

2.2 The two systems commonly used for cell-free translation

At present the two major cell-free systems are derived from wheat germ (WG) extracts and from rabbit reticulocyte lysates (RRL). Other less widely used systems include extracts of mouse ascites tumor cells, yeast, HeLa cells, and E. coli. RRL is geared up for globin synthesis but represents a system in the process of shutting down (i.e. the reticulocyte is a cell in the process of maturation to an erythrocyte that involves throwing away the nucleus and mitochondria, etc. and circulating for 3 months until gobbled up by the spleen). In contrast, wheat germ embryos are just getting started, so to speak, and in fact are dormant at the time of extraction. The full implications of this difference are not clear but it is important to keep in mind. An operational difference which may or may not be related to the previous point is that WG -at least as conventionally prepared - is largely free of "cytosolic" or "free" Signal Recognition Particle (SRP) while RRL has large quantities of free SRP. It is also observed that the "elongation arrest" property of SRP is exaggerated in WG relative to that in RRL. What any of these points means for physiology is still unclear but they have important operational consequences and hence should be remembered. For example, salt-washed (SRP depleted) microsomal membranes are typically competent for translocation in RRL - but not in WG - while the addition of purified SRP to WG - but not to RRL - typically abolishes synthesis of secretory products as a consequence of SRP arrest of the nascent chains.