What is I M R D Anyway?

N	E	E	I
T	T	S	S
R	H	U	C
O	O	L	U
D	D	T	S
U	S	S	S
C			I
T			O
I			N
O			
N			

Susan Ruff

20.109

Spring 2007

Lab Reports (& Research Articles) tell a story.

INTRODUCTION What you studied, and why it's important.

METHODS How you studied it.

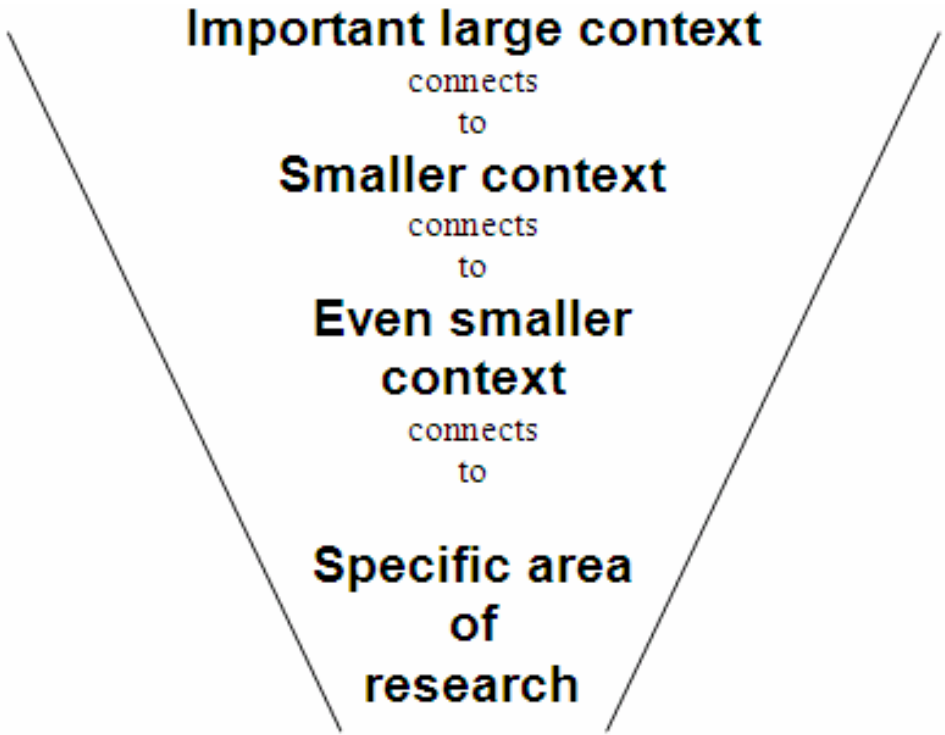
RESULTS What you saw.

DISCUSSION Your interpretation of what you saw.

All examples are taken from or based on "The Deubiquitylation Activity of Ubp8 Is Dependent upon Sgf11 and Its Association with the SAGA Complex" by Kenneth K. Lee, et al., *Molecular and Cellular Biology*, Feb. 2005, p. 1173-1182.

INTRODUCTION What you studied, and why it's important.

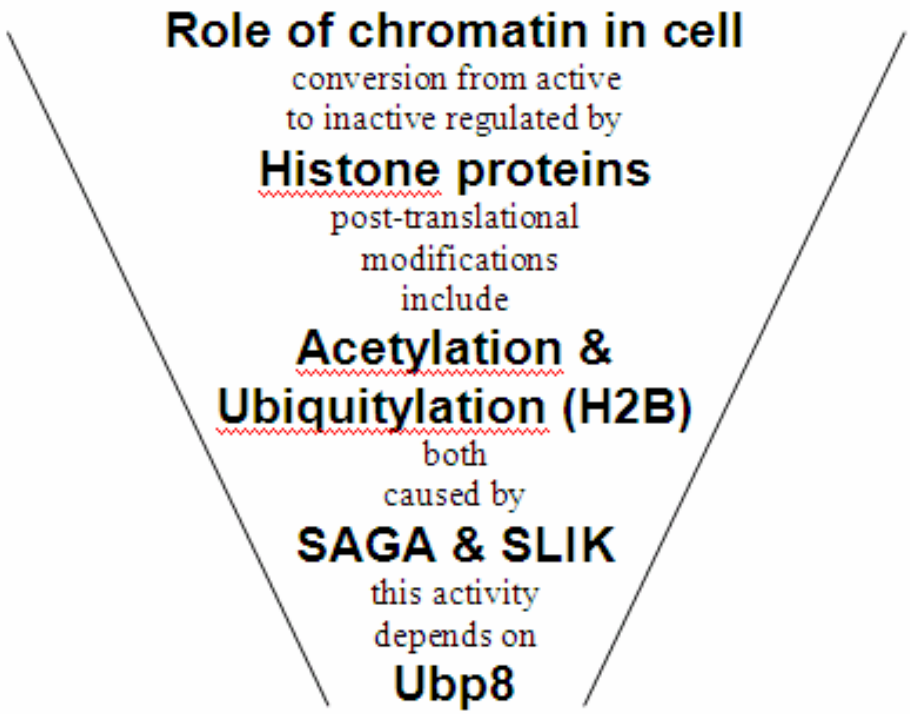
Background funnel:



GAP:	What is not known about the specific area of research?
Purpose:	What is the purpose of this research (and how does it help to fill the identified gap?)
Approach:	What is done to achieve the stated purpose?

INTRODUCTION What you studied, and why it's important.

Background funnel:



GAP:	Little is known about UBPs.
Purpose:	Understand how Ubp8 deubiquitylates H2B in context of SAGA.
Result:	Identified Sgf11
Revised purpose:	Understand relationship between Ubp8 & Sgf11 & how it relates to SAGA & SLIK.
Approach:	[See article for two specified tests.]

MATERIALS & METHODS How you studied it.

Professionally written:

***S. cerevisiae* strains.** The genotypes of strains used for this study are listed in Table 1. Individual TAP-tagged strains and deletion strains were obtained from Open Biosystems. TAP-tagged strains with deletions were obtained by crossing and dissecting individual TAP-tagged strains and deletion strains.

Less professional:

We first obtained individual TAP-tagged strains and deletion strains from Open Biosystems. These strains are listed in Table 1. Then we crossed and dissected these strains to obtain TAP-tagged strains with deletions, which are also listed in Table 1.

Write methods topically, not chronologically.
Past tense passive voice is acceptable.

RESULTS What you saw.

Tell your story, but avoid interpreting results.

…The purification of Ubp8 looked similar to purifications of SAGA (Fig. 1A, compare lanes 2 and 3). In order to confirm this, we performed mass spectrometry analysis (MudPIT) of the purification product and confirmed that Ubp8 is only associated with proteins that were previously identified in the SAGA, SLIK, and ADA HAT complexes (Fig. 1C) (22)…

DISCUSSION Your interpretation of what you saw.

…One possibility is that SPT20 may help to tether Ubp8 and Sgf11 to SAGA, since the loss of SPT20 increases H2B ubiquitylation levels, but not quite to the extent of a Ubp8 deletion (10).

If another reasonable researcher could dispute a claim, that claim belongs in the Discussion.

Design figures and tables carefully.

Readers skim by looking only at figures, tables, and captions. Tell your story.

Introduce each figure by number in the text.

The figure legend explains each part of the figure.

