The 'Expansion Switch,' the 'Kill Switch,' and NF1

Andrea J. Alveshere [a]* and Vincent M. Riccardi [b]

a Department of Sociology and Anthropology, Western Illinois University, Macomb, IL, USA, *a-alveshere@wiu.edu; **b** The Neurofibromatosis Institute, La Crescenta, CA,USA

Introduction: In this study, we apply an anthropological perspective to explore how human evolutionary developments relate to the unique human *NF1* locus wild-type, variant and mutant allele functioning, and their respective phenotypes. Studies of related biochemical pathways provide potential explanations for the fixity of the unique human neurofibromin variant in the modern human population, the fact that the NF1 syndrome has never been observed in non-human primates, NF1 symptom onset and progression, a potential relationship between childhood diet and lifetime tumor burden, and the difficulty in developing model organisms for NF1 and many cancers.

NF1 Background: In 2018, we reported that the wild-type human variant of the *NF1* gene encodes a unique neurofibromin protein that has a single amino acid difference from the chimpanzee protein.[1] Other primates all share the chimp-like residue at that position; however, there are other residue differences that distinguish most primate species, suggesting a possible role for neurofibromin in speciation events. The chimp-like variant has never been documented among modern humans, aside from somatic mutations in tumor tissue. Therefore, the human wild-type *NF1* allele is derived compared to all other primates, and is fixed in the modern human population. The available Neanderthal, Denisovan, and early modern human sequences are also homozygous for the wild-type human *NF1* allele, implying that this allele was fixed in these hominin populations by at least 120,000 years ago.

No human live births have ever been documented to carry pathogenic mutations on both copies of the chromosome suggesting that at least one copy of the wild-type human allele is required for normal human development. Many *NF1* heterozygous individuals do not exhibit many (or even any) symptoms until late childhood or early adulthood, indicating that haploinsufficiency can be well-tolerated, at least early in life. The NF1 disorder has never been documented in non-human primates. Do *NF1* mutations not affect them? Or is *NF1* haploinsufficiency embryonically-lethal in non-human primates?

Some NF1 disorder symptoms, such as certain learning challenges, appear to arise from *NF1* haploinsufficiency. The neurofibroma lesions themselves, however, require a secondary somatic mutation causing a complete knockout of the *NF1* gene in the individual cell. The *NF1* gene product, neurofibromin, down-regulates RAS. RAS activates the cellular proliferation/differentiation ERK/MAPK signal transduction cascade. Loss of *NF1* over-activates RAS and the ERK/MAPK cascade. This mechanism is known to be associated with neurofibroma development. Other mutations in this pathway are responsible for the majority of cancers.

Compared to the chimp form, the human wild-type *NF1* allele is predicted to elicit a slight increase in overall phosphorylation potential. Increasing the phosphorylation potential of neurofibromin may make it less available for the inhibition of RAS. If the wild-type human *NF1* allele is predicted to facilitate a slight up-regulation of RAS activity, compared to that of chimpanzees, could this enable a relative increase – or "expansion" – in the growth potential of human neural cells? Why is this up-regulation not only tolerated, but *fixed*, in the modern human population? Is it fixed through genetic drift, and maintained by pure chance in this massive, easily-mutated gene? Or, did it provide such an adaptive advantage that hominins with the mutation out-competed the rest?

Related Pathway Research: In 2009, Arora et al. observed that "increased cognitive ability in humans compared to chimps may be explained, at least in part, by a reduction in the level of programmed cell death in the brain" and hypothesized that "a byproduct of natural selection for increased brain size and cognitive ability in humans was an elevated propensity for cancer and other diseases associated with reduced apoptotic function."[2] They identified patterns of gene expression consistent with the general hypothesis, but not the specific pathway changes that may have facilitated this developmental shift.

In 2018, Nyce identified a primate-specific cellular "kill switch," catalyzed upon inactivation of the "genome guardian" *TP53*, in the presence of adequate dehydroepiandrosterone sulfate (DHEAS).[3] *TP53* encodes the human form of the tumor suppressor protein p53. DHEAS is the sulfated, inactive form of DHEA, which circulates at high levels in the human bloodstream. The sex hormones, estrogen and testosterone, are also produced from DHEA. DHEA also has a role in the making of insulin growth factor 1 or IGF-1. Cells with inactivated p53 act as a sink for circulating DHEAS, which drives uncompetitive inhibition of G6PD to irreversibility, and results in a catastrophic increase in reactive oxygen species, resulting in cell death. Figure 1 illustrates the average circulating DHEAS levels (the blue and gray lines) over the human lifespan. The blue box indicates the average human reproductive years, which coincide with peak DHEAS levels. The high lifetime cancer risk faced by humans today (indicated in red), as compared to other animals (indicated in green) results from diminished "kill switch" function as DHEAS levels drop, combined with the extreme longevity of today's humans, well beyond our reproductive years.

