行政院國家科學委員會專題研究計畫　期中進度報告

子計畫一：自體免疫疾病中蛋白質精胺酸甲基化之蛋白質體研究(1/3)

計畫類別：整合型計畫
計畫編號：NSC93-2745-B-040-002-URD
執行期間：93 年 08 月 01 日至 94 年 07 月 31 日
執行單位：中山醫學大學生物醫學科學學系

計畫主持人：李娟

計畫參與人員：張宏豪、洪銓錨、李宜娟、洪健仁等

報告類型：精簡報告

處理方式：本計畫可公開查詢

中　華　民　國 94 年 5 月 27 日

行政院國家科學委員會補助專題研究計畫 □ 成果報告 □期中進度報告

自體免疫疾病中蛋白質精胺酸甲基化之蛋白質體研究(1/3)

Proteomic analyses of methylarginine modification in autoimmune diseases

計畫類別：□ 個別型計畫 ■ 整合型計畫
計畫編號：NSC93-2745-B-040-002-URD
執行期間： 93 年 8 月 1 日至 96 年 7 月 31 日
計畫主持人：李娟
計畫參與人員： 張宏豪、洪銓錨、李宜娟、洪健仁等

成果報告類型(依經費核定清單規定繳交)：■精簡報告 □完整報告

本成果報告包括以下應繳交之附件：
□赴國外出差或研習心得報告一份
□赴大陸地區出差或研習心得報告一份
□出席國際學術會議心得報告及發表之論文各一份
□國際合作研究計畫國外研究報告書一份

處理方式：除產學合作研究計畫、提升產業技術及人才培育研究計畫、列管計畫及下列情形者外，得立即公開查詢
□涉及專利或其他智慧財產權，□一年□二年後可公開查詢

執行單位：中山醫學大學生物醫學科學學系

一、 摘要

在此研究計劃中，我們想要探討蛋白質精胺酸甲基化(分為對稱型或不對稱型双甲基精胺酸)和自體免疫疾病的關係。許多蛋白質包含了甲基精胺酸，如：fibrillarin、部分的hnRNP 蛋白、myelin basic protein 以及SmD1、D3 蛋白，而這些蛋白也已被發現是許多自體免疫疾病的自體免疫抗原。在這個計劃中，我們首先從紅斑性狼瘡病人中取得anti-Sm 和 anti-RNP 自體免疫抗體血清及獲取其他的免疫抗體血清，尋找抗血清中是否有區分蛋白質精胺酸甲基化之抗體，再利用蛋白質體學方法找出有甲基化自體免疫抗原。我們在一位病人血清作西方點墨法時，發現低甲基化之細胞萃取蛋白和正常甲基化組比較，有數調訊號減低情況。我們將相關蛋白點挖出，以蛋白脢水解後作質譜分析，有二蛋白質含有典型精胺酸甲基化蛋白質中出現之 RGG 序列，是否此自體免疫抗體所辨識的自體免疫抗原被辨認的位置與精胺酸甲基化有關正在研究中。

Abstract

In this project we are going to find out the relationship between protein methylation, mostly protein *N*-arginine methylation to form symmetric or asymmetric N^G, N^G –dimethylarginines (sDMA or aDMA), and autoimmune disease. Many methylarginine containing proteins such as fibrillarin, several hnRNP, myelin basic protein and SmD1 and D3 are known to be autoantigens of different autoimmune diseases. Sm protein D1 and D3 were reported to contain symmetric di-methylarginines and a few different anti-Sm autoantisera recognized only the sDMA peptide of SmD1 and D3 but not unmethylated or asymmetric dimethylarginine peptides. We thus examined if the anti-Sm sera from local SLE patients also preferentially recognize the methyl-modified Sm D proteins and if there are other proteins that can be differentially recognized by the anti-Sm sera due to their methylation states. We treated HeLa cells with adenosine dialdehyde (AdOx), an inhibitor of protein methylation. Anti-Sm autosera from three different SLE patients were used in western blot analyses of AdOx-treated (proteins presumably at hypomethylation state) and untreated (proteins at normal methylation states) HeLa cell extracts. Reduced signals between molecular mass of 18 to 21 kDa and about 31 kDa were consistently detected from cell extracts treated with AdOx compared to the ones without AdOx treatment by one-dimensional SDS-PAGE analyses. However, there were no significant differences between the signals corresponding to SmD1 in samples of different methylation status. By two-dimensional electrophoresis, the differentially detected signals were further pinpointed and putative spots were picked by comparing the western signals with the protein sypro ruby stain, and digested by trypsin. The peptide fragments were analyzed by MS and MS/MS analyses. Interestingly, two polypeptides contain typical arginine and glycine (RGG) sequences of the arginine

methyltransferase substrates are identified. The confirmation of the proteins differentially detected by anti-Sm due to their methylation status are under investigation.

