

Dartmouth College

## Dartmouth Digital Commons

---

Dartmouth Scholarship

Faculty Work

---

10-1995

### The Nascent-Polypeptide-Associated Complex: Having a "NAC" for Fidelity in Translocation.

William Wickner  
*Dartmouth College*

Follow this and additional works at: <https://digitalcommons.dartmouth.edu/facoa>



Part of the [Medical Biochemistry Commons](#)

---

#### Dartmouth Digital Commons Citation

Wickner, William, "The Nascent-Polypeptide-Associated Complex: Having a "NAC" for Fidelity in Translocation." (1995). *Dartmouth Scholarship*. 1618.  
<https://digitalcommons.dartmouth.edu/facoa/1618>

This Article is brought to you for free and open access by the Faculty Work at Dartmouth Digital Commons. It has been accepted for inclusion in Dartmouth Scholarship by an authorized administrator of Dartmouth Digital Commons. For more information, please contact [dartmouthdigitalcommons@groups.dartmouth.edu](mailto:dartmouthdigitalcommons@groups.dartmouth.edu).

## Commentary

### The nascent-polypeptide-associated complex: Having a “NAC” for fidelity in translocation

William Wickner

Department of Biochemistry, Dartmouth Medical School, 7200 Vail Building, Hanover, NH 03755-3844

Intracellular protein targeting and membrane translocation are highly regulated events, catalyzed by a cascade of specific interactions and coordinated with protein folding. On pages 9435–9439 of this issue of the *Proceedings*, Lauring *et al.* (1) show that the nascent-polypeptide-associated complex (NAC) plays a prominent role in regulating the association of signal recognition particle (SRP) with nascent chains and in preventing the binding of ribosomes with nonsecretory nascent chains to the endoplasmic reticulum. These studies, together with a recent dazzling array of biochemical and genetic studies in canine pancreas, yeast, *Escherichia coli*, and plant chloroplasts, give us added appreciation for the versatility of SRP function in different organisms and circumstances. SRP is not just for cotranslational targeting anymore but may be viewed as a chaperone and as a GTP-linked system for translocational fidelity.

SRP was first purified as a translocation factor from a salt extract of canine microsome (2, 3). It binds with micromolar affinity to ribosomes (2, 3) but with nanomolar affinity to ribosomes with nascent chains bearing a signal sequence. SRP binds signal sequences via its 54-kDa subunit (4, 5). Once bound, SRP slows or even arrests the growth of the nascent chain (3) until the complex binds to the SRP receptor of the endoplasmic reticulum (6, 7). This association triggers GTP hydrolysis by SRP and the SRP receptor and SRP dissociation from both the nascent chain and the receptor itself (8–12). The effects of SRP on chain growth provide a means for promoting cotranslational translocation, whereas targeting may be achieved by the GTP-regulated cycle with its receptor. However, SRP can function late in chain growth (13, 14) or even with full-length, ribosome-bound chains (15), and its capacity to slow translation is not needed for *in vitro* reconstitution of translocation (16–18).

Each aspect of SRP function was first elucidated by careful biochemistry of the mammalian system and then tested in the genetically tractable translocation reactions of *Saccharomyces cerevisiae* and *E. coli*. These studies not only have confirmed the importance of SRP for translocation but also have shed new light on its role. In yeast, SRP and its receptor are

required for normal growth rates, although the cells can adapt to their absence (19–22). Several secreted proteins depend strongly on SRP for translocation, others are strongly stimulated by its presence, whereas others apparently translocate normally in its absence (19).

*E. coli* SRP is also essential for growth (23, 24) but has no evident effect on the secretion of many proteins and only a minor kinetic effect on others (23–25). This SRP does, however, have a large effect on the secretion of pre- $\beta$ -lactamase, a well-studied periplasmic enzyme (25, 26). Strikingly, pre- $\beta$ -lactamase is exported entirely posttranslationally (27, 28), showing that SRP can function in posttranslational translocation. Nevertheless, its function may have to begin with short nascent chains, as indicated in elegant crosslinking studies (29). Recent studies by J. Lührink and colleagues (J. Lührink, personal communication) have shown that ribosome-bound *E. coli* SRP and trigger factor compete for nascent chain association in a manner reminiscent of mammalian NAC and SRP (see below). Trigger factor can associate with the GroEL chaperone (O. Kandror, M. Sherman, and A. L. Goldberg, personal communication), and this association can greatly enhance the capacity of GroEL to bind certain unfolded proteins. Thus, trigger factor might deliver nascent cytoplasmic proteins to this chaperone. *E. coli* SRP can associate with the SRP receptor homolog FtsY and activate a signal peptide-sensitive GTPase activity (30), but translocation is believed to proceed via SecA and SecYEG, the *E. coli* preprotein translocase (31).

