Department of Electronic & Telecommunication Engineering

University of Moratuwa

BM 2101 – Analysis of Physiological Systems

ASSIGNMENT 03

PROPERTIES OF HODGKIN-HUXLEY EQUATIONS

Name	**Index number**
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for the module BM 2101 – Analysis of Physiological Systems.

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Question 01

Continue to bisect the amplitude interval for sub-threshold and supra-threshold stimulating currents and obtain an estimate of the threshold stimulating current amplitude to two decimal places.

Solution 01

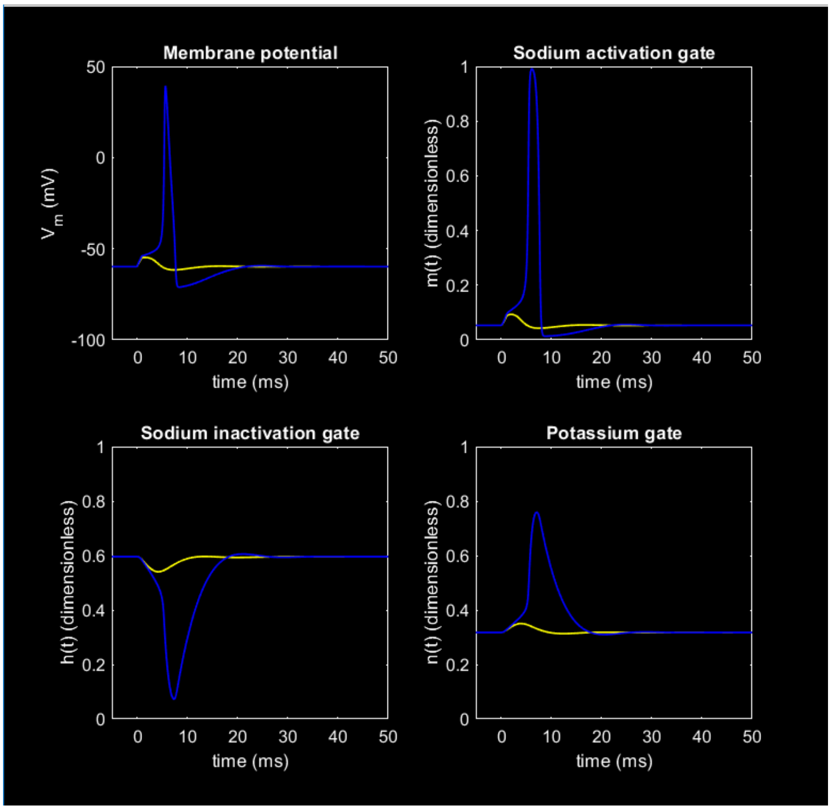
- From the curves we obtained for the cases; amp1 = 6 μAcm^{-2} and amp1 = 7 μAcm^{-2}, it is clear that the subthreshold is greater than 6 μAcm^{-2} and the supra-threshold is less than 7 μAcm^{-2}.
- Then, using the numerical bisection algorithm, we can narrow down the region between the sub-threshold and supra-threshold.
- The implementation is as follows. (The corresponding plots can be found in Plots.mlx)

```
%no action potential

amp1 = (6+7)/2;

hhmplot(0,50,0);

%no action potential
amp1 = (6.5+7)/2;
hhmplot(0,50,0);

%no action potential
amp1 = (6.75+7)/2;
hhmplot(0,50,0);

%no action potential
amp1 = (6.875+7)/2;
hhmplot(0,50,0);

%action potential is generated.
amp1 = (6.9375+7)/2;
hhmplot(0,50,0);

%no action potential
amp1 = (6.9375+6.9688)/2;
hhmplot(0,50,0);

%action potential is generated
amp1 = (6.9532+6.9688)/2;
hhmplot(0,50,0);
```

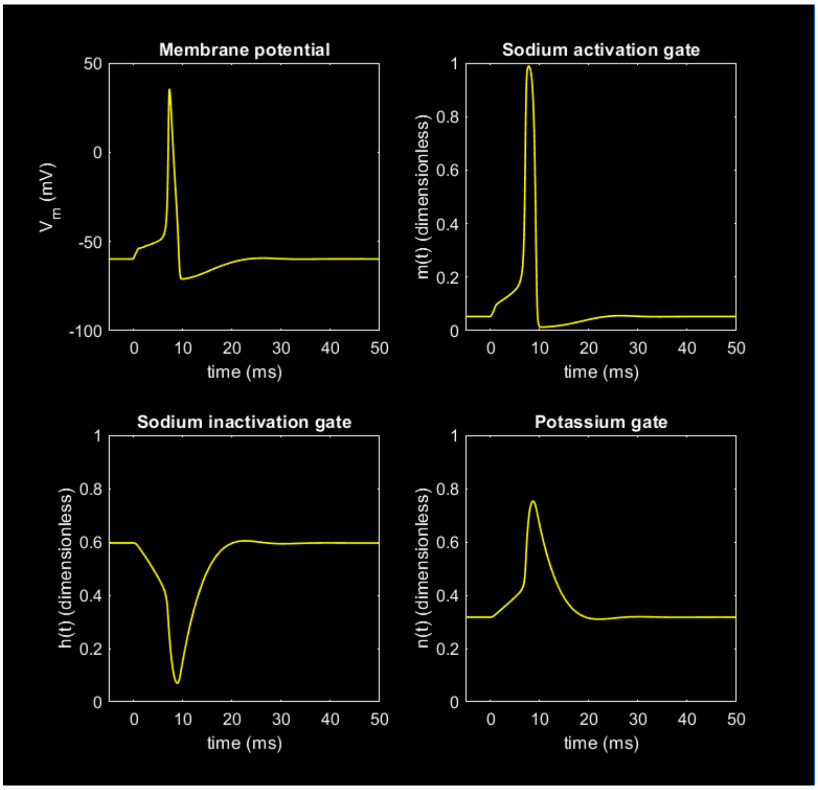
```
%no action potential
amp1 = (6.9532+6.9610)/2;
hhmplot(0,50,0);


%action potential is generated
amp1 = (6.9571+6.9610)/2;
hhmplot(0,50,0);

%no action potential is generated
amp1 = (6.9571+6.9590)/2;
hhmplot(0,50,0);

%action potential is generated
amp1 = (6.9581+6.9590)/2;
hhmplot(0,50,0);

%action potential is generated
amp1 = (6.9581+6.9586)/2;
hhmplot(0,50,0);

%action potential is generated
amp1 = (6.9581+6.9584)/2;
hhmplot(0,50,0);

%action potential is generated
amp1 = (6.9581+6.9583)/2;
hhmplot(0,50,0);
```

- Considering values up to two decimal points, it was shown that amp1 = = 6.95 μAcm^{-2} fails to generate an action potential whereas amp1 = = 6.96 μAcm^{-2} generates an action potential.
- Thus, we can conclude that the subthreshold is 6.95 μAcm^{-2} and the supra-threshold is 6.96 μAcm^{-2} .
- Therefore, the **threshold stimulating current amplitude = $\mathbf{6.96\ \mu Acm^{-2}}$**
- The relevant plots are shown below.

