The native state of the antibodies to ferritin obtained by affinity chromatography was checked by immunoelectrophoresis, which gave only one precipitation arc with the antigen.

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RAPID AUTOMATED SYNTHESIS OF OLIGODEOXYRIBONUCLEOTIDES USING A NEW CONDENSING REAGENT

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Continuing investigations in the field of the solid-phase synthesis of oligonucleotides and the preparation of artificial DNA fragments from them [1-4], we have considered the possibility of a rapid automated synthesis of oligodeoxyribonucleotides using a new condensing reagent — a mixture of 2,4,6-triisopropylbenzenesulfonyl chloride (TPS) and 4-dimethylaminopyridine 1-oxide (DMAPO), the efficacy of which has been demonstrated in the synthesis of oligonucleotides in solution.

The solid-phase synthesis of undecathymidylate and of three oligomers with a length of 9-11 units was performed on a "Viktoriya-2" automatic apparatus with a modified hydraulic system. As the polymeric support we used one based on Silochrom silica gel C-80 [3]. For chain growth we used a 0.1-0.15 M solution of protected mono-, di-, and trinucleotide blocks in absolute pyridine activated with a mixture of TPS (3 equivalents) and DMAPO (7 equivalents). The excess of the nucleotide component calculated on the first nucleoside was 5-6 equivalents. The sequence of washings of the support was similar to that described previously [4] but their total duration was decreased to 22 minutes. The time of condensation with the mononucleotides amounted to 2-3 min, and with the di- and trinucleotide blocks to 5-6 min, so that one cycle of chain growth on the support did not exceed 30 min, regardless of the length of the nucleotide block. The average yield per condensation stage determined from the dimethoxytrityl carbinol liberated [3] was 80-86%. After detachment from the support and complete deblocking of the reaction mixtures by the action of concentrated ammonia (at 45°C for 16 h), the following were isolated by high-performance liquid chromatography under the conditions described in [3]: 28.4% of d(TTTTTTTTTT), 17.7% of d(GATCTCAGC), 22% of d(GCGTTCCTTC), and 11.5% of d(AATTGGATCAT). The sequences of the nucleotides in the oligomers synthesized were confirmed by the Maxam-Gilbert method.

Thus, in the present work it has been shown that the use of the new condensing agent — a mixture of TPS and DMAPO — leads to a considerable acceleration of the condensation stage in the solid-phase synthesis of oligodeoxyribonucleotides and permits the desired oligomers to be obtained rapidly in high yield.

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