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Is Dr. Connor's Regenerative Transformation Possible?

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Abstract
In *The Amazing Spiderman*, Dr Connors injects himself with a regenerative serum containing lizard DNA which causes an amputated arm to regrow overnight. This article explores the possibility of utilizing signal molecules, found in both humans during embryotic development and the healing response of animals with high regenerative capacity, for the regeneration of human limbs. The treatment is found to be plausible but the timeframe for the regrowth of Connor's arm is unrealistic.

Introduction
In *the Amazing Spiderman,* Peter Parker visits Dr. Curt Connors, a scientist who studies the regenerative properties of lizards [1]. During this visit Parker provides Connors with the missing algorithm necessary for his regenerative serum [1]. Soon after, Connors injects himself with a lizard DNA serum and regenerates his amputated arm overnight [1]. Unfortunately, the serum had unintended side effects and Connors mutates into an evil lizard [1]. Although this scenario is fictional, one is left to wonder if it is scientifically possible. The story poses a real question for regenerative medicine; is it feasible for humans to regenerate missing limbs?

Limb Regeneration in Nature
Humans have a natural healing capacity, although the human regenerative response is comparatively limited [2]. Although mammals lack the ability to regenerate limbs, they are capable of undergoing healing for damaged skin, muscles, bone tissue, tendons and ligaments [2, 3]. Urodeles (an order of amphibians) and zebrafish are typically used to study limb regenerative capacity, providing valuable insight [4]. The limb regeneration process typically occurs in three steps — wound healing, dedifferentiation, and redevelopment (as seen in Figure 1) [4]. Wound healing involves the formation of an epithelial cell layer over the amputation plane and the initiation of the inflammatory response [2]. Dedifferentiation involves the induction of regeneration-specific genes, and the formation of an

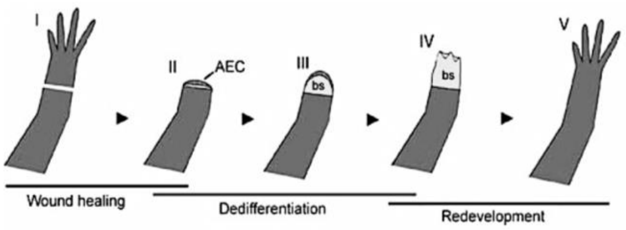

Figure 1: Steps of Limb Regeneration including wound healing, dedifferentiation, and redevelopment [4].

apical epithelia cap (AEC) [4]. Dedifferentiation also involves the creation of blastema (bs); a mass of undifferentiated stem cells that can undergo redevelopment [3]. Redevelopment involves the growth of the blastema, creating new limbs distal to its site of origin and re-differentiation of cells. This process also involves close interactions between the blastema and AEC, likely mediated by growth factors such as fibroblast growth factors (FGFs), used in limb elongation during embryonic development [4].

Signals Involved in Initiation of Limb Regeneration
Although limb regeneration is a three-step process, many believe that the signal molecules associated with the early processes ultimately decide the final outcome [4]. Some of these signal molecules include matrix metalloproteinases (MMPs), fibroblast growth factors (FGFs), and molecules of the Wnt/Beta-catenin pathway [3, 4]. MMPs are enzymes activated during inflammation and wound healing and are also unregulated early on in limb regeneration [4]. Other molecules vital to the initiation process are Wnt proteins and Beta catenin is Wnt/ β-catenin, which are proteins that control

cell proliferation through the regulation of target gene expression [4]. In fact, when organisms were treated with a MMP inhibitor or a Wnt/ β-catenin inhibitor following amputation, limb regeneration was partially inhibited [4]. The Wnt/Beta-catenin pathway also induces the expression of the *fgf-10* gene. These FGF molecules maintain interactions between the ACE and the blastema, necessary for limb outgrowth [4]. Studies show that two *fgf* genes are expressed after the amputating of a regenerative limb and neither is expressed after amputating a non-regenerative limb [4]. Moreover, it was shown that when treated with exogenous β-catenin or FGF-10, the regenerative capacity in non-regenerative limbs was improved [4].

Was Dr. Curt Connor's Transformation Feasible?

