Retrotransposons as Engines of Human Bodily Transformation

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Abstract — The historical literature suggests that there are unusual physical, as well as psychological, consequences in humans to the attainment of the exalted state of mind known as enlightenment, nirvana or samadhi. These reported changes include, but are not limited to, sudden reversal of aging, emergence of a light body and observed bodily ascension into the sky. This paper proposes a "jumping DNA" or transposon-mediated mechanism to explain rapid and large-scale cellular changes associated with human bodily transformation.

Only 3% of human DNA encodes the physical body. The remaining 97% of the 3 billion base pair genome contains over a million genetic structures, called transposons, that have the capacity to jump from one chromosomal location to another. Transposons that jump to a new location via an RNA intermediate are known as retrotransposons.

The three main classes of documented or putative retrotransposons in human cells are SINEs, LINEs and HERVs. SINEs and LINEs have been unambiguously shown to transpose in humans and there is indirect evidence that HERVs are active. A 1,700 base pair DNA sequence was isolated from purified activated human T cells (Kelleher *et al.*, 1996). The sequence of this DNA contains a novel combination of all three transposon families (SINEs, LINEs and HERVs) arranged like "beads on a string." I describe its structure and I propose that this DNA sequence, because of its cassette like configuration and its transcriptional expression and regulation, would be an effective participant in large scale transposon mediated genetic change that eventually results in transformation of the human body.

The hypothesis is testable by using the DNA sequence as a molecular probe to monitor transposon activity in the blood cells of individuals undergoing profound psychological transformation as a result of advanced meditation, near death experience (NDE) or close encounter experiences with UFOs. The relevance of these proposed experiments to the study of survival of human consciousness after death is discussed.

Keywords: mobile DNA — transposon — HERV — Alu — line — NDE — enlightenment

Introduction

In the future, attention undoubtedly will be centered on the genome, with greater appreciation of its significance as a highly sensitive organ of the cell that monitors genomic activities and corrects common errors, senses unusual and unexpected events, and responds to them, often by restructuring the genome. — Barbara McClintock, Nobel prize acceptance speech (McClintock, 1984).

In his book, *The Future of the Body*, author Michael Murphy states: "While most religions have supported body-denying attitudes and practices, they have also given rise to myths and doctrines of physical transfiguration. Some of these, it seems to me, point towards real possibilities. The Christian doctrine of glorification (of the body) and Taoist legends about holy flesh, for example, might express premonitions of something that could actually happen" (Murphy, 1992).

Many of the descriptions in the literature associate a sudden reversal of aging or the emergence of a light body with the attainment of "enlightenment." This phenomenon appears to be mostly, but not always, associated with death. For example: "Very early in the morning of the 25 of October 1419 the 62 year old Tsong Khapa made a series of inner offerings although no one present could understand why. Then his breathing ceased and his body regained the vibrancy of a 16 year old. Many disciples present witnessed the emission of varjegated light rays from his body, which substantiated the belief that Tsong Khapa entered the realm of enlightened beings" (Blackman, 1997). There have been similar descriptions in the native American/Mexican culture, notably Carlos Castenada's account of the exit of the nagual/sorcerer Don Juan Matus from the physical world by "burning the fire from within" (Castenada, 1981, 1984). According to Castenada, Don Juan chose both the time and place to accomplish this feat. "The warriors of Don Juan's party... vanished into the total light. I saw Don Juan take the lead. And then there was only a line of exquisite lights in the sky. Something like a wind seemed to make the cluster of lights contract and wiggle. There was a massive glow on one end of the line of lights where Don Juan was" (Castenada, 1981).

The specifics of a master selecting the method, the time and the place of his or her exit from the physical world is common to many cultures and religious traditions and is often associated with the emergence of a light body (Blackman, 1997). Exercising choice in this manner lies outside the normal experience of most human beings who are overtaken by death.

The death of the Tibetan master Milarepa generated reports of (i) two identical bodies that were being worshipped at two different places (an example of bilocation), (ii) a body that was impervious to burning on a funeral pyre and (iii) a body "transformed into a radiant celestial body, as youthful as a child eight years old" (Lhalungpa, 1982). The above reports all pertain to the question of survival after bodily death. They imply that the human body, under certain rare circumstances, is capable of generating a corporeal entity that survives the cessation of life on earth.

There are many additional references and anecdotal reports to suggest that transformation of the body can happen independently of death. Blofeld (1979) described a Chinese scholar who believed he would transform his flesh into a

"shining, adamantine substance, weightless yet hard as jade." Early Chinese religion speaks often of a form of shamanic bodily "ascent" (Paper, 1995). Paper says : "From the end of the Han period, the term *xian* denotes one who has achieved material longevity (> 200 years)...." Further on, in the Zhuangzi, the term *xian* refers to "a person who has the power to ascend (as a bird) but not a spirit." The disciples of the Indian adept Sri Ramakrishna Paramahansa (1836–1886) reported multiple physical changes, among which were the lengthening of his spine by almost an inch (Isherwood, 1959). In addition, the Catholic beatification process which insists on documentary evidence to support canonization status contains many written records of physical transformation associated with religious ecstasy (Thurston, 1952). These reports can be viewed as credible since the Catholic church authorities established stringent proof requirements for the canonization of individuals (see Murphy, 1992 and references therein). Thurston (1952) described multiple instances of bodily elongation by Catholic nuns as a consequence of religious ecstasy. Finally, Sri Auribindo espoused that succeeding lifetimes occur in the physical world and the soul can progress towards a luminous embodiment involving the divinization of living matter (Auribindo, 1970).

