Chloroplasts communicate their redox needs by altering protein import, according to Michael Küchler, Jürgen Soll, and colleagues (Munich University, Munich, Germany). They found that an electron transport component and an import protein bind to each other and may communicate to control the rate of protein import.

The group’s original aim was to find other components in the complex that translocates proteins across the chloroplast inner membrane. Tic62 fit the bill, as it co-purified, colocalized, and communoprecipitated with known translocone components. It also bound to two interesting molecules: NAD, and a ferredoxin-NAD(P)$^+$ oxidoreductase (FNR). FNR acts at the end of the photosynthetic electron transport chain to transfer electrons from ferredoxin to NAD(P), thus creating NAD(P)H.

Increasing the level of NAD with either an analogue or an alternative electron sink resulted in increased translocation of FNR-L1, one isoform of FNR. Thus, if the last step of electron transport is insufficient to meet the demands of the chloroplast, it is capable of replenishing itself. Soll is now testing to see whether the Tic62-bound FNR regulates this process directly by transferring electrons to the NAD bound to Tic62.

Reference: Küchler, M., et al. 2002. EMBO J. 21:6136–6145.

An exit strategy for the clean-up crew

Macrophages race in to gobble up bacterial invaders, but what happens to the destroyers once the feast is over? Geoffrey Bellingan, Geoffrey Laurent (University College, London, UK), and colleagues find that the macrophages can slip back into the immune system because regulated adhesion molecules direct the macrophages to cells overlying lymphatic vessels.

The first attackers arriving at the scene of an infection in the abdominal cavity are neutrophils with a later peak in monocytes and macrophages. As the infection wanes, the neutrophils apoptose and are engulfed by macrophages. “The foot soldiers all shrivel up and die,” says Bellingan, “but no one really addressed how these other guys [the macrophages] would be cleared.”

Macrophages depart through lymphatics.

The London team found that the macrophages departed through the lymphatics, with the activated inflammatory macrophages (rather than the resident macrophages) being cleared preferentially. The activated macrophages adhered only to areas overlying the lymphatics, and both the adhesion and the exit were inhibited by agents that block β1-integrin-dependent adhesion.

Similar inhibitors have been used preclinically to slow influx of leukocytes. But Bellingan points out that such agents, if administered too late, may prolong inflammation by blocking macrophage exit. As to what triggers the exit, Bellingan guesses that ingestion of either apoptotic neutrophils or other targets may cause a switch in the activity of macrophage adhesion molecules, thus initiating the exit procedure.

Reference: Bellingan, G.J., et al. 2002. J. Exp. Med. 196:1515–1521.

14–3–3, set me free

The protein that does everything, 14–3–3, has now been shown to direct traffic. Ita O’Kelly, Steve Goldstein, and colleagues (Yale University, New Haven, CT) find that 14–3–3 can displace β-COP from various proteins, thus freeing them from retention in the ER and leading to surface expression.

Goldstein set out to find proteins interacting with the COOH terminus of a K$^+$ leak channel, KCNK3, and came up with a surprise: 14–3–3β. The group also found that KCNK3 binds β-COP, the COP1 retrieval protein, via a known dibasic motif. Binding of 14–3–3β and β-COP to KCNK3 was mutually exclusive. Deletion of the last residue of the 14–3–3β binding site led to retention of all KCNK3 protein in the ER, but surface expression was rescued by a further mutation of the dibasic β-COP binding sequence.

A similar system was demonstrated for another leak channel, an acetylcholine receptor subunit, and an MHC-associated protein. Others had individual clues in these systems about trafficking and the binding of 14–3–3 and β-COP, but Goldstein’s group is the first to put the whole story together.

For KCNK3, hormonal signals that turn on PKA may trigger the binding of 14–3–3β to the phosphorylated channel subunit, thus increasing surface expression and decreasing the excitability of the cell. Goldstein now wants to know if such a mechanism for controlling surface expression levels is common, and what proteins act with 14–3–3 to release the grip of ER retention.

Reference: O’Kelly, I., et al. 2002. Cell. 111:577–588.

Reference: Küchler, M., et al. 2002. EMBO J. 21:6136–6145.