venous occlusion including two acute cerebrovascular episodes. Gut involvement was manifested as dysmotility of the oesophagus with reflux oesophagitis in 5% of the patients. The lung was not frequently involved, but mild pleurisy and interstitial involvement were present in 7% of the patients. It should be noted that this data is probably underestimated since few patients underwent specific functional tests. Central and peripheral nervous system involvement was documented in 12 patients in the form of psychosis (2 cases), seizures (1 case), acute cerebrovascular accidents (2 cases) and polyneuropathy (7 cases). The kidney was spared. Haematological disorders were detected in 32 patients (19%).

Immunological parameters at inclusion
ANA, as expected, were the principal abnormalities present in 97 patients (58%), with homogeneous and speckled patterns in 45 (27%) and 40 (24%) cases respectively. Antibodies to dsDNA were detected in 9 patients (5% of the cases). Anti-ENA antibodies were documented in 46 patients (27%), with the most frequent pattern being anti-SSA/Ro (14%). Other immunological parameters, such as RF, a-CL and ANCA were detected in only a minority of patients.

Clinical and serological features after 5 years of follow-up
During the 5-year follow-up period, 10 out of 165 patients developed a defined autoimmune disease. The clinical and serological features of the remaining 155 cases were similar to those of the original group and remained quite stable during the 5 year follow-up period (Figs. 1 and 2). Nineteen of them were and remained asymptomatic.
Clinically we detected a slight increase in the number of cases with exocrine gland (9 pts.), articular (6 pts.), vascular (4 new cases of RP), haemopoietic (8 pts.) and gastrointestinal involvement (4 cases with oesophageal dysfunction). We also evaluated signs of permanent damage and found two cases with a new onset psychosis, one with seizures lasting

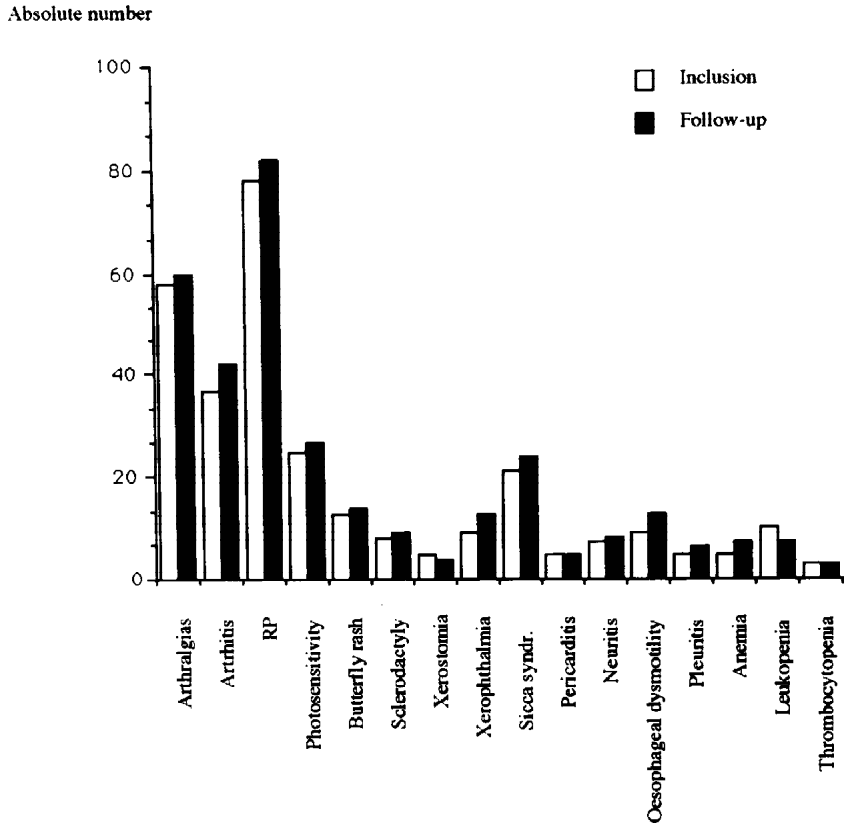

Fig. 1. Clinical features at inclusion and after the five-year follow-up in 155 UCTD patients.