Thus, in several different cultures and religions, references can be found to a dormant capacity of the human body that appears to emerge after a period of extended spiritual evolution. As a consequence of sustaining this exalted inner state over a considerable period of time, the living human flesh can transform.

The purpose of this article is, in line with the historical literature, to propose that "enlightenment," in addition to being a state of mind, may also have physical consequences that have a direct bearing on survival of consciousness after death. Second, it is proposed that the effect of "enlightenment" on the body may be physiologically no different from other cellular changes in the body that arise from mutation of DNA, for example cancer. Third, I propose a molecular biological hypothesis for the mechanism of this transmutation process. The hypothesis makes a prediction, which is experimentally testable by using standard laboratory techniques currently in use in molecular biology.

Background

Mobile DNA in Humans

Contrary to popular belief, chromosomal DNA is not a static structure that is transmitted unchanged from generation to generation. Barbara McClintock won the Nobel prize in 1983 for showing that certain genetic sequences jump from one chromosomal location to another (for an excellent discussion, see McClintock, 1984). These structures, known as transposons, are found in the genome of every life form on this planet, from bacteria to humans.

If one were to hypothesize a transmutation of the human body, it would be necessary to orchestrate a change, cell by cell, involving the simultaneous silencing of hundreds of genes and the activation of a different set of hundreds

C. A. Kelleher

more. A transposition burst is a plausible mechanism at the DNA/RNA level that could accomplish such a genome wide change. Transposition bursts comprise the concerted movement of multiple mobile DNA elements from different genetic locations to new positions, sometimes on different chromosomes. The only bursts so far documented in the fruit fly Drosophila (Gerasimova, *et al.*, 1984, 1985), were lethal. This does not necessarily mean that transposition bursts are rare, since without specifically looking for their genetic consequences, they would be difficult to find. Human DNA contains an abundance of the necessary genetic structures to accomplish a transposition burst involving hundreds, or even thousands, of genes.

The physical human body is encoded by only 3% of the genetic infomation in each cell. A significant percentage of the remaining 97% arose in evolution, by retrotransposition (Weiner *et al.*, 1986). Retrotransposition is defined as the movement of a DNA sequence to a different chromosomal location, by means of transcription of that element, followed by reverse transcription into cDNA (complementary-DNA) and finally insertion back into the host genome. The exact locus of insertion determines whether or not the transposition results in a genetic change in the cell.

There are three major categories of sequences in human DNA that are documented or putative retrotransposons: these are SINEs (<u>short interspersed ele-</u> ments), LINEs (<u>long interspersed e</u>lements) and HERVs (<u>human endogenous</u> retroviral <u>s</u>equences) (Weiner *et al.*, 1986).

The most common SINEs are known as Alu elements. Alu elements are present at 10^5-10^6 copies per human cell (Deininger, 1989). *De novo* Alu transpositions, which resulted in neurofibromatosis type I (Wallace *et al.*, 1991) and haemophilia B (Vidaud *et al.*, 1993), have been reported in humans. The movement of a genetic element to a new chromosomal location that results in a mutation is known as insertional mutagenesis.

The L1 group is the largest known of the families of LINE like elements, consisting of approximately 10–50,000 copies per human cell. LINE elements have also been shown to undergo insertional mutagenesis resulting in disease, causing haemophilia A (Kazazian *et al.*, 1988), breast carcinoma (Morse *et al.*, 1988) and colon cancer (Miki *et al.*, 1992).

The third category remain unproven as retrotransposons. They are multiple families of HERVs, all of which have similar gene structures to infectious retrovinuses, including HIV. HERVs are represented in the genome in copy numbers ranging from one to several thousands (Wilkinson *et al.*, 1994).

Therefore, in total, the three types of retrotransposons described above are present in each human cell, both germline and somatic, at copy numbers of greater than a million. However, it must be emphasized that in humans only the disease-causing consequences of transposition have so far been found, since all discoveries of transpositions have been the result of researching the genetic origin of a particular disease, for example Haemophilia or cancer. Second, the huge copy number per cell of each of these transposon families makes it very difficult to catch an element "in the act" of moving to a different chromosomal location. To find a single element jumping would have a very low probability since the DNA probes used to target a transposition event rely on sequence homology and are therefore present at hundreds or thousands of copies. A single jump, using Southern blotting or more modern polymerase chain reaction (PCR) techniques would be extremely difficult to find unless one had precise information on the integration point of the transposon. Therefore, most of these transpositions have been found retrospectively and usually semendipitously.

The above described existing data on insertional mutagenesis in humans are important because they argue for the continued ability, in recent times, of transposons to jump from one location to another and, in so doing, to alter gene function.

Each cell has between 70,000 and 100,000 genes (Rowen *et al.*, 1997), whose combined expression makes up the physical human body. Hence, the ratio of the copy number per cell of human retrotransposons (1 million) to human genes (one hundred thousand) is approximately 10 to 1. Even if, as has been claimed, approximately 90% of human retrotransposons are defective, this still leaves about a 1:1 ratio of transcriptionally active retrotransposons and human genes. This ratio is important for the hypothesis in this paper.