二、 研究目的與背景

A common feature of autoimmune diseases such as systemic lupus erythematosus (SLE), systemic sclerosis, Sjögren’s disease (SD), and mixed connective tissue disease is the breakdown of tolerance to self antigens, leading to the production of antibodies reactive with multiple self proteins. Recently, the possibility of posttranslational modification to create new self antigens or mask antigens normally recognized by the immune system has been discussed (Utz et al., 2000; Doyle and Mamula, 2001; Doyle and Mamula 2002). Neo-Ags originating from these modified proteins might find their way into the extracellular milieu where they can be taken up by Ag-processing cells and presented to T cells. In experimental systems, this mechanism can initiate an immune response to a protein that is ignored otherwise. Recently, studies have shown that self proteins are cleaved differently in apoptotic cells versus nonapoptotic cells, thus creating new autoAgs. Cells undergoing apoptosis are also subject to a number of protein modifications including, phosphorylation, transglutamination, ubiquitination and citrullination (Utz et al., 2000; Zampieri et al., 2001). Apoptotic cells have been considered a source of autoantigens in the induction of SLE (Utz et al., 1997;). Even though apoptotic cells remain intact, certain antigens that are common targets of autoantibody responses in SLE patients localize to surface blebs of apoptotic cells (Casciola-Rosen et al., 1994) and mice immunized with apoptotic Jurkat cells develop antibodies to multiple autoantigens and autoantigen complexes associated with SLE (Gensler et al., 2001). It is reasonable to speculate that these proteins on the surface of apoptotic cells have undergone post-translational modifications that affect the recognition and processing of Ag thus leading to an immune response.

Protein *N*-arginine methylation is an irreversible modification on the guanidino nitrogen of the arginyl residues that accounts for the majority of stable protein methylation events in cells (Li et al., 1998; Najbauer et al., 1993). Most of the methylarginines in the proteins identified appear to be N^G–monomethylarginine (MMA) and asymmetric N^G, N^G –dimethylarginines (aDMA) in various RNA binding proteins within the Arg-Gly-Gly context (Najbauer et al., 1993). These modifications were catalyzed by the type I arginine methyltransferase (Gary and Clarke, 1998). Another type II methyltransferase helps to modify some other methylaccepting proteins such as myelin basic protein (Baldwin and Carnegie, 1971) and core snRNP SmD1, D3 (Brahms et al, 2000), B/B' and one of the Sm-like proteins LSm4 (Brahms et al., 2001) to form MMA and symmetric N^G, N'^G–dimethylarginine (sDMA).

The type I enzymes include PRMT1 that appears to be the predominant methyltransferase in the mammalian system (Lin et al., 1996, Tang et al., 2000; Pawlak et

al., 2000), the zinc-finger containing PRMT3 (Tang et al., 1998), the coactivator-associated arginine methyltransferase CARM1/PRMT4 (Chen et al., 1999) and the newly identified PRMT6 (Frankel et al., 2002). Another PRMT-1-like PRMT gene (HRMT1L3,) with 80% sequence identity with PRMT1 was identified in human and vertebrates but with no reported function yet (Zhang and Cheng, 2003, Hung and Li, 2004). The only type II PRMT identified is the Janus kinase-binding protein JBP1/PRMT5 (Pollack et al., 1999; Roe et al., 2001; Branscombe et al., 2001), which was first identified in *Schizosaccharomyces pombe* as an Shk1 kinase binding protein skb1 (Gilbreth et al., 1996) and a *Saccharomyces seveviciae* histone synthetic lethal gene hsl7 (Ma et al., 1996). Another recently identified PRMT7 gene p82/p77 is a genetic suppressor element mediates cell sensitivity to DNA-damaging agents. Evidence of in vitro methylation of MBP, a typical type-II substrate, had been demonstrated for the p82/p77 immunoprecipitants (Gros et al., 2003). In yeast *S. seveviciae*, RMT1/HMT1 corresponds to the type I methyltransferase activity (Gary et al., 1996; Henry and Silver, 1996) while Hsl7p is responsible for the type II activity (Lee et al., 2000). The survival of yeast HMT1 or Hsl7 mutants indicates that they are not essential genes.

