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A Computational Network Model of Protein Kinases for Cell Survival/Death and its congruent tanner circuit

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Abstract

Computational modeling is useful as a means to assemble and test what we know about proteins and networks. The network model of EGFR pathway for cell survival/death has been implemented using SPICE simulator and Matlab. The sub pathways in EGFR pathways have been simulated using Tanner EDA. The proteins in the pathway are combined and the further simulation is done using the Neural Network Toolbox in Matlab. The ability of quinazoline to compete with the ATP to bind to the EGFR protein has been extensively studied and found out that mutated EGFR proteins have higher affinity for quinazoline which is having specific anti-tumor activity.

Keywords: Matlab, quinazoline, EGFR pathway.

Contents

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1. Introduction

1.1 Protein Kinases modeling

A protein kinase is a kinase enzyme that modifies other proteins by chemically adding phosphate groups to them (phosphorylation). Phosphorylation usually results in a functional change of the target protein by changing enzyme activity, cellular location, or association with other proteins (Manning, *et al*., 2002). Eukaryotic protein kinases are enzymes that share a conserved catalytic core. Phosphorylation of activation loops in protein kinases is a characteristic activation step for many different protein kinases (Figure 1). Most kinases act on both serine and threonine, others act on tyrosine, and a number of dual-specificity kinases act on all three. Up to 30% of all human proteins may be modified by kinase activity, and kinases are known to regulate the majority of cellular pathways, especially those involved in signal transduction. Protein kinases belong to a diverse family of enzymes that have various regulatory roles, yet function similarly by catalyzing the transfer of the phosphate of adenosine triphosphate

(ATP) to an enzyme-specific protein substrate (Gago, *et al*., 2008). Protein kinase cascades feature in many signal transduction pathways. A single upstream protein kinase appears to be responsible for the control of multiple downstream targets (Parker, *et al*., 2001). Because protein kinases have profound effects on a cell, their activity is highly regulated. Protein kinases have merged as key regulators of all aspects of neoplasia, including proliferation, invasion, angiogenesis and metastasis, hence making cancer fundamentally a disease of aberrant protein kinase activity and signal transmission (Dhanasekaran, *et al*., 1998).

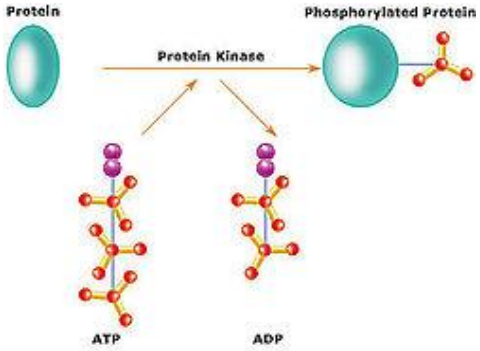


Figure 1: Action of Protein kinase (Manning, *et al*., 2002).

1.2 Tyrosine Kinases-EGFR signaling pathway network model construction

Of the protein kinases, approximately 90 are tyrosine kinases, which are enzyme that can transfer a phosphate group from ATP to a protein where the phosphate group is attached to the amino acid tyrosine on the protein. It functions as an "on" or "off" switch in many cellular functions. The tyrosine kinases are important mediators of the signaling cascade, determining key roles in diverse biological processes like growth, differentiation, metabolism and apoptosis in response to external and internal stimuli (Hanks, *et al*., 1988). The receptor tyrosine kinases function in transmembrane signaling, whereas tyrosine kinases within the cell function in signal transduction to the nucleus. Tyrosine kinase activity in the nucleus involves cell-cycle control and properties of transcription factors. Cellular growth and reproduction may rely in some part on tyrosine kinase. Tyrosine kinase function has been observed in the nuclear matrix, which is comprised not of chromatin, but of the nuclear envelope and a “fibrous web” that serves to physically stabilize DNA. Lyn and Src family tyrosine kinases in general have been known to function in signal transduction pathways (Radha, *et al*., 1996).

The epidermal growth factor receptor (EGFR) pathway is one of the best-studied signal transduction systems. EGFR is a trans membrane receptor with a tyrosine kinase domain protruding into the cytoplasm. They contain an extracellular domain and an intracellular protein TK core and must form dimers to initiate signaling (James, *et al*., 2006). The growth-stimulatory signal of epidermal growth factor is mediated by the trans membrane EGF receptor. The binding of EGF at the cell surface induces dimerization of EGFR, which results in the activation of EGFR tyrosine kinase activity and *trans*-autophosphorylation (Wang, *et al*., 2002). The EGFR pathway plays an important role during the growth and development of a number of organs including the heart, the mammary gland, and the central nervous system. They also play a significant role in regulating cell division, cellular differentiation, and morphogenesis (Carpenter, 2000). In the EGFR signaling pathway activated EGFR undergoes nuclear translocalization and subsequently regulates gene expression and potentially mediates other cellular processes. Transcriptional activity of nuclear EGFR appears to depend on its C-terminal trans activation domain and its physical and functional interaction with other transcription factors that contain DNA binding activity (Lo, *et al*., 2006).