Retrotransposons and Evolution

It is now commonly accepted that transposons play a significant role in evolution, both micro and macro (Finnegan, 1989; Ratner & Vasil' eva, 1992; Mc-Donald, 1990, 1993; Kloeckener-Gnuissem & Freeling, 1995). Others go further and propose that at least in some species, there is evidence that transposon activity is directly correlated with speciation (the emergence of new species) (Ginzburg *et al*, 1984; Wisotzkey *et al.*, 1997). The reason that transposons are such effective drivers of evolution is that a single transposition event can upregulate, downegulate, interrupt gene expression, create new fusion genes, or delete a gene all depending upon where precisely in the new gene locus they insert (see Figure 1).

Dramatic physical change, for example an old person who suddenly appears to observers with the body of a youth (Lhalungpa, 1982; Blackman, 1997), or the appearance of a light body as a result of attaining enlightenment, could be described as the emergence of a new species in a single generation from humanity. I propose that a synchronized, non-random transposition burst is the most simple molecular mechanism to account for the required new configuration. Transposon activity is known to be present in normal human immune cells at a very low level (Kelleher *et al.*, 1990, 1996; Wilkinson *et al.*, 1990, Medstand *et al.*, 1992, Krieg *et al.*, 1992; Brodsky *et al.*, 1993). I propose that a large increase in this activity, under rare circumstances, culminates in the generation of a new body from the existing body. This final emergence

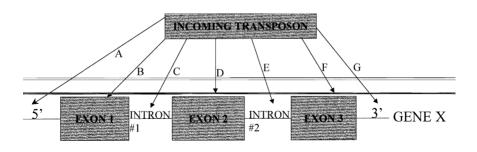


Fig. 1. Schematic shows the effect of a transposon insertion into a generic gene X. A typical gene comprises a 5' region which contains the regulatory elements necessary for transcription, the coding regions or exons which are separated like beads on a string by non coding regions or introns. The 3' region also contains regulatory sequences as well as processing instructions. If an incoming transposon inserts into the 5' region (A) it may disrupt or enhance transcription. Insertion into any exons (B, D or F) will create an altered gene product which may comprise either a truncated protein, or a larger altered protein with potentially different biological properties. Insertion into any intron (C or E) may have no effect or it may disrupt processing of the gene or it may disrupt transcription leading to a smaller protein. Finally, insertion of the transposon into the 3' region (G) may disrupt regulation, processing or it may yield a longer protein.

requires a burst that results in the silencing of hundreds of genes and the activation of hundreds more.

As stated above, the *non-coding* part of human DNA, comprising approximately 97% of human genetic information, has all of the required transposons to accomplish this. Further, both Alu and LINE elements have already been shown to transpose in human cells, HERVs have only partially been caught in the act (Mager & Goodchild, 1989).

Results/Experimental Work

Transcription of Transposons in Human Cells

If human retrotransposons are capable of transposition, then by definition they must be transcriptionally active. This was originally tested in human cells using HERV-H as a model (Kelleher *et al.*, 1990; Wilkinson *et al.*, 1990). HERV-H is the largest family of endogenous retrovirus sequences in the human genome (Wilkinson *et al.*, 1994). They are represented by approximately 100 full length 8,400 base pair sequences (Hirose *et al.*, 1993), a further 900 truncated 5,600 base pair sequences and a further 1,000 short sequences called long terminal repeats (LTR) (Mager & Henthorn, 1984); Mager & Freeman, 1987). HERV-H family members have the main structural features of retroviruses (Feuchter & Mager, 1990). In fact it has been proposed (Temin, 1974), although it is controversial (Roswitha *et al.*, 1996), that HERVs are the evolutionary precursors of modern day retroviruses such as HIV. The important point is that HERVs use almost identical mechanisms as retroviruses to insert into a new genetic location. While retroviruses infect neighboring cells, HERVs "infect" neighboring chromosomes within a cell.

By isolating and purifying mRNA from normal human blood donor peripheral blood mononuclear cells (PBMCs) and cancer cells, we (Kelleher *et al.*, 1990; Wilkinson *et al.*, 1990) and others (Johansen *et al.*, 1989, Liu & Abraham, 1991) used Northern blot analysis to show that HERV-H sequences are indeed transcribed in a variety of human cancer cells as well as in some normal cells. The highest level of transcription in normal cells occurred in full term placenta with some low level transcription in peripheral blood mononuclear cells (PBMC) (Kelleher *et al.*, 1990, Wilkinson *et al.*, 1990). Subsequently, others have confirmed our finding that there is transcription, albeit at a low level, of HERV sequences in PBMCs from normal individuals (Medstand *et al.*, 1992; Krieg *et al.*, 1992; Brodsky *et al.*, 1993). Therefore the first requirement for the hypothesis in this paper has been met.

Transcription of a Transposon Cassette in Human Cells

In order for a human transposon burst to be most easily facilitated, all three main families of transposons should be capable of simultaneous transcription in the same population of cells. This would greatly increase the probability for multiple genes being concurrently altered as a result of the burst. The targeting could result in enhancement of current gene function, activation of new gene transcription, silencing of current gene transcription, the creation of new mRNA by means of fusion transcripts or novel splicing mechanisms or the deletion of existing genes (see Figure 1). It should be noted that all of the above transposition mediated changes have been described for mammalian genomes (Amariglio & Rechavi, 1993, and references therein).

To examine the structure of the transcripts that are present at low levels in normal human PBMCs we needed to significantly boost their transcription. Highly purified T cells from a number of different normal donors were artificially activated and it was shown that the transcription level of the HERV-containing mRNA was dramatically boosted (Kelleher *et al.*, 1996).