Plots for currents of 6 μAcm^{-2}(yellow) and 7 μAcm^{-2}(blue)



Plots for stimulating current of 6.96 μAcm^{-2}



Question 02

If the integration time interval $[t_o; t_f]$ contains only one action potential, as it should in the above examples, by executing the following command for any amplitude of the stimulating current,

```
>> [qna,qk,ql] = hhsplot(0,50);
```

What in general will be the relationship between $\int J_{ei} dt$ and $\int \sum J_k dt$ (allow for some numerical error)?

Solution 02

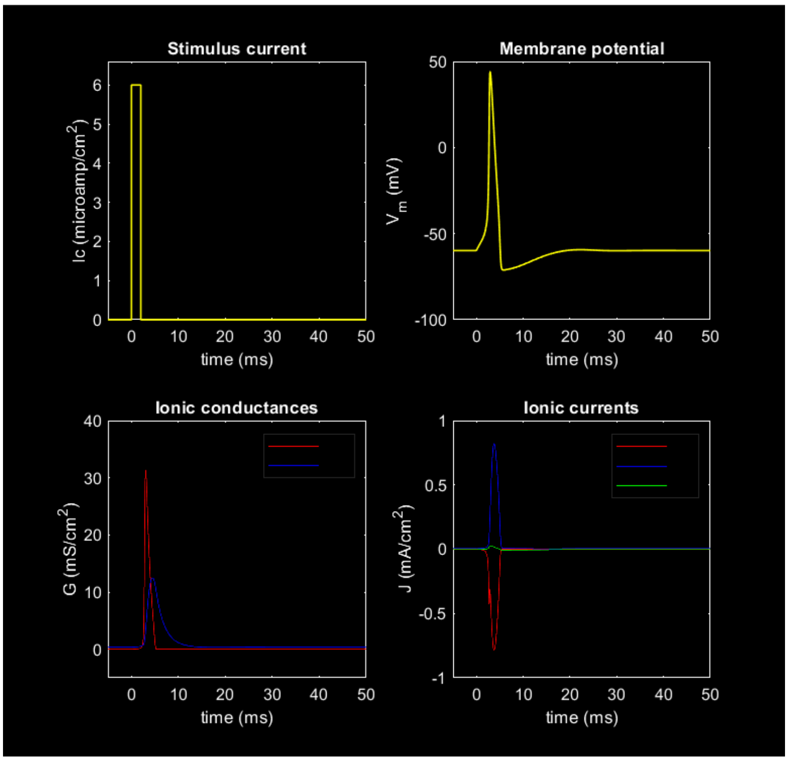
The following table summarizes the results obtained for J_{ei} = 6, 7, 8, 9, 9.8, 10 and widths = 1, 1.5, 2, 3.

J_{ei} $(\mu A cm^{-2})$	$width$ (ms)	$\int J_{ei}\, dt$	$qna + qk + ql$	$\int \sum J_k\, dt$	$\int \sum J_k\, dt$ (Approximation)
6	1	6	5.9997	5.9997	≈ 6
6	2	12	11.9998	11.9998	≈ 12
7	1	7	7.0014	7.0014	≈ 7
7	2	14	14.0000	14.0000	≈ 14
8	1	8	7.9989	7.9989	≈ 8
8	2	16	15.9998	15.9998	≈ 16
9	1	9	8.9984	8.9984	≈ 9
9	2	18	17.9993	17.9993	≈ 18
9.8	1.5	14.7	14.6993	14.6993	≈ 14.7
10	3	30	29.9990	29.9990	≈ 30

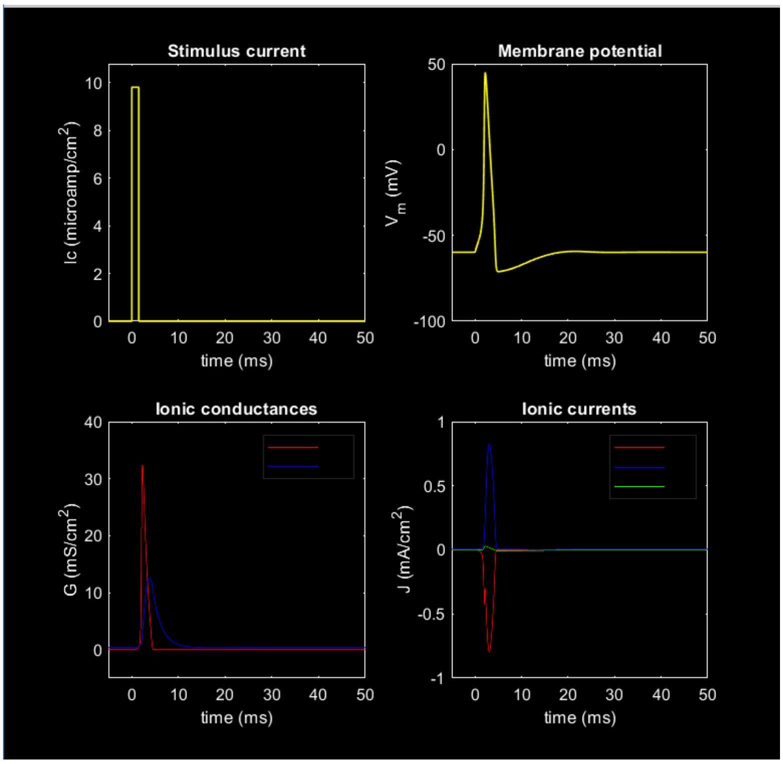
- Note that $\int J_{ei}\, dt$ represent the net inward current within a specified period of time and $\int \sum J_k\, dt$ represents the summation of the currents through the ionic gates within the same time duration.
- From the above table, we can conclude that,

$$\boxed{\int_{t_0}^{t_f} J_{ei}\, dt \approx \int_{t_0}^{t_f} \sum J_k dt}$$

- Given below are some of the plots obtained for the values mentioned above. (All plots can be found in the Plots.mlx).



$$J_{ei} = 6\mu Acm^{-2}$$
$$width = 2ms$$



$$J_{ei} = 9.8\mu Acm^{-2}$$
$$width = 1.5ms$$

Question 03

By setting delay2 successively to 20, 18, 16, 14, 12, 10, 8 and 6 ms, adjust amp2 to an accuracy of 0.1 µAcm^{-2} so as to just elicit an action potential. The amplitude so obtained, I_{2th}, will correspond to the current threshold amplitude for a second pulse as a function of the inter-stimulus interval.