The most striking confirmation of the chaperone role of SRP comes in studies of chloroplast protein import (32). The nuclear-encoded precursor form of the light-harvesting chlorophyll *a/b* protein is synthesized on cytoplasmic polysomes and imported across two membranes into the matrix. There, it associates with a complex that contains the chloroplast homolog of the 54-kDa subunit of SRP, which is required for subsequent import of the precursor into the thylakoid. This study, in agreement with other model studies (33, 34), clearly establishes the capacity of SRP to support posttranslational translocation in a nonribosomal context.

The elegant studies of Wiedmann and colleagues provide an important piece of the puzzle. They have found that NAC is a heterodimeric protein (35), which forms the shared “exit tunnel” from the ribosome (36). It competes with SRP for association with nascent chains; SRP has a higher affinity for nascent chains with a signal sequence, whereas NAC has a lower affinity for these same sequences. The association of a nascent chain with NAC not only prevents SRP association, which may otherwise lead to mistargeting and aberrant translocation, but also blocks ribosome association with the endoplasmic reticulum membrane (1). Indeed, in the absence of NAC, the system becomes independent of SRP and SRP receptor, and the affinity of ribosomes for the Sec61 complex is sufficient to target a nascent chain to the translocation site (37, 38). Translocation in this case is still strongly enhanced by a functional signal sequence, suggesting additional steps of SRP-independent signal sequence recognition (38). However, both the Wiedmann and Rapoport laboratories have shown that the ribosome is a targeting factor of major importance and that SRP and NAC regulate this targeting capacity.

SRP and NAC may be viewed as a versatile targeting chaperone team. Both SRP and NAC are ribosomally bound and may compete for signal sequences. SRP, which acts at a particular step of the translation cycle (39), has a higher signal sequence binding affinity. Although the role of SRP in mammalian translocation is not entirely tied to its capacity to slow polypeptide elongation (16–18), this property helps to ensure a cotranslational character. Delivery of the nascent chain complex to the endoplasmic reticulum membrane is blocked by NAC (for nonsecretory proteins); it is not yet known whether NAC has a direct role in delivering these proteins to cytosolic chaperones. The delivery of presecretory nascent chain complexes bearing SRP to the endoplasmic reticulum membranes may rely on three binding affinities: that of SRP for the SRP receptor, that of the ribosome for the Sec61 complex (40), and that of the signal sequence itself for the Sec61 complex (38) and for lipid (41, 42). The regulation, hierarchy of binding affinities, and temporal order of these three inter-

actions are still under study. The interaction of SRP with its receptor triggers GTP binding and hydrolysis and the release of SRP from both the nascent chain and the SRP receptor (11, 12). NAC adds selectivity to the association of SRP with signal sequences and the association of ribosomes with the Sec61 complex; its discovery and characterization by the Wiedmann group has added an important dimension to our understanding of translocation.