Interestingly, we also found (Kelleher *et al.*, 1996) that following the activation of T cells, the transcription of the cassette was very tightly temporally controlled. Within 3–4 hours of the cell stimulus, transcription was markedly increased, reaching a maximum after 8 hours and then declining to basal levels within 24 hours.

Genetic Structure of the Transposon Cassette from Human T cells

Using standard molecular techniques we isolated and analyzed a 1,700 base pair region of the transcript from human PBMC (Kelleher *et al.*, 1996). We then fully sequenced the region using DNA sequencing methodology.

Figure 2 shows that the transcript is not composed of a single family of transposons which we had originally envisaged. Rather, it has a novel "beadson-a-string" structure comprising *all three major families* of retrotransposons (SINEs, LINEs and HERVs) that are found in humans in a single mRNA. The transcript contains SINE, LINE and two different types of HERV sequences, all tandemly linked (Kelleher *et. al.*, 1996). This was the first description of the structure of a human transposon cassette, although cassette like modular configurations of regulatory genes in mRNA have been described in *Strongy* - *locentrotus purpuratus*, the sea urchin (Nemer *et al.*, 1993).

Another noteworthy structure in the cassette, also shown in Figure 2, is the presence of an "A-T" rich region of approximately 214 bases. Interspersed in this region are six ATTTA motifs. These ATTTA motifs are known to be important in regulating mRNA stability (Peltz *et al.*, 1991). The presence of so many unmutated ATTTA motifs in such a short region (214 base pairs) strongly suggests that the levels of this unusual transposon cassette are tightly regulated in the cell.

The isolation and characterization of this transposon cassette from human immune cells showed that all three families of human retrotransposons can be simultaneously transcribed in T cells. Thus, a second prerequisite for the hypothesis in this paper has been fulfilled.

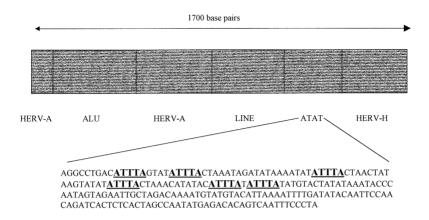


Fig. 2. The 1700 base pair sequence contains two different families of HERVs (called HERV-A and HERV-H) a LINE sequence and a SINE (Alu) sequence. In addition it contains an "A-T" rich region which has six separate repeats of the "ATTTA" motif which is known to regulate stability of RNA (Peltz *et al.*, 1991).

Identifying Locations of Transposon Target Sequences in Human Structural Genes

In order for a final large-scale transposition burst to occur, transcription of appropriate genes that play an important role in the structure of the human body would need to be silenced. This silencing could occur either preceding. or simultaneously with, a second wave of transposition that would activate a battery of novel genes. Type I collagen is the most abundant extracellular protein of bones, and is essential for bone strength. Collagen is also a major constituent of tendons, ligaments and skin. Twenty percent of the chemical constituents of muscle is protein, of which the most abundant is myosin. Actin is a major constituent of the thin filaments of striated muscle and tropomyosin and troponin complexes comprise the remaining third of thin filament mass. By searching the Genbank (www.ncbi.nlm.nih.gov/Web/Search/index.html) and Genemap (www.ncbi.nlm.nih.gov/SCIENCE96/ResTools.html) databases and utilizing the BLAST nucleotide/protein search algorithms (Altschul et al., 1990, 1997) the chromosomal locations and repetitive DNA content of the main structural genes in the human body were identified. These genes are collagen, myosin, actin, tropomyosin, troponin, keratin and tubulin. Table 1 shows that all the structural genes examined contain bona fide Alu transposon sequences, embedded in different regions of the genes. This result suggests that in a future transposition burst involving Alu elements, transcription of all of the above structural genes could theoretically be silenced by means of insertion of the Alu element into the gene near the already embedded Alu, or else by simple deletion of the whole gene by Alu mediated homologous recombination. Such a transposition burst involving every cell of the body would cease new muscle/bone/tendon/skin/hair production.

Discussion

Spiritual Evolution, the Emergence of a Light Body, and Survival After Death

In this paper, I first propose a transposon mediated molecular mechanism for human bodily transformation. Second, in this and a previous paper (Kelleher *et al.*, 1996), the structure of an unusual transposon cassette, isolated from the human immune system, is described. Third, I propose to test the hypothesis that human bodily transformation can proceed *via* transposon activation by monitoring cassette transcription in human populations who are intermediate in the enlightenment process.

To date, all of the documented descriptions of the emergence of the light body have been in humans who have attained spiritual mastery. The transformation appears to be the culmination of an unusually sustained focus, sometimes for a lifetime, on the goal of spiritual evolution. Importantly, this phenomenon appears to be independent of culture and religious belief.

In addition, many, but not all, of these descriptions refer to human

C. A. Kelleher

TABLE 1

The summary of transposon sequences in human structural proteins (as of March 1998) used Gene Map, GenBank and BLAST searches (see text). Chromosome location refers to which of the 23 pairs of chromosomes the genes (or gene families) are located. Since the databases are being updated daily, the above data will be in need of future revision. Because of the preliminary nature of the chromosomal assignments and the genome sequencing effort, some data on pseudogenes may be included.