Solution 03

```
%delay = 25

amp1 = 26.8;
width1 = 0.5;
delay2 = 25;
amp2 = 13.7;
width2 = 0.5;
hhsplot(0,40);

%delay = 20
amp1 = 26.8;
width1 = 0.5;
delay2 = 20;
amp2 = 11.6;
width2 = 0.5;
hhsplot(0,40);

%delay = 18
amp1 = 26.8;
width1 = 0.5;
delay2 = 18;
amp2 = 11.3;
width2 = 0.5;
hhsplot(0,40);

%delay = 16
amp1 = 26.8;
width1 = 0.5;
delay2 = 16;
amp2 = 12.7;
width2 = 0.5;
hhsplot(0,40);

%delay = 14
amp1 = 26.8;
width1 = 0.5;
delay2 = 14;
amp2 = 17.0;
width2 = 0.5;
hhsplot(0,40);
```

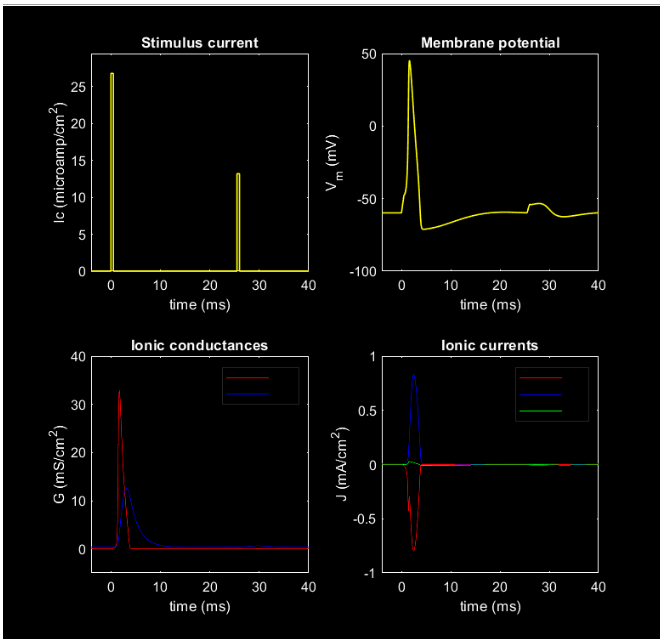
```
%delay = 12

amp1 = 26.8;
width1 = 0.5;
delay2 = 12;
amp2 = 25.5;
width2 = 0.5;
hhsplot(0,40);

%delay = 10
amp1 = 26.8;
width1 = 0.5;
delay2 = 10;
amp2 = 40.8;
width2 = 0.5;
hhsplot(0,40);

%delay = 8
amp1 = 26.8;
width1 = 0.5;
delay2 = 8;
amp2 = 70.1;
width2 = 0.5;
hhsplot(0,40);

%delay = 6
amp1 = 26.8;
width1 = 0.5;
delay2 = 6;
amp2 = 145.2;
width2 = 0.5;
hhsplot(0,40);
```

- The supra-threshold current of an action potential for a single pulse is $I_{1th} = 13.4\ \mu Acm^{-2}$.
- The magnitude of the first stimulating current is set to twice the value of this threshold.
- That is, $26.8\ \mu Acm^{-2}$. Moreover, the width of the pulse is 0.5ms.

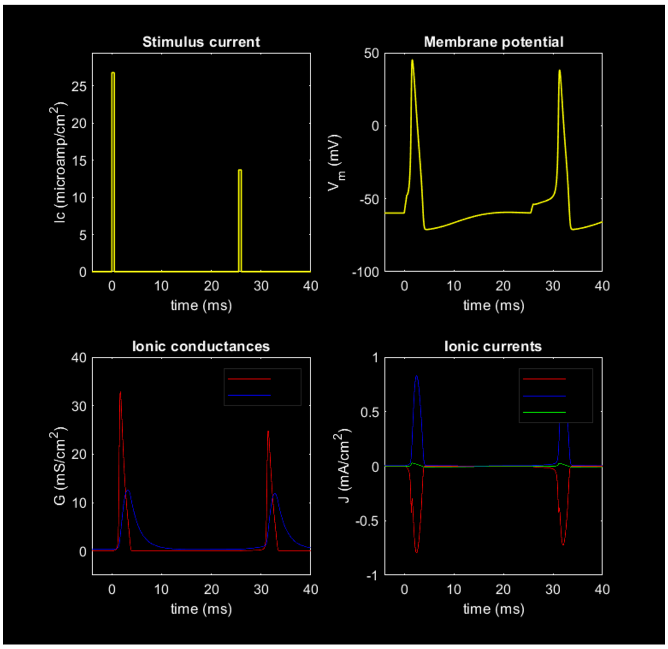
- Then, the minimum amplitude of the second stimulating current pulse is found by trial and error.
- The following table summarizes the variation of the threshold of the second pulse, with different delays.
- The width of the second pulse is set to 0.5ms in all cases.

$\boldsymbol{Delay\ (ms)}$	25	20	18	16	14	12	10	8	6
$\boldsymbol{I_{2th}(\mu A cm^{-2})}$	13.7	11.6	11.3	12.7	17.0	25.5	40.8	70.1	145.2

- The following curves represent two cases where the second pulse manages generate an action potential where as the other fails to do so.