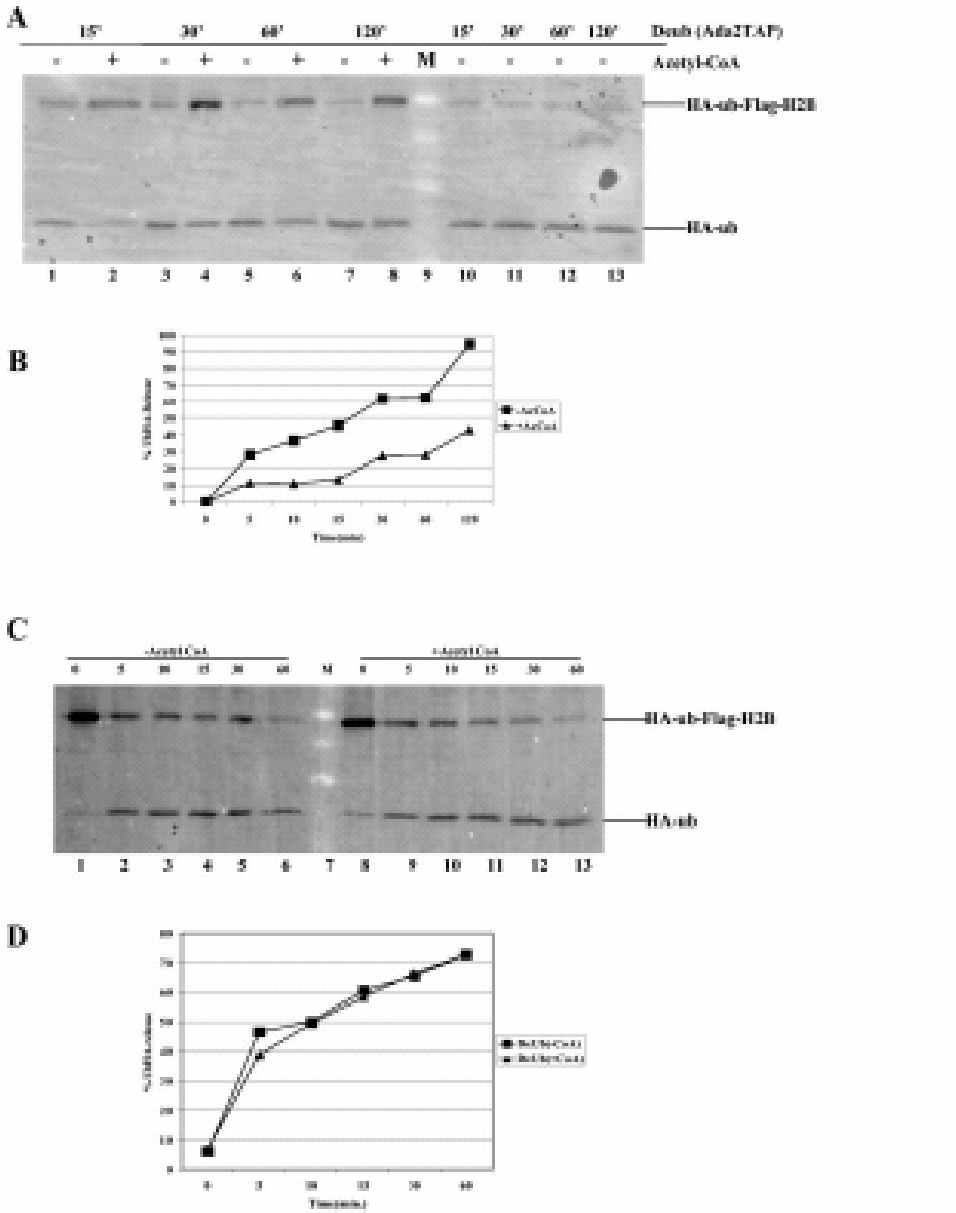


FIG. 5. Effect of acetylation on deubiquitylation in vitro. (A) Purified FLAG-H2B (modified and unmodified) was acetylated with Ada2-TAP gcn5Δ for 60 min prior to being subjected to deubiquitylation with wild-type SAGA for the indicated times. HAT assays were done either in the presence or in the absence of acetyl-CoA. Lanes 10 to 13 did not have acetyl-CoA and Ada2-TAP gcn5Δ as a control for deubiquitylation. (B) Semiquantitative analysis of the effects of preacetylating the FLAG-H2B template prior to deubiquitylation. (C) Simultaneous acetylation and deubiquitylation of purified FLAG-H2B with wild-type SAGA. Lanes 1 to 6, deubiquitylation in the absence of acetyl-CoA; lane 7, molecular weight marker; lanes 8 to 13, deubiquitylation in the presence of acetyl-CoA. (D) Semiquantitative analysis of the effect of simultaneous acetylation and deubiquitylation by SAGA. Acetylation was monitored by the incorporation of [^{3}H]acetyl-CoA, and deubiquitylation was monitored by the release of HA-ubiquitin from FLAG-ub-H2B-HA.

Design figures and tables carefully.