Protein	Function (Chromosome location (Gene Map Search)	SINE, LINE or HERV? (GenBank/Entrez Search)	GenBank Accession#
Collagen	Bone, tendons, Ligaments, skin	1,2,6,7,9,11,12 13, 21, X	Alu in COL α1 (VII) Alu, LINE in COL1A2 (Intron 40)	AA789067 AF004877
			Alu in COL 4A3 Alu in COL 3A1	S78699 M26939
Myosin	Muscle	2,3,12,14,16,17 18	Alu, LTR in Myosin heavy chain, smooth muscle isoform	AA465475
			Alu in Myosin heavy chain, Skeletal muscle Alu in Myosin heavy chain,	AA211627 AA180111
			Cardiac muscle β isoform Alu in Myosin light chain 3	W84601
			Skeletal muscle isoform	
Actin	Muscle	2,7,10,15	Aluin chromosome 7 β actin	U19757
Tropomyosin	Muscle	9,15	Alu in Tropomyosin,cytoskeletal Alu in Tropomyosin α Skeletal muscle	AA180752 AA180999
			Alu, PTR7 in Tropomyosin Fibroblast Isoform TM3	AA075778
Troponin	Muscle	1	Alu in 3' intron of slow skeletal Troponin T (TNNT1)	
			Alu in fast skel. Troponin T	AA194223
Keratin	Hair, teeth	5, 12,17	Alu in Keratin 18	M24842, M19353, X12799
			Alu in Type I epidermal Keratin	J00124
Tubulin	Cytoskeleton	6,8,11,12,22	10 Alu elements in 5β tubulin (Intron III)	X00734

consciousness that survives exit from the physical world. Therefore, as a topic in the field of survival of consciousness research, the emergence of a light body deserves further study.

It is possible that there are stages on the road to the attainment of enlightenment, and that these stages are experienced by a great number of ordinary people. A testable prediction in this paper is that transcription of the transposon cassette should be increased in people who are experiencing intermediate stages in spiritual evolution. One such stage may be meditation and deep religious observance. Other stages may include the near death experience as well as UFO encounters. Interestingly, there are psychological data to suggest that the behavioral and psychosocial consequences of NDEs and close encounter experiences are remarkably similar (Ring, 1992, and references therein) and that they involve profound changes in attitude and behavior. Indeed, Ring (1992) has proposed that the NDE and close encounter experiences are triggers for spiritual development.

Cancer as a Model for Accumulation of Transposon Generated Mutations

In her discussion on "astounding" intricacies of plant genome reprogramming, McClintock (1984) said: "But this (plant genome reprogramming) is no more astounding, it would seem, than the sharing of a single genome by two brilliantly designed organisms, the caterpillar and the moth. It is becoming increasingly apparent that we know little of the potentials of a genome. Nevertheless, much evidence tells us that they must be vast." A tumor cell is one of the better known examples in humans of a cell that carries a reprogrammed genome.

The development of a tumor, which evolves from a single cell to become a disease that can engulf the human body, in most cases is a slow and gradual process (Vogelstein & Kinzler, 1993). Several distinct mutations (three to six) accumulate over time, sometimes decades (Vogelstein & Kinzler, 1993). As already described, in some cases these steps involve transposon mediated insertional mutagenesis (Morse *et al.*, 1988; Miki *et al.*, 1992).

It can be proposed that the attainment of enlightenment is also a multi-step process, proceeding over time from a series of experiences, including the near death experience, meditation, and other stimuli, each accompanied by transposon activation. The culmination of this process after many years of dedicated discipline, which the vast majority of humans never reach, is a massive transposition burst associated with the attainment of enlightenment.

Just as many of the tests for cancer in the laboratory involve testing for gene rearrangements, translocations, duplications, deletions, inversions or amplifications, testing for transposon activity could be done at the RNA level, since transcription is the first step in the mechanism of retro-transposition. Transpositions occurring in the brain or in other organs during a person's lifetime would be difficult to detect because of the dangers and inconveniences of biopsy sampling. In contrast, retrotransposon activity in the immune system would be readily detectable because of the ease of blood sampling. As stated earlier, many of the *de novo* transposition events documented in humans have been those found retrospectively in tumor cells. Therefore, the multi step nature, the genetic basis and the long time period for cancer evolution all suggest parallels for modeling the molecular steps on the road to enlightenment. Further, the development of cancer from a single or a few neoplastic cells to multiple metastases in distant regions and organs of the body is an everyday example of the capacity of the human genome to effect large-scale, body-wide somatic change in a single generation.

The Transposon Cassette as a Molecular Marker in Humans

It is known in many religious disciplines, that meditative practices are common steps on the way to enlightenment. Although some of the physical and psychological effects of meditation are well known (Murphy & Donovan, 1997), the physical effects of the enlightenment process, because of its rarity, remain at the anecdotal level. Dramatic metabolic and electroencephalographic changes have been seen during Buddhist meditation (Benson *et al.*, 1982, 1990). But changes to gene transcription did not feature in these studies. In his book *Living with Kundalini* (Krishna, 1993), Gopi Krishna describes his subjective experience following the rise of the kundalini: "...my whole organism was reacting to a new situation created inside by an altered activity of the vital organs to adjust itself to the changed environment within. Undoubtedly the disorder in my body was caused by the rapid passage of the luminous vital energy from cell to cell."