Figure 2 depicts the relative circulating DHEAS levels in various primates, dogs, and mice, as presented in a follow-up article by Nyce (2020).[4] With human concentrations normalized to 1.0, this shows chimpanzees having 55% of human levels, gorillas only 19%, and mice less than 0.01%, indicating a dramatic functional difference between humans and other organisms that are often used as models for tumor disorders. This article also identifies the time depth of the various mutations that improve the "kill switch" mechanism, and postulates that fire-using hominins' increased exposure to carcinogenic polycyclic aromatic hydrocarbons (PAHs) provided evolutionary pressure to develop such dramatically high, protective DHEAS levels.

In 2020, Campbell reported findings linking juvenile prefrontal cortex and temporoparietal brain development stages to increases in DHEA production, and proposed a mechanism linking childhood meat consumption and neuronal activity.[5] IGF-1 increases in positive correlation with animal protein consumption, and prevents apoptosis of cells within the adrenal gland, increasing the centripetal movement of cells into the zona reticularis. Increasing IGF-1 during brain development is predicted to result in a thicker zona reticularis and greater lifetime production of DHEAS. DHEAS crosses the blood-brain barrier and enters into neurons, activating the Sigma-1 receptor, which increases energy production and alleviates stress-related production of oxygen free radicals, resulting in the increased production and release of neurotransmitters.

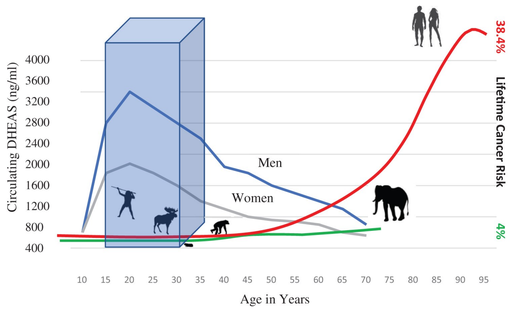

Figure 1. Human circulating DHEAS levels ; human and non-human lifetime cancer risk. Figure 4 in Nyce, J.W. (2018).[3]

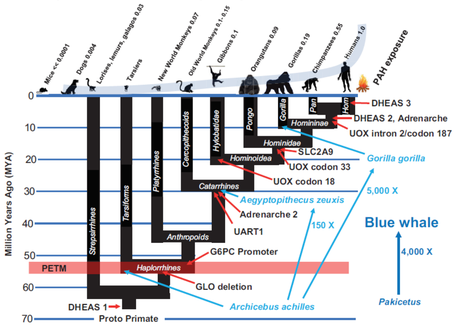

Figure 2. Relative DHEAS levels among primates, dogs and mice, and evolutionary timing of changes to loci in related pathways. Figure 8 in Nyce, J. W. (2020).[4]

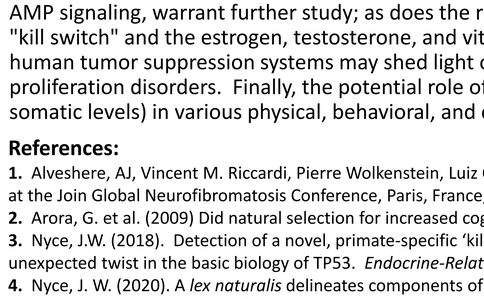

Figure 3. Disease manifestations in patients with neurofibromatosis type 1: epochs in which they develop. Figure 2 in Ratner and Miller (2015).[6]

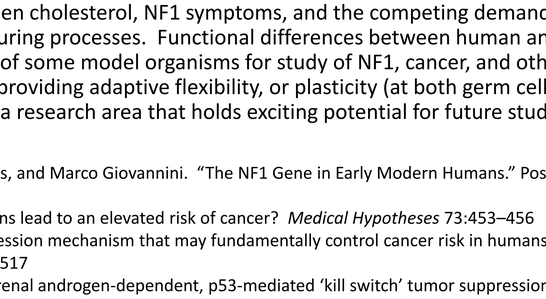

Figure 4. NF1 regulates cell proliferation, differentiation, survival and growth through the MAPK, INK4/ARF and mTOR pathways and cAMP signalling. Figure 1 in Helfferich et al. (2016).[7]

Discussion: Among NF1 patients, development of most benign neurofibromas, and risk of the rarer malignant peripheral nerve sheath tumors, tend to initiate slowly around the same time as DHEAS production, but continue to rise dramatically as DHEAS drops, forming an overall curve for NF1 tumor burden (Figure 3) that is roughly inverse to the modern human DHEAS curve, and rises much earlier in life than the typical human cancer curve. Levels of animal protein in the childhood diet may affect one's lifetime DHEAS production potential. Impacts on neural development, attention/behavior and executive function issues, deficits in spatial reasoning, and challenges with speech and musicality in *NF1* heterozygous individuals, all point to a wild-type human *NF1* allele optimized, in homozygous form, for memory and learning, communication, and problem-solving. Short stature, bone growth and healing challenges, and low circulating vitamin D concentration, may be associated with high demand for cholesterol in *NF1* heterozygous individuals, which is required for production of DHEA, estrogen, testosterone, vitamin D, and the nerve sheath itself. On the other hand, high cholesterol has been observed in some NF1 individuals, and cholesterol-lowering statin drugs appear to improve learning disabilities including verbal and non-verbal memory in some children with NF1. Increased neurofibroma growth during puberty and pregnancy may also reflect competition for DHEA or cholesterol.

Reduced risks of diabetes and obesity in *NF1* heterozygotes may involve interactions between DHEA and IGF-1. Reduced risks of alcohol and opiate addiction in *NF1* heterozygous individuals may reflect differences in neurotransmitter production, reception, or reuptake abilities, and could possibly reflect an ability of ancient wild-type *NF1* homozygous hominins to experience greater neurochemical rewards for engaging in certain evolutionarily-advantageous activities.

If the *NF1* wild-type human allele, in combination with the protective effects of the "kill switch," and the uniquely-high human DHEAS levels, *did* provide a strong adaptive advantage, that advantage may not only be linked to the RAS/RAF/MEK/ERK pathway. *NF1* regulates both RAS and the cyclic AMP signaling pathway, and RAS also regulates the mTOR proliferation cell growth pathway, and the INK4/ARF cell cycle control pathway, that includes the p53 tumor suppressor gene (Figure 4).

Conclusions: The combination of the human wild-type *NF1* allele with the DHEAS-mediated "kill switch" and soaring human DHEAS levels, may have acted as a neuronal "expansion switch," providing ancient hominins and modern humans with a suite of new or enhanced abilities that conveyed a strong adaptive advantage. Influence of the human wild type allele on cell growth, cell cycle control, and cyclic AMP signaling, warrant further study; as does the relationship between cholesterol, NF1 symptoms, and the competing demands of the "kill switch" and the estrogen, testosterone, and vitamin D manufacturing processes. Functional differences between human and non-human tumor suppression systems may shed light on the limitations of some model organisms for study of NF1, cancer, and other cell-proliferation disorders. Finally, the potential role of the *NF1* gene in providing adaptive flexibility, or plasticity (at both germ cell and somatic levels) in various physical, behavioral, and cognitive traits, is a research area that holds exciting potential for future study.

References:

1. Alveshere, AJ, Vincent M. Riccardi, Pierre Wolkenstein, Luiz Oswaldo C. Rodruigues, and Marco Giovannini. "The NF1 Gene in Early Modern Humans." Poster presented at the Join Global Neurofibromatosis Conference, Paris, France, 11/03/2018.
2. Arora, G. et al. (2009) Did natural selection for increased cognitive ability in humans lead to an elevated risk of cancer? *Medical Hypotheses* 73:453–456
3. Nyce, J.W. (2018). Detection of a novel, primate-specific 'kill switch' tumor suppression mechanism that may fundamentally control cancer risk in humans: an unexpected twist in the basic biology of TP53. *Endocrine-Related Cancer* 25, R497–R517
4. Nyce, J. W. (2020). A *lex naturalis* delineates components of a human-specific, adrenal androgen-dependent, p53-mediated 'kill switch' tumor suppression mechanism, *Endocrine-Related Cancer*, 27(2), R51-R65
5. Campbell B. (2020). DHEAS and Human Development: An Evolutionary Perspective. *Frontiers in Endocrinology*, 11:101.
6. Ratner N, Miller SJ. A RASopathy gene commonly mutated in cancer: the neurofibromatosis type 1 tumour suppressor. *Nature reviews. Cancer.* 2015 May;15(5):290-301.
7. Helfferich, J. et al. (2016) Neurofibromatosis type 1 associated low grade gliomas: A comparison with sporadic low grade gliomas, *Critical Reviews in Oncology/Hematology* 104:30-41