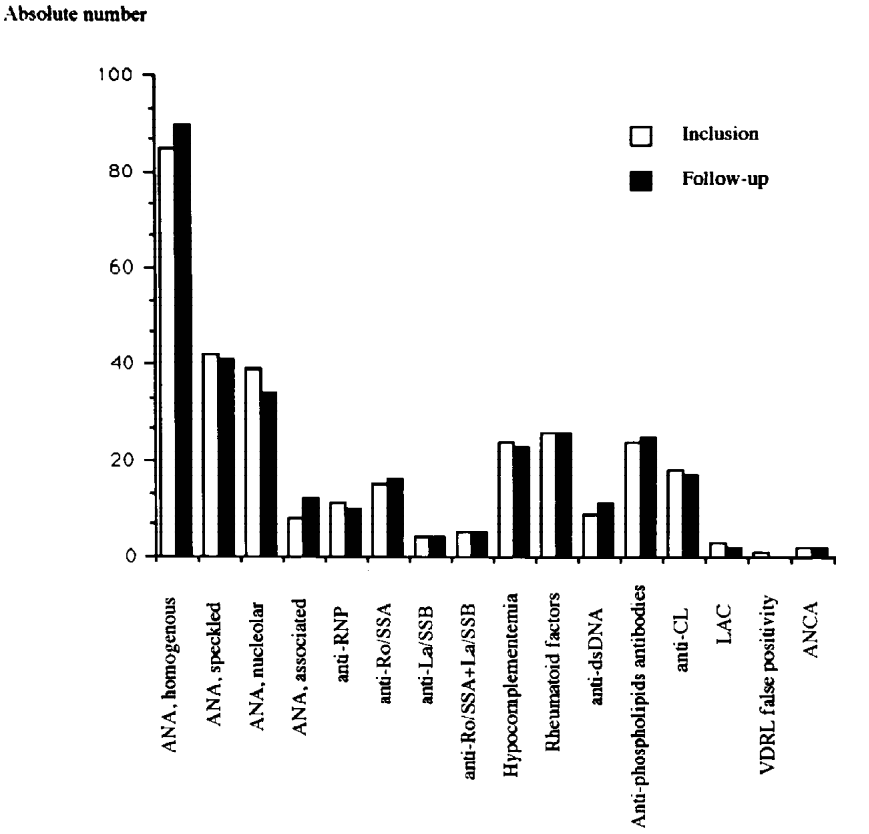

Fig. 2. Immunologic parameters at inclusion and after the five-year follow-up in 155 UCTD patients (data provided by the references centre).

more than 6 months and requiring specific therapy, one with lung fibrosis and six cases of arthritis with initial erosions. No significant variations were observed in the serological patterns with respect to the onset (Fig. 2).

When the correlations between clinical and immunological signs were studied, it was found that among 57 patients with arthralgias, 35 (60%) presented ANA positivity with a homogeneous pattern and 28 (48%) ANA positivity with a speckled pattern. In the patients with arthritis these percentages were lower. RF was present in 17 patients (29%) with arthralgias and only in 7 (19%) with arthritis. RP was associated with ANA (22/78 with a homogeneous, 20/78 with a speckled and 4/78 with a nucleolar pattern, respectively). Anti-SSB/La antibodies (either isolated or associated with SSA/Ro) were positive in 2 and 8 cases, respectively, and showed a poor correlation with sicca syndrome. Anti-cardiolipin antibodies were detected in 18 patients (11%), whereas among 5 patients with thrombotic events (2 with cerebrovascular accident and 3 with peripheral arterial/venous occlusion) none presented anti-phospholipid immunity.

Patients received standard symptomatic treatments for their articular complaints and RP; in only a few cases was immunosuppressive therapy employed.

Evolution of UCTDs after a 5 year follow-up period (Table II)

At the end of the follow-up period, 10 out of the 165 cases had evolved into a clearly defined autoimmune disease. SLE in 5 patients (in one associated with APS), SS in 4 patients, and DM-PM and MCTD in one case each, were diagnosed following established criteria (Table II). In four patients the diagnosis was confirmed following the manifestation of a striking clinical feature (sicca syndrome in three cases and arthritis and pleuropericarditis in one). In one patient the detection of anti-dsDNA antibodies led to a diagnosis of SLE, while in another the finding of a-CL led to the diagnosis of SLE/APS. In the remaining four cases the diagnosis of a defined CTD was based on the association of both clinical and immunological parameters (see Table II for details).