Interestingly, many type I or type II methylaccepting proteins such as fibrillarin, several hnRNPs, myelin basic protein and SmD1 and D3 are known to be autoantigens of different autoimmune diseases (please see the following table and Boekel and Venrooij, 2002).

Autoantigen/ arginine methyl accepting protein	autoimmune disease	methylarginine
Fibrillarin	scleroderma	aDMA
Nucleolin	SLE (systematic lupus erythematosus)	aDMA
HnRNPA1	connective tissue disease (SLE, RA, MCTD)	aDMA
Sm D1, D	SLE	sDMA
Myelin basic protein	multiple sclerosis	sDMA

Ten different autoantisera recognize only the sDMA peptide of SmD1 and D3 but not unmethylated or aDMA peptides (Brams et al., 2000), indicating methylarginine modification can be important for autoantibody recognition. Furthermore, peptides with aDMA modification were identified as natural MHC class I ligand, indicating that specific cytotoxic T-cell response against cells presenting aDMA modified cells can be elicited (Yague et al., 2000). Whether abnormal arginine methylation can be correlated with the formation of autoimmune disease worth further investigation. In this project we are going to investigate the involvement of type I and type II protein arginine methylation in autoimmune disease, most specifically SLE. Even though by now there were no reports on type I modified aDMA residues as specific sites for autoantibody recognition, it is rather widely distributed in known autoantiagens involved in a wide spectrum of autoimmune disease. Different from phosphorylation that is irreversible, arginine protein methylation is

irreversible and most likely to occur soon after protein synthesis, thus it probably can be viewed as a normal process of protein maturation. However, involvement of protein arginine methylation with signal transduction has been suggested. How specific arginine modification in the nuclear protein such as Sm D1 will elicit the production of specific autoantibodies is an interesting question. Moreover, the arginyl residues not only can be modified by methylation, in some autoantigens they undergo citrullination by peptidylarginine deiminase (Moscarello, 2002). In multiple sclerosis, MBP methylation has been reported to reduce arginine deimination, which has been directly related to the disease. Citrulination of fillagrin has been shown to be the key factor in rheumatoid arthritis (Girbal-Neuhauser et al., 1999). Filaggrin is a large protein with mutiple RGGR sequences dispersed in its 3870 amino acid residues (Gan et al., 1990). It is interesting whether this protein can be methylated, and whether SmD with sDMA modification might undergo citrullination in some fraction. We will cooperate with Project 4 on this issue.

Finally, posttranslationally modified self-antigens may play a role in regulating autoimmune disease through suppression of autoreactive T cells. St. Louis *et al.* found that a palmitoylated PLP peptide (139-151) completely suppressed, or considerably reduced the acute and chronic relapsing stages of EAE in SJL mice. Adoptive transfer of lymphocytes from mice treated with the palmitoylated PLP peptide also delayed the onset of disease in recipient mice. These studies offer the intriguing idea that posttranslationally modified peptides can act as altered peptide ligands, and thus may be useful in the treatment of autoimmune diseases (Louis et al., 2001).