The second pulse has failed to generate an action potential



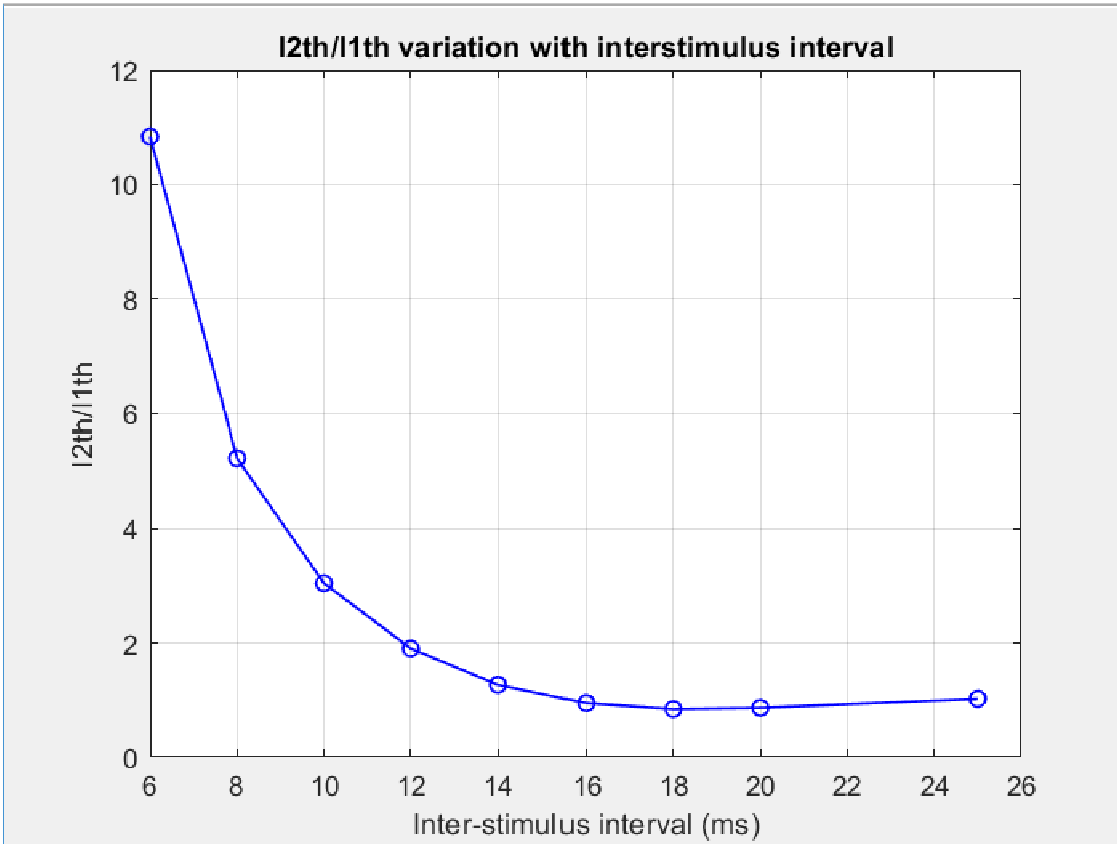
The second pulse has generated an action potential

Question 04

By plotting the ratio I_{2th}/I_{1th} as a function of inter-pulse interval estimate the absolute and relative refractory periods.

Solution 04

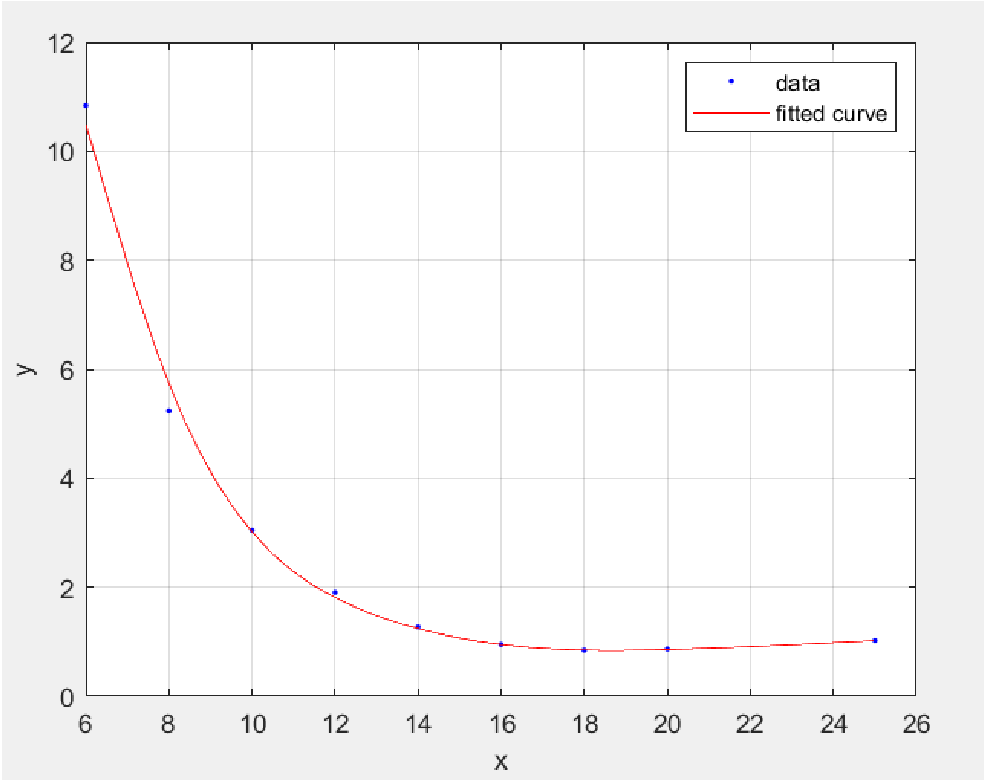
- The curve representing the ratio I_{2th}/I_{1th} as a function of inter-stimulus interval is given below.



- For a smoother curve, we can use 'fit' option in MATLAB

```
%Obtaining a smoother curve
f = fit(delay', ratio', 'smoothingspline');
plot(f,delay',ratio')
grid on
```

- The resulting curve is as follows.