Table II. Main clinical and serological features of the 10 patients who developed a defined autoimmune disease after a 5-year follow-up.

Pt.	Sex/age at inclusion	Clinical and serological features at inclusion	Clinical and serological features emerging during the follow-up	Evolution into defined CTD	Evolution time (mos.)
1	F / 64	RP, sclerodactyly, leucopenia, thrombocytopenia, RF	Arthralgias, sicca syndrome	Sjögren's	26
2	F / 38	Photosensitivity*, butterfly rash*, xerophtalmia, anti-SSA/Ro, anti-SSB/La, Coombs +ve	Xerostomia	Sjögren's	24
3	F / 32	RP, oral erosion and aphthae, anti-SSA/Ro, anti-SSB/La, RF	Sicca syndrome	Sjögren's	9
4	F / 40	Arthralgias, photosensitivity*, leucopenia*, ANA/ homogeneous pattern, RF	Arthritis, sicca syndrome, parotid gland swelling, anti-SSA/Ro antibodies	SLE/ Sjögren's	34
5	F / 50	Arthralgias, pericarditis, ANA/homogoneus pattern	Photosensitivity, butterfly rash, anti-dsDNA antibodies	SLE	25
6	F / 38	arthralgias, RP photosensitivity, livedo reticularis, ANA/ speckled pattern	Butterfly rash, xerostomia, anti-dsDNA antibodies	SLE	7
7	F / 64	Photosensitivity, pleuritis, peri-myocarditis, RF	Arthritis, sicca syndrome, ANA/homogeneous pattern, anti-dsDNA antibodies,	SLE	12
8	M/ 30	Peripheral arterial occlusions, pleuritis, ANA/homogenous	Arthritis, a-CL	SLE-APS	12
9	M / 29	Arthralgias, RP, sclerodactyly, leucopenia	Arthritis, pleuritis, pericarditis, anti-RNP antibodies	MCTD	3
10	F / 31	RP, sclerodactyly, RF	Myositis, anti-Jo 1 antibodies	PM	12

*data recorded by the medical history.

Due to the small number of cases which evolved into a major disease, it was not possible to perform a statistical analysis on the predictive value of the different clinical and immunological parameters studied.

Discussion

In recent years there has been growing concern regarding the diagnosis of incomplete forms of the autoimmune diseases. While the clinical and serological manifestations present can lead to a specific diagnosis in most cases, sometimes the patient's symptoms and signs do not fulfill all of the established criteria. Moreover, there remain many situations in which only a few clinical or serological manifestations are present, and where objective criteria for the diagnosis are lacking. In such cases, there is no general agreement on the nature and denomination of the condition. For the connective tissue diseases the terms undifferentiated CTD and early UCTD have been used by different authors (7-10).

We report here our analysis of 165 patients from 10 different clinics, mainly the Internal Medicine Departments of university hospitals (i.e., tertiary referral centres) in Italy who were diagnosed as having a UCTD and who were followed for 5 years.

Like many other autoimmune diseases, UCTD affects predominantly females (12: 1 over males), generally in their fourth decade of life. In our series the most common findings of UCTD included articular and mucocutaneous features and RP, in agreement with other reports (9, 21-26). Despite the documented lower incidence of lung, kidney and heart involvement in the UCTDs compared to the defined connective tissue diseases, we found a relatively high prevalence of permanent major organ damage.

Other reports have focused their attention on the UCTDs. Alarcón *et al*. studied 213 patients with early UCTD, diagnosed by the presence of: 1) isolated keratoconjunctivitis sicca; 2) unexplained polyarthritis; and 3) at least three selected clinical and laboratory criteria (9). In this group, patients with MCTD were also enrolled. After a 5-year follow-up the authors could identify some clinical and laboratory features which were predictive of the subsequent development of a defined CTD (22-24). Mosca *et al*. evaluated the clinical and serological profiles of 91 UCTD patients who presented at least one clinical sign suggestive of CTD and one non-organ-specific autoantibody, and followed them for at least 1 year (25).

In a previous work, some of the present authors described the natural history and evolution of 84 patients initially diagnosed as having early UCTD according to the definition of Alarcón *et al*. (26). Multivariate analysis allowed us to select those variables correlating with the evolution into a particular CTD. For the present multicentre study, we enrolled patients with signs and symptoms of proven or hypothesised autoimmune aetiology that could be identified as a clear autoimmune entity. The discrepancies to be found in the literature reflect the different inclusion (classification and/or

diagnosis) criteria used and underlines once again the necessity of reaching a general agreement in this field. A given symptom or sign, even if relatively specific for autoimmune disease, does not always represent a homogeneous feature and can have a different meaning in various clinical settings.