1. Luring, B., Kreibich, G. & Wiedmann, M. (1995) *Proc. Natl. Acad. Sci. USA* **92**, 9435–9439.
2. Walter, P. & Blobel, G. (1980) *Proc. Natl. Acad. Sci. USA* **77**, 7112–7116.
3. Walter, P. & Blobel, G. (1981) *J. Cell Biol.* **91**, 557–561.
4. Krieg, U. C., Walter, P. & Johnson, A. E. (1986) *Proc. Natl. Acad. Sci. USA* **83**, 8604–8608.
5. Kurzchalia, T. V., Wiedmann, M., Girshovich, A. S., Bochkareva, E. S., Bielka, H. & Rapoport, T. A. (1986) *Nature (London)* **320**, 634–636.
6. Gilmore, R., Walter, P. & Blobel, G. (1982) *J. Cell Biol.* **95**, 470–477.
7. Meyer, D. I., Krause, E. & Dobberstein, B. (1982) *Nature (London)* **297**, 647–650.
8. Römisch, K., Webb, J., Herz, J., Prehn, S., Frank, R., Vingron, M. & Dobberstein, B. (1989) *Nature (London)* **340**, 478–482.
9. Bernstein, H. D., Poritz, M. A., Stub, K., Hoben, P. J., Brenner, S. & Walter, P. (1989) *Nature (London)* **340**, 482–486.
10. Connolly, T. & Gilmore, R. (1989) *Cell* **57**, 599–610.
11. Connolly, T., Rapiejko, P. J. & Gilmore, R. (1991) *Science* **252**, 1171–1173.
12. Connolly, T. & Gilmore, R. (1993) *J. Cell Biol.* **123**, 799–807.
13. Ainger, K. J. & Meyer, D. I. (1986) *EMBO J.* **5**, 951–955.
14. Siegel, V. & Walter, P. (1988) *EMBO J.* **7**, 1769–1775.
15. Perara, E., Rothman, R. E. & Lingappa, V. R. (1986) *Science* **232**, 348–352.
16. Siegel, V. & Walter, P. (1986) *Nature (London)* **320**, 81–84.
17. Siegel, V. & Walter, P. (1988) *Cell* **52**, 39–49.
18. Lipp, J., Dobberstein, B. & Haeuptle, M.-T. (1987) *J. Biol. Chem.* **262**, 1680–1684.
19. Hann, B. C. & Walter, P. (1991) *Cell* **67**, 131–144.
20. Hann, B. C., Stirling, C. J. & Walter, P. (1992) *Nature (London)* **356**, 532–533.
21. Ogg, S. C., Poritz, M. A. & Walter, P. (1992) *Mol. Biol. Cell* **3**, 895–911.
22. Brown, J. D., Hann, B. C., Medzihradsky, K. F., Niwa, M., Brulingame, A. L. & Walter, P. (1994) *EMBO J.* **13**, 4390–4400.
23. Ribes, V., Römisch, K., Giner, A., Dobberstein, B. & Tollervey, D. (1990) *Cell* **63**, 591–600.
24. Poritz, M. A., Bernstein, H. D., Stub, K., Zopf, D., Wilhelm, H. & Walter, P. (1990) *Science* **250**, 1111–1117.
25. Phillips, G. J. & Silhavy, T. J. (1992) *Nature (London)* **359**, 744–746.
26. Luring, B., ten Hagen-Jongman, C. M., van der Weijden, C. C., Oudega, B., High, S., Dobberstein, B. & Kusters, R. (1994) *EMBO J.* **13**, 2289–2296.
27. Koshland, D. & Botstein, D. (1980) *Cell* **20**, 749–760.
28. Koshland, D. & Botstein, D. (1982) *Cell* **30**, 893–902.
29. Luring, B., High, S., Wood, H., Giner, A., Tollervey, D. & Dobberstein, B. (1992) *Nature (London)* **359**, 741–743.
30. Miller, J. D., Bernstein, H. D. & Walter, P. (1994) *Nature (London)* **367**, 657–659.
31. Brundage, L., Hendrick, J. P., Schiebel, E., Driessen, A. J. M. & Wickner, W. (1990) *Cell* **62**, 649–657.
32. Li, X., Henry, R., Yuan, J., Cline, K. & Hoffman, N. E. (1995) *Proc. Natl. Acad. Sci. USA* **92**, 3789–3793.
33. Crooke, E., Guthrie, B., Lecker, S., Lill, R. & Wickner, W. (1988) *Cell* **54**, 1003–1011.
34. Sanz, P. & Meyer, D. (1988) *EMBO J.* **7**, 3553–3557.
35. Wiedmann, B., Sakai, H., Davis, T. A. & Wiedmann, M. (1994) *Nature (London)* **370**, 434–440.
36. Wang, S., Sakai, H. & Wiedmann, M. (1995) *J. Cell Biol.* **130**, 519–528.
37. Luring, B., Sakai, H., Kreibich, G. & Wiedmann, M. (1995) *Proc. Natl. Acad. Sci. USA* **92**, 5411–5415.
38. Jungnickel, B. & Rapoport, T. A. (1995) *Cell* **82**, 261–270.
39. Ogg, S. C. & Walter, P. (1995) *Cell* **81**, 1075–1084.
40. Kalies, K.-U., Görlich, D. & Rapoport, T. A. (1994) *J. Cell Biol.* **126**, 925–934.
41. Martoglio, B., Hofmann, M. W., Brunner, J. & Dobberstein, B. (1995) *Cell* **81**, 207–214.
42. Siegel, V. (1995) *Cell* **82**, 167–170.