- It's hard to determine precise values for the absolute refractory period and relative refractory period as the curve is less smooth due to low no. of data points we have used.
- But, we can assume approximate values for ARP and RRP, using the curve plotted by applying smoothing spline technique on the data points.
- The graph has a steep slope within 6ms – 18ms. That is, within this region, the threshold value increases exponentially as the delay is decreased from 18ms to 6ms.
- In other terms, the relative refractory period is denoted by the region in which the ratio between I_{2th} and I_{1th} is significantly greater than 1.
- Therefore,

$$\textbf{\textit{Relative refractory period}} \approx \mathbf{12\textit{ms}}$$

- The threshold is really large when the time at which the second pulse is given is really close to the end of the absolute refractory period (i.e. smaller delay).
- According to the graph we have, we can say that the absolute refractory period is less than 6ms.
- As an estimation, we can say that,

$$\textbf{\textit{Absolute refractory period}} \approx \mathbf{5\textit{ms}}$$

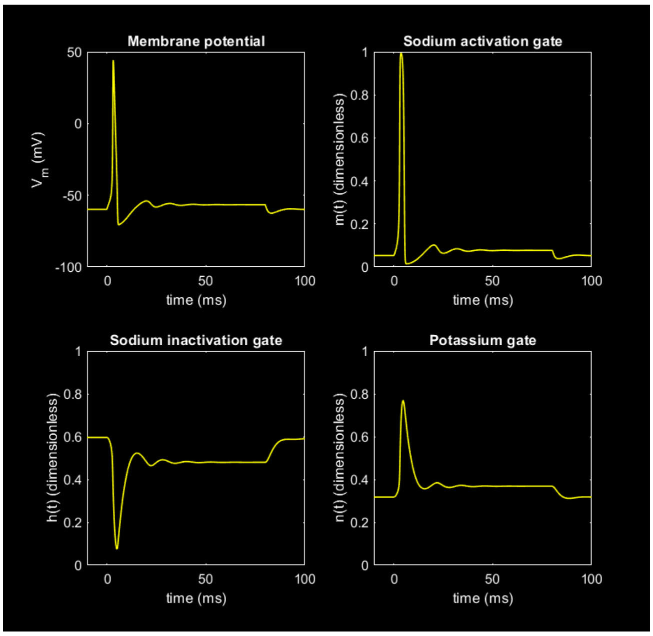
Question 05

By using either hhsplot or hhmplot, estimate the number of action potentials per second by applying single 80 ms wide stimulus currents of 5, 10, 20, 30, 50, 70 and 100 μAcm^{-2}. Plot action potential frequency as a function of stimulating current amplitude. e.g.

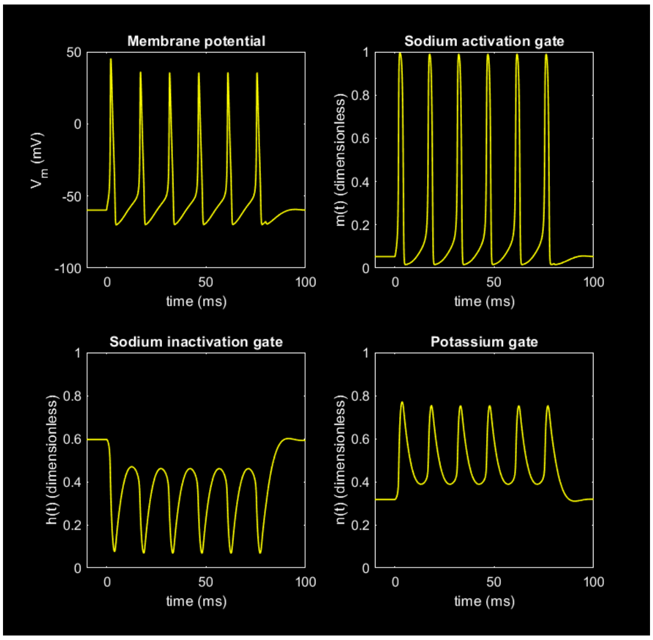
```
>> amp1 = 5;
>> width1 = 80;
>> delay = 0;
>> amp2 = 2;
>> width2 = 0;
>> hhmplot(0,100,0);
```

What changes do you notice in the amplitude of the action potentials as a function of stimulus intensity amplitude?