三、 結果與討論

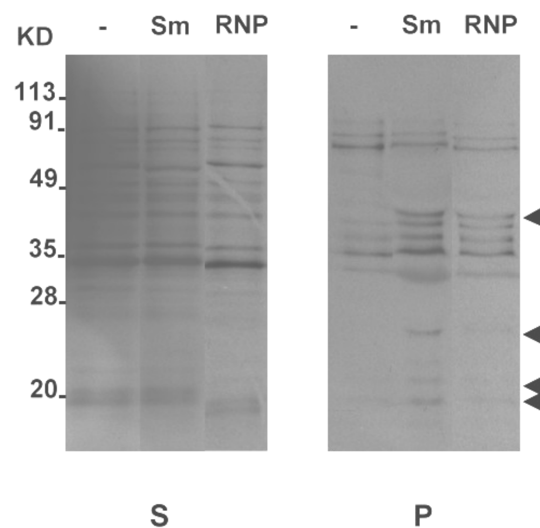

Fig. 1. Immunoprecipitation of in vitro methylated proteins by anti-Sm or anti-RNP autoantibodies. Lymphoblastoid cell extracts (containing 100 g of total protein) prepared from cells grown in the presence of 20 M of AdOx for 72 h were methylated *in vitro* with 0.75 µCi of [^{3}H]-SAM for 1 h at 37C. Anti-Sm or anti-RNP antisera from autoimmune patients were incubated with protein A/G-agarose beads (1:2) overnight at 4℃. The in vitro methylated proteins were then incubated with the agarose beads incubated with anti-Sm, anti-RNP or no antisera (-). The methylated proteins were separated by SDS-PAGE and visualized by fluorography.
Left panel: the supernatant after immunoprecipitation. Right panel: the immunoprecipitated proteins.

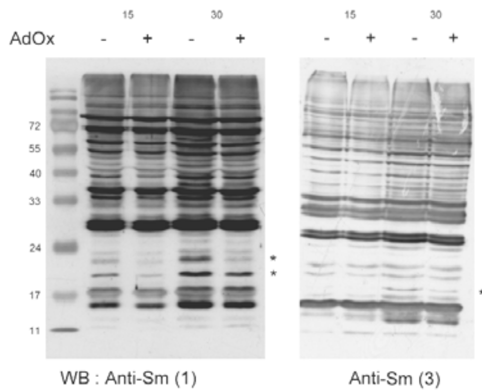

Fig. 2. Differential recognition of proteins due to methylation status by anti-Sm autoantibodies. HeLa cell extracts (containing 20 g of total protein) were prepared from cells grown in the presence or absence of 20 M of AdOx for 24 h. The cell extracts were separated by SDS-PAGE and transferred onto NC membrane. Western blot analysis was performed using SYM 11 Anti-dimethyl-Arginine, symmetric) for control (a), anti-Sm autoantibody from SLE patient 1(b), anti-Sm autoantibody from SLE patient 3 (c), andanti-RNP autoantibody from SLE patient 5 (d).

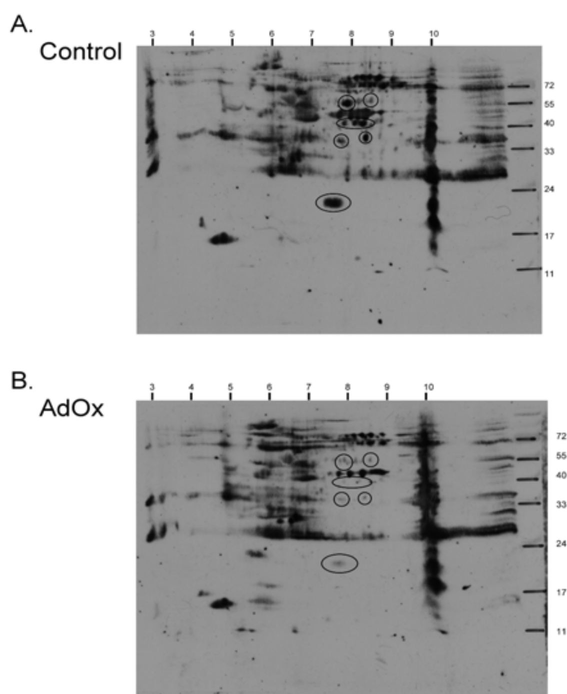

Fig. 3. Differential recognition of proteins due to methylation status by anti-Sm autoantibodies analyzed by two-dimensional electrophoresis. HeLa cell extracts (containing 250 g of total protein) were prepared from cells grown in the absence (A) or presence (B) of 20 M of AdOx for 24 h. The cell extracts were separated by SDS-PAGE and transferred onto NC membrane. Western blot analysis was performed anti-Sm autoantibody from SLE patient 1. The signals that are differentially recognized by the antisera were circled.

四、 成果自評

In the first nine months of the three-year project, we are glad that we were able to obtain some interesting results to contribute the connection of autoimmune disease SLE with protein methylation, mainly arginine methylation. We have identified two putative protein-arginine methyl-accepting proteins that have not been reported to be autoantigens or protein-arginine methyl-accepting protein. The results also indicate that the system is well-functioning and we will apply it to the studies of other autoimmune diseases.