A mutation that causes certain EGFR pathway to be constitutively active has been associated with tumorigenesis of the breast, ovaries, brain, and prostate gland. Most oncogenes encode components of cell signaling pathway. Abnormal cell components in cancer include growth factors and their receptors, signaling molecules and transcription factors. Aberrant cell signaling causes uncontrolled cell proliferation and survival, the hallmarks of cancer. Constitutive oncogenic activation in cancer cells can be blocked by selective tyrosine kinase inhibitors and thus considered as a promising approach for innovative genome based therapeutics (Paul, *et al*., 2005).

1.3 Formulation of problem

Knowledge on molecular biological systems is increasing at an amazing pace. It is becoming harder to intuitively evaluate the significance of each interaction between molecules of the complex biological systems. Hence it is essential to develop an efficient computational method to explore the biological mechanism. Many diseases including cancer, autoimmune disorders, cardiac disease and diabetes are associated with defects in protein phosphorylation and there are over 500 protein kinases in the human genome, and it has emerged as a major target in

the design and development of small molecule inhibitors (Gago, *et al*., 2008). The role of tyrosine kinases in the patho-physiology of cancer has obtained much interest in the area of cancer research. The emphasis is to be in the field of parameters to disrupt receptor tyrosine kinases (RTK) signaling pathways for cancer therapy which include antigrowth factor antibodies, receptor antagonists, antireceptor monoclonal antibodies, antisense, and small molecule tyrosine kinase inhibitors which are considered as promising approaches for innovative genome based therapeutics. Quinazoline analogs are potential inhibitors of tyrosine kinase pathway which can be used for the treatment of cancer with much higher inhibitory activities (Hennequin, *et al*., 2002).

A short introduction into systems biology researchers will be able to develop and validate qualitative models of biological pathways in a systematic manner using the well-established C-mos technology. The candidate signal transduction pathways including function-unknown proteins should be organized based on the proteomic data. The data of tyrosine kinase pathway obtained by *in vivo* studies is the relative quantitative data which should be implemented in the simulation work to obtain the computational network model of EGFR pathway as well as to obtain the clinical efficacy of different quinazoline analogs like imatinib, gefitinib, erlotinib, lapatinib and many more analogs which act on our pathway of interest and are also undergoing clinical trials. The numerical models of complex biological processes such as signal transduction is in its infancy and faces challenges. The area of research comprises simulation and stepwise modeling, animation, model validation as well as qualitative and quantitative analysis for behavior prediction. The ways in which the proteins network to process and transduce signals are poorly understood. The objective of this study to construct a computational network model of protein kinases for cell survival/death and action of Quinazoline on EGFR pathway for the better prediction of clinical efficacy against tumor activity.

2. Methodology

Computational modeling is useful as a means to assemble and test what we know about proteins and networks. This work represents an application of Very Large Scale Integration (VLSI) in System Biology. Inspired by the computational feasibility of Simulation Program For Integrated Circuit Emphasis (SPICE) a systematic signaling network has been built that would enable the predictive signal of cell survival/death.

2. 1 Simulation using T-SPICE simulator

The EGFR signaling pathway is sub divided into JAK/STAT pathway, ERK pathway, PI3K pathway, p38 pathway and JNK pathway. The proteins involved in these pathways are taken as the input for simulation. The parameters in this model are to be tuned manually based on biological knowledge available in the literature (Fujii, *et al.*, 2010). SPICE (Simulation Program with Integrated Circuit Emphasis) is a general-purpose open source electronic circuit simulator. It is a powerful program that is used in integrated circuit to check the integrity of circuit designs and to predict circuit behavior (Nagel, *et al*., 1971). The software used for simulation of EGFR signaling pathway is Tanner Electronic Design Automation. Tanner EDA provides a complete line of software solutions that catalyze innovation for the design, layout and verification of analog and mixed-signal integrated circuits (ICs). It is widely used for the applications in areas such as power management, displays and imaging, automotive, consumer electronics, life sciences, and RF devices (Warwick, *et al*., 2009). Here the T-SPICE is used to represent the biological model by using digital signals (Jain, *et al*., 2010)

2.2 Composite protein data by using Matlab

Neural networks are composed of simple elements operating in parallel. These elements are inspired by biological nervous systems. Neural networks have been trained to perform complex functions in various fields, including pattern recognition, identification, classification and control systems. Neural networks can also be trained to solve problems with a huge amount of data that are difficult for conventional computers or human beings. The toolbox emphasizes the use of neural network paradigms that build up to or are themselves used in engineering, and other practical applications. Typically, neural networks are adjusted, or trained, so that a particular input leads to a specific target output (Siegelmann, *et al*., 1991). Another incentive for these abstractions is to reduce the amount of computation required to simulate networks, so as to allow one to experiment with larger networks and train them on larger data sets (Balabin, et al., 2009).