It is important to note that the transposon mediated transformation does not explain all of the phenomena associated with enlightenment, for example levitation and bilocation. The extraordinary physical effects of human beings coming to enlightenment may be an extension of those experienced during meditation (Murphy, 1992). Therefore, testing the transcription levels of transposons in the blood cells of individuals who are pursuing meditation will test the hypothesis of whether the cassette described in this paper could be used as a molecular marker for people on the road to transformation. In addition, testing transposon transcription in those people who have recently undergone near death experiences or close encounters with UFOs would considerably strengthen the hypothesis. Previous work (Kelleher et al., 1990, 1996; Wilkinson et al., 1990) and the work of others (Medstand et al., 1992; Krieg et al., 1992; Brodsky et al, 1993) has gone some way towards establishing a baseline for HERV transcription in normal individuals. The data in this paper would predict a higher transposon transcription level in the PBMCs of those undergoing deep meditation practice, in those who have recently experienced NDEs or close encounters with UFOs. Such a test is routine for most molecular biological laboratories.

The Placement of Transposon Sequences in the Human Genome

Some of the main gene families associated with the fundamental structure of the human body are given in Table 1. The genes for collagen, myosin, actin, tropomyosin, troponin, keratin and tubulin are scattered on multiple chromosomes. Table 1 shows that each of these genes has at least one Alu element located within its genomic configuration.

To accommodate the hypothesis of a future transposition burst, a series of integration markers in the human genome would be already positioned within these main structural genes to guide incoming transposons to their target. The majority of transposons in the human genome are non-functional and contain multiple stop codons. Although two decades ago widely dismissed as evolutionary junk or "selfish DNA" (Orgel & Crick, 1980) the presence of widely dispersed, non-functional transposons in the human genome may facilitate an upcoming transposition burst. For example, it has been shown in yeast that transposon integration hotspots contain copies of similar sequences (Ji et al., 1993). Likewise, in humans HERVs have been shown to target deletion of sequences containing same family HERVs by homologous recombination (for example. Mager & Goodchild, 1989) and similarly Alu sequences target other Alus for deletion (Nystrom-Lahti *et al.*, 1995). Therefore, the data in Table 1 suggest that each of the structural genes in human DNA contain the necessary transposon target sequences that are necessary to halt production of that protein

From an evolutionary standpoint, it makes sense that a positional transposon marker is non-functional, otherwise its position may change over time. Its purpose is to provide the necessary sequence homology that will guide the successful targeting, followed by integration, of a fully functional incoming transposon. It is outside the scope of this paper to discuss whether the placement in evolutionary history of SINE, LINE and HERV transposon sequences in the human genome is random or non-random. Since transposon sequences are present in every form of life on the planet, from bacteria to humans, and are established drivers of evolution (Finnegan, 1989; Ratner & Vasil' eva, 1992; McDonald 1990, 1993; Kloeckener-Gnuissem & Freeling 1995), and possibly speciation (Ginzburg *et al.*, 1984; Wisotzkey *et al.*, 1997), the transformative mechanism in humans associated with enlightenment could be just one of many transposon mediated pathways in evolution.

Regardless of the evolutionary mechanisms used for their placement, the preliminary data on their position in the human genome (Table 1) suggests the possibility of deletion, and hence inactivation, as a result of a future transposition burst. It is the positioning of these retrotransposon sequences, relative to the genes, which should determine whether, in the future they will be activated or repressed during a transposition burst. Such a non-random transposition burst could fall into the evolutionary mechanism called adaptive mutation described for bacteria originally by Cairns *et al.*, (1988) and subsequently by Hall (1998 and references therein). Second, it is of interest that others have independently noted long range order in the non coding (intron), but not in the coding (exon) sequences of DNA (Peng *et al.*, 1992), although the biological function, if any, of this order has not been elucidated.

So far only about 2-4% of the human genome has been sequenced, (Rowen *et al.*, 1997). Therefore, there is insufficient data to judge the total pattern of the placement of transposon sequences in the human genome. This will be

remedied in the year 2005, the proposed date for the completion of the human genome sequencing project. At that date, the location of the proposed transposon "guide sequence" in each of the 100,000 genes in the genome can determine whether that particular gene will be transcriptionally silenced or activated as a result of a transposition burst. Because such a small percentage (2-4%) of the genome has been sequenced, it is impossible in 1999 to predict which genes would be activated to generate a light body. Suffice it to say when the full sequence of the yeast *Saccharomyces cerevisiae* was published, fully 50% of the approximately 6000 genes had unknown function (Goffeau *et al.*, 1996). Nevertheless, the prediction that the genes for each of the main structural components of the human body should contain target sequences for transposition insertion has been validated (Table 1). Further, according to the data in Table 1, each of the genes encoding the main structural proteins could be deleted during a transposition burst. This would lead to a complete stop in the production of these proteins in every cell of the body.

In summary, the falsifiable hypothesis in this paper is that the transposon cassette described herein could be used as a molecular marker in human subpopulations undergoing *intermediate steps* on the path to enlightenment. These human subpopulations include, but are not limited to: NDEers, abductees, contactees, meditation practitioners, and spiritual adepts.

