Commentary
The RNA interference pathway: a new target for autoimmunity

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Abstract

Many intracellular macromolecular complexes that are involved in the production or degradation of RNAs are targeted by auto-antibodies in systemic autoimmune diseases. RNA interference (RNAi) is a recently characterized gene silencing pathway by which specific mRNAs are either degraded or translationally suppressed. In a recent issue of *Arthritis Research and Therapy*, Andrew Jakymiw and colleagues reported that the enigmatic Su auto-antigen complex contains key components of the RNAi machinery. Anti-Su autoantibodies from both human patients with rheumatic diseases and a mouse model of autoimmunity recognize the endo-nucleolytic Argonaute and Dicer proteins, both crucial enzymes of the RNAi pathway. These data raise the question of how the anti-Su response is triggered. So far, it is unknown whether molecular modifications may be involved, as has been proposed for other intracellular autoantigens. The implication of RNAi in anti-viral defence may suggest a role for virus infection in this process.

Many key regulators of gene expression were previously shown to be targeted by the immune system in a variety of autoimmune diseases. Patients with systemic autoimmune diseases commonly produce antibodies against specific classes of evolutionarily conserved nucleic acid-protein complexes. Most frequently, the targeted proteins are either RNA-binding themselves or associated with RNA-binding proteins, rather than proteins associated with DNA. In many cases, the autoantibodies were used as a tool for the identification of the corresponding autoantigens and to characterize their structure and function [1]. Frequently, the prototypical autoantigens were designated with the first two characters of the patient's name in which autoantibodies against them were first detected. Examples are Sm, Ro, La, and Th/To, which represent autoantigenic targets associated with spliceosomal U small nuclear (sn) ribonucleoprotein particles (RNPs), the Y RNA-containing Ro RNPs, the RNA-binding La protein, and the RNase MRP/RNase P endo-ribonucleases, respectively. These ribonucleoprotein particles accumulate in different subcellular compartments and play important roles in a variety of RNA metabolic processes (Table 1) [2]. The recent work by Jakymiw and colleagues [3] shows that components of the RNA interference machinery are also recognized by autoantibodies, referred to as anti-Su antibodies.

RNA interference (RNAi) is a recently identified mechanism by which gene expression can be controlled post-transcriptionally [4]. RNAi is mediated by small RNA molecules, 21 to 25 nucleotides in length, that guide protein complexes to complementary mRNA targets, whose expression is then silenced. These small RNAs can be either small interfering RNAs (siRNAs) or microRNAs (miRNAs), which derive from long double-stranded (ds)RNA and from stem-loop structures in long, largely unstructured transcripts, respectively. The production and function of siRNAs and miRNAs requires a common set of proteins, including a protein called Dicer, which acts as a dsRNA-specific endonuclease and is involved in the maturation of these RNAs, and the family of Argonaute (Ago) proteins, small RNA-binding, nucleolytic enzymes mediating the degradation of the targeted mRNA. Ago and the siRNA or miRNA form the core of the RNA-induced silencing complex (RISC) [5].

The molecular identity and biological function of the Su autoantigen remained elusive for many years. A protein of about 100 kDa co-immunoprecipitating with GW182, a recently identified protein required for RNAi, was hypothe-sized by Jakymiw and colleagues [3] to be the Su auto-antigen, which was known to migrate as a 100/102 kDa doublet in SDS-PAGE [6]. Using a combination of immuno-cytochemical and immunoprecipitation experiments, they demonstrated that the Su autoantigen represents a macromolecular complex associated with GW182 and the RNAi pathway. The anti-Su autoimmune sera recognize

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Ago = Argonaute; ds = double-stranded; miRNA = microRNA; RISC = RNA-induced silencing complex; RNAi = RNA interference; siRNA = small interfering RNA; RNP = ribonucleoprotein particle.