After the simulation of EGFR sub pathways using T-Spice, the decomposed EGFR pathway is joined together by taking almost 8 proteins which should be present for the cell to survive. They are EGFR, PI3K, Akt, p38, Ras, ERK, JAK, JNK. Since 8 proteins are taken as the input there will be a huge amount of data produced in the truth table (Table 1 – 7) as the values are calculated using the formula 2^n where n is the number of inputs into the truth table, here the value of n is 8 and there will be 256 output values produced. So this huge amount of input and output is arranged into an excel sheet by which the Neural Network toolbox in Matlab is trained. The test set is considered as any of the input conditions taken.

2.3 Computational Network Model of EGFR pathway using Tanner

The goal was to construct a computational network model of EGFR signaling pathway for cell survival/death and also to find out the affinity of quinazoline analogs towards the EGFR pathway. The area of research comprised

simulation, modeling, model validation as well as qualitative and quantitative analysis for behavior prediction. The complex EGFR pathway had been decomposed into several sub pathways as the number of entities involved was large. In this study the digital analog 1 represents cell survival and 0 represents cell death as in truth table (Table 1 – 7) and their respective tanner code (Tanner Code 1-7) as circuit shown in Figure 2 – 8.

Truth Table-1

PI3K	AkT	p53	O/P
1	1	1	1
1	0	0	0
1	0	1	0
1	1	0	0
0	1	1	0
0	0	1	0
0	1	0	0
0	0	0	0

Karnaugh Map-1

PI3K \ AkT, p53	00	11	10	01
0	0	0	0	0
1	0	1	0	0

Boolean Expression = PI3K . AkT . p53

Tanner Code-1

```
M1 N3 PI3K N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N7 AkT N13 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 Output N3 Gnd NMOS L=2u W=120u AD=66p PD=24u AS=66p
M4 N13 p53 Gnd NMOS L=2u W=10u AD=66p PD=24u AS=66p
M5 N3 PI3K Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Vdd p53 N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 Vdd AkT N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M8 Output N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
V9  p53Gnd pulse(0.0 5.0 0 .1n .1n 133n 266n)
v10 AkT Gnd pulse(0.0 5.0 0 .1n .1n 421n 842n)
v11 PI3K Gnd pulse(0.0 5.0 0 .1n .1n 109n 218n)
.model pmos pmos
.model nmos nmos
.tran 5n 1u
.print v (Output) v (p53) v (AkT) v (PI3K)
```

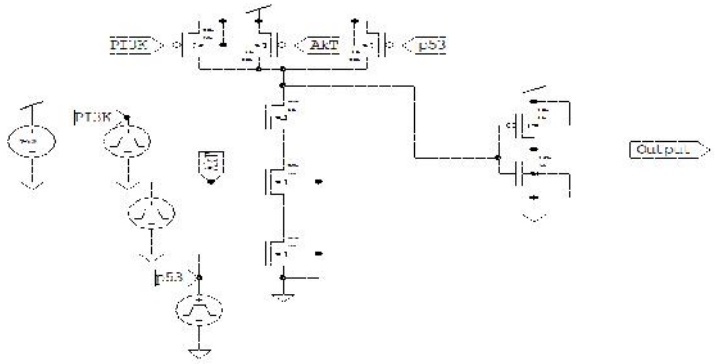


Figure 2: Tanner Circuit-1

Truth Table-2

SOS	p38	MK2	O/P
1	1	1	1
1	0	0	0
1	0	1	0
1	1	0	0
0	1	1	0
0	0	1	0
0	1	0	0
0	0	0	0

Karnaugh Map-2

SOS \ p38, MK2	00	11	10	01
0	0	0	0	0
1	0	1	0	0

Boolean Expression = SOS . p38 . MK2

Tanner Code-2

```
M1 N3 SOS N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N7 p38 N13 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 Output N3 Gnd NMOS L=2u W=120u AD=66p PD=24u AS=66p
M4 N13 p38 Gnd NMOS L=2u W=10u AD=66p PD=24u AS=66p
M5 N3 MK2 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Vdd SOS N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 Vdd MK2 N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M8 Output N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
V9  SOS Gnd pulse(0.0 5.0 0 .1n .1n 133n 266n)
v10 p38 Gnd pulse(0.0 5.0 0 .1n .1n 421n 842n)
v11 MK2 Gnd pulse(0.0 5.0 0 .1n .1n 109n 218n)
.model pmos pmos
.model nmos nmos
.tran 5n 1u
.print v(Output) v(MK2) v(p38) v(SOS)
```

