Magnesium-dependence of *in vitro* translation programmed by gene-specific mRNAs

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In vitro translations programmed by mRNAs to single genes, prepared in large quantities by *in vitro* transcription in the SP6 system (1), have revealed efficient translation of particular mRNA species may depend on capping of mRNA (1) or pottasium concentration (2). Efficiency of translation *in vitro* in a wheatgerm system, with ^{35}S-methionine and programmed by mRNAs to subunits of glutamine synthetase (GS)(3,4) or poly-A enriched RNA from mature leaves of *Phaseolus vulgaris* were compared. Optimizing the magnesium concentration for translation revealed the fidelity of translation increased for high molecular weight mRNAs as magnesium concentration decreased with both total mRNA and GSo mRNA (Fig. 1). Leaf RNA translated to give equal TCA precipitable counts regardless of magnesium (data not shown), however the fidelity of chain elongation clearly improved to an optimum at 2.0 mM. Translation of GSo mRNA gave two distinct polypeptides: The higher molecular weight protein did not respond to the magnesium concentration, it may be a product of inefficient chain termination. The lower moleclar weight protein, which comigrates with native GSo, showed a 100-fold improvement of translation efficiency from 3.5 to 1.5 mM by autoradiography (fig.1) and TCA precipitation (data not shown). Purified RNAs to GS subunits o, o and o have the same optimum. Purified pre-plastocyanin (5) mRNA had an optimum at 3.5 mM magnesium in the same system. Capping the GS RNAs had no detectable effect on translation efficiency, raising pottasium concentrations to 100 and 150 mM inhibited translation 5- and 10-fold respectively (data not shown). The sharp magnesium optimum, its effects on fidelity of chain elongation and termination may devalue studies correlating mRNA concentration with changes in polypeptides by *in vitro* translation of several different samples of undialysed poly-A enriched RNA, a technique common in molecular biology.

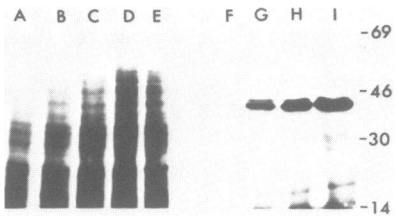

Figure 1. Fluorograph of SDS-PAGE gel showing translation products of *P.vulgaris* poly-A enriched RNA (Lanes a - e) and GSo subunit mRNA (Lanes f -i) at the following magnesium concentrations (mM): 3.5:- lane a, 3.0:- lanes b and f, 2.5:- lanes c and g, 2.0:- lanes d and h, 1.5 lanes e and i. Markers are kd.

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