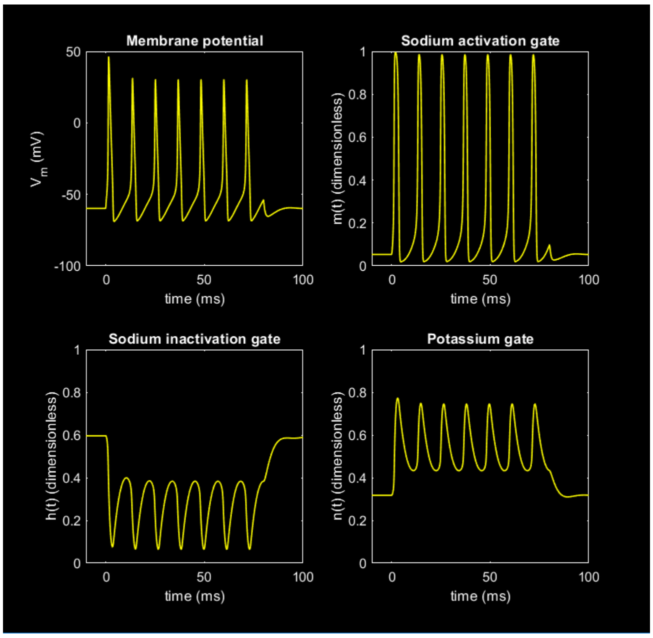
Solution 05



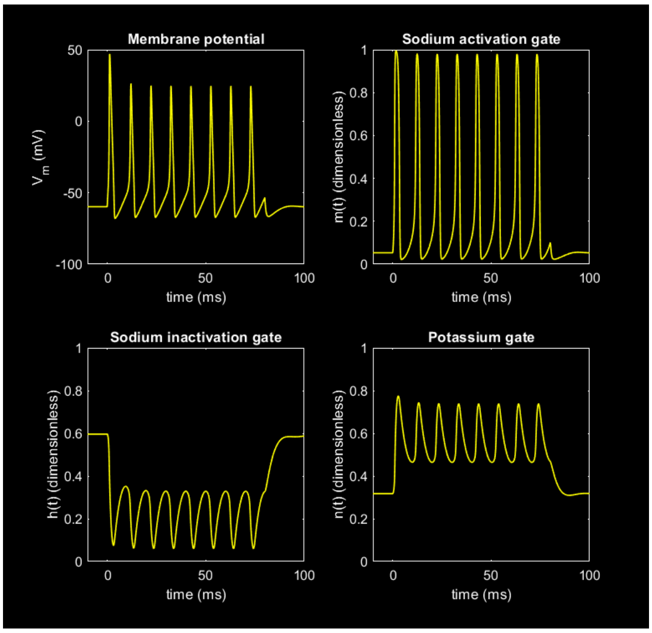
$(5\ \mu Acm^{-2} \qquad 1\ AP)$



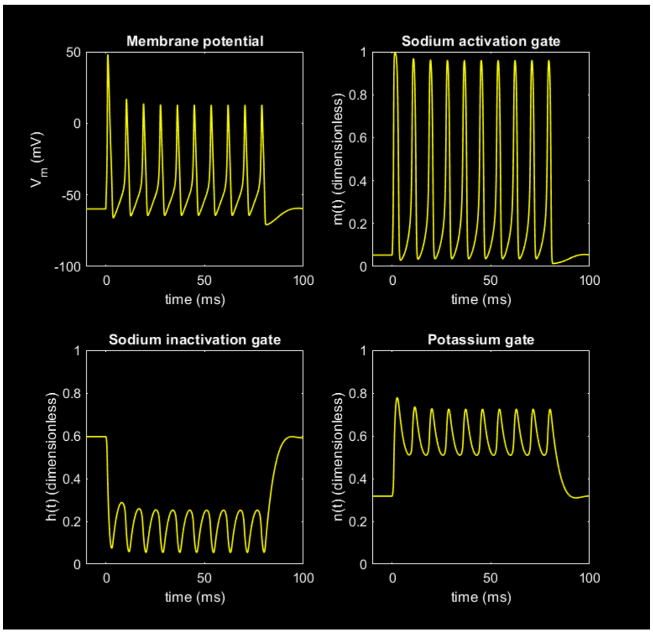
$(10\ \mu Acm^{-2} \qquad 6\ APs)$



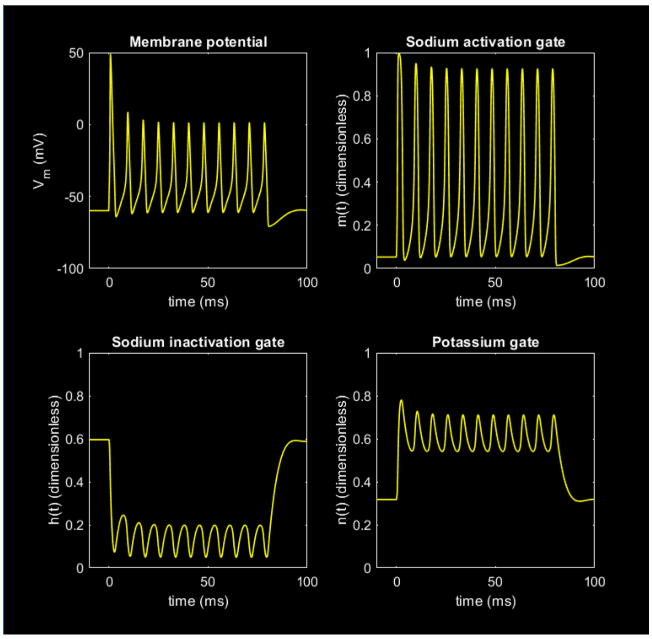
$(20\ \mu Acm^{-2} \qquad 7\ APs)$



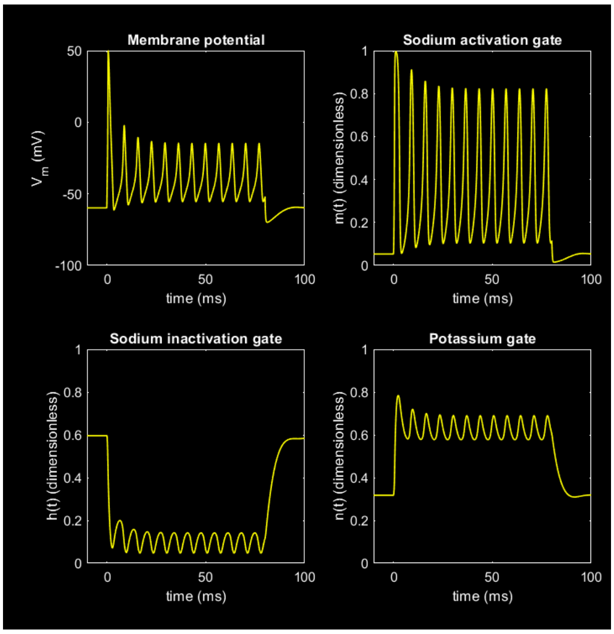
$(30\ \mu Acm^{-2} \qquad 8\ APs)$



$(50\ \mu Acm^{-2} \qquad 10\ APs)$



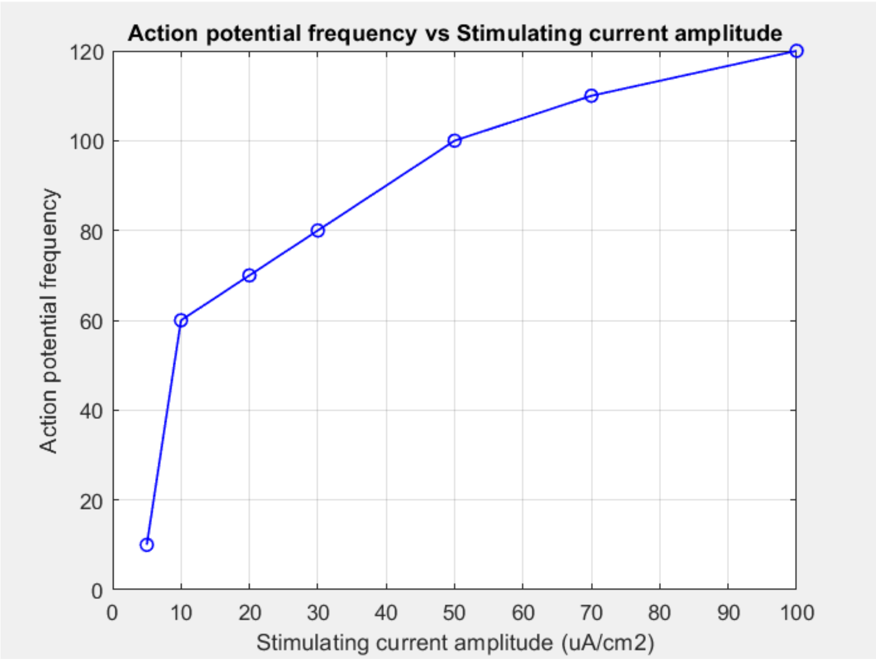
$(70\ \mu Acm^{-2} \qquad 11\ APs)$



$(100\ \mu Acm^{-2} \qquad 12\ APs)$

- The above APs are counted for a time span of 100ms. Thus, to find the frequency, we should multiply the APs by 10.