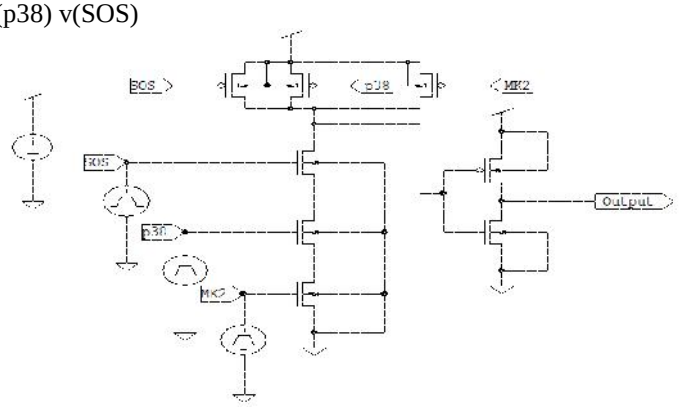


Figure 3: Tanner Circuit-2

Truth Table-3

RAS	RAF	ERK	O/P
1	1	1	1
1	0	0	0
1	0	1	0
1	1	0	0
0	1	1	0
0	0	1	0
0	1	0	0
0	0	0	0

Karnaugh Map-3

RAS \ RAF, ERK	00	11	10	01
0	0	0	0	0
1	0	1	0	0

Boolean Expression = RAS . RAF . ERK

Tanner Code-3

```
M1 N3 RAS N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N7 RAF N13 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 Output N3 Gnd NMOS L=2u W=120u AD=66p PD=24u AS=66p
M4 N13 RAF Gnd NMOS L=2u W=10u AD=66p PD=24u AS=66p
M5 N3 ERK Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p PS=24u
M6 Vdd RAS N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 Vdd ERK N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M8 Output N3 Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
V9  RAS Gnd pulse(0.0 5.0 0 .1n .1n 133n 266n)
v10 RAF Gnd pulse(0.0 5.0 0 .1n .1n 421n 842n)
v11 ERK Gnd pulse(0.0 5.0 0 .1n .1n 109n 218n)
.model pmos pmos
.model nmos nmos
.tran 5n 1u
.print v(Output) v(RAS) v(RAF) v(ERK)
```

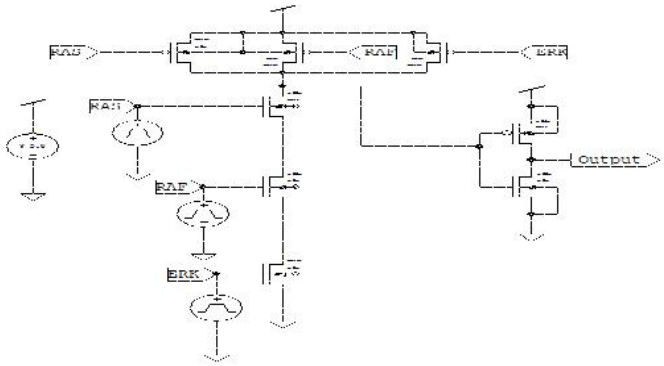


Figure 4: Tanner circuit-3

Truth Table-4

RAS	MEKK	JNK	O/P
1	1	1	1
1	0	0	0
1	0	1	0
1	1	0	0
0	1	1	0
0	0	1	0
0	1	0	0
0	0	0	0

Karnaugh Map-4

RAS \ MEKK, JNK	00	11	10	01
0	0	0	0	0
1	0	1	0	0

Boolean Expression = RAS. MEKK . JNK

Tanner Code-4

```
M1 N3 RAS N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N7 MEKK N13 NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 Output N3 Gnd NMOS L=2u W=120u AD=66p PD=24u AS=66p
M4 N13 RAS Gnd NMOS L=2u W=10u AD=66p PD=24u AS=66p
M5 N3 JNK Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Vdd MEKK N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 Vdd JNK N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M8 Output N3 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
V9  RAS Gnd pulse(0.0 5.0 0 .1n .1n 133n 266n)
v10 MEKK Gnd pulse(0.0 5.0 0 .1n .1n 421n 842n)
v11 JNK Gnd pulse(0.0 5.0 0 .1n .1n 109n 218n)
.model pmos pmos
.model nmos nmos
.tran 5n 1u
.print v(Output) v(RAS) v(MEKK) v(JNK)
```

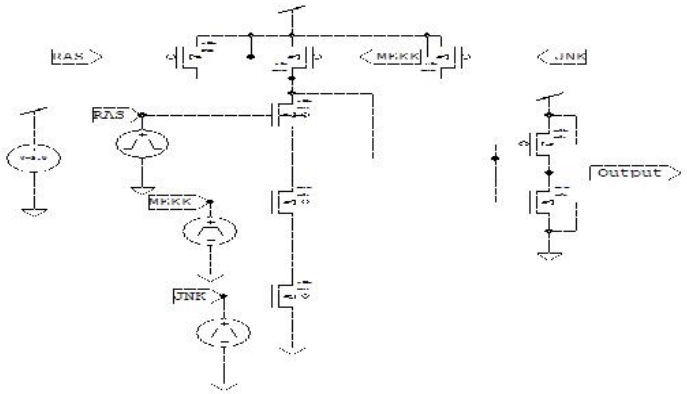


Figure 5: Tanner circuit-4

Truth Table-5

JAK	STAT	O/P
0	0	0
0	1	0
1	0	0
1	1	1

Karnaugh Map-5

JAK \ STAT	0	1
0	0	0
1	0	1

Boolean Expression = JAK. STAT

Tanner Code-5

M1 Output N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N6 STAT Gnd N16 NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 N7 JAK N6 N16 NMOS L=2u W=22u AD=66p PD=24u AS=66p
M4 N7 JAK Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M5 Vdd STAT N7 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Output N7 Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
v7 Vdd Gnd 5.0
v8 JAK Gnd pulse(0.0 5.0 0 10n 10n 100n 200n)
v9 STAT Gnd pulse(0.0 5.0 0 10n 10n 100n 200n)
.model pmos pmos
.model nmos nmos
.tran 5n 1u
.print v(Output) v(STAT) v(JAK)