五、 參考文獻

Abramovich C, Yakobson B, Chebath J, Revel M. A protein-arginine methyltransferase binds to the intracytoplasmic domain of the IFNAR1 chain in the type I interferon receptor. EMBO J. 1997 Jan 15;16(2):260-6.

Ai LS, Lin CH, Hsieh M, Li C. Arginine methylation of a glycine and arginine rich peptide derived from sequences of human FMRP and fibrillarin. Proc Natl Sci Counc

Repub China B. 1999 Oct;23(4):175-80.
Aletta JM, Cimato TR, Ettinger MJ. Protein methylation: a signal event in post-translational modification. Trends Biochem Sci. 1998 Mar;23(3):89-91.
Baldwin GS, Carnegie PR. Specific enzymic methylation of an arginine in the experimental allergic encephalomyelitis protein from human myelin. Science. 1971 Feb 2;171(971):579-81.
van Boekel MA, van Venrooij WJ. Modifications of arginines and their role in autoimmunity. Autoimmun Rev. 2003 Mar;2(2):57-62.
Brahms H, Meheus L, de Brabandere V, Fischer U, Luhrmann R. Symmetrical dimethylation of arginine residues in spliceosomal Sm protein B/B' and the Sm-like protein LSm4, and their interaction with the SMN protein. RNA. 2001 Nov;7(11):1531-42.
Branscombe TL, Frankel A, Lee JH, Cook JR, Yang Z, Pestka S, Clarke S. PRMT5 (Janus kinase-binding protein 1) catalyzes the formation of symmetric dimethylarginine residues in proteins.J Biol Chem. 2001 Aug 31;276(35):32971-6.
Casciola-Rosen LA, Anhalt G, Rosen A: Autoantigens targeted in systemic lupus erythematosus are clustered in two populations of surface structures on apoptotic keratinocytes. J Exp Med 1994, 179:1317–1330.
Chen D, Ma H, Hong H, Koh SS, Huang SM, Schurter BT, Aswad DW, Stallcup MR. Regulation of transcription by a protein methyltransferase. Science. 1999 Jun 25;284(5423):2174-7.
Doyle HA, Mamula MJ. Post-translational protein modifications in antigen recognition and autoimmunity.Trends Immunol. 2001 Aug;22(8):443-9.
Frankel A, Yadav N, Lee J, Branscombe TL, Clarke S, Bedford MT. The novel human protein arginine N-methyltransferase PRMT6 is a nuclear enzyme displaying unique substrate specificity. J Biol Chem. 2002;277(5):3537-43.
Friesen WJ, Massenet S, Paushkin S, Wyce A, Dreyfuss G. SMN, the product of the spinal muscular atrophy gene, binds preferentially to dimethylarginine-containing protein targets. Mol Cell. 2001 May;7(5):1111-7.
Gahring LC, Carlson NG, Meyer EL, et al.: Granzyme B proteolysis of a neruonal glutamate receptor generates an autoantigen and is modulated by glycosylation. J Immunol 2001, 166:1433–1438.
Gan SQ, McBride OW, Idler WW, Nedialka M, Steinert PM. Organization, structure, and polymorphisms of the human profilaggrin gene. Biochemistry 1990;29:9423
Gary JD, Clarke S. RNA and protein interactions modulated by protein arginine methylation. Prog Nucleic Acid Res Mol Biol. 1998;61:65-131.
Gary JD, Lin WJ, Yang MC, Herschman HR, Clarke S. The predominant protein-arginine methyltransferase from Saccharomyces cerevisiae. J Biol Chem. 1996 May 24;271(21):12585-94.
Gensler TJ, Hottelet M, Zhang C, et al.: Monoclonal antibodies derived from BALB/C mice immunized with apoptotic Jurkat T cells recognize known autoantigens. J Autoimmun 2001, 16:59–69.
Girbal-Neuhauser E, Durieux J-J, Arnaud M, et al. The epitopes targeted by the rheumatoid arthritis-associated antifilaggrin autoantibodies are posttranslationally generated on various sites of (pro) filaggrin by deimination of