Stimulating current amplitude(μAcm^{-2})	5	10	20	30	50	70	100
Action Potential Frequency (Per second)	10	60	70	80	100	110	120

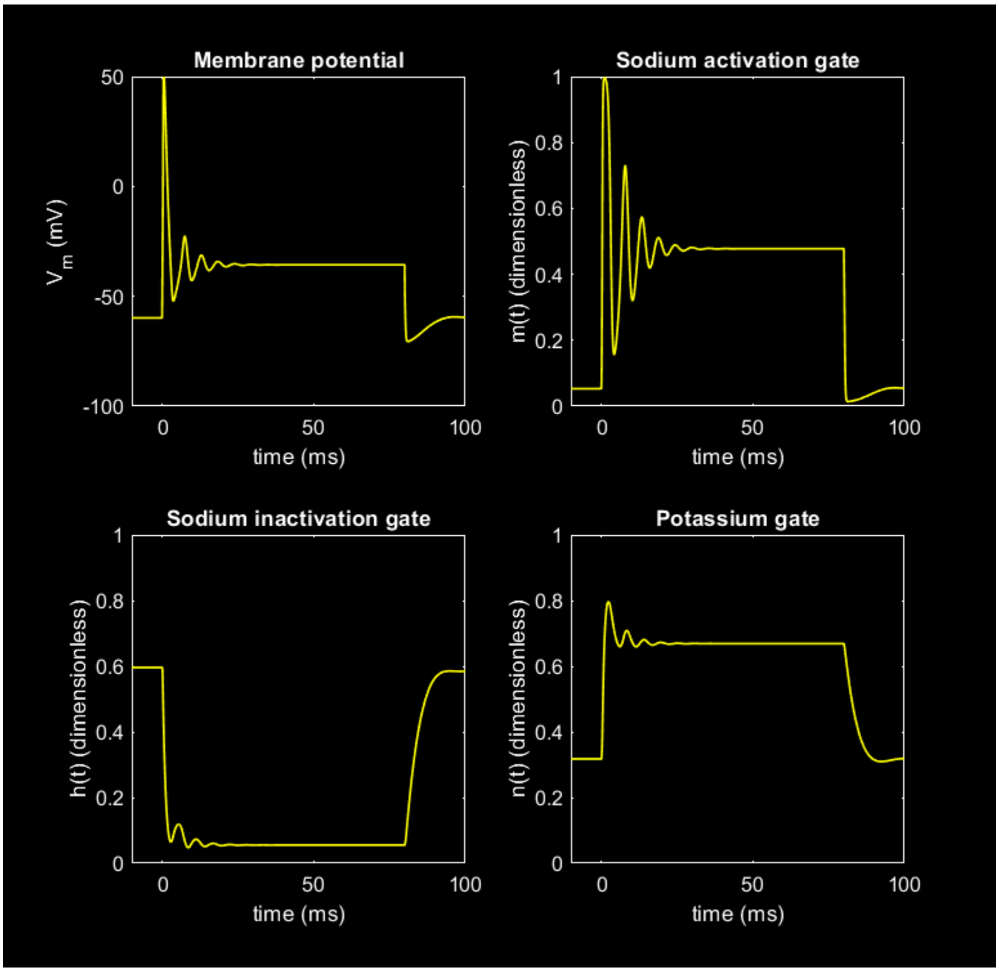


- From the above graph, we can clearly see that the action potential frequency increases with increasing amplitudes of the stimulating current.
- The curve replicates a variation of decreasing gradients with increasing current amplitudes.
- Note that, despite the variation in the stimulating current amplitudes, the peak of the first AP has remained more or less the same, in all cases.
- On the other hand, the peaks of the successive APs (from the 2nd peak onwards) have dropped when the stimulating current amplitude is increased.

<u>Question 06</u>

Set the stimulating current amplitude to 200 μAcm^{-2}. What do you notice? This result is known as a depolarisation block. Can you think of an explanation of the results of Question 5 and Question 6 in terms of the voltage dependence of the h and n factors of the Hodgkin-Huxley equations?

<u>Solution 06</u>



$(Stimulating\ current\ amplitude = 200\ \mu Acm^{-2}\ \ and\ the\ width = 80ms)$

Second order differential equation of Hodgkin and Huxley model

$$\frac{1}{2\pi a(r_0+r_i)}\frac{d^2V(z,t)}{dt^2}=C_m\frac{dV}{dt}+\overline{G_{K+}}n^4(V-V_{K+})+\overline{G_{Na+}}m^3h(V-V_{Na+})+\overline{G_L}(V-V_L)$$

- We know that,

$$n=n(V,t)$$

$$m=m(V,t)$$

$$h=h(V,t)$$

- Hence, it is clear that n, m and h are voltage dependent parameters.
- In the cases mentioned in question 05, we can see that the damping effect on the oscillatory response increases with increasing stimulating currents.
- That is, the amplitudes of the pulses will drop at a faster rate when the stimulating current is high.
- Yet, in those cases, the repetitive activity of the AP can be observed clearly, despite the damping effects.
- Question 06 corresponds to a case where the stimulating current is really high, which implies that the damping effect on the oscillatory response is more significant.
- Moreover, we know that 'n' corresponds to the K^+ gates where as 'h' corresponds to the inactivation gates of Na^+.
- When a significant depolarization current is given over a period of time, initially there will be an efflux of K^+ ions via the leak channels which is then followed by a transient influx of Na^+ ions.
- This results in the action potentials. But, when Na^+ flows in, the voltage gated K^+ channels would open and allow the flow of K^+ ions through them.
- Due to the constant depolarizing current, the K^+ ions will continue to flow out.
- Thus, the plateau level is slightly above the initial level.