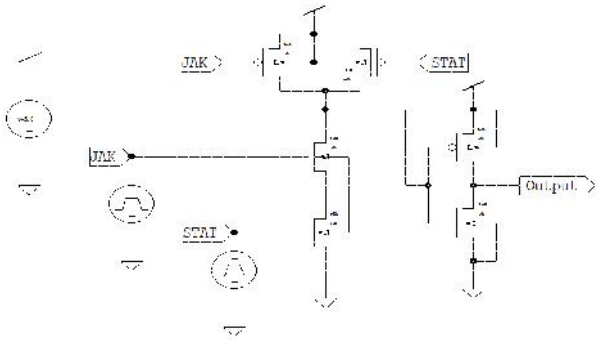


Figure 6: Tanner circuit-5

Truth Table-6

MK2	AP1	O/P
0	0	0
0	1	0
1	0	1
1	1	0

Karnaugh Map-6

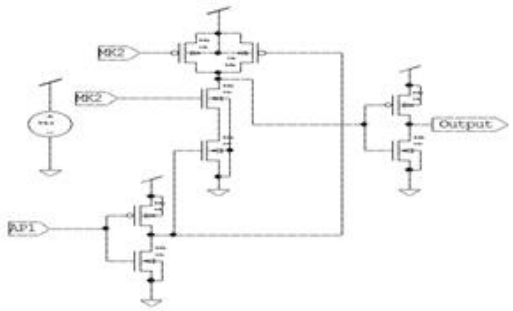
MK2 \ AP1	0	1
0	0	0
1	1	0

Boolean Expression = MK2. $\overline{AP1}$

Tanner Code-6

M1 N1 MK2 N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M2 N15 AP1 Gnd Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 N7 N15 Gnd Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M4 Output N1 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p

```
M5 N1 MK2 Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Vdd N15 N1 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 N15 AP1 Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M8 Output N1 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
v9 Vdd Gnd 5.0
.model pmos pmos
.model nmos nmos
.tran 40n 400n
v1 MK2 gnd bit ({0011})
v2 AP1 gnd bit ({0101})
.print tran  Output MK2 AP1
```



Truth Table-7

ERK	AP1	O/P
0	0	0
0	1	0
1	0	1
1	1	0

Karnaugh Map-7

ERK \ AP1	0	1
0	0	0
1	1	0

Boolean Expression = ERK. $\overline{AP1}$

Figure 7: Tanner circuit-6

Tanner Code-7

```
M1 N1 ERK N7 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p M2
N15 AP1 Gnd Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M3 N7 N15 Gnd Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M4 Output N1 Gnd NMOS L=2u W=22u AD=66p PD=24u AS=66p
M5 N1 ERK Vdd Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M6 Vdd N15 N1 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
M7 N15 AP1 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p PS=24u
M8 Output N1 Vdd PMOS L=2u W=22u AD=66p PD=24u AS=66p
v9 Vdd Gnd 5.0
.model pmos pmos
.model nmos nmos
.tran 40n 400n
v1 ERK gnd bit ({0011})
v2 AP1 gnd bit ({0101})
.print tran Output ERK AP1
```

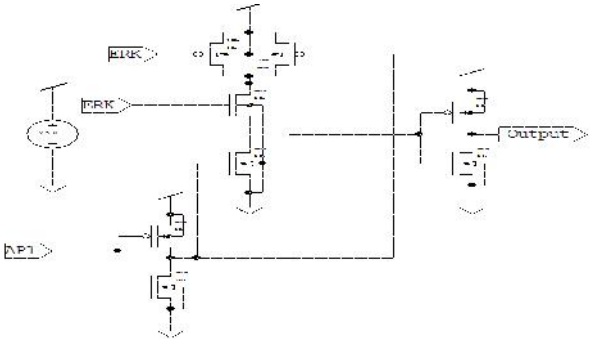


Figure 8: Tanner circuit-7

2. 4 Neural Network computational model construction of EGFR pathway

From the decomposition of complex EGFR signaling pathway it was possible to detect the essential proteins for the cell survival. They are EGFR, PI3K, AkT, RAS, p38, ERK, JNK, JAK. These eight proteins are required for the cell to survive. The truth table (Table 1 - 7) produced using these eight proteins contain 256 conditions which is a huge data when applied in T-SPICE simulator. So the Neural network toolbox in Matlab is used to do the computational network model construction. The Neural Network was trained using the 256 conditions in the Matlab.