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References

- Altschul, S. F., Gish, W., Miller, W., Myers, E. W., and Lipman, D. J. (1990). Basic local alignment search tool. *Journal of Molecular Biology*, 215, 403.
- Altschul, S. F., Madden, T. L., Schaffer, A. A., Zhang J., Zhang Z. et al. (1997). Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Re*search, 25, 3389.
- Amariglio, N. & Rechavi, G. (1993). Insertional mutagenesis by transposable elements in the mammalian genome. *Environ. Mol. Mutagen*, 21, 212.
- Auribindo, S. (1970). The Life Divine. Vol. 19 in The Collected Works, pp. 802-803.
- Benson, H., Lehmann, J. W., Malhotra, M. S., Golman, R. F., Hopkins, J. and Epstein, M. D. (1982). Body temperature changes during the practice of g Tum-mo yoga. *Nature*, 295, 234.
- Benson, H., Malhotra, M. S., Goldman, R. F., Jacobs, G. D., and Hopkins, P. J. (1990). Three case reports of the metabolic and electroencephalographic changes during advanced Buddhist meditation techniques. *Behav. Medicine*, 16, 90.
- Blackman, S. (1997). Graceful exits: How Great Beings Die. New York, Tokyo: Weatherhill.
- Blofeld, J. (1979). Taoism: The Road to Immortality. Massacheussets: Unwin.
- Brodsky, I., Foley, B., Haines, D., Johnston, J., Cuddy, K., and Gillespie, D. (1993). Expression of HERV-K provinuses in human leukocytes. *Blood*, 81, 2369.
- Cairns, J., Overbaugh, J. and Miller, S. (1988). The Origin of Mutants. Nature, 335, 142.
- Castenada, C. (1984). The Fire From Within. New York: Simon & Schuster.
- Castenada, C. (1981). The Eagle's Gift. New York: Simon & Schuster.

- Deininger, P. L. (1989). SINES: short interspersed repeat DNA elements in higher eukaryotes. In Mobile DNA, pp 619–636. Edited by D. E. Berg & M. M. Howe. Washington DC: American Society for Microbiology.
- Feuchter, A. & Mager, D. L. (1990). Functional heterogeneity of a large family of human LTRlike promoters and enhancers. *Nucleic Acids Research*, 18, 1261.
- Finnegan, D. J. (1989). Eukaryotic transposable elements and genome evolution. Trends in Genet ics, 5, 103-107.
- Gerasimova, T., Mizrokhi, L., and Goegiev, G. (1984). Transposition bursts in genetically unstable Drosophila Melanogaster. *Nature*, 309, 714.
- Gerasimova, T., Matjunima, L., Mizrokhi, L., and Georgiev, G. (1985). Successive transposition explosions in Drosophila Melanogaster and reverse transpositions of mobile dispersed genetic elements. *EMBO J.* 4, 3773.
- Ginzburg, L. R., Bingham, P. M., and Yoo, S. (1984). On the theory of speciation induced by transposable elements. *Genetics*, 107, 331.
- Goffeau, A., Barrell, B.G., Bussey, H. et al. (1996). Life with 6000 Genes. Science, 274, 546.
- Hall, B. G. (1998). Activation of the Bgl Operon by Adaptive Mutation. Mol. Biol. Evol. 15, 1.
- Hirose, Y., Takamatsu, M., and Harada, F. (1993). Presence of *env* genes in members of the RTVL-H family of human endogenous retrovirus like elements. *Virology*, 192, 52.
- Isherwood, C. (1959). Ramakrishna and his disciples. New York: Simon & Schuster.
- Ji, H., Moore, D. P., Blomberg, M. A., Braiterman, L. T., Voytas, D. F., Natsoulis, G., and Boeke, J. D. (1993). Hotspots for unselected Ty 1 transposition events on yeast chromosome III are near tRNA genes and LTR sequences. *Cell*, 73, 1007.
- Johansen, T., Holm, T., and Bjorklid, E. (1989). Members of the RTVL-H family of human endogenous retrovirus-like elements are expressed in placenta. *Gene*, 79, 259.
- Kazazian, H. H., Wong, C., Youssoufian, H., Scott, A. F., Phillips, D. G., and Antonirakis, S. E. (1988). Haemophilia A resulting from de novo insertion of L1 reveals a novel mechanism for mutation in man. *Nature*, 332, 164.
- Kelleher, C. A., Wilkinson, D. A., Freeman, J. D., Mager, D. L., and Gelfand, E. W. (1996). Expression of novel transposon-containing mRNAs in human T cells. *Journal of General Virolo* gy, 77,1101.
- Kelleher, C. A., Wilkinson, D. A., and Mager, D. L. (1990). Abnormal expression of a novel transcript polyadenylated by a human endogenous long terminal repeat (LTR) in primary human leukemic marrow and blood cells. *Blood*, 76, 234a.
- Kloeckener-Gruissem, B., & Freeling, M. (1995). Transposon-induced promoter scrambling: a mechanism for the evolution of new allelles. *Proceedings of the National Academy of Science*, 92, 1836.
- Krieg, A., Gourley, M. F., Klinman, D. M., Perl, A. and Steinberg, A.D. (1992). Heterogeneous expression and coordinate regulation of endogenous retroviral sequences in human peripheral blood mononuclear cells. *AIDS Research and Human Retrovinuses*, 8, 1991.
- Krishna, G. (1993). In Living With Kundalini: Autobiography of Gopi Krishna. (Ed.) Shepard, L. Boston & London: Shambala.
- Lhalungpa, L. P. (1982). *The Life of Milarepa. A New Translation from the Tibetan*. Boulder, Colorado: Prajna Press.
- Liu, A. Y. & Abraham, B. A. (1991). Subtractive cloning of a hybrid human endogenous retrovirus and calbindin gene in the prostate cell line PC3. *Cancer Research*, 51, 4107.
- Mager, D. L. & Freeman, J. D. (1987). Human endogenous retroviruslike genome with type C pol sequences and gag sequences related to human T cell lymphotropic viruses. Journal of Virolo gy, 73, 2463.
- Mager, D. L. & Goodchild, N. L. (1989). Homologous recombination between the LTRs of a human retrovirus-like element causes a 5-kb deletion in two siblings. *American Journal of Human Genetics*, 45, 848.
- Mager, D. L., & Henthom, P. S. (1984). Identification of a retrovirus-like repetitive element in human DNA. Proceedings of the National Academy of Science, 81, 7510-7514.
- McClintock, B. (1984). The significance of responses of the genome to challenge. *Science*, 226, 792.
- McDonald, J. F. (1990). Macroevolution and retroviral elements. Bioscience, 40, 183-191.
- McDonald, J. F. (1993). Evolution and consequences of transposable elements. Current Opinion in Genetics and Development, 3, 855.