TABLE 1. *S. cerevisiae* strains used for this study

Strain	Genotype	Reference or source
By4741	*his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*	Open Biosystems
YKL101	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Ada2-TAP::*HIS3 MX6*	Open Biosystems
YKL134	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Ada2TAP::*HIS3 MX6 ubp8Δ*::*KANMX6*	This study
YKL128	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Ada2-TAP::*HIS3 MX6 sgf11Δ*::*KANMX6*	This study
YKL117	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Ubp8-TAP::*HIS3 MX6*	Open Biosystems
YKL132	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Ubp8-TAP::*HIS3 MX6 sgf11Δ*::KANMX6	This study
YKL138	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0*:Sgf11-TAP::*HIS3 MX6 ubp8Δ*::KANMX6	This study
YKL60	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:gen5Δ*::KANMX6	Open Biosystems
YKL120	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:ubp8Δ*::KANMX6	Open Biosystems
YKL97	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:sgf11Δ*::KANMX6	Open Biosystems
YKL136	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:gen5Δ*::KANMX6 *ubp8Δ*::KANMX6	This study
YKL116	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:gen5Δ*::KANMX6 *sgf11Δ*::KANMX6	This study
YKL137	*mata his3Δ1 leu2Δ0 met15Δ0 ura3Δ0:ubp8Δ*::KANMX6 *sgf11Δ*::KANMX6	This study
YKH045	*MATa ura3-1 leu2,3,-112 his3-11,-15 trp1-1 ade2-1 htb1-1 htb2-1* pRS314 (Flag-HTB1-CEN-TRP1) pRG145 (GAPDHprom-3HA-UB14-URA3 integrative)	10
YKH046	*MATa ura3-1 leu2,3,-112 his3-11,-15 trp1-1 ade2-1 htb1-1 htb2-1* pRS314 (Flag-htb1K123R-CEN-TRP1) pRG145 (GAPDHprom-3HA-UB14-URA3 integrative)	10
YKH047	*MATa ura3-1 leu2,3,-112 his3-11,-15 trp1-1 ade2-1 htb1-1 htb2-1* pRS314 (Flag-HTB1-CEN-TRP1) *ubp8Δ*::KanMx pRG145 (GAPDHprom-3HA-UB14-URA3 integrative)	10
YKL142	*MATa ura3-1 leu2,3,-112 his3-11,-15 trp1-1 ade2-1 htb1-1 htb2-1* pRS314 (Flag-HTB1-CEN-TRP1) *sgf11Δ*::*LEU2* pRG145 (GAPDHprom-3HA-UB14-URA3 integrative)	This study
YKL143	*MATa ura3-1 leu2,3,-112 his3-11,-15 trp1-1 ade2-1 htb1-1 htb2-1* pRS314 (Flag-HTB1-CEN-TRP1) *gen5Δ*::*LEU2* pRG145 (GAPDHprom-3HA-UB14-URA3 integrative)	This study
Fy2034	*MATa* HA-SPT7-TAP::TRP1 *ura3Δ0 leu2Δ1 his3Δ200 gen5*::*HIS3 trp1Δ63 lys2-173R2*	33
YJW589	*MATa* HA-SPT-TAP::TRP1 *ura3Δ0 leu2Δ1 his3Δ200 gen5DBr*::KANMX6 *trp1Δ63 lys2-173R2*	Mark Chandy

Unlike for figures, the table number and title go on top of the table.

Lab Reports (& Research Articles) tell a story.

INTRODUCTION	What you studied, and why it's important.
METHODS	How you studied it.
RESULTS	What you saw.
DISCUSSION	Your interpretation of what you saw.

The story should be cohesive.

The title and abstract must attract your audience.

Title About 10 words long. Include important keywords.

Abstract Less than 250 words.

Introduction

Purpose

Results

Conclusion

Covalent modifications of the histone tails and the cross talk between these modifications are hallmark features of gene regulation. The SAGA histone acetyltransferase complex is one of the most well-characterized complexes involved in these covalent modifications. The recent finding that the removal of the ubiquitin group from H2B is performed by a component of SAGA, Ubp8, is intriguing as it assigns two posttranslation modification processes to one complex. In this work, we characterize the association of Ubp8 with SAGA and the effect that acetylation and deubiquitylation have on one another in vitro and in vivo. We found not only that Ubp8 is a part of the SAGA complex, but also that its deubiquitylation activity requires Ubp8's association with SAGA. Furthermore, we found that the Ubp8 association with SAGA requires Sgf11 and that this requirement is reciprocal. We also found that the acetylation and deubiquitylation activities of SAGA are independent of one another. However, we found that preacetylating histone H2B inhibited subsequent deubiquitylation. Additionally, we found that increasing the ubiquitylation state of H2B inhibited the expression of the ARG1 gene, whose repression was previously shown to require the RAD6 ubiquitin ligase. Taken together, these data indicate that the expression of some genes, including ARG1, is regulated by a balance of histone H2B ubiquitylation in the cell.

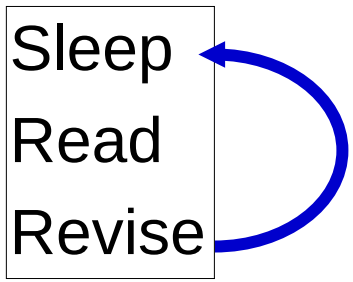
Do not write the sections in order.

Order of sections in paper	Possible order of writing
Title	Figures and Tables
Authors	Results
Abstract	Discussion
Introduction	Materials and Methods
Materials and Methods	Introduction
Results (inc Figs & Tables)	References
Discussion	Authors
Acknowledgements	Acknowledgements
References	Abstract
	Title

Plan time to set the paper aside.

Allow time over several days.

Write.



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http://web.mit.edu/due/handbook.pdf

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Harlan Breindel breindel@mit.edu