Matlab Code

```
train= xlsread('D:\matlab_pgms\work.xls',1);
trainout= xlsread('D:\matlab_pgms\work.xls',2);
test= xlsread('D:\matlab_pgms\work.xls',3);
traint=train';
testt=test';
trainoutt=trainout'
```

Table 8: Truth table for Training set containing 256 conditions

EGFR	PI3K	AkT	RAS	p38	JAK	JNK	ERK	EGFR	PI3K	AkT	RAS	p38	JAK	JNK	ERK
0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
0	0	0	0	0	0	0	1	1	0	0	0	0	0	0	1
0	0	0	0	0	0	1	0	1	0	0	0	0	0	1	0
0	0	0	0	0	0	1	1	1	0	0	0	0	0	1	1
0	0	0	0	0	1	0	0	1	0	0	0	0	1	0	0
0	0	0	0	0	1	0	1	1	0	0	0	0	1	0	1
0	0	0	0	0	1	1	0	1	0	0	0	0	1	1	0
0	0	0	0	0	1	1	1	1	0	0	0	0	1	1	1
0	0	0	0	1	0	0	0	1	0	0	0	1	0	0	0
0	0	0	0	1	0	0	1	1	0	0	0	1	0	0	1
0	0	0	0	1	0	1	0	1	0	0	0	1	0	1	0
0	0	0	0	1	0	1	1	1	0	0	0	1	0	1	1
0	0	0	0	1	1	0	0	1	0	0	0	1	1	0	0
0	0	0	0	1	1	0	1	1	0	0	0	1	1	0	1
0	0	0	0	1	1	1	0	1	0	0	0	1	1	1	0
0	0	0	0	1	1	1	1	1	0	0	0	1	1	1	1
0	0	0	1	0	0	0	0	1	0	0	1	0	0	0	0
0	0	0	1	0	0	0	1	1	0	0	1	0	0	0	1
0	0	0	1	0	0	1	0	1	0	0	1	0	0	1	0
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0	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1

Table 9: Truth table for Test set 1 containing 5 conditions

EGFR	PI3K	AkT	RAS	p38	JAK	JNK	ERK
1	1	1	0	1	1	1	1
0	0	0	0	0	1	0	1
0	0	0	0	0	1	1	0
1	1	1	1	0	1	0	1
1	1	1	1	1	1	1	1

Table 10: Truth table for Test set 2 containing 5 conditions

EGFR	PI3K	AkT	RAS	p38	JAK	JNK	ERK
1	1	1	0	1	1	1	1
1	1	1	1	1	1	1	1
0	0	0	0	0	1	1	0
1	1	1	1	0	1	0	1
1	0	1	1	1	0	1	1

3. Results and Discussion

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the proteins PI3K, AkT and p53 are present, it leads to the cell survival condition.

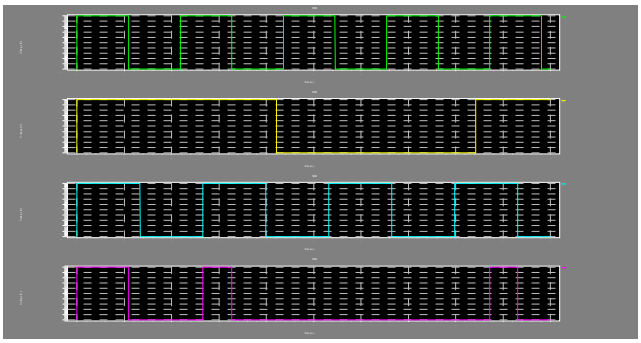

Figure 9: Tanner Output-2 represents 1 for cell survival and 0 for cell deat

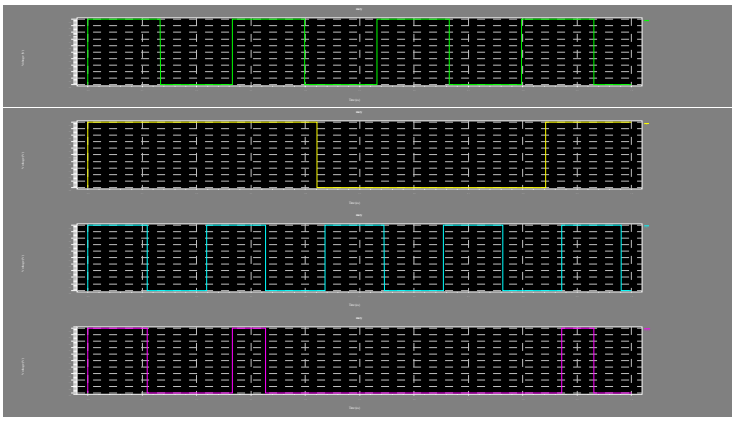

Figure 10: Tanner Output-1 which represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the proteins SOS, p38 and p53 are present, it leads to the cell survival conditio

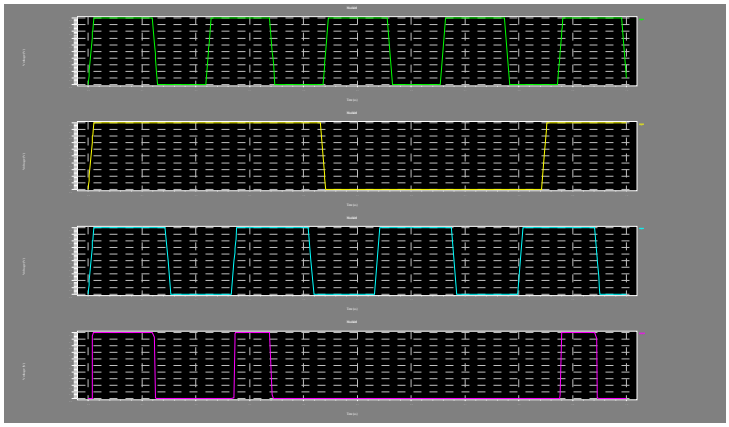

Figure 11: Tanner Output-3 represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the proteins RAS, RAF and ERK are present, it leads to the cell survival condition.