C. A. Kelleher

- Medstand, P., Lindeskog, M., and Blomberg, J. (1992). Expression of human endogenous retroviral sequences in peripheral blood mononuclear cells of healthy individuals. *Journal of Gener* al Virology, 73, 2463.
- Miki, Y., Nishisho, I., Horii, A., Miyoshi, Y., Utsunomiya, J., Kinzler, K.W., Vogelstein, B., and Nakamura, Y. (1992). Disruption of the APC gene by retrotranposal insertion of L1 sequence in a colon cancer. *Cancer Research*, 52, 643.
- Morse, B., Rothberg, P. G., South, V. J., Spandorfer, J. M. and Astrin, S. M. (1988). Insertional mutagenesis of the myc locus by a LINE-1 sequence in a human breast carcinoma. Nature, 333, 87.
- Murphy, M. (1992). The Future of the Body: Explorations into the Further Evolution of Human Nature. New York: Tarcher-Putnam.
- Murphy, M. & Donovan, S. (1997). *The Physical and Psychological Effects of Meditation*. Institute for Noetic Sciences, Sausalito, CA.
- Nemer, M., Bai, G., and Stuebing, E. W. (1993). Highly identical cassettes of gene regulatory elements, genomically repetitive and present in RNA. *Proceedings of the National Academy of Science*, 90, 10, 851.
- Nystrom-Lahti, M., Kristo, P., Nicolaides, N. C. et al, .(1995). Founding mutations and Alu-mediated recombination in hereditary colon cancer. Nat Med, 1, 1203.
- Orgel, L. E. & Crick, F. H. C. (1980). Selfish DNA: the ultimate parasite. Nature, 284, 604.
- Paper, J. D. (1995). *The Spirits are Drunk: Comparative Approaches to Chinese Religion*. Albany: State University of New York Press.
- Peltz, S. W., Brewer, G., Bernstein, P., Hart, P. A. and Ross, J. (1991). Regulation of mRNA turnover in eukaryotic cells. *Critical Reviews in Eukaryotic Gene Expression*, 1, 99.
- Peng, C-K., Buldyrev, S.V., Goldberger, A. L., Havlin, S., et. al. (1992). Long range correlations in nucleotide sequences. *Nature*, 356, 168. Commentary by Maddox J. (1992) *Nature*, 358, 103.
- Ratner, V. A. & Vasil' eva, L. A. (1992). The role of mobile genetic elements (MGE) in microevolution. *Genetika*, 28, 5.
- Ring, K. (1984). Heading Towards Omega. New York: William Morrow.
- Ring, K. (1992). The Omega Project: Near Death Experiences, UFO Encounters, and Mind at Large. New York: William Morrow.
- Roswitha, L., Lower, J. and Kurth, R. (1996). The viruses in all of us: characteristics and biological significance of human endogenous retrovirus sequences. *Proceedings of the National Academy of Science*. 93, 5177.
- Rowen, L., Mahairas, G., and Hood, L. (1997). Sequencing the human genome. *Science*, 278, 605.
- Temin, H. M. (1974). On the origin of RNA tumor viruses. Harvey Lect., 69, 173.
- Thurston, H. (1952). The Physical Phenomena of mysticism. Burns Oates.
- Vidaud, D., Vidaud, M., Bahnak, B. R. et.al. (1993). Haemohilia B due to a de novo insertion of a human specific Alu subfamily member within the coding region of the factor IX gene. Euro pean Journal of Human Genetics, 1, 30.
- Vogelstein, B. & Kinzler K. W. (1993). The multistep nature of cancer. Trends Genetics, 9, 138.
- Wallace, M. R., Anderson, L. B., Saulino, A. M., Gregory, P. E., Glover, T. W., and Collins, F. S. (1991). A de novo Alu insertion results in neurofibramotosis type I. Nature, 353, 864.
- Weiner, A. M., Deininger, P. and Efstratiadis, A. (1986). Nonviral retroposons: genes, pseudogenes and transposable elements generated by the reverse flow of genetic information. *Annual Review of Biochemistry*, 55, 631.
- Wilkinson, D. A., Freeman, D. J., Goodchild, N. L., Kelleher, C. A., and Mager, D. L. (1990). Autonomous expression of RTVL-H endogenous retroviruslike elements in human cells. *Journal* of Virology, 64, 2157.
- Wilkinson, D. A., Mager, D. L., and Leong, J. C. (1994). Endogenous human retroviruses. In *The Retroviridae*, pp. 465-535. Ed. J.A. Levy. New York: Plenum Press.
- Wisotzkey, R. G., Felger, I., and Hunt, J. A. (1997). Biogeographic analysis of the Uhu and LOA elements in the Hawaiian Drosophila. *Chromosoma*, 106, 465.