For example, the overall prevalence and related specificity of ANA in our cohort of patients is analogous to previous series on autoimmune rheumatic diseases, mainly UCTD (21, 27). Nevertheless, we documented some conflicting correlations between serologic abnormalities, the actual clinical features and the subsequent disease evolution. RF positivity did not correlate with the presence of joint involvement and anti-SSA/Ro and anti-SSB/La antibody positivity did not correlate with the presence of sicca syndrome. We found a slight association between articular involvement and RF, which was present in 29% of the patients with arthralgias and only in 19% of those with evident arthritis. Anti-SSB/La antibodies, either alone or in association with SSA/Ro, were found in only a minority of cases (10 of the original group of 165 patients). This finding is in accordance with some reports (21, 28), but is at variance with some other studies. Gerli *et al*. demonstrated that anti-SSB/La antibodies correlate significantly with the degree of inflammatory infiltration of the salivary glands in SS (29). Mosca *et al*. confirmed in their work that anti-SSA/Ro was correlated with xerostomia and xerophthalmia (25). Thus, the various immunologic specificities detected in our patients suggests either that the syndrome comprises different forms of early CTD or that only an association among different specificities is indicative of a given disease. Clearly, it is important to use validated and standardised techniques for the serological analyses in patients with UCTDs. We detected an overall discordance of 15% in the immunologic determinations between individuals and reference centres (data not shown).

During the five-year follow-up period, 10 of the 165 (6%) patients studied developed a well-established autoimmune disease, SLE (5 cases, in one associated with APS), SS (3 cases), MCTD and PM (1 case each). These cases were reviewed to determine whether an earlier diagnosis might have been made, but it was confirmed that only the appearance of new clinical or immunologic signs allowed the final diagnosis. This further emphasises the fact that autoimmune diseases, in particular the CTDs, present distinctive features only when fully developed, whereas at the onset the immunologic abnormalities often present a non-specific and elusive clinical picture.

Gladman *et al.* described a group of patients with features suggestive of SLE, not fulfilling the ACR classification criteria and whose condition she defined as "latent lupus". Thirty percent of these patients developed full blown SLE during the five-year follow-up period. There were no clinical or laboratory features which distinguished this subgroup from the remaining cases with persistent latent lupus, however (30).

Other studies have shown that about one-third of patients with RP and UCTD will develop a well-established CTD, mainly a SSc-CREST, within 2 years (31-33). Presumably the patients included in these series represented cases of RP associated with clinical or laboratory features relatively specific for CTD. When considering patients with true primary RP, i.e. without features of latent systemic involvement, this percentage falls to 5%, which is consistent with the data of others (31) as well as that presented here. Concerning RA, 39 of our patients had arthritis at the onset, but only one went on to develop a full blown MCTD. Thus, our patients are not similar to those described by Alarcón *et al.* (23), in 20% of whom the disease evolved to RA within five years. Even in our one case, the discrepancy could be ascribed to differences in the inclusion criteria and the length of the follow-up period.

Which variables might correlate with the evolution into a defined autoimmune disease could not be determined due to the insufficient number of cases. However, patients with a constellation of signs and symptoms suggestive of an autoimmune origin should be reassured as to the very low possibility of their disorder eventually developing into a major disease.

At the end of the 5-year follow-up period, 19/155 (12%) patients remained asymptomatic. This underlines the fact that the UCTDs can be considered as a milder form or an early phase of classic autoimmune diseases, primarily of the rheumatic type, with low rate of evolution into a defined autoimmune disease. Further studies will help us to create a scoring system, based on clinical and laboratory parameters, for the clinical evaluation, including the possible outcome, of patients with UCTDs. Such a screening tool would allow physicians to distinguish patients with undefined forms of CTD from those those with pre-CTD (or latent CTD), since a higher percentage of patients with latent or evident major organ involvement will develop a CTD within a relatively short period of time. As long as the pathogenesis of the autoimmune diseases remains unknown, and specific methods to identify susceptible subjects are not available, an epidemiologic approach remains the only possible route for risk estimation.

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