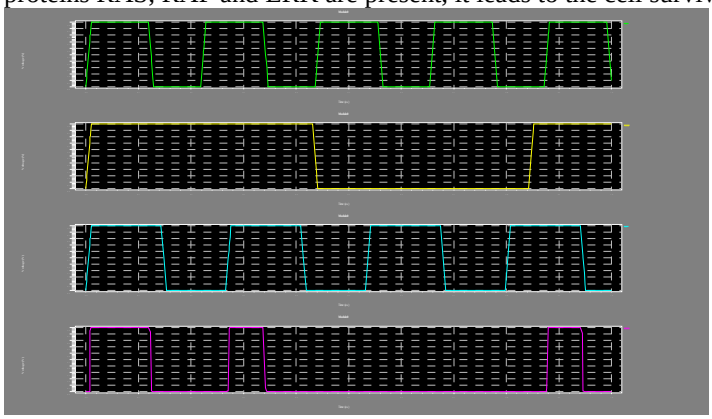

Figure 12: Tanner Output-4 represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos

circuit and its output shows that when the proteins RAS, MEKK and JNK are present, it leads to the cell survival condition.

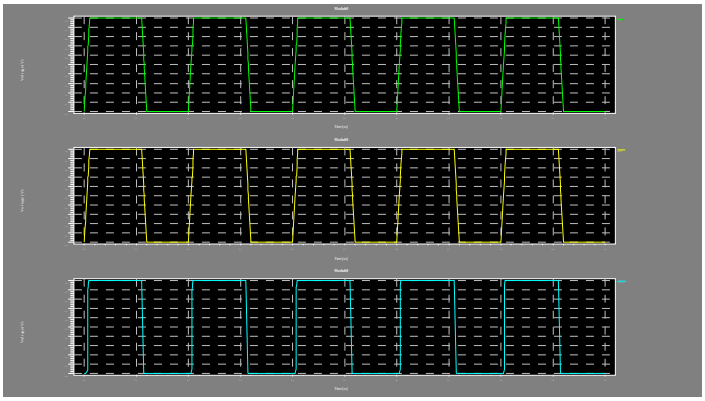

Figure 13: Tanner Output-5 represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the proteins JAK and STAT are present, it leads to the cell survival condition.

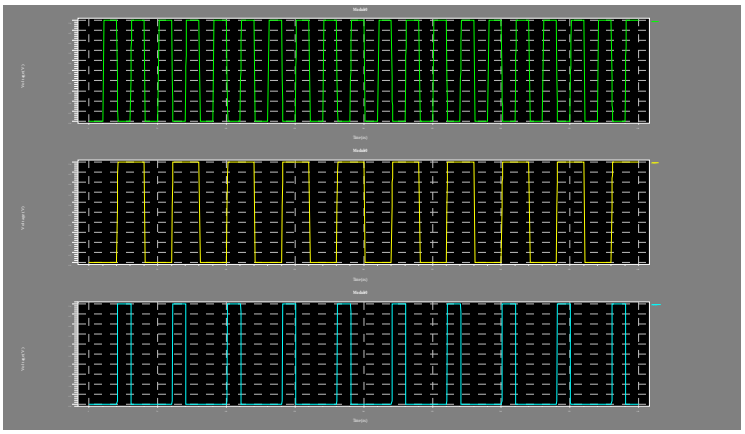

Figure 14: Tanner Output-6 represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the protein AP1 is not activated by MK2 the cell survives and the activation of AP1 leads to apoptosis.

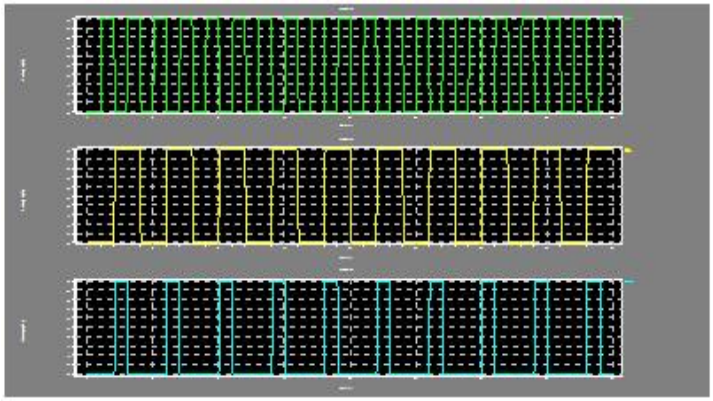

Figure 15: Tanner Output-7 represents 1 for cell survival and 0 for cell death

The output of the SPICE simulation is in the wave form with time in the X-axis and voltage applied in the Y-axis. When the wave is in the raised position it represents 1 which indicates cell survival and when wave is in down position it represents 0 which indicates cell death. This Karnaugh map, Boolean expression, C-Mos circuit and its output shows that when the protein AP1 is not activated by MK2 the cell survives and the activation of AP1 leads to apoptosis. In accordance with the previous work done the decomposed EGFR pathway has been successfully modeled using the T-SPICE simulator and five cell survival conditions as well as two cell death conditions have been shown (Figure 9 -15). Which is similar model to (Jain, *et. al.,* 2009, 2010).

