



Figure 4-5. HPLC purification of [³H]progesterone aryl azide labeled fragments from trypsin digest of β -subunit

A, B. nAChR β -subunit isolated from nAChR-rich membranes photolabeled with [³H]progesterone aryl azide was digested with trypsin. The digest was initially fractionated by Tricine SDS-PAGE. Two bands containing ³H (Band 2, 4-8 kD (A); Band 1, 2-4 kD (B)) were applied to a Brownlee Aquapore C4 column and fractionated by reverse-phase HPLC. ³H elution profiles (●) are from scintillation counting of 10% of each fraction. Fluorescence (·····) and absorbance profiles (—) were measured as described in methods. For Band 2 (A), material in fractions 31 and 24 (circled) was sequenced, and for Band 1 (B), fractions 22, 25, 26, and 28 was sequence (see Table 4-1).