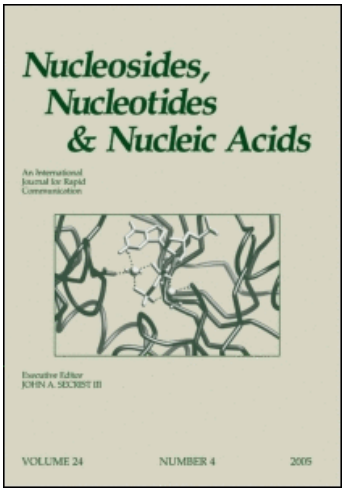

Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

Interaction of Oligonucleotide Derivativts with Animal Cells

V. V. Vlassov[a]; E. A. Deeva[a]; M. N. Nechaeva[a]; E. N. Rykova[a]; L. A. Yakubov[a]
[a] Novoetbirsk Institute of Bioorganic Chemistry, Siberian Division of the USSR Academy of Sciences, Novosibirsk, USSR

To cite this Article Vlassov, V. V. , Deeva, E. A. , Nechaeva, M. N. , Rykova, E. N. and Yakubov, L. A.(1991) 'Interaction of Oligonucleotide Derivativts with Animal Cells', Nucleosides, Nucleotides and Nucleic Acids, 10: 1, 581 — 582
To link to this Article: DOI: 10.1080/07328319108046535
URL: http://dx.doi.org/10.1080/07328319108046535

NUCLEOSIDES & NUCLEOTIDES, 10(1-3), 581-582 (1991)

INTERACTION OF OLIGONUCLEOTIDE DERIVATIVES WITH ANIMAL CELLS

V.V.Vlassov, E.A.Deeva, M.N.Nechaeva, E.N.Rykova, *L.A.Yakubov
Novosibirsk Institute of Bioorganic Chemistry, Siberian Division of the USSR Academy of Sciences, 630090 Novosibirsk, USSR

Abstract. Existence of specific oligonucleotide-binding proteins was demonstrated in cell lines obtained from mice, rats, monkeys using alkylating derivatives of ^{32}P labeled oligonucleotide pT_{16}.

Recently it has been shown that oligonucleotide uptake by mammalian cells goes via endocytosis which seems to be mediated by specific membrane receptors[1,2]. The details of the uptake mechanism still remain unclear. In this paper we have demonstrated the existence of oligonucleotide-binding receptors on different types of mammalian cells.

Cell lines (cos-1 and vero cells, L-671 mouse myoblasts, mouse and mink fibroblasts, Ag 17.1 and CHO hamster fibroblasts) were maintained as in[2].Hepatocytes were obtained by homogenization of perfused livers of BALB/c mice, cell suspension was washed twice in phosphate buffered saline (PBS) and used in experiments immediately. The oligonucleotide pT_{16} and its alkylating derivative, 4(N-methyl-N-2-chloroethylamino)benzyl-5'-phosphoramide, $ClR(pT)_{16}$, were synthesized and purified as described[3]. To label the receptors, the cells were incubated with ^{32}P-labeled $ClR(pT)_{16}$ (0.5 μM) at 37° C in 5% CO_2 atmosphere for 30 min. The cells were collected, lysed and the proteins were electrophoresed in 9-15% gradient PAAG according to Laemmly[4]. Gels were dried and autoradiographed.

The results are shown in FIG 1. The labeling with oligonucleotide derivatives reveals two peptides (78 and 80 kDa) in most of the cells tested . In the cases of the cos-1 and Ag 17.1 cells two additional proteins (82 and 84 kDa)

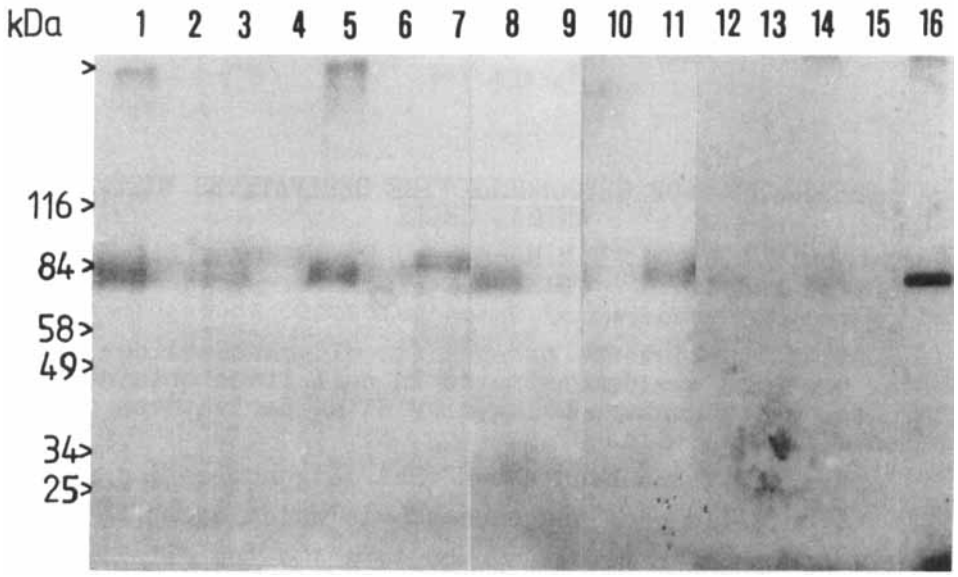

FIG.1.Oligonucleotide-binding proteins of mammalian cells. 1, A9; 2, A9 with 5 μM pT_{16} competition; 3, cos-1; 4, cos-1 with competition; 5, L671; 6, L671 with competition; 7, mice hepatocytes; 8, CHO; 9, CHO with competition; 10, vero with competition; 11, vero; 12, mink fibroblasts; 13, same with competition; 14, AG17.1; 15, AG17.1 with competition;16, A9.

are labeled. They are the only ones attacked in the case of mouse hepatocytes. The specificity of labeling was proved by competitive experiments, similar to the procedure[2].

The results suggest that the oligonucleotide-binding receptors are ubiquitous and offer a possibility to deliver oligonucleotide derivatives into different cells. The intriguing question about the biological role of the proteins remains to be answered.

REFERENCES

1. Goodchild J, Letsinger R.L.,Searin P.S.,Zamecnik M.,Zamecnik P.C.(1988) Human Retroviruses, Cancer and AIDS: Approaches to Prevention and Therapy. (Liss, New York) pp423-438.
2. Yakubov L.A., Deeva E.A., Zarytova V.F., Ivanova E.M., Ryte A.S.,Yurchenko L.V., Vlassov V.V. (1989) Proc.Natl.-Acad.Sci.USA,86, 6454-6458.
3. Knorre D.G., Vlassov V.V., Zarytova V.F.,Karpova G.G. (1985) Adv.Enzyme Regul. 24,277-300.
4.Laemmly U.K. (1970) Nature 2,680-685.