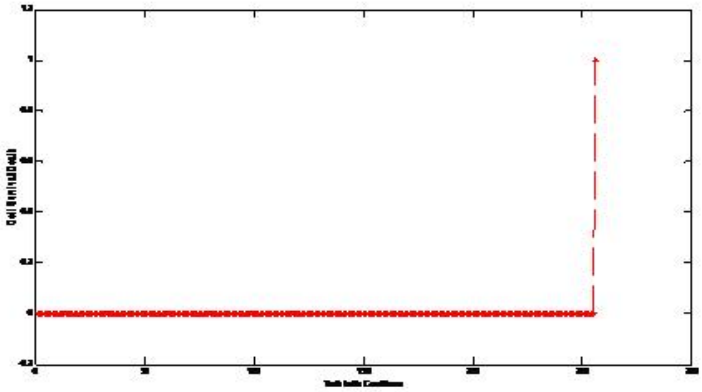

Figure 16: Graph for Training Set of Neural Network

The graph shows (Figure 16) the 256 conditions in truth table (Table 8) by which the Neural Network toolbox of Matlab has been trained. Since the eight proteins should be present for the cell to survive the truth table represented an AND gate and results congruence to (Jain *et. al.,* 2009).

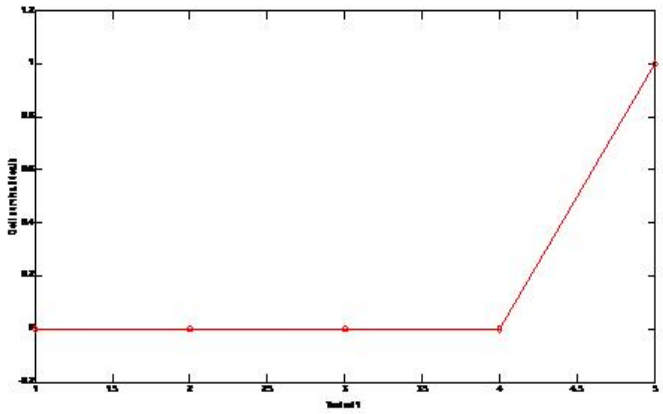

Figure 17: Graph for Test Set 1 of Neural Network

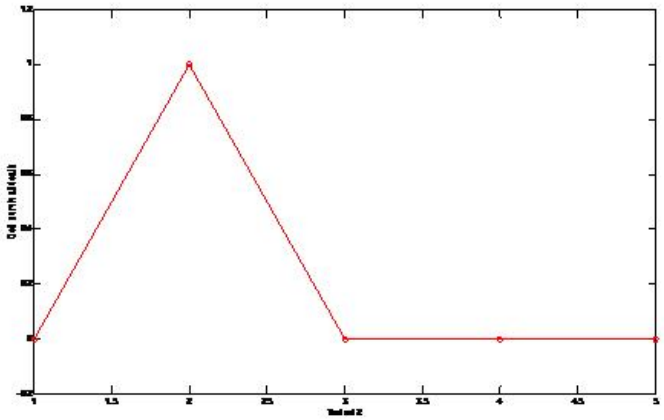

Figure 18: Graph for Test Set 2 of Neural Network

The graph (Figure 17) for test sets (Table 9) show the different conditions experimented with the Neural Network toolbox of Matlab which had been trained using the 256 conditions. The graph (Figure 18) for test sets (Table 10)

also shows that each of the eight proteins should be present for the cell to survive. It also confirmed that quinazoline have a great affinity towards mutated proteins when compared with the normally active protein (Kotra *et al*., 2008).

4. Conclusion

The computational network model of protein kinases for cell survival/death had been constructed by using the proteins involved in EGFR pathway. With that model the truth table, boolean expression and C–Mos circuit had been made for each possible pathway. Then each sub pathway had been combined and simulated using the Neural Network Toolbox in Matlab. The action of quinazoline drug on EGFR protein had been studied and found out that the EGFR proteins having mutation has higher affinity towards quinazoline than the normally active protein. It was also found out that each quinazoline derivatives have specific action on different mutations which eventually lead to different kinds of cancer. This study helped to guide the use of currently available EGFR inhibitors and provided new direction for the design and development of even more potent inhibitors that are tailored to specific EGFR mutants. We conclude that it is possible to build self consistent prediction model that can computationally yield important insights into the control of cell survival/death responses. This work also helped in the better predicament of clinical efficacy in terms of binding efficacy of quinazoline derivatives with EGFR protein.

5. Acknowledgements

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