

QUANTITATIVE EXTRACTION OF REMAINING AMOUNTS OF SOLUBLE PROTEINS
FROM PEA SEEDS UNDER ALKALINE CONDITIONS

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The extractable proteins of the garden pea are separated according to their solubility into two classes, the water soluble albumins and the salt soluble globulins. To extract proteins completely, pulverized seeds are treated with salt solution (NaCl) buffered with phosphate (pH 7). However, even after repeated extractions, the residue still contains some seed protein. Therefore, after extracting in salt solutions, the seed flour was treated under alkaline conditions (0.04 m NaOH) with small amounts (0.5%) of sodium dodecyl sulfate (SDS). By this method additional soluble protein could be extracted from the seeds.

The polypeptide composition of the seed proteins was determined by SDS-Polyacrylamide-Gel-Electrophoresis (PAGE). The banding pattern of the alkaline extract was compared with the polypeptides of the albumins and globulins from var. 'Dippes Gelbe Viktoria' and mutant 1000 of Gottschalk's collection. As shown in the pherogram, the albumin fractions (Fig. 1a) of both genotypes differed distinctly from the globulin fractions (Fig. 1b) and the alkaline fractions (Fig. 1c). However, the globulins were similar to the alkaline-extracted proteins. Only two small polypeptides in the alkaline fraction were lacking, as indicated in Fig. 1. Thus, the third alkaline-extracted fraction did not represent an independent protein fraction. It seems to be a contradiction to the classification of seed proteins because all types of globulin are regarded as being soluble in salt solutions. However, the finding is in agreement with the fact that the proteins of Pisum sativum could be subdivided exclusively in albumins and globulins. Nevertheless, this method allows one to extract additional amounts of globulins from ripened seeds not extractable under conditions mentioned above.

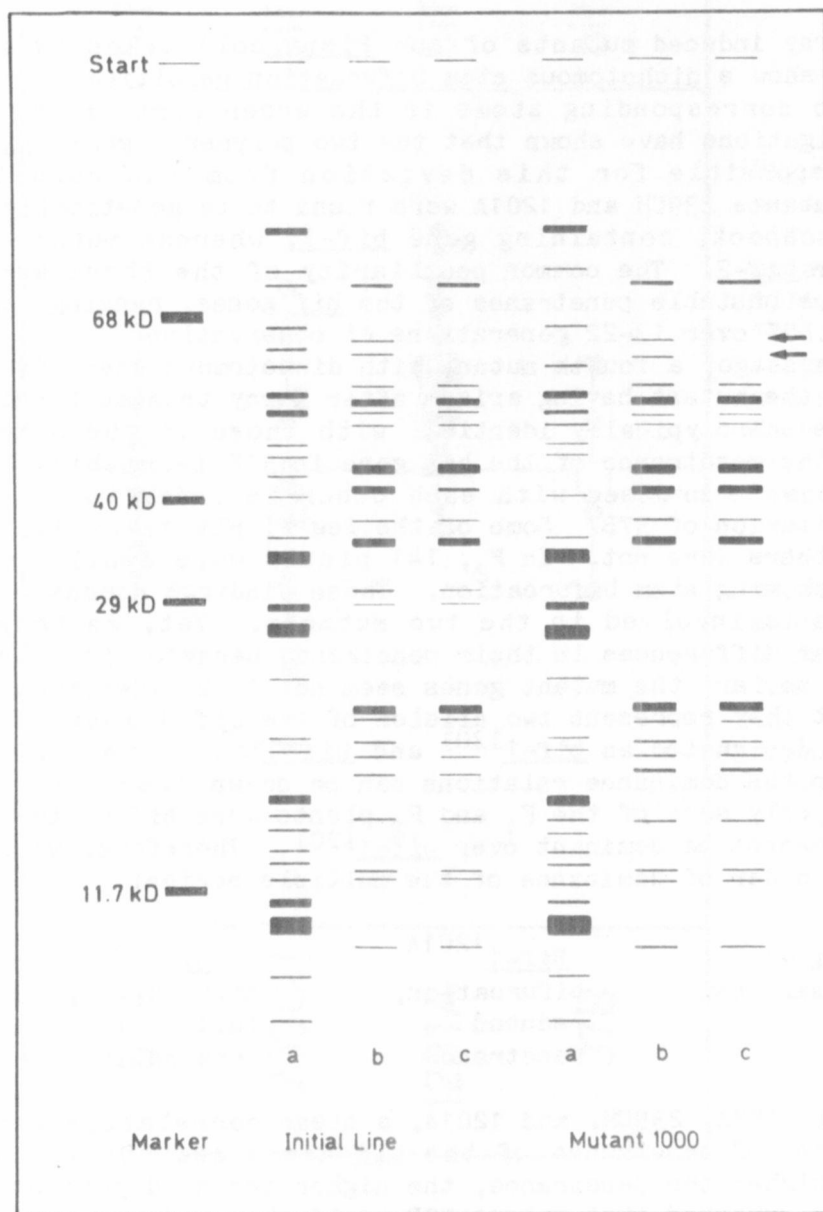


Fig. 1. Polypeptide composition of albumin fraction (a), globulin fraction (b), and the alkaline extracted proteins (c) of the initial line and mutant 1000 compared with a marker set. Arrows indicate deviations in banding pattern of the alkaline fractions.

kD = molecular weight in kilo Daltons