success telling everyone how good was Demeterios' method for isolation of chromosomes. Unfortunately he returned to the States shortly after that. The only time I heard from him was in May 1978 when he called me on the 'phone and asked me whether I could send him some cells (which I did).

I first became familiar with the precipitation of DNA by calcium in phosphate buffers in 1970 when I was an undergraduate student in chemistry at the Aristotelian University of Thessaloniki and in parallel I was working in the Virology and Biochemistry Laboratories of the Theagenion Cancer Institute in Thessaloniki, Greece, under the direction of Joyce Taylor-Papadimitriou (now head of a group at the Imperial Cancer Research Fund Laboratories in London, UK) and Prof John Georgatsos (now at the Dept of Biochemistry, University of Thessaloniki, Greece). I was supported from a grant from the Damon Ranyon Foundation. I was working on nucleases present in preparations of interferon. Optimizing conditions for their assay involved the use of various cations and buffers. I then realised that in phosphate buffers and in the presence of calcium the DNA precipitated.

When in 1973 at McGill University in Montreal as a graduate student working on reoviruses in Prof Angus F Graham's laboratory I saw the publication of Frank Graham and Alex Van der Eb (Graham, F.L. and Van der Eb, A.J. A new technique for the assay of infectivity of human adenovirus 5 DNA. Virology 52, 456-463, 1973) on the use of the calcium phosphate technique for transfer of viral genes, I realised the significance of my observations. So when looking for more efficient methods for gene transfer in 1976 in Toronto I knew the work of Graham and Van der Eb and I was confident that I could precipitate DNA with $CaCl_2$ in phosphate buffers. The experiments with the calcium phosphate technique and the purified visibly intact chromosomes were, not surprisingly as we know now, an immediate success. The experiments were reproducible and worked for more than one marker, ie methotrexate or amanitin resistance (Spandidos, D.A. and Siminovitch, L. Transfer of codominant markers by isolated metaphase chromosomes in Chinese hamster ovary cells. Proc. Natl. Acad. Sci. USA 74, 3480-3484, 1977).

However, the whole procedure involved the mastering of the isolation of chromosomes and the subsequent gene transfer. Because chromosomes could not be stored the whole procedure involved long hours and very meticulous handling. By the second year of my postdoc the work had advanced significantly with the transfer of biochemical markers and I had absolute confidence in the technique.

When Frank Graham (the co-discoverer of the calcium phosphate technique) who had now moved from the Netherlands to the McMaster University in Hamilton, Ontario, Canada, visited me at the lab in the summer of 1977, he told me that he was surprised but very glad that it had worked but he could not understand how. The same year he had published a paper together with his wife Sylvia (Baechetti, S. and Graham, F.L. Transfer of the gene for thymidine kinase to thymidine kinase-dificient human cells by purified herpes simplex viral DNA. Proc. Natl. Acad. Sci. USA 74, 1590-1594, 1977) where he had calculated the efficiency of transfer of cellular genes would be so low (about 1 transformant per 2×10^9 recipient cells per 10 mg of cellular DNA) as to be impractical. Thus the successful transfer of cellular genes using the calcium phosphate technique was unexpected. Even today we do not understand the observed and well established high efficiency of gene transfer (about 1 transformant per 5×10^5 recipient cells per 10 mg of cellular DNA) using chromosomes or DNA. Perhaps repetitive DNA sequences