arginine residues. J Immunol 1999;162:585
Huang HM, Tam MF, Tam TC, Chen DH, Hsieh M, Li C. Proteomic analysis of stable protein methylation in lymphoblastoid cells. J Biochem (Tokyo). 2002 Nov;132(5):813-8.
Hung MC, Li C. Identification and phylogenetic analyses of the protein arginine methyltransferase gene family in *Fugu rubripes* and *Ciona intestinalis*: Biochem Biophy Res Commu (manuscript in preparation) 2004
Kaufmann H, Bailey JE, Fussenegger M. Use of antibodies for detection of phosphorylated proteins separated by two-dimensional gel electrophoresis. Proteomics. 2001 Feb;1(2):194-9.
Kim S, Merrill BM, Rajpurohit R, Kumar A, Stone KL, Papov VV, Schneiders JM, Szer W, Wilson SH, Paik WK, Williams KR. Identification of N(G)-methylarginine residues in human heterogeneous RNP protein A1: Phe/Gly-Gly-Gly-Arg-Gly-Gly-Gly/Phe is a preferred recognition motif. Biochemistry. 1997 Apr 29;36(17):5185-92.
Klein S, Carroll JA, Chen Y, Henry MF, Henry PA, Ortonowski IE, Pintucci G, Beavis RC, Burgess WH, Rifkin DB. Biochemical analysis of the arginine methylation of high molecular weight fibroblast growth factor-2. J Biol Chem. 2000 Feb 4;275(5):3150-7.
Kujubu DA, Stimmel JB, Law RE, Herschman HR, Clarke S. Early responses of PC-12 cells to NGF and EGF: effect of K252a and 5'-methylthioadenosine on gene expression and membrane protein methylation. J Neurosci Res. 1993 Sep 1;36(1):58-65.
Kzhyshkowska J, Schutt H, Liss M, Kremmer E, Stauber R, Wolf H, Dobner T. Heterogeneous nuclear ribonucleoprotein E1B-AP5 is methylated in its Arg- Gly-Gly (RGG) box and interacts with human arginine methyltransferase HRMT1L1. Biochem J. 2001 Sep 1;358(Pt 2):305-14.
Li C, Ai LS, Lin CH, Hsieh M, Li YC, Li SY. Protein N-arginine methylation in adenosine dialdehyde-treated lymphoblastoid cells. Arch Biochem Biophys. 1998 Mar 1;351(1):53-9.
Lin CH, Hsieh M, Li YC, Li SY, Pearson DL, Pollard KM, Li C. Protein N-arginine methylation in subcellular fractions of lymphoblastoid cells. J Biochem (Tokyo). 2000 Sep;128(3):493-8.
Lin CH, Huang HM, Hsieh M, Pollard KM, Li C. Arginine methylation of recombinant murine fibrillarin by protein arginine methyltransferase. J Protein Chem. 2002 Oct;21(7):447-53.
Lin WJ, Gary JD, Yang MC, Clarke S, Herschman HR. The mammalian immediate-early TIS21 protein and the leukemia-associated BTG1 protein interact with a protein-arginine N-methyltransferase. J Biol Chem. 1996 Jun 21;271(25):15034-44.
Lischwe MA, Cook RG, Ahn YS, Yeoman LC, Busch H. Clustering of glycine and NG,NG-dimethylarginine in nucleolar protein C23. Biochemistry. 1985 Oct 2;24(22):6025-8.
Lischwe MA, Ochs RL, Reddy R, Cook RG, Yeoman LC, Tan EM, Reichlin M, Busch H. Purification and partial characterization of a nucleolar scleroderma antigen (Mr = 34,000; pI, 8.5) rich in NG,NG-dimethylarginine. J Biol Chem. 1985 Nov 15;260(26):14304-10.
Liu Q, Dreyfuss G. In vivo and in vitro arginine methylation of RNA-binding proteins. Mol Cell Biol. 1995 May;15(5):2800-8.
Louis JS, Uniyal S, Xu L, et al.: Tolerance induction by acylated peptides:suppression of EAE in the mouse with palmitoylated PLP