Question 07

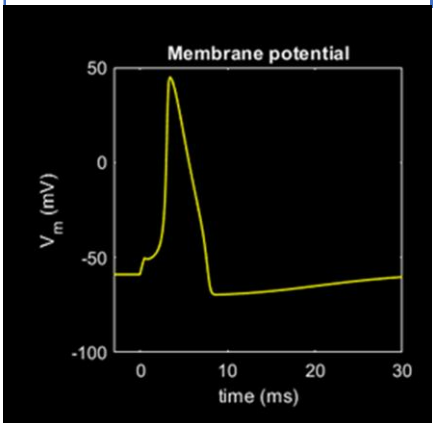
By using a single current pulse of intensity 20 µAcm^{-2} and 0.5ms in width, observe the effects of the following temperatures on the duration and amplitude of the resulting action potential: 0, 5, 10, 15, 20, 24, 25, 26 and 30^0C.

```
>> vclamp = 0;
>> amp1 = 20;
>> width1 = 0.5;
>> tempc = 0;
>> hhmplot(0,30,0);
>> hhsplot(0,30);
```

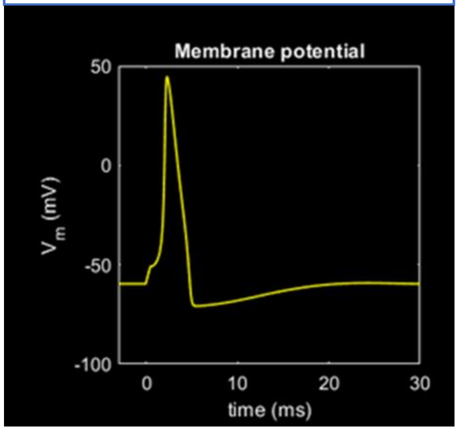
In general, what features of the action potential are affected by increasing temperature?

Solution 07

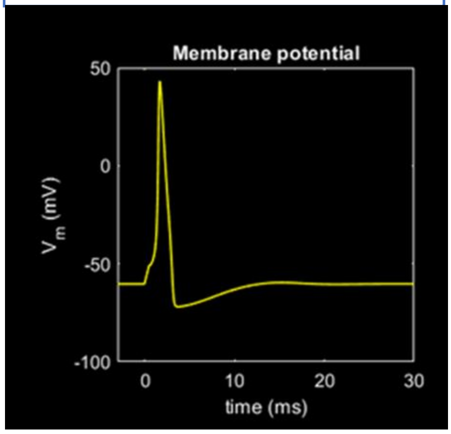
0 ^{0}C



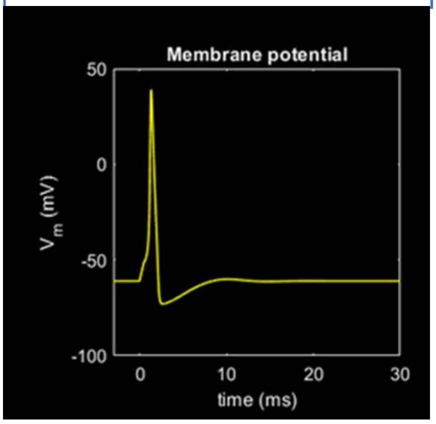
5 ^{0}C



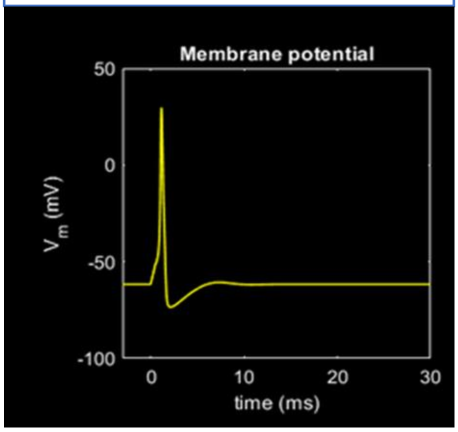
10 ^{0}C



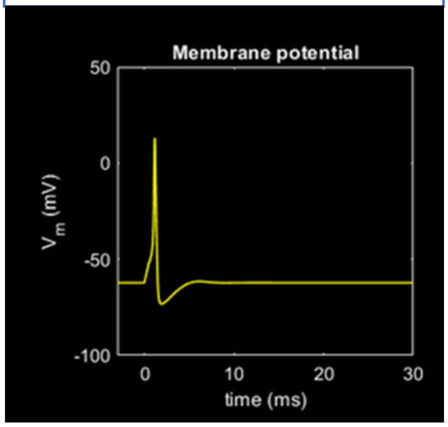
15 ^{0}C



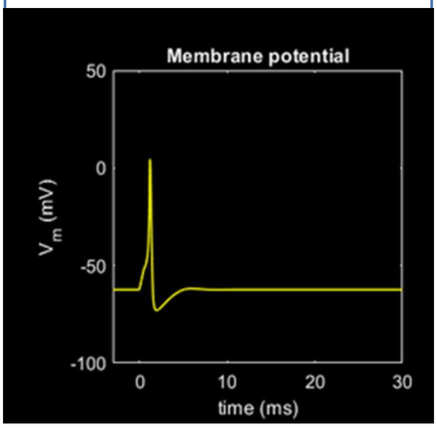
20 ^{0}C



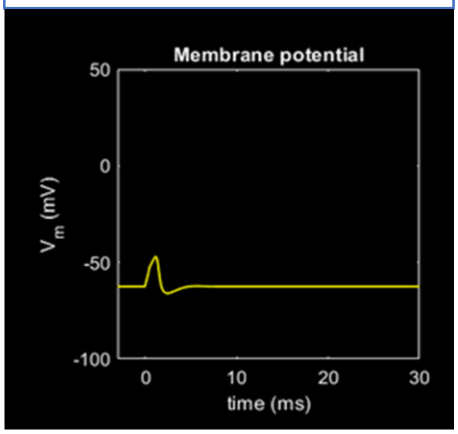
24 ^{0}C



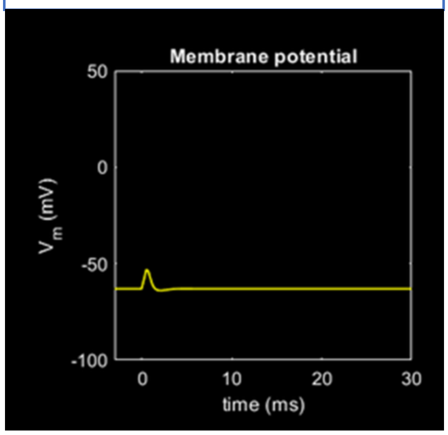
25 ^{0}C



26 ^{0}C



30 ^{0}C



From the above graphs, we can observe that when the temperature is increased,

- The duration of the action potential decreases. As a result, when the temperature is increased, the membranes will attain the resting potential quickly.
- Delay in opening the voltage gated K^+ channels depends on the temperature. When the temperature is high, this delay is reduced.

- Thus, the peak level of the action potential decreases when the temperature is increased.
- The reason behind this is that, when the delay in opening the K^+ gates is reduced, the inward current of Na^+ will be nullified by the outward flow of K^+ at an early stage of the action potential.
- We can observe that the maximum amplitude falls below V_{th} when the temperature is above 26^0C. Thus, an action potential won't be generated in such temperatures.