Table 1

Examples of autoantigens involved in RNA metabolic processes

Autoantigen	Ribonucleoprotein complex	Target protein(s)	Subcellular localization	Process
Sm	U snRNP	Sm proteins	Nucleoplasm	Pre-mRNA splicing
Ro	Y RNP	Ro60	Cytoplasm	RNA quality control
La	La RNP	La protein	Nucleoplasm	Maturation RNA Pol III transcripts
Th/To	RNase MRP, RNase P	Rpp38, Rpp25, Rpp20	Nucleoli	Pre-rRNA processing
Su	RISC	Ago, Dicer	Cytoplasm	RNAi

RISC = RNA-induced silencing complex; RNP = ribonucleoprotein particle.

several members of the Ago protein family, which are known to associate into RISC and which have a high degree of sequence identity. Targeting of the RNAi machinery by anti-Su autoimmune sera was further supported by the identification of a 200 kDa protein associated with the Su antigenic complex as Dicer, the enzyme that converts dsRNA precursors into functional siRNAs and miRNAs [5].

The disease specificity of anti-Su antibodies has been addressed in only a few studies so far. Although they were initially reported to be specifically associated with systemic lupus erythematosus [7], a later study showed that auto-antibodies to the Su antigen are found in a variety of systemic rheumatic diseases [6]. The elucidation of the molecular identity of the Su antigen will facilitate the determination of the diagnostic and/or prognostic value of anti-Su antibodies.

Why are so many protein and RNA-protein complexes that are involved in the synthesis, processing and degradation of various classes of RNA targeted by the immune system in systemic autoimmune diseases? One explanation might be that under certain circumstances (e.g., during cell death) relatively large amounts of these key components of RNA metabolic processes are chemically modified, escape from the cell (e.g., during necrosis) and are exposed to the immune system. The molecular modifications may be recognized as non-self and elicit a primary immune response in individuals with a 'proper' genetic context. Environmental factors like trauma, drugs, irradiation or viruses may be responsible for a massive induction of cell death, either by apoptosis or (secondary) necrosis. Especially during programmed cell death, key regulators of gene expression are inactivated by proteolytic cleavage or other types of modifications [8,9], most likely to prevent them from counteracting the execution of cell death. Via intra- and intermolecular epitope spreading, a primary immune response targeting the modified antigen may spread to genuine autoantigenic epitopes, the recognition of which does not require molecular modifications. The diversity of autoantibody specificities and their association with particular diseases may be related to the combination of genetic and environmental factors that are involved. This hypothesis is supported by several studies demonstrating

that the molecular modifications are essential for the early recognition by the immune system and that epitope spreading with autoantigenic proteins/complexes occurs in mammals [10-12].

This raises the question of whether a similar mechanism may be involved in the anti-Su autoimmune response. Future studies will have to clarify whether one or more of the Ago and Dicer proteins undergo molecular changes in dying cells and, if so, whether the resulting neo-epitopes are recognized by early anti-Su sera.

Virus infections have been long associated with autoimmune diseases and various mechanisms implicating viruses in the etiology of these diseases have been proposed [13]. An intriguing aspect of autoimmune targeting of the RNAi machinery is that RNAi was initially recognized as an antiviral mechanism in plants and certain invertebrates [14]. The evolutionary conservation of RNAi suggests a similar role for RNAi in mammals, which is indeed supported by the production of suppressors of RNAi by mammalian viruses [15]. Based upon these phenomena and additional evidence for the linkage of RNAi to virus-induced cellular events, Jakymiw and colleagues [3] argue that it is not surprising that the components of the RNAi machinery develop into targets of autoimmunity. On the one hand, the induction of apoptosis as a result of the infection may lead to molecular modification of host components, including Ago and/or Dicer. On the other hand, the association of virus-like particles with components of the RNAi machinery [16] may promote the development of autoimmunity.

Conclusion

The identification of components of the RNAi machinery as targets of the anti-Su autoantibody system provides another example of an autoimmune response directed at a macromolecular complex that plays a key role in the post-transcriptional regulation of gene expression. Future studies will have to reveal how the generation of anti-Su autoantibodies is initiated, whether a viral factor is involved in this process, and whether testing for these autoantibodies is clinically relevant.

Competing interests

Acknowledgements

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