peptides. J Neuroimmunol 2001, 115:79–90.
McBride AE, Silver PA. State of the arg: protein methylation at arginine comes of age. Cell. 2001 Jul 13;106(1):5-8.
Moscarello MA, Pritzker L, Mastronardi FG, Wood DD. Peptidylarginine deiminase: a candidate factor in demyelinating disease. J Neurochem 2002;81:335
Miklos GL, Maleszka R. Integrating molecular medicine with functional proteomics: realities and expectations. Proteomics. 2001 Jan;1(1):30-41.
Mowen KA, Tang J, Zhu W, Schurter BT, Shuai K, Herschman HR, David M. Arginine methylation of STAT1 modulates IFNalpha/beta-induced transcription. Cell. 2001 Mar 9;104(5):731-41.
Mamula MJ, Gee RJ, Elliott JI, Sette A, Southwood S, Jones PJ, Blier PR. Isoaspartyl post-translational modification triggers autoimmune responses to self-proteins.J Biol Chem. 1999 Aug 6;274(32):22321-7.
Najbauer J, Johnson BA, Young AL, Aswad DW. Peptides with sequences similar to glycine, arginine-rich motifs in proteins interacting with RNA are efficiently recognized by methyltransferase(s) modifying arginine in numerous proteins. J Biol Chem. 1993 May 15;268(14):10501-9.
Nichols RC, Wang XW, Tang J, Hamilton BJ, High FA, Herschman HR, Rigby WF. The RGG domain in hnRNP A2 affects subcellular localization. Exp Cell Res. 2000 May 1;256(2):522-32.
Nollau P, Mayer BJ. Profiling the global tyrosine phosphorylation state by Src homology 2 domain binding. Proc Natl Acad Sci U S A. 2001 Nov 20;98(24):13531-6.
Oh JM, Brichory F, Puravs E, Kuick R, Wood C, Rouillard JM, Tra J, Kardia S, Beer D, Hanash S. A database of protein expression in lung cancer. Proteomics. 2001 Oct;1(10):1303-19.
Pandey A, Mann M. Proteomics to study genes and genomes. Nature. 2000 Jun 15;405(6788):837-46.
Pellizzoni L, Baccon J, Charroux B, Dreyfuss G. The survival of motor neurons (SMN) protein interacts with the snoRNP proteins fibrillarin and GAR1. Curr Biol. 2001 Jul 24;11(14):1079-88.
Pellizzoni L, Charroux B, Dreyfuss G. SMN mutants of spinal muscular atrophy patients are defective in binding to snRNP proteins. Proc Natl Acad Sci U S A. 1999 Sep 28;96(20):11167-72.
Rajpurohit R, Lee SO, Park JO, Paik WK, Kim S. Enzymatic methylation of recombinant heterogeneous nuclear RNP protein A1. Dual substrate specificity for S-adenosylmethionine :histone-arginine N-methyltransferase. J Biol Chem. 1994a Jan 14;269(2):1075-82.
Scott HS, Antonarakis SE, Lalioti MD, Rossier C, Silver PA, Henry MF. Identification and characterization of two putative human arginine methyltransferases (HRMT1L1 and HRMT1L2). Genomics. 1998 Mar 15;48(3):330-40.
Schurter BT, Koh SS, Chen D, Bunick GJ, Harp JM, Hanson BL, Henschen-Edman A, Mackay DR, Stallcup MR, Aswad DW. Methylation of histone H3 by coactivator-associated arginine methyltransferase 1. Biochemistry. 2001 May 15;40(19):5747-56.
Tang J, Frankel A, Cook RJ, Kim S, Paik WK, Williams KR, Clarke S, Herschman HR. PRMT1 is the predominant type I protein arginine methyltransferase in mammalian cells.

J Biol Chem. 2000a Mar 17;275(11):7723-30.
Tang J, Kao PN, Herschman HR.
Protein-arginine methyltransferase I, the predominant protein-arginine methyltransferase in cells, interacts with and is regulated by interleukin enhancer-binding factor 3. J Biol Chem. 2000b Jun 30;275(26):19866-76.
Tang J, Gary JD, Clarke S, Herschman HR.
PRMT 3, a type I protein arginine N-methyltransferase that differs from PRMT1 in its oligomerization, subcellular localization, substrate specificity, and regulation. J Biol Chem. 1998 Jul 3;273(27):16935-45.
Utz PJ, Gensler TJ, Anderson P. Death, autoantigen modifications, and tolerance. Arthritis Res. 2000;2(2):101-14.
Utz PJ, Hottelet M, Schur PH, et al.: Proteins phosphorylated during stress induced apoptosis are common targets for autoantibody production in patients with systemic lupus erythematosus. J Exp Med 1997, 185:843–854.
Zampieri S, Degen W, Ghirardello A, et al.: Dephosphorylation of autoantigenic ribosomal P proteins during Fas-L induced apoptosis: a possible trigger for the development of the autoimmune response in patients with systemic lupus erythematosus. Ann Rheum Dis 2001, 60:72–76.