First of all, is it possible for humans to undergo limb regeneration despite a relatively low regenerative potential? The fact that the key genes expressed during limb regeneration in animals with high regenerative potential are identical to the genes expressed during human development supports the idea that humans possess the appropriate genetic machinery [5]. It has been suggested that by reinforcing the positive feedback loop between signal molecules such as *fgf* genes, the regenerative capabilities of non-regenerative vertebrate limbs may be enhanced [4]. If the serum used allowed the human body to access mechanisms associated with certain signal molecules, limb regeneration would be possible. An important second question to ask is whether or not Connor's transformation occurred in a realistic timeframe. To perform such a calculation, one must assume that there is a constant growth rate and is dependent on the length of the regenerate limb (distance) and time. The equation for the rate of growth is therefore:

$$\text{Rate of Growth} = \frac{\text{Length of Limb to Regenerate}}{\text{Time}}$$

With an associated standard error defined as:

$$\Delta Z = Z\sqrt{\left(\frac{\Delta A}{A}\right)^2 + \left(\frac{\Delta B}{B}\right)^2}$$

where Z is equal to rate of growth, A corresponds to the length of limb to regenerate, B corresponds to the time, and the ΔZ, ΔA, and ΔB correspond to the associated errors. For this calculation, time is set to 8 ± 2 hours, assuming normal sleep patterns. Based on Figure 2 and the assumption that his arm is approximately 0.75 m long, the length of the amputated arm is estimated to be 0.50 ± 0.02 m long. Therefore, the length of regenerate limb would simply be the difference between the arm length and the amputated arm. Using these values, Connor's growth rate is estimated to be 1500 ± 380 mm/day. Although Connors utilizes lizard DNA in his serum, his transformation was compared to that of a zebrafish because of the nature of the serum [6]. Due to the fact that there is a lack of well-developed molecular and genetic methods for manipulating genes in amphibians, they are typically not used in experiments involving signal molecules [6]. However, zebrafish are amenable to standard molecular and genetic manipulations such as mutagenesis screens, where specific genes necessary for limb regeneration have been identified [7]. In addition, the zebrafish has one of the fastest regrowth rates in nature, so it sets the relative limit for rates found in nature [6]. This comparison is also more effective because it compares Connor's transformation to organisms shown to express similar genes that would theoretically be used in a serum [6]. When compared to the growth rate of a zebrafish (0.1 to 0.4 mm/day) following fin amputation, the rate of growth for Connor's seems highly unrealistic [7].

Figure 2: A modified image depicting Dr. Curt Connor's amputated arm and the associated length [4].

Conclusion

In essence, limb regeneration seen in the movie *the Amazing Spiderman* is a very real possibility in the nearby future assuming that such a gene therapy passes necessary ethical and medical criteria. Such treatment would aim to enhance the inherent regenerative abilities of the human genome and associated cellular mechanisms, rather than creating a human-animal hybrid. In addition, even if a functional regenerative serum were made, the growth rate of 1500 ± 380 mm/day seen in the film would not be feasible. So, although the side effects of a future limb regeneration serum may hold equally dangerous results, no one needs to worry about a lizard villain with regenerative capabilities roaming the streets.

References

[1] Columbia Pictures, Marvel Entertainment, 2012. *The Amazing Spiderman* (film).
[2] Han, M., Xiaodong, Y., Taylor, G., Burdsal, C.A, Anderson, R.A., and Muneoka, K., 2005. *Limb Regeneration in Higher Vertebrates: Developing a Roadmap*, The Anatomical Record **287B,** 14-24.
[3] Kumar, A., Godwin, J.W., Gates, P.B., and Brockes, J.P., 2014. *Molecular Basis for the Nerve Dependence of Limb Regeneration in an Adult Vertebrate,* Science **318**, 772-777.
[4] Yokoyama, H., 2008. *Initiation of limb regeneration: The critical steps for regenerative capacity*, Develop. Growth Diff. **50**, 13-22.
[5] Marvel, n.d., Curtis Connors, Available at: http://marvel-movies.wikia.com/wiki/Curtis_Connors.
[6] Poss, K.D, Keating, M.T, Nechiporuk, A., 2003. *Tales of Regeneration in Zebrafish*, Developmental Dynamics **226**, 202-210.
[7] Lee, Y., Grill, S., Sanchez, A., Murphy-Ryan, M., Poss, K.D, 2005. *Fgf signalling instructs position-dependent growth rate during zebrafish fin regeneration,* Development